Decontamination of Corn Containing Aflatoxin by Treatment with Ammonia

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ABSTRACT

Corn containing aflatoxin can be effectively decontaminated by treatment with gaseous ammonia at atmospheric pressure. Preliminary studies were made of the effect of ammonia level, corn moisture, temperature, and time on the aflatoxin level. Ammonia tolerance tests were carried out on laying hens as were acceptance tests by swine. Toxicity feeding trials with ducklings, broiler chicks, and trout confirmed that the process inactivates the aflatoxin. The process was then applied under farm scale conditions to produce material for feeding trials with swine, poultry, and cattle under FDA protocols. The process can reduce aflatoxin levels from 1000 parts per billion (ppb) to within the FDA action level of 20 ppb.

INTRODUCTION

The severe economic impact of contamination of agricultural food and feed products by aflatoxin is not only that the products themselves are lost but also that the contaminated products must be disposed of. The value of the crop does not just drop to zero but actually becomes a liability that must be buried or otherwise eliminated. Consequently, the development of processes by which aflatoxin-contaminated agricultural products can be reclaimed for at least limited use, such as animal feed, has been given high priority at the Northern and Southern Regional Research Centers. A variety of crops are affected by aflatoxin. Corn, peanuts, and cottonseed are major crops which are subject to contamination, but each of these agricultural commodities presents specific problems in reducing or eliminating the aflatoxin level. A process developed for one commodity cannot necessarily be carried over directly to another.

In developing a decontamination process for reducing aflatoxin levels in corn, close liaison with the Food and Drug Administration was necessary. While acceptable limits of aflatoxin are set for feed use, it is not enough to reach these limits in a decontamination process. It is necessary to show, through animal feeding studies, that the process for reducing the assayed aflatoxin level does not produce deleterious compounds either from the chemical products from aflatoxin or from chemical changes in the corn itself. In preparing the decontaminated corn, therefore, the details of the process must be clearly specified and adhered to.

Various approaches to reducing aflatoxin levels in agricultural commodities have been investigated. A direct route would be blending of aflatoxin-containing corn, for example, with "clean" corn, but in general this is not permitted by FDA. However, an exception was made by FDA recently which would permit the blending of aflatoxin-contaminated corn with "clean" corn to achieve acceptable animal feeds in certain states (1). This is for poultry, swine, and beef cattle, and the exemption applies only to corn harvested in 1977 in Alabama, Florida, Georgia, Mississippi, North and South Carolina, and Virginia. When the aflatoxin level of the contaminated corn is at 1000 parts per billion (ppb), it is obvious that a very large amount of good corn is needed to reduce the average contamination level of the blend below 20 ppb.

Other ways for removal or inactivation of aflatoxin based on physical, chemical, or biological means have been tried. Physical separation methods are highly desirable, being relatively inexpensive, but appear to be limited to rather special situations in which the contamination is confined to a relatively small proportion of the seed with sufficient difference in seed properties due to contamination to make the separation possible (2). For corn, neither the wet milling nor dry milling process removed or inactivated the aflatoxin (3-5). While aflatoxins are relatively stable to heat, roasting of peanuts lowered the aflatoxin content as did the cooking of oilseed meals, but not below the level needed to meet the FDA tolerance limit (6). The roasting of corn after treatment with aqua ammonia has been reported to enhance aflatoxin inactivation, but the work is still in a preliminary stage (7). Solvent extraction of oilseed meals has been reported but is subject to economic and other limitations (8,9).

In considering approaches to decontamination or inactivation of aflatoxin in corn, the methods described above seemed inappropriate and chemical means were considered. Ammonia had been found to be one of the more effective reagents for treatment of cottonseed and peanut meals (10) and offers a number of potential advantages for corn. First, it is relatively inexpensive and is available in the potentially large quantities required. Second, ammonia is well known to the farmer, and its use for an "on-farm" process is thus quite feasible. Finally, although there are hazards in the use of ammonia, as with any chemical, these hazards are well known but can be controlled.

In developing a large scale process for on-farm or elevator use to detoxify corn containing aflatoxin, the following steps were followed:

1. Preliminary studies on a laboratory and pilot plant scale were conducted to investigate the parameters, such as corn moisture content, ammonia concentration, and temperature, which could affect the effectiveness of the inactivation process.

2. Preliminary studies of the pilot scale decontaminated corn were performed, using rainbow trout, ducklings, and broiler chicks, to ensure that the decrease in chemically assayed level of aflatoxin was actually matched by a decrease in biological activity. This preliminary bioassay information was essential before embarking on a scale-up of the process and the commencement of large scale feeding trials under FDA protocols.

3. Animal acceptance and utilization trials were carried out to ensure that the process developed actually produced corn that would be usable in final full scale feeding trials.

4. Treated and control corn were prepared on a 1000-bushel scale for use in feeding trials to be carried out under separate FDA protocols on poultry, swine, and cattle.

The work described here was carried out over a period of years, but the final step 4, above, was carried out in two phases. In 1975 the pilot scale process was scaled up to prepare material for swine and poultry feeding trials. In 1977, treated corn (with controls) was prepared for cattle feeding trials. In both phases the aflatoxin levels in the corn were reduced from 1000 ppb to less than 5 ppb, well within
the FDA guidelines of 20 ppb.

EXPERIMENTAL PROCEDURES

Preliminary Studies

Both yellow and white corn from several locations were used, with aflatoxin levels between 30 and 1200 μg/kg (ppb). For a number of experiments, 3.6 kg of the corn were placed in double plastic bags (2 mil. polyethylene) and aqua ammonia was added. The contents were mixed manually, then stored for specified times at temperatures from -18 to 60 C (11).

Ammonia tolerance tests were carried out on laying hens by C.C. Calvert, Feed Energy Conservation Laboratory, SEA-FR, Beltsville, Md., using standard rations but with corn ammoniated up to the 2.6% level. Feed consumption, egg production rate, and taste tests of both scrambled and hard boiled eggs were carried out. Acceptance tests of ammoniated corn by swine were carried out by Jensen et al. (12). Toxicity feeding trials to confirm that the aflatoxin was biologically inactivated were carried out with ducklings, broiler chicks, and trout.

Large Scale Decontamination

A blended lot of yellow dent corn, U.S. Sample Grade, naturally contaminated with 900 μg/kg total aflatoxin (750 B1, 90 B2, 40 G1 and 20 G2) was used. Under the FDA protocol, four sets of samples for feeding were used: uncontaminated corn; uncontaminated corn ammoniated; aflatoxin-containing corn; and ammoniated (decontaminated) corn. No detectable levels of aflatoxin, ochratoxin, or zearalenone were found in the uncontaminated corn.

Cleaning, Blending, and Tempering of Corn

The corn was cleaned in one pass through a rotating, single action corn cleaner to remove fines and broken kernels.

The corn (ca. 12% moisture content) was adjusted to ca. 17.5% moisture by a two-step addition of tap water in approximately equal proportions. A grain auger fitted with spray nozzels served as the tempering apparatus. The two-step procedure minimized any nonuniformity in the tempering and was necessary because the surface of freshly wetted kernels can only retain ca. 3% water. Between the two temper steps, the corn was held for a period of 5-8 hr. It was blended in the blending bin for 5 hr or longer after the last water addition before the transfer to the ammoniation bin. Usually, 15-20 hr elapsed between last addition of water and first addition of NH3.

Ammonia. The liquid anhydrous ammonia used was agricultural grade of minimum purity 99.5%.

Grain bins. Standard Butler bins of a capacity ca. 2500 bushels were used. These bins had the conventional elevated, slotted metal grain-drying floors with 15 cm diameter under-the-floor grain unloading auger. In the blending bin, there was also a single vertical screw bin stirring device used to blend the corn.

The galvanized surface of the metal bins is readily attacked by ammonia vapor, so that all interior surfaces of the bins used for ammoniation or for drying of the corn were coated with an epoxy paint, which very effectively protected the metal bins. In the ammoniation bin, it was also necessary to caulk the seams to contain the ammonia vapor. An all-purpose sealing compound (Nashu Corp., No. 101, Duct Sealer) served this purpose. All seams were also coated internally with roofing cement. Cellular foam weather stripping was used as a gasket for the roof access door.

A 5-6 KW blower, delivering 0.5 m3 air/sec, at 50 cm water column pressure, and 20 cm diameter polyvinyl chloride (PVC) plastic piping were used to recycle the ammonia-air mixture through the ammoniation bin.

Ammoniation. The bin used for ammoniation is shown in Figure 1. Liquid ammonia from the storage tank was led to a heat exchanger, vaporized, and added to the air stream entering the plenum chamber under the slotted floor of the bin. NH3 vapor concentration in the air stream climbed progressively as ammonia was added to the plenum chamber, passed up through the corn, and recycled back to the plenum chamber from the top of the bin through the PVC tubing evident in Figure 1. NH3 concentration was monitored carefully and allowed to reach a maximum level in the air of only 75% of the lower explosive limit (LEL) concentration (15 volume percent for dry NH3-air mixtures at 25 C). To ensure uniform distribution of ammonia in corn bed, the NH3-air mixture was recycled for a total of 48 hr from the time of the initial ammonia addition. The corn remained in the bin another 11 days and then was transferred to the drying bin (the second bin in Figure 1).

Drying of Ammoniated Corn

The corn was dried to ca. 10% moisture with air heated to an average temperature of 40 C. At this moisture level, no ammonia was detected.

Sampling

Safety considerations. The usual precautions in using liquid ammonia were observed. The major hazard in the operation, however, involved the handling of corn at high aflatoxin levels. Here, dust is a major concern, and masks were routinely used even at low dust levels; and where dust was a major problem, as in grain cleaning, it was necessary to enclose the grain cleaner within a polyethylene-covered wooden framework. During such dusty operations, respirators were worn. Contaminated cleanings were buried with lime. Protective clothing was also worn, showers were available, and chlorine solution was used to decontaminate equipment exposed to aflatoxin-containing dust.

RESULTS AND DISCUSSION

Preliminary Studies

Four major factors determined the effectiveness of the ammonia in reducing aflatoxin in corn: the aflatoxin level, the moisture level, temperature, and time. Figure 2 shows the pronounced effect of temperature on the change of aflatoxin level with time. At 0.5% NH3 and a 15% moisture