Aflatoxin Content of Peanut Hulls

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ABSTRACT

The degree of aflatoxin contamination in peanut hulls was determined by analyzing inoculated hand-shelled hulls and hulls from peanuts known to contain aflatoxin. Hulls adjusted to 20% moisture, inoculated with *Aspergillus flavus,* and incubated 7 days at 25 C supported growth of *A. flavus* but not aflatoxin production. Peanuts from 20 selected Segregation III (visible *A. flavus*) lots contained 13-353 ppb of aflatoxin. The machine-shelled hulls from these lots were analyzed and 3 lots contained no detectable aflatoxin, 13 lots contained 4-88 ppb and 4 lots contained >116 ppb. Aflatoxin concentrations of 53-87 ppb were detected in hulls when peanuts containing relatively high levels of aflatoxin (up to 26.8 ppm in damaged kernels) were carefully machine-shelled. Hulls from the same samples obtained by hand-shelling contained no detectable aflatoxin. When machine-shelled hulls were screened through successively smaller screens, the aflatoxin concentration of the smallest fraction (<3.18 mm) was always highest and indicated that small peanut kernels and peanut parts in the hulls actually contained the aflatoxin. Separating hulls over a 4.76 mm round-hole screen appeared to provide a means of removal of most aflatoxin in peanut hulls. No aflatoxin was found in hulls from uncontaminated peanuts.

INTRODUCTION

United States peanut production in the past few years has approached 2 million tons, with the potential for more than 400,000 tons of peanut hulls each year. Disposal of peanut hulls is generally a problem, and the greatest usages are as burnable fuel and cattle feed extenders in bulk or pelletized form. Uses such as charcoal, paraffin-based logs, kitty litter, mulch and flour for dietary fiber have received some attention. The potential for aflatoxin in peanuts is well documented; this potential may be extended to peanut hulls. Peanuts are analyzed to provide consumer protection and thus reduce the aflatoxin contamination potential. Hulls, however, may be utilized as cattle feed with little regard for potential aflatoxin contamination. Because of these facts, we have evaluated peanut hulls as a substrate for aflatoxin production, analyzed hulls from contaminated peanuts for aflatoxin content and determined the source of contamination in mill-run hulls.

MATERIALS AND METHODS

To determine the potential for aflatoxin production on peanut hulls, 25 g of dried, hand-shelled hulls were adjusted to approximately 20% moisture and inoculated with an isolate of *Aspergillus flavus* known to produce aflatoxin. The inoculum was 1 ml of a spore suspension (10^4 spores/ml) in sterile distilled water containing 0.025% Tween-20. Ten replicates in 250-ml flasks were incubated 10 days at 25 C, then assayed for aflatoxin by minicolumn chromatography (3,4,5).

In 1980, several loads of farmers stock Segregation III (visible *A. flavus*) peanuts were sampled with a Federal-State Inspection Service (FSIS) pneumatic sampler (1) to obtain samples of approximately 61 kg. Approximately 30 kg were separated on an FSIS farmers stock divider, and foreign material and loose shelled kernels were removed. The peanuts were shelled with a Model 4 sample sheller that has shelling characteristics closely related to commercial shelling operations (2). The shelled peanuts were analyzed and 20 lots were selected containing a range of aflatoxin from 13-353 ppb. Hulls from these 20 lots were analyzed for aflatoxin by minicolumn chromatography. Due to the absorbent nature of peanut hulls, the minicolumn extraction technique for peanuts (3,4,5) was modified by using a 7:1 (v/w) ratio of methanol/water to hulls for the initial extraction. This modification resulted in a dilution factor of 3.5, which was used in the calculation of all results. This factor must be considered in negative results (0 ppb), since the lower limit of detection was raised by the dilution. The minicolumns used for hull analysis contained double the normal amount of alumina to insure elimination of fluorescent contaminants normally found in hulls. Aflatoxin in several samples was verified by TLC comparison with standard aflatoxins. Total oil on all samples was determined on ground blended hulls by a method similar to AOCS Method Ab 3-49 (6), except that extraction thimbles were used in Soxhlet extractors.

In 1981 and 1982, peanuts were grown in various environmental control plots to study temperature and moisture stress effects on preharvest occurrence of aflatoxin (7,8). From these studies, hulls were obtained from the Model 4 sheller or hand-shelled from 1-kg samples to exclude peanuts or peanut parts. Hulls from the 1982 study were screened through the following standard slotted (S) and round (R) hole screens: 7.94 mm (5/16 in.) × 19.05 mm (3/4 in.), 7.14 mm (9/32 in.) × 19.05 mm, 6.35 mm (1/4 in.) × 19.05 mm, 4.76 mm (3/16 in.) round hole, 3.18 mm (1/8 in.) round hole. The six resulting fractions were designated as 7.94 mm R, 7.14 mm R, 6.35 mm R, 4.76 mm S, 3.18 mm S, and –3.18 mm S. The various fractions, except the smallest (–3.18 mm R), were carefully examined to determine the quantity of various peanuts and peanut parts included in each fraction. Before the screened 1982 samples were ground and blended for oil analysis, all fractions were combined.

RESULTS AND DISCUSSION

Aflatoxin was not found in hand-shelled hulls that had been inoculated with a toxigenic strain of *A. flavus.* Inoculated hulls supported growth of the fungus but not the production of aflatoxin. This indicated that hull contamination per se may not occur and suggested that hull contamination may result from extraneous material in the hulls. Mayne et al. (9) reported low levels of aflatoxin on cottonseed hulls inoculated with *A. flavus,* but did not indicate a complete separation of meats and hulls.

Analysis of the machine-shelled hulls from 20 lots of 1980 Segregation III peanuts revealed a wide range of aflatoxin concentrations (Table I). Three hull samples contained no detectable aflatoxin, 5 samples contained 4-18 ppb, 8 samples contained 25-88 ppb and 4 samples contained >116 ppb. Four of the hull samples contained more aflatoxin than the peanuts from the same load. This could occur if the hulls contained many small shriveled pods, small peanuts, and/or damaged kernels that are associated with the highest levels of contamination (11). The samples were shelled using standard shelling practices, and no special care was taken to remove and hand-shell small shriveled pods which became part of the hull components. These 20 samples represent possibly the worst situation for aflatoxin in hulls that may be encountered under similar shelling parameters. They were from peanuts selected for analysis because they had been identified not

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only as having *A. flavus* growth, but also various levels of aflatoxin contamination. In shelling, broken peanuts and small shriveled pods recur in the hulls; therefore, a relationship may exist between aflatoxin content of peanuts and aflatoxin content of resulting hulls. This relationship would be influenced by shelling efficiency as related to the quantity of peanuts, peanut parts and shriveled pods in the hulls.

Total oil data (Table I) of the 20 samples, taken as a measure of the quantity of peanuts in hulls, indicates that shelling efficiency was relatively good since most samples (80%) contained less than 1.5% total oil. This compares to total oil content of hand-shelled hulls of ca. 0.4-0.7%. The correlation coefficient of aflatoxin content of hulls with total oil per cent in hulls was 0.37. Aflatoxin content of peanuts from the 20 loads ranged from 13 to 353 ppb (Table I), and the correlation coefficient with aflatoxin content of hulls was 0.57. These data indicate that in addition to the financial factors of peanut loss in shelling, high efficiency must be maintained to keep potential aflatoxin contamination of hulls to a minimum. This concept would be of greatest importance in years characterized by late season drought when more aflatoxin contamination usually occurs (10,11) or when Segregation III peanuts are shelled.

In 1981 and 1982, lots of aflatoxin free peanuts from the environmental control plots resulted in aflatoxin free hulls, and further discussion of these lots is omitted. In 1981, hull lots from the plots contained up to 87 ppb aflatoxin, while the aflatoxin concentration in some peanuts was as high as 26 ppm (Table II). Care was exerted in shelling these samples and most contamination of these lots must have come from small peanuts and peanut pieces, since small shriveled pods, which often contain highly contaminated kernels (10,11), generally were removed and hand-shelled into various seed categories. Hulls from a 1-kg hand-shelled sample from each lot were all negative for aflatoxin, providing further evidence that aflatoxin contamination of hulls is related to peanuts in the hulls. These data also point out the need for careful shelling procedures to reduce the potential for aflatoxin contamination of peanut hulls.

TABLE I

Aflatoxin and Oil Content of Machine Shelled Hulls and Aflatoxin Content of Peanuts from 20 Lots of 1980 Segregation III Peanuts

Sample no.	Hulls		Peanuts
	Aflatoxin (ppb)	Total oil (% D.W.)	Aflatoxin (ppb)
1	18	1.55	13
2	10	0.63	17
3	0	0.42	17
4	4	1.34	21
5	7	0.81	22
6	0	0.57	52
7	28	0.88	54
8	0	0.73	76
9	77	0.61	85
10	88	1.02	88
11	53	1.10	105
12	25	1.30	105
13	18	0.79	151
14	175	1.54	163
15	235	0.55	217
16	350	1.98	220
17	63	1.85	254
18	63	0.29	277
19	116	0.59	281
20	77	1.38	353

TABLE II

Aflatoxin Content of Peanuts and Hulls from Environmental Studies, 1981

Seed category	Sample lot				
	A	B	C	D	E
	Aflatoxin ppb				
Jumbo	315	0	0	0	0
Medium	816	72	0	0	0
#1	135	trace	0	0	0
Other edible	620	4	0	0	0
Oil stock	3400	980	0	0	0
LSK[a]	1350	1680	119	270	0
Damaged	26,800	5000	4500	815	200
Hulls	77	87	87	53	0

[a]LSK = loose shelled kernels.

Machine-shelled hulls from 1982 were separated over various screens, and the percentage distributions of hulls in various size fractions among lots were similar (Table III). Loose peanuts and peanut parts generally were found in each fraction examined. The few small, shriveled pods containing peanuts generally were restricted to the largest screen size. The per cent peanuts, as determined by carefully picking peanuts and peanut parts from the hulls after screening, indicated (Table III) that most fractions contained less than 1% (wt) visible peanuts. The various −3.18 R fractions were not separated visually due to the extremely small component sizes. However, total oil percentages (Table III) indicated that the smallest fraction contained proportionally more peanuts than other fractions. Extrapolation of per cent oil data to peanut weight suggests

TABLE III

Distribution, Total Oil and Aflatoxin Concentration of Peanut Hulls in Various Screen Sizes from 1982 Environmental Studies

Screen size		Sample lot			
		A	B	C	D
7.94 mm S	% hulls	58.3	58.1	56.6	62.9
	% oil	1.0	0.4	0.4	0.4
	ppb	0	0	0	0
7.14 mm S	% hulls	5.9	5.2	6.0	5.1
	% oil	0.9	0.8	0.4	0.3
	ppb	0	0	0	0
6.35 mm S	% hulls	7.2	6.9[a]	7.2	5.4
	% oil	0.6	0.9	0.4	0.6
	ppb	0	0	0	0
4.76 mm R	% hulls	11.4	12.4[a]	11.8	11.6
	% oil	0.4	1.0	0.6	0.7
	ppb	7.0	0	0	T
3.18 mm R	% hulls	8.7	8.4[a]	7.4	6.2[a]
	% oil	0.5	1.2	0.9	1.1
	ppb	25.0	0	0	0
−3.18 mm R	% hulls	10.4	9.0	11.2	8.8
	% oil	2.6	2.4	3.9	3.2
	ppb	81.0	11.0	14.0	14.0

S = X 19.05 mm slotted screen, R = round hole screen.

[a]Contained >1% (w/w) peanut parts (−3.18 R was not visually examined).

Total oil and aflatoxin data are the mean of duplicate analyses (T = trace).

that the −3.18 R fractions may have contained 6-8% peanuts and/or peanut parts. Aflatoxin levels for 1982 plots were lower overall than in 1981. Plot A (Table III) was by far the most heavily contaminated, with 6500 ppb of aflatoxin in the damaged seed category. Plots B-D were less contaminated than A and did not contain enough seed in the damage category for an aflatoxin analysis. Aflatoxin was detected in peanuts from the 4 sample lots, and highest concentrations in edible peanuts from these lots were 3100, 250, 330 and 20 ppb, respectively (8). In each lot where aflatoxin had been detected, hulls in the smallest fraction contained the most aflatoxin (Table III). No aflatoxin was detected in hulls riding a 6.35 mm S or larger screen. Aflatoxin in peanuts and peanut parts thus appeared to be the source for aflatoxin found in peanut hulls. Lee et al. (12) separated cottonseed into hulls, fines (small, dry particles of kernels) and meats. Assays for aflatoxin indicated a marked concentration of aflatoxin in the fines and revealed an avearage 17-fold difference between fines and meats. A similar situation appears to exist in peanut hulls.

The data indicate that aflatoxin may be found in peanut hulls due to inclusion of aflatoxin containing peanuts and peanut parts in the hulls. In a worst case situation, as presented in Table I, unacceptably high aflatoxin levels may result when heavily contaminated peanuts are shelled. However, equal consideration must be given to the fact that no aflatoxin was found in hulls when aflatoxin-free peanuts were shelled. This represents by far the majority of peanuts. The risk associated with use of peanut hulls in animal feed should be relatively low. Segregation III peanuts generally account for less than 3% of national production (1970-1980 average) and are shelled distinctly separate from Segregation I peanuts. Feeding hulls from Segregation I peanuts should involve no greater risk than that presently associated with peanuts using existing U.S. grading methods. Use of peanut hulls in dairy operations, wherein contamination may be transmitted to milk, may warrant analysis of suspect hulls.

REFERENCES

1. Dickens, J.W., Trans. Amer. Soc. Agric. Eng. 7:384 (1964).
2. Davidson, J.I.; R.F. Hudgins and C.T. Bennett, Oleagineux 36:433 (1981).
3. Holaday, C.E., and J.A. Lansden, J. Agric. Food Chem. 23: 1134 (1975).
4. Holaday, C.E., JAOCS 53:603 (1976).
5. Shotwell, O.L., and C.E. Holaday, J. Assoc. Off. Anal. Chem. 64:674 (1981).
6. Official and Tentative Methods of the American Oil Chemists' Society, AOCS, Champaign, IL, 1969, Method Ab 3-49.
7. Blankenship, P.D.; R.J. Cole, T.H. Sanders and R.A. Hill, *Mycopathologia 58:69* (1984).
8. Sanders, T.H.; R.J. Cole; R.A. Hill and P.D. Blankenship, Proc. Amer. Peanut Res. Educ. Soc. 15:90 (1983).
9. Mayne, R.Y.; W.A. Pons Jr.; A.O. Franz Jr. and L.A. Goldblatt, JAOCS 43:251 (1966).
10. Sanders, T.H.; R.A. Hill; R.J. Cole and P.D. Blankenship, JAOCS 58:966A (1981).
11. Hill, R.A.; P.D. Blankenship; R.J. Cole and T.H. Sanders, Appl. Environ. Microbiol. 45:628 (1983).
12. Lee, L.S.; S.P. Koltun and S. Buco, JAOCS 60:1548 (1983).

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☘ Variability for Oil and Fatty Acid Composition in Castorbean Varieties

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ABSTRACT

Thirty-six castorbean varieties were surveyed for oil and fatty acid composition, in order to determine variability of these seed compounds. A large variability of seed oil percentage was observed, ranging from 39.6 to 59.5%. Concerning the fatty acids, little variability was observed for ricinoleic acid, which was the most abundant in the oil, ranging from 83.65 to 90.00%. The other fatty acids appeared in small concentrations and showed a small range: 0.87 to 2.35, 0.68 to 1.84, 2.96 to 5.64, 3.19 to 5.98, and 0.34 to 0.91%, for palmitic, stearic, oleic, linoleic, and linolenic acid, respectively. Non-significant correlations were observed between fatty acids and seed oil percentage. However, significant correlations were observed among fatty acid concentrations: positive and negative ones. These significant correlations could be associated with the biosynthetic pathways of the fatty acids, which are not fully elucidated. They suggest, however, that selection for a particular fatty acid will tend to increase those positively correlated, and decrease those negative ones. Selection and plant breeding techniques could then be applied to modify the oil content of the castorbean seeds, considering the variability observed. For the fatty acid composition, however, the variability was not large enough to make substantial changes in their concentrations by selection procedures. More varieties should be surveyed to find out if such variability is available.

INTRODUCTION

Variability in seed oil content and fatty acid composition is known for several oilcrops, and this knowledge has been used in breeding programs aiming to adjust the food properties of oilseeds (1).

The castorbean oil, however, is used only for industrial purposes (2). Although the chemical compositions of the castorbean and the oil are known (3,4,5,6), information concerning the germplasm variability for fatty acid composition is lacking, except when affected by different environmental conditions (7).

Therefore, 36 castorbean varieties were surveyed for chemical composition in order to find out the variability for fatty acid and oil content, for breeding purposes.

MATERIALS AND METHODS

Seeds of 36 castorbean varieties (*Ricinus communis, L*) from the collection of the Instituto Agronomico de Campinas, São Paulo, Brasil, were analyzed for oil content and for fatty acid composition.

These varieties were grown under field conditions in the Campinas Experimental Center in 1979. Samples of the seeds were taken from the first racemes when they reached the harvest point for chemical analysis. Other plant characteristics, including height, color, bloom, earliness, seed color and size, also were determined in these varieties. However, they were not used for comparisons in this work.