

# Effect of Propionic Acid on Aflatoxin Levels of Stored Georgia 1981 Dent Corn Samples<sup>1</sup>

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## ABSTRACT

In 1981, 75 corn samples (ca. 60 ears per sample) were collected from 4 areas in Georgia to study methods for handling freshly harvested samples. The collection was not representative of the Georgia corn crop. Every sixth ear was husked and dried the day of collection. The rest of the corn was shipped, without husking, to Peoria in cardboard boxes. When the remaining undried, unhusked samples arrived in Peoria, they were husked and randomly separated into 5 equivalent samples. One set of 75 samples was shelled and dried as soon as possible to avoid further aflatoxin formation; another set of 75 samples was stored for 6 weeks before shelling and drying. The remaining 3 sets were placed in plastic bags with 5%, 2% and 1% Monoprop (1 part propionic acid plus 1 part Versite) and stored for 6 weeks before shelling and drying. The samples dried in Georgia had an average total aflatoxin level of 199 ng/g. Samples shelled and dried immediately after arriving in Peoria had an average level of 640 ng/g. Samples shelled and dried after a 6-week storage period had average aflatoxin levels of 601 ng/g. Samples stored 6 weeks with 5%, 2% and 1% Monoprop (2.5%, 1% and 0.5% propionic acid) had average aflatoxin levels of 340 ng/g, 351 ng/g and 376 ng/g, respectively. Monoprop cannot be used to maintain the integrity of high-moisture corn samples collected for aflatoxin analysis.

## INTRODUCTION

Aflatoxin levels can increase in field-contaminated corn if the corn is stored after harvest at high moisture. One report indicated a ca. 10-fold increase in aflatoxin levels (from 200 to 2,300 ng/g), in a 3-day period after harvest, in high-moisture corn that was not dried (1). In 1979, freshly harvested dent corn was collected in Georgia at maturity and separated into two sets of 57 matched samples (2). One set was dried in Georgia the day of collection; the other set was shipped to Peoria (NRRC) before drying. The mean total aflatoxin level in the samples dried in Georgia was 36 ng/g; the mean level in the matched samples shipped to Peoria before drying was 78 ng/g. Aflatoxin formed during shipment of the freshly harvested undried corn from Georgia. Volatile organic acids, such as acetic or propionic acids, have been suggested to prevent mold growth and aflatoxin formation in corn samples stored at high moisture (1). However, when we attempted to prevent mold growth and aflatoxin formation during storage of undried corn samples in plastic bags with 5% Monoprop, aflatoxin levels decreased (3). Monoprop is a mixture of 1 part propionic acid plus 1 part Versite. Versite, a horticultural vermiculite, serves as an inert support facilitating the handling of propionic acid in the field.

We tried the storage of freshly harvested 1981 corn samples in the presence of lower levels of Monoprop—1% and 2%—as well as 5% to see if low levels could be used to prevent aflatoxin formation after collection. We are now reporting the results of that study.

## MATERIALS AND METHODS

### Sample Collection and Handling

Samples of 1981 mature dent ear corn (about 60 ears per location) were harvested from July 29 to September 7 in

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each of 75 locations in the coastal plain region of Georgia. One set of subsamples consisted of every sixth ear of corn from a lot of about 60 ears that was husked and placed in a forced-air oven at 60 C on the day of collection. The moisture of the samples was not determined. Drying took 72 hr, after which the samples were shipped to NRRC in Peoria to be shelled, prepared for analysis and analyzed for aflatoxin. The remaining unhusked ears of corn, which had not been dried, were placed in corrugated cardboard boxes for shipment to NRRC. Samples were in transit for an average of 7 days. The average temperature to be expected in the areas through which the samples were shipped is 74 F. In these areas, over the years, the average daily maximum temperature has been 84 F; the average daily minimum temperature has been 64 F. On arrival in Peoria, the husks were removed from the ears and the 75 samples were separated into 5 sets of matched subsamples. One of the 5 subsamples was shelled and dried as soon as possible (overnight at 80 C). The other 4 sets of subsamples were stored for 6 weeks at 75-85 F: one was stored untreated in paper sacks before shelling and drying. The remaining 3 sets of subsamples were placed in airtight plastic bags with 3 different levels of Monoprop—5% (5% of the weight of the corn or 2.5% propionic acid), 2% and 1%. At the end of 6 weeks, the untreated subsamples and the 3 subsamples treated with 5%, 2% and 1% Monoprop were shelled and dried. Samples were dried to less than 14% moisture.

### Preparation of Samples for Analysis

Each subsample of shelled, dried corn was coarsely ground in a Straub disc mill. The coarsely ground corn was finely ground to pass through a No. 20 sieve in a 6-in. Raymond hammer mill fitted with a screen with 1/8-in. perforations before being blended in a Hobart planetary mixer for 15 min.

### Analysis for Aflatoxin

Subsamples (50 g, ground and blended) were assayed for aflatoxins by the method designated as the CB (Contaminants Branch) method approved for corn by both the Association of Official Analytical Chemists (4) and the American Association of Cereal Chemists (5). Selected samples that had large decreases in aflatoxin levels after treatment and storage with 1%, 2% or 5% Monoprop were analyzed by the method approved by the AOAC for cottonseed (6), the method developed for aflatoxin in corn using thin-layer chromatography (TLC) or high-performance liquid chromatography (HPLC) (7) and a method for aflatoxin in mixed feeds (8).

## RESULTS AND DISCUSSION

Aflatoxin incidences and levels in the 6 subsamples—the set dried in Georgia, the set dried in Peoria after shipping, the set stored for 6 weeks, and the 3 sets stored for 6 weeks with varying amounts of Monoprop—are summarized in Table I. The samples collected were not representative of the 1981 corn harvested in Georgia or any one area of Georgia. Samples were collected for this study from farms within 140 miles of Tifton, Georgia. Four areas were represented (Table II). Group 1 and 2 samples were collected

## EFFECT OF ACID ON AFLATOXIN IN CORN

TABLE I

Comparisons of Aflatoxin Levels in 1981 Corn Samples Dried in Georgia and Peoria After Shipping (in Husks), After Storage or Treatment with Monoprop<sup>a</sup>

Total aflatoxin (ng/g)	Dried in Georgia		Shipped to NRRC in husks before drying		Ears with husks removed stored 6 weeks before drying							
	No. of samples	%	No. of samples	%	Treated with Monoprop							
					Untreated		1%		2%		5%	
					No. of samples	%	No. of samples	%	No. of samples	%	No. of samples	%
ND <sup>b</sup>	11	14.7	11	14.7	11	14.7	12	16.0	11	14.7	9	12.0
<20	13	17.3	9	12.0	3	4.0	9	12.0	5	6.7	8	10.7
20-99	16	21.3	13	17.3	14	18.7	13	17.3	19	25.3	22	29.3
100-499	21	28.0	16	21.3	22	29.3	24	32.0	25	33.3	18	24.0
≥500	14	18.7	26	34.7	25	33.3	17	22.7	15	20.0	18	24.0
% Incidence		85.3		85.3		85.3		84.0		85.3		88.0
% ≥20		68.0		73.3		81.3		72.0		78.6		77.3
% ≥100		46.7		56.0		62.6		54.7		53.3		48.0
Average level (ng/g), all samples	199		640		601		376		351		340	
Average level (ng/g), positive samples	233		750		697		448		410		388	

<sup>a</sup>Six sets of 75 matched samples.

<sup>b</sup>ND = Nondetected.

TABLE II

Total Aflatoxin in Samples of 1981 Georgia Dent Corn

Group	Number of samples	Total aflatoxin	
		Mean	Range
1	15	222.1	8-694
2	25	431.0	19-1112
3	23	37.6	0-128
4	12	5.4	0-61

near Tifton. Samples for group 3 came from a location southwest of Tifton, and those for group 4 came from a location northwest of Tifton. The samples collected further north in the 1981 collection were practically free of aflatoxin. The set of samples that was dried in Georgia best represents aflatoxin occurrence at harvest in the corn used in this study. The incidence of samples with detectable aflatoxin was 85.3%; the incidence of samples with aflatoxin at levels equal to or greater than 20 ng/g (the Food and Drug Administration guideline) was 68%. The average total aflatoxin level in samples collected and dried immediately for this study was 199 ng/g.

In 1981, the undried corn samples were not husked in Georgia before being shipped to NRRC. When the samples arrived in Peoria, all husks were removed immediately. A comparison of the results of aflatoxin analysis of subsamples dried in Georgia before shipping and those obtained of subsamples dried in Peoria on arrival revealed no difference in the incidence of nondetectable aflatoxin (Table I). However, the mean level of aflatoxin (199 ng/g) of the samples dried in Georgia the day of collection was significantly lower than the mean level of 640 ng/g in the set dried on arrival in Peoria. The incidence of aflatoxin levels of equal to or greater than 500 ng/g was 18.7% in the samples dried in Georgia and 34.7% in samples dried in Peoria. The husks probably prevented moisture from escaping; thus little drying could take place during the ca. 7 days in transit. In 1980, when husks were removed from all corn samples immediately after collection, no difference was found in mean levels of aflatoxin between samples dried in Georgia immediately after collection and those shipped without

husks and dried on arriving in Peoria (3). We recommend that husks be removed from corn samples at time of collection for aflatoxin analysis.

The mean aflatoxin levels in the untreated husked corn samples stored for 6 weeks did not increase (Table I). No significant difference was found between the mean toxin levels in corn samples dried in Peoria immediately after arrival (640 ng/g) and those stored in Peoria for 6 weeks before drying (601 ng/g). The samples treated with 1%, 2% and 5% Monoprop had significantly lower mean levels of aflatoxin—376, 351 and 340 ng/g—than the untreated samples stored for 6 weeks, and significantly higher levels than the samples dried in Georgia. Although the mean level of aflatoxin appeared to decrease as the percentage of Monoprop increased, the differences in the mean levels were not statistically significant. Two sample means, whose ratio exceeds the least significant ratio (for these samples, LSR = 1.43), differ significantly at the .05 level. The change in aflatoxin levels between the untreated samples and treated samples stored for 6 weeks was most noticeable in the incidence of samples containing toxin at levels equal to or greater than 500 ng/g. Of the untreated samples stored for 6 weeks, 33.3% had levels equal to or greater than 500 ng/g. The samples stored for 6 weeks with Monoprop had a 20-24% incidence of samples with levels equal to or greater than 500 ng/g.

No reports were found in the literature of aflatoxin destruction in corn caused by propionic acid. The hemiacetal or water adduct of aflatoxin B<sub>1</sub>, B<sub>2a</sub>, does form slowly in acidic aqueous or acetone solutions (9,10). No evidence of the presence of aflatoxin B<sub>2a</sub> in extracts of corn obtained by the CB method (4,5) was discovered. We analyzed selected samples, in which aflatoxin decreased markedly during the 6-week storage period in the presence of Monoprop, by 3 other methods using more polar extracting solvents. The 3 methods and the solvents for extraction were the official method adopted for aflatoxin in cottonseed (solvent, acetone/water [85:15]) (6), a method developed for corn (solvent, methanol/water [80:20]) (7) and a method developed for aflatoxin B<sub>1</sub> in mixed feeds (solvent, dichloromethane/20% citric acid [20:1]) (8). Aflatoxin B<sub>2a</sub> was not detected in the partially purified extracts obtained by any of the 3 methods. If B<sub>2a</sub> was formed, it

may have formed a complex with proteins in corn, causing it to be insoluble in solvents used for extraction.

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## ☘ Tofu and Tempeh as Potential Protein Sources in the Western Diet<sup>1</sup>

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#### ABSTRACT

In recent years, a growing interest in foods of plant origin, especially plant protein foods, has become evident. A large number of low-technology, Oriental soybean foods have appeared on the American market outside the Oriental community. The most popular one is tofu. Tempeh, another soybean protein food, also has attracted a lot of attention. Tempeh has not yet been accepted to the extent that tofu has, but it is becoming a hamburger-alternative for vegetarians, now numbering 10-15 million in the United States. According to information published by the Soyfoods Center of California, the number of tofu producers in North America rose from 50 in 1975 to 182 in 1983, and annual production has increased from 13,250 tons in 1979 to 27,500 tons in 1983. The annual tempeh production was estimated at 500 tons in 1982. Changes in the perceptions of soybean foods as they pertain to health and social status seem to be the determining factors behind the momentous expansion of Oriental soybean foods. Therefore, the growth trend is expected to be continuing for some time into the future. Tofu is made by curdling the protein with a calcium or magnesium salt from a water extract of whole soybeans. It is a highly hydrated, gelatinous product with a soft, smooth texture and a bland taste. Therefore, tofu can be easily incorporated with other foodstuffs and used in nearly every culinary context, from salad to dessert and from breakfast foods to burgers. Tempeh is made by fermenting boiled soybeans with *Rhizopus oligosporus*. After 20-24 hr at 30 C, the beans are covered with white mycelium that binds the beans together to form a firm cake. It can be seasoned, and cooked by frying, roasting or baking — just like meat.

#### INTRODUCTION

In recent years, a growing interest in foods of plant origin, especially plant protein foods, has become evident. A large number of low-technology soybean foods that have been an important protein source in the far eastern countries for centuries have appeared on the American market outside the Oriental community. The most popular ones are tofu, or bean curd, and tempeh. These products are basic protein foods that are prepared by traditional methods. Westerners know very little of these products and their processing technology.

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#### TRADITIONAL SOYBEAN FOODS

Traditional soybean foods may be categorized as either nonfermented or fermented. Fresh green soybeans, soybean sprouts, soybean milk, protein-lipid film, soybean curd and soybean flour are the principal nonfermented ones (1) (Table I). Of these, soybean curd, or tofu, is the most widely consumed. Despite the fact that to make tofu and protein-lipid film one first must make soybean milk, consumption of traditional soybean milk was popular only among the Chinese. Today, it is not unusual to find soybean sprouts and soybean milk in health food stores in the United States, and tofu has become a regular item in many supermarkets around the country.

Of the great number of fermented soybean foods consumed in the Far East, only a few have been studied in the West; these are described in Table II (1). Some of these products are used primarily as flavoring agents that also add some protein or amino acid to generally bland and low-protein diets. Others, such as tempeh and natto, are served as staples. Because of its universally acceptable flavor, texture and high protein, tempeh has been the hamburger alternative for vegetarians in the United States since the early 1970's.

#### TRENDS IN MARKET GROWTH FOR TOFU AND TEMPEH

According to industry statistics gathered by Shurtleff and Aoyagi (2) of The Soyfoods Center in California, tofu has been made commercially by Asian immigrants in the United States since 1904. By 1975, there were more than 50 tofu producers, all of whom were Asian-Americans. Tofu was then an ethnic food, available mostly in Chinatown and other ethnic food stores, or at supermarkets in California and Hawaii where large Oriental populations live. Publication of "The Book of Tofu" by William Shurtleff in 1975, undoubtedly set in motion the Western tofu market (Table III). About 70 tofu shops, mostly run by Caucasian-Americans, were opened in the 3 years after the publication, and the number has continued to grow. Annual production has increased from 13,250 tons in 1979 to 27,500 tons in 1983. The retail value of tofu was estimated at \$50 million