PILOT-PLANT DRY MILLING OF CORN CONTAINING AFLATOXIN[1]

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ABSTRACT

Three lots of corn (one yellow, two white) naturally contaminated with different levels of aflatoxin (13, 160, and 510 p.p.b. of B-1) were dry-milled to determine distribution of the toxin among product fractions. No problems were encountered in the milling steps. Product yields and fat contents were fairly typical of those for a normal dent corn. Aflatoxin level was always lowest in the grits and highest in the germ, hull, or degermer fines, and varied with the contamination level of corn being milled. Proportion of aflatoxin B-1 in the prime product mix (i.e., grits, low-fat meal, and low-fat flour) amounted to only 7 to 10% of total quantity of B-1 in all products. Concentrations of aflatoxin in degermer fines, germ, and hull exceeded that of the corn milled. Aflatoxin B-1 level in endosperm-derived products correlated with their fat content. Yield of prime product mix, based on all products recovered, varied between 49 and 60% in these tests.

In 1971, the Food and Drug Administration ordered a recall of corn meal allegedly tainted with aflatoxin (1). Such action raised questions about relative distribution of aflatoxin among various product fractions if a contaminated corn should perchance be milled, and about possible correlation between level of contamination in corn and in product fractions. Experimental work was undertaken at the Northern Laboratory to answer these questions.

MATERIALS AND METHODS

Contaminated Corn

One lot of cleaned yellow corn, U.S. Grade No. 1 (identified as lot A), containing 13 p.p.b. of B-1 aflatoxin, and two lots of cleaned white corn, U.S. Grade Nos. 1 and 5, with 160 (lot B) and 510 (lot C) p.p.b. of B-1, respectively, were milled. The lots were naturally contaminated and came from three different states. Lot A came from North Carolina, lot B from Arizona where it had been grown under irrigation, and lot C was grown in Missouri on land with the water table near the soil surface.

Details of cleaning the three lots in a fanning mill (a simple milling separator) have been described previously (2). The cleaning operation removed all but a trace of broken corn-foreign material (BCFM). After cleaning, the yellow corn contained 2% total damaged kernels (DKT), and the white corn, lots B and C, had a trace and 15%, respectively. Damage in lot C was principally in the germ area. All three lots had test weights of 57 to 59 lb./bu. at 11 to 12% moisture.

Milling Equipment, Conditions, and Procedures

Three hundred pounds of corn from lot A[3] and 17 from lot B were pretempered overnight from approximately 10 to 15% moisture, then given a first temper to

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[3]Lots A, B, and C in this paper correspond to lots H, A, and E, respectively, in reference 2.

21% for 1.75 and 2.25 hr., respectively. Three percent more moisture was added as a dehulling temper about 20 min. ahead of the dehulling-degerming step. For lot C, 200 lb. was given a first temper from 12 to 20% moisture with a holding time of 1.2 hr. With these latter conditions, the hull could be readily removed so the dehulling temper was omitted. The corn was tempered at room temperature (65° to 85°F.) with tap water in equipment described previously (3).

After tempering, lots A and C were processed in a Beall degermer-dehuller, size No. 0, at motor loads of 15 and 10 hp., respectively. The degermer was fitted with a "blunt" studded rotor operated in a 50% closed position at 840 r.p.m. Three screens, approximately 0.135 in. thick, with 15/64-in. round-hole perforations (r.h.p.), and two grinding plates made up the degermer cage. Tail gate was adjusted to give an approximate 2:1 ratio of through stock to tail stock.

Tempered corn from lot B was dehulled and degermed in an experimental horizontal drum degermer fitted with a 20/64-in. r.h.p. screen (3). The rotor operated at 1,750 r.p.m. for a net motor load of about 0.4 hp.

Each degermer stock was split in a Boerner sampler to give a representative sample which was dried immediately to 17 to 18% moisture content in a forced circulation tray dryer operated at 180° F. air temperature, and then fractionated by a rolls-and-grading system into typical dry-milled products. Samples weighing roughly 4.5 lb. each of degermer stock from lots B and C were fractionated using the equipment, equipment settings, and flowsheet described previously (4). For lot A, 42 lb. of degermer stock was fractionated; this larger quantity made it necessary to substitute a vibratory sifter (Sweco) for the small box sifter and to use a 20-mesh sieve instead of the 25-mesh sieve. Fractionation of the larger quantity was delayed with the dried stock held overnight at 34° F. Upon completion of the fractionation step, the products were dried to 12% moisture or less under same conditions used for the degermer stock.

TABLE I
Yields and Fat Contents of Products

Product	Yield, %			Fat, % d.b.		
	A	B	C	A	B	C
Grits	30	45	48	0.8	1.0	0.9
Low-fat meal	16	9	9	1.0	1.3	1.2
Low-fat flour	3	4	3	2.4	1.7	NA[a]
Mix No. 1	49	58	60	0.9[b]	1.1[b]	...
High-fat meal	11	7	9	3.9	1.9	4.0
High-fat flour	2	2	2	4.3	1.4	NA
Mix No. 2	13	9	11	4.0[b]	1.8[b]	...
Hull	8	6	5	2.8	1.5	NA
Bran meal	8	9	8	3.6	3.0	NA
Germ	21	17	13	15.4	23.7	NA
Degermer fines	1	1	3	6.0	3.6	NA
Recoverable oil, lb. per cwt. corn	2.18	2.70	...			

[a]Not analyzed.
[b]Weighted average.

Before each milling run, all equipment was cleaned mechanically and decontaminated by washing it with either a 1% solution of sodium hypochlorite, or 70% ethanol. The decontamination step may not have been necessary. After milling lot B, the equipment was cleaned mechanically, but not decontaminated, for a succeeding run made on noncontaminated corn to check on possible carry-

TABLE II
Aflatoxin Content of Products

Product	B-1, p.p.b.		
	A	B	C
Corn	13	160	510
Degermer stock	13	NA	NA
Grits	Tr	12	50
Low-fat meal	2	21	80
Low-fat flour	9	50	150
Mix No. 1[a]	2	17	60
High-fat meal	16	120	670
High-fat flour	23		480
Mix No. 2[a]	17	120	640
Hull	50	380	380
Bran meal	12	90	640
Germ	18	230	1,520
Degermer fines	60	300	1,210
Composite (cal'd.)[a]	12	90	410

[a]Weighted average.

TABLE III
Distribution of Aflatoxin among Products[a]

Product	B-1		
	A	B	C
Grits	2	6	6
Low-fat meal	3	2	2
Low-fat flour	2	2	1
Mix No. 1	7	10	9
High-fat meal	14		15
High-fat flour	4	12	2
Mix No. 2	18	12	17
Hull	31	24	5
Bran meal	7	9	12
Germ	30	41	49
Degermer fines	7	4	8

[a]Expressed as weight % of aflatoxin found in all products.

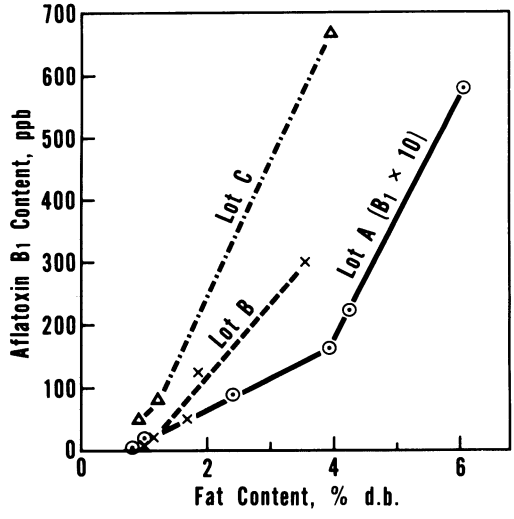

Fig. 1. Correlation of B-1 aflatoxin content of individual endosperm-derived products with their fat content. A, B, and C represent different lots of corn that were milled. Aflatoxin content of fractions from lot B was one-tenth of reading as plotted on graph.

over of the toxin. No aflatoxin B-1 was found in the prime products from the noncontaminated corn. The only aflatoxin found was a trace in the germ and hull fractions. This level could have resulted from the concentration effect when these fractions were analyzed individually in contrast to analysis of the whole corn which had a nondetectable level of the toxin.

Sample Preparation and Analytical Methods

Samples for analysis were taken of the corn being milled, degermer stock, and product fractions. Fifty-pound samples of the blended corn were ground to −20 mesh, blended, and a 2.2-lb. (1 kg.) sample was set aside for analyses. For lot A, 10 lb. of degermer stock and 175- to 350-g. portions of the milled products were taken except for the low-fat flour where only 95 g. was available. For lots B and C, 100- to 400-g. samples were taken of the milled products for aflatoxin assays with the exception of degermer fines and some flour fractions where the available quantity was in the 35- to 50-g. range.

Aflatoxin determinations were made on 50-g. portions, if available, of material (ground to −20 mesh) by the method recommended for corn (5,6). Thin-layer chromatographic plates were read on a Schoeffel SD-3000 Spectrodensitometer, with a SDC 300 Density Computer®. Values reported for the milled fractions from lots A and B are the average of duplicate determinations, and in some cases the average of three. Three and five determinations were made on corn from lots B and C, respectively. For lot A, one determination was made on the corn, two on the degermer stock, and singles on the milled fractions. For a single determination made on whole corn, the relative standard error (RSE) has been reported as 37% (7). With two determinations, the RSE decreased to 25% and for three, to 20%. Aflatoxin values above 50 p.p.b. have been rounded to the nearest multiple of 10.

Fat contents of germ fractions and whole grain were determined by Butt extraction procedure with a pentane-hexane mixture as the solvent. Other fractions were analyzed for their fat contents by a gas-liquid chromatographic method (8) which gives values some 15 to 20% higher than those of the Butt method on fractions containing about 2% fat or less.

Corn samples were graded by USDA standard procedure (9).

RESULTS AND DISCUSSION

Product Yields and Fat Contents

No problems were encountered in milling any of the lots. Yields and fat contents of most products and yield of recoverable oil were fairly typical of those for a normal dent corn (Table I). The yellow corn, lot A, presumably had a relatively low amount of vitreous endosperm, based upon the low yield of grits. Yield of degermer fines from this corn was low and germ fraction high. These yields are attributed to use of the vibratory sifter instead of the box sifter. The latter subjects degermer stock to a more thorough separation.

The relatively low fat contents of the high-fat meal and flour from lot B are typical of products from the horizontal drum degermer.

Yield of the recycle fraction (+3.5 mesh, i.e., essentially whole kernels), based on recovered products, was 8, 2, and 3% for lots A, B, and C, respectively.

Aflatoxin Contents

Among the product fractions, concentration of aflatoxin B-1 was always lowest in the grits (Table II). The level varied with that in the corn being milled, and was one-tenth or less of that in the corn. Low-fat meal had a higher aflatoxin level than the grits, followed next by low-fat flour, these levels always being considerably less than that in the corn. More specifically, B-1 level of the meal was 13 to 16% of that in the corn, while for flour, the level was about 30% for lots B and C and 70% for lot A.

As a group, the prime products (i.e., Mix No. 1), which often are used in foods, contained only 6 to 10% by weight of all aflatoxin B-1 found in the products (Table III) although the prime products represented 49 to 60% of the output in terms of product yield.

Aflatoxin concentration in the high-fat meal and flour varied on either side of that for the corn, and the quantity in these fractions represented 10 to 18% of total aflatoxin B-1 found in the products. B-1 content of bran meal was much like that in the corn.

Highest levels of aflatoxin occurred in germ, hull, and degermer fines fractions. These levels usually exceeded those in the corn, sometimes by severalfold. Together, the hull and germ fractions accounted for 54 to 72% of all aflatoxin, the major portion usually being in the germ. In milling aflatoxin-contaminated rice, Schroeder et al. (10) found 60 to 80% of the toxins by weight were in the combined bran and polish fraction removed in the dry-milling process.

For individual lots, aflatoxin B-1 content of products derived from the endosperm correlated directly with their fat content (Fig. 1). This correlation suggests that part of the aflatoxin found in these fractions resulted from contamination either by portions of the germ not being completely removed

from the grit and meal particles during the degerming and roller milling steps or by finely ground pieces of germ appearing in the flour and fines. The following data also support the hypothesis that germ can have a relatively high content of aflatoxin. A sample of essentially pure germ particles prepared later from lot C and recovered from a first break mill stream relatively rich in germ assayed 940 p.p.b. of aflatoxin B-1 (average of two determinations). These germ pieces had been prepared by cutting and scraping off adhering pieces of hull, tip cap, and endosperm with a dissecting scalpel. Most of the germ pieces appeared sound, but some were brownish in color which would be indicative of mold damage and possible contamination with aflatoxin. The mixture of hull fragments, tip cap, and endosperm removed from the above germ pieces had a lower aflatoxin content, namely, 200 p.p.b. of B-1. A third sample consisting of residual pieces of hull and endosperm and undoubtedly some small pieces of adhering germ and taken directly from the mill stream had an intermediate level of aflatoxin B-1; namely, 380 p.p.b. (Single determinations were made on the latter two samples.)

The hull fraction from lots A and B contained a significantly higher portion of total aflatoxin than the hull from lot C (Table III). Since lots A and B were pretempered and no dehulling temper was used for lot C, the question arises whether or not some aflatoxin might have diffused from other parts of the kernel, principally the germ region, into the hull during the tempering period. Because diffusion from the germ region into the hull would oppose the generally accepted direction for moisture movement within the kernel during tempering, diffusion of aflatoxin into the hull does not appear likely. Furthermore, aflatoxins are only slightly soluble in water. Another possibility for the variation noted in distribution of the toxin among our fractions is that aflatoxin content of the hull, or of kernel components comprising the hull fraction as recovered by dry milling, exceeds that of the germ region during initial or lower levels of contamination of the kernel. Also, the problem may be one of sampling. No logical explanation is now apparent for the variations in distribution of aflatoxin between germ and hull fractions.

Recovery of aflatoxin B-1 varied considerably among the three milling runs. The variations in recovery may be due in part to problems of sampling and sample size, and in part to high relative standard deviations of the assay method (7,11). Use of a larger sample in the roller milling operation presumably improved the recovery for lot A. Roller milling of samples weighing 20 lb. or more is suggested for future studies.

Bright Greenish-Yellow Fluorescence

Our milled products were examined for presence of particles exhibiting the bright greenish-yellow (BGY) fluorescence associated with presence of aflatoxin when corn is examined under a high-intensity long-wave (365 nm.) ultraviolet (UV) light (12,13). Samples were spread on a surface, as for a dark-speck count (14), illuminated with UV and the number of BGY particles counted per 10 sq. in. of sample surface. Fluorescing particles were observed in all endosperm fractions. The count usually increased in going from grits to meal to flour. However, the high-fat meal and flour fractions did not always have a higher count than their low-fat counterparts. Count for the degermer fines equalled or exceeded that of any other fraction. Few or no particles were observed in any of the germ or bran meal products in spite of their high aflatoxin contents, and none was found in the hull fractions.

In no case was the correlation within any product between BGY count and aflatoxin content adequate to permit use of this procedure as a guide to aflatoxin content of milled fractions from corn known to be contaminated. At most, BGY fluorescence must be used only as a presumptive test for aflatoxin, to be followed by a quantitative analysis specific for aflatoxin. The BGY fluorescence is not attributed to aflatoxin itself but to a compound formed from kojic acid or other metabolites of the mold (13). Enzymes of the germ, probably peroxidases, were reported by Fennell et al. (13) apparently to be involved in formation of the BGY compound. Therefore, regions of the corn kernel lacking the necessary enzymes presumably could support growth of the mold and aflatoxin formation without production of BGY fluorescing material. Our results on bran and hull fractions presumably reflect such a situation.

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