Natural Occurrence of Fumonisins in Corn from Iran

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Corn collected in the Mazandaran and Isfahan Provinces of Iran was analyzed for fumonisin B_1 (FB_1), fumonisin B_2 (FB_2), and fumonisin B_3 (FB_3). The samples from Mazandaran Province, situated on the Caspian littoral of Iran, were random samples from farmers' corn lots collected in September 1998, whereas those from Isfahan Province, situated further south in the center of Iran, were bought as corn cobs in the local retail market during October 1998. All 11 samples from Mazandaran showed high levels of fumonisin contamination with FB_1 levels between 1.270 and 3.980 μg/g, FB_2 levels between 0.190 and 1.175 μg/g, and FB_3 levels between 0.155 and 0.960 μg/g. Samples from Isfahan showed lower levels of contamination with eight of eight samples having detectable FB_1 (0.010−0.590 μg/g), two of eight samples having detectable FB_2 (0.050−0.075 μg/g), and two of eight samples having detectable FB_3 (0.050−0.075 μg/g). This is the first report of fumonisin contamination of corn from Iran, in which samples from the area of high esophageal cancer on the Caspian littoral have been shown to contain high levels of fumonisins.

Keywords: *Fumonisins; Iran; esophageal cancer; Fusarium; mycotoxins; corn*

INTRODUCTION

The fumonisin mycotoxins are produced in corn (*Zea mays* L.) by a small number of related *Fusarium* species, of which *F. verticillioides* (Sacc.) Nirenberg (= *F. moniliforme* Sheldon) and *F. proliferatum* (Matsushima) Nirenberg are the most important as they are common contaminants of corn in many areas of the world (Marasas, 1996). Although an increasing number of fumonisin analogues have been isolated from *Fusarium* culture material (Musser and Plattner, 1997), the main forms present in naturally contaminated corn are fumonisins B_1 (FB_1), B_2 (FB_2), and B_3 (FB_3) (Shephard et al., 1996). Fumonisins have been shown to produce a wide range of pathological effects in animals, including the economically important syndromes of leukoencephalomalacia in horses (Kellerman et al., 1990) and pulmonary edema in swine (Harrison et al., 1990). In addition, they are nephrotoxic to rats (Riley et al., 1994) and exhibit toxic effects to turkey poults (Weibking et al., 1993) and broiler chicks (Ledoux et al., 1992). Of particular concern to human health is the finding that they are hepatocarcinogenic in experimental rats (Gelderblom et al., 1991). In addition, they have been statistically associated with the high incidence of esophageal cancer (EC) in certain areas of the Transkei region of the Eastern Cape Province, South Africa (Rheeder et al., 1992). More recent work has shown an association with primary liver cancer in China (Ueno et al., 1997). On the basis of toxicological evidence, the International Agency for Research on Cancer (IARC) has declared the "toxins produced by *F. moniliforme*" to be possibly carcinogenic to humans (class 2B carcinogens) (Vainio et al., 1993).

Since their original discovery in 1988 (Gelderblom et al., 1988), fumonisins have been found as natural contaminants of corn and corn-based foods and feeds in many areas of the world. A survey published in 1996 listed 35 countries worldwide in which fumonisin contamination had been described (Shephard et al., 1996). Studies on Transkeian corn collected in high and low EC incidence areas during 1985 and 1989 showed some of the highest levels of contamination recorded in corn intended for human consumption (up to 117.5 μg/g) and focused attention on the possible role of these carcinogenic mycotoxins in the etiology of EC (Rheeder et al., 1992; Sydenham et al., 1990). Although EC is a disease of worldwide occurrence, its incidence is subject to considerable geographical variation (Marasas, 1994). In particular, extremely high incidence rates (>50 per 100000 population per year) have been reported from the Centane (Kentani) district of the southwestern Transkei region in South Africa (Makaula et al., 1996), from Linxian and Cixian Counties of Henan Province, China (Li et al., 1980), and from the Caspian littoral of Iran (Kmet and Mahboubi, 1972; Hormozdiari et al., 1975; Joint Iran−IARC Study Group, 1977). Relatively high rates have also been reported among African-Americans living around Charleston, SC (NCI, 1990) and among the population of northeastern Italy (Franceschi et al., 1990). EC is a multifactorial disease in which a number of etiological factors have been proposed. These include smoking, alcohol consumption, dietary

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10.1021/jf991196t CCC: $19.00
Published on Web 04/22/2000

deficiencies, and the consumption of corn (Xia, 1984; Jaskiewicz et al., 1988; Brown et al., 1988; Franceschi et al., 1990). In the Caspian littoral of Iran, lower consumption of some anticarcinogenic trace elements (Azin et al., 1998), consumption of a special diet for pregnant women composed of sour pomegranate seeds, black pepper, and garlic (Ghadirian, 1987), consumption of bread contaminated with silica fiber (O'Neill, 1980), very little fruit and vegetable consumption, long lasting and daily thermal irritation of the esophagus with very hot tea (Ghadirian, 1987), and environmental exposure to some carcinogenic trace elements (Azin et al., 1998) have all been considered to play a role in the development of EC.

In addition to the results reported for the Transkei (Rheeder et al., 1992; Sydenham et al., 1990), high levels of fumonisins have been reported in corn from high EC incidence areas in China (Chu and Li, 1994; Zhang et al., 1997). The presence of fumonisins has also been recorded in corn from relatively high incidence areas in South Carolina (Sydenham et al., 1991) and Italy (Doko and Visconti, 1994). This study is the first to report fumonisin contamination of corn from the high EC incidence area in Mazandaran Province on the southern shore of the Caspian Sea.

EXPERIMENTAL PROCEDURES

Corn Samples. During September 1998, 11 samples of farmers' corn lots (from the fields of Dasht-e-Naz Sari city), intended for animal feed, were collected at random from consignments sold to the Iranian Agriculture Office in Mazandaran Province. During October 1998, corn ears (eight samples) intended for human consumption were bought at different periods from a local retail market in Isfahan Province. All of the samples were dried, ground to a meal, and stored at 4 °C prior to dispatch to the Programme on Mycotoxins and Experimental Carcinogenesis (PROMEC), South Africa, for fumonisin determination.

Determination of FB_1, FB_2, and FB_3. Fumonisins were isolated at PROMEC according to the method of Cawood et al. (1991), and a standard solution containing 50 μg/mL of each analogue was used for analysis. Each sample was analyzed for the presence of FB_1, FB_2, and FB_3 using the method of Sydenham et al. (1996) with minor modification. In brief, subsamples (20 g) of ground corn were extracted with methanol/water (70:30; 100 mL). Aliquots of the filtered extracts were cleaned up on strong anion exchange (SAX) solid-phase extraction cartridges. Fumonisins were eluted from the SAX cartridges with acetic acid (1%) in methanol and the eluates dried under nitrogen. Residues were redissolved in methanol, and an aliquot was derivatized with *o*-phthaldialdehyde (OPA) prior to separation on a reversed-phase HPLC system using fluorescence detection. The detection limit of the method was of the order of 0.01 μg/g for all three fumonisin analogues.

Statistics. Statistical analyses of results were made according to standard analysis of variance (ANOVA) using the Systat 8 software package (SPSS Inc., Chicago, IL).

RESULTS AND DISCUSSION

The relative geographical positions of Mazandaran and Isfahan provinces are shown in Figure 1. The fumonisin levels determined in the corn sampled in Mazandaran Province during 1998 are shown in Table 1. High levels of all three fumonisin analogues were found in all samples analyzed, with FB_1 ranging between 1.270 and 3.980 μg/g. Total fumonisins ranged from 1.625 to 6.115 μg/g, the latter being above the maximum level of 5 μg/g recommended for corn used as horse feed (Joint Mycotoxin Committee, 1994; Miller et al., 1996). As observed in most naturally contaminated corn samples elsewhere in the world (Shephard et al., 1996), FB_1 was the most abundant analogue and accounted on average for 74% (range = 65−79%) of the total fumonisins in each sample.

Figure 1. Map of Iran, showing relative positions of Mazandaran and Isfahan provinces.

Table 1. Fumonisin Levels in Corn Samples from Mazandaran Province, Iran

sample	fumonisins (μg/g) FB_1	FB_2	FB_3	total fumonisins (μg/g)
1	2.960	0.665	0.450	4.075
2	3.360	0.940	0.550	4.850
3	3.160	0.870	0.490	4.520
4	1.975	0.335	0.190	2.500
5	1.565	0.310	0.225	2.100
6	1.270	0.190	0.165	1.625
7	1.765	0.315	0.260	2.340
8	3.980	1.175	0.960	6.115
9	1.270	0.210	0.155	1.635
10	1.765	0.275	0.270	2.310
11	1.890	0.345	0.260	2.495
mean	2.269	0.512	0.361	3.142
(median)	(1.890)	(0.335)	(0.260)	(2.495)

Table 2. Fumonisin Levels in Corn Samples from Isfahan Province, Iran

sample	fumonisins (μg/g) FB_1	FB_2	FB_3	total fumonisins (μg/g)
1	0.545	0.050	0.050	0.645
2	0.090	nd[a]	nd	0.090
3	0.020	nd	nd	0.020
4	0.590	0.075	0.075	0.740
5	0.035	nd	nd	0.035
6	0.010	nd	nd	0.010
7	0.040	nd	nd	0.040
8	0.025	nd	nd	0.025
mean	0.169	0.063	0.063	0.201
(median)[b]	(0.038)	(0.063)	(0.063)	(0.038)

[a] nd, not detected (<0.01 μg/g). [b] Mean and median of positive samples.

Mazandaran Province lies along the southern shore of the Caspian Sea, an area previously identified as one of the world's "hot spots" for EC (Joint Iran−IARC Study Group, 1977). A recent esophageal balloon cytology screening and surveillance program in Bandar Torkaman, a port in Golestan province, over the period 1995−1997 has confirmed a high incidence of EC in the southeastern Caspian littoral of Iran, which contrasts with the low incidence of EC found in Isfahan Province (Dr. F. Saidi, Shaheed Beheshti University of Medical

Table 3. Fumonisin Levels in Corn from High EC Risk Areas in China, Iran, Italy, South Africa, and the United States

sample type	origin	cancer area	date	FB_1 (μg/g) mean[a]	FB_1 (μg/g) range	FB_2 (μg/g) mean[a]	FB_2 (μg/g) range	FB_3 (μg/g) mean[a]	FB_3 (μg/g) range	reference
China										
corn	Linxian County	high EC	1989	0.87 (13/27)	<0.10−2.96	0.45 (3/27)	<0.10−0.55	no data		Yoshizawa et al., 1994
	Shangqiu County	low EC	1989	0.89 (5/20)	<0.10−1.73	0.33 (2/20)	<0.10−0.45	no data		Yoshizawa et al., 1994
fine corn	Linxian County	high EC	1991	30.4 (7/7)	19.8−60.0	no data		no data		Chu and Li, 1994
	Cixian County	high EC	1991	39.5 (8/8)	30.3−47.9	no data		no data		Chu and Li, 1994
moldy corn	Linxian County	high EC	1991	49.3 (9/9)	17.9−118.4	no data		no data		Chu and Li, 1994
	Cixian County	high EC	1991	93.8 (10/10)	28.4−154.9	no data		no data		Chu and Li, 1994
corn	Haimen	high liver	1993	5.11 (37/40)	<0.05−25.97	2.02 (25/40)	<0.10−6.77	1.12 (25/40)	<0.10−4.13	Ueno et al., 1997
	Penlai	low liver	1993	0.65 (16/40)	<0.05−3.19	0.50 (7/40)	<0.10−1.19	0.18 (4/40)	<0.10−0.27	Ueno et al., 1997
corn	Linxian County	high EC	1994	2.73 (27/34)	0.08−21.00	0.70 (20/34)	0.05−4.35	0.38 (15/34)	0.06−1.66	Gao and Yoshizawa, 1997
	Shangqiu County	low EC	1994	2.70 (10/20)	0.08−8.47	0.59 (8/20)	0.09−1.22	0.31 (7/20)	0.09−0.58	Gao and Yoshizawa, 1997
corn	Linxian, Cixian, and Anyang Counties	high EC	1995/6	total fumonisins (ELISA test kit): mean of all samples = 0.70 μg/g (106/164); range = <0.5−16.0						Zhang et al., 1997
	Fanxian and Yanqing Counties	low EC	1995/6	total fumonisins (ELISA test kit): mean of all samples = 0.20 μg/g (23/82); range = <0.5−1.5						Zhang et al., 1997
Iran										
corn	Mazandaran	high EC	1998	2.27 (11/11)	1.27−3.98	0.51 (11/11)	0.19−1.18	0.36 (11/11)	0.15−0.96	this study
corn	Isfahan	low EC	1998	0.17 (8/8)	0.01−0.59	0.06 (2/8)	<0.01−0.08	0.06 (2/8)	<0.01−0.08	this study
Italy										
corn			1992/3	2.81 (7/7)	0.10−5.31	0.84 (7/7)	0.03−1.48	no data		Doko and Visconti, 1994
polenta, corn flour, grits			1992/3	2.53 (8/8)	0.42−3.76	0.58 (8/8)	0.80−0.91	no data		Doko and Visconti, 1994
Transkei, Eastern Cape Province, South Africa										
good corn	Bizana	low EC	1985	0.38 (2/12)	<0.05−0.55	0.08 (3/12)	<0.05−0.15	no data		Rheeder et al., 1992
	Centane	high EC	1985	1.60 (12/12)	0.05−7.90	0.61 (10/12)	<0.05−2.25	no data		Rheeder et al., 1992
moldy corn	Bizana	low EC	1985	6.52 (11/11)	0.45−18.90	2.50 (11/11)	0.15−6.75	no data		Rheeder et al., 1992
	Centane	high EC	1985	23.90 (12/12)	3.45−46.90	7.55 (12/12)	0.90−16.30	no data		Rheeder et al., 1992
good corn	Bizana	low EC	1989	0.67 (6/8)	<0.05−3.31	0.52 (2/8)	<0.05−0.97	no data		Rheeder et al., 1992
	Centane	high EC	1989	1.84 (5/6)	<0.05−5.38	0.51 (5/6)	<0.05−1.32	no data		Rheeder et al., 1992
moldy corn	Bizana	low EC	1989	4.05 (7/7)	0.11−11.34	1.28 (6/7)	<0.05−3.70	no data		Rheeder et al., 1992
	Centane	high EC	1989	53.74 (6/6)	3.02−117.5	13.68 (6/6)	0.75−22.96	no data		Rheeder et al., 1992
United States										
cornmeal/grits	Charleston, SC	high EC	1990/1	0.64 (7/7)	0.11−1.92	0.18 (6/7)	<0.05−0.46	no data		Sydenham et al., 1991

[a] Means represent the mean of all positively contaminated samples.

Sciences and Health Services, Tehran, Iran, personal communication). Previous studies on fumonisin contamination of corn in areas of high EC risk are summarized in Table 3, which contains results for China (in areas of both low and high EC and primary liver cancer risk), the Transkei region of South Africa, Italy, and the United States. Some of the highest levels of fumonisins recorded in corn for human consumption have been found in moldy corn in the high EC risk areas of Cixian County, China (154.9 μg/g; Chu and Li, 1994) and in the Centane district, Transkei region of South Africa (117.5 μg/g; Rheeder et al., 1992). However, the mean FB_1 level (2.269 μg/g) in the samples from Mazandaran is comparable to the mean reported for a limited number of Italian corn products (Doko and Visconti, 1994), as well as to the mean FB_1 levels found in "good corn" in the high EC risk area of Centane, Transkei, during 1985 and 1989 (1.60 and 1.84 μg/g, respectively; Rheeder et al., 1992); it is considerably higher than the mean of 0.64 μg/g (range = 0.11−1.84 μg/g) observed in various corn products purchased in Charleston, SC, in 1990/1991 (Sydenham et al., 1991). Fumonisin contamination levels in corn from high EC risk areas of China show a seasonal variation. Corn sampled in Linxian County in 1989 showed a mean FB_1 level of 0.87 μg/g, which was not statistically significantly different from the mean of 0.89 μg/g in the low EC risk area of Shangqiu County (Yoshizawa et al., 1994), whereas samples of fine corn from Linxian and Cixian Counties in 1991 had means of 30.4 and 39.5 μg/g, respectively, with even higher levels being recorded in samples of moldy corn (means of 49.3 and 93.8 μg/g, respectively; Chu and Li, 1994). Corn collected during 1994 from Linxian and Shangqiu Counties had mean FB_1 levels of 2.73 and 2.70 μg/g, respectively (Gao and Yoshizawa, 1997), which are comparable to the levels observed in Mazandaran for the samples in this study. All samples from Mazandaran were contaminated with fumonisins, whereas the fraction contaminated in the high EC risk area of Linxian (27/34) was higher than in the low EC risk area of Shangqiu (10/20). A similar difference in contamination rate between these two areas had previously been observed in 1989 (13/27 compared to 5/20, respectively; Yoshizawa et al., 1994). Differences in mean total fumonisin levels determined by enzyme-linked immunosorbent assay (0.70 μg/g for all samples compared to 0.20 μg/g) and fumonisin contamination rates (106/164 compared to 23/82) between high (Linxian, Cixian, and Anyang Counties) and low (Fanxian and Yanqing Counties) EC incidence areas were again noted in 1995/1996 (Zhang et al., 1997).

In contrast to the corn from Mazandaran, the corn kernels bought in markets in Isfahan Province showed far lower levels of fumonisin contamination (Table 2). Indeed, the mean FB_1 level was only 0.169 μg/g as opposed to the 2.269 μg/g in samples from the former area. Although all samples from Isfahan showed some FB_1 contamination, most levels were below 0.1 μg/g, except for two samples with levels of 0.545 and 0.590 μg/g. These latter samples were the only samples from Isfahan that showed contamination with the other fumonisin analogues. Although sample numbers are low, the results clearly show a considerable difference in fumonisin occurrence in corn grown in these two separate areas. Both the levels of the most abundant analogue, FB_1, and the levels of total fumonisins showed statistically significant ($p < 0.0001$) differences between the areas. It is of interest to compare these results with fumonisin levels determined in corn elsewhere in the world (Table 3). Clearly, the levels found in corn kernels from Isfahan (mean FB_1 levels of 0.169 μg/g) compare favorably with levels previously reported for fumonisins in agricultural crops from other areas in the world (Shephard et al., 1996) and lie within the limit of 1 μg/g for combined FB_1 and FB_2 proposed in Switzerland for corn intended for human consumption (FAO, 1997).

These results have identified high levels of fumonisin contamination in corn from the high EC incidence area along the Caspian littoral of Iran. Thus, high fumonisin levels have now been found in corn from all areas previously identified as world "hot spots" for EC. Given the seasonal variation that can occur in fungal and mycotoxin contamination, further studies of corn from northern Iran are clearly of importance.

ACKNOWLEDGMENT

We thank Dr. J. P. Rheeder for statistical comparison of results.

LITERATURE CITED

Azin, F.; Raie, R. M.; Mahmoudi, M. M. Correlation between the Levels of Certain Carcinogenic and Anticarcinogenic Trace Elements and Esophageal Cancer in Northern Iran. *Ecotoxicol. Environ. Saf.* **1998**, *39*, 179−184.

Brown, L. M.; Blot, W. J.; Schumann, S. H.; Smith, V. M.; Ershow, A. G.; Marks, R. D.; Fraumeni, J. F. Environmental Factors and High Risk of Esophageal Cancer among Men in Coastal South Carolina. *J. Natl. Cancer Inst.* **1988**, *80*, 1120−1125.

Cawood, M. E.; Gelderblom, W. C. A.; Vleggaar, R.; Behrend, Y.; Thiel, P. G.; Marasas, W. F. O. Isolation of the Fumonisin Mycotoxins: A Quantitative Approach. *J. Agric. Food Chem.* **1991**, *39*, 1958−1962.

Chu, F. S.; Li, G. Y. Simultaneous Occurrence of Fumonisin B_1 and Other Mycotoxins in Moldy Corn from the People's Republic of China in Regions with High Incidences of Esophageal Cancer. *Appl. Environ. Microbiol.* **1994**, *60*, 847−852.

Doko, M. B.; Visconti, A. Occurrence of Fumonisins B_1 and B_2 in Corn and Corn-based Human Foodstuffs in Italy. *Food Addit. Contam.* **1994**, *11*, 433−439.

FAO (Food and Agricultural Organization). *Worldwide Regulations for Mycotoxins 1995. A Compendium. Food and Nutrition Paper*; FAO Food and Nutrition Paper 64; Food and Agricultural Organization of the United Nations: Rome, Italy, 1997.

Franceschi, S.; Bidoli, E.; Baron, A. E.; La Vecchia, C. Maize and Risk of Cancers of the Oral Cavity, Pharynx, and Esophagus in Northern Italy. *J. Natl. Cancer Inst.* **1990**, *82*, 1407−1411.

Gao, H.-P.; Yoshizawa, T. Further Study on *Fusarium* Mycotoxins in Corn and Wheat from a High-Risk Area for Human Esophageal Cancer in China. *Mycotoxins* **1997**, *45*, 51−55.

Gelderblom, W. C. A.; Jaskiewicz, K.; Marasas, W. F. O.; Thiel, P. G.; Horak, R. M.; Vleggaar, R.; Kriek, N. P. J. Fumonisins—Novel Mycotoxins with Cancer-promoting Activity Produced by *Fusarium moniliforme*. *Appl. Environ. Microbiol.* **1988**, *54*, 1806−1811.

Gelderblom, W. C. A.; Kriek, N. P. J.; Marasas, W. F. O.; Thiel, P. G. Toxicity and Carcinogenicity of the *Fusarium moniliforme* Metabolite, Fumonisin B_1, in rats. *Carcinogenesis* **1991**, *12*, 1247−1251.

Ghadirian, P. Food Habits of the People of the Caspian Littoral of Iran in Relation to Esophageal Cancer. *Nutr. Cancer* **1987**, *9*, 147−157.

Harrison, L. R.; Colvin, B. M.; Green, J. T.; Newman, L. E.; Cole, J. R. Pulmonary Edema and Hydrothorax in Swine

Produced by Fumonisin B_1, a Toxic Metabolite of *Fusarium moniliforme*. *J. Vet. Diagn. Invest.* **1990**, *2*, 217−221.

Hormozdiari, H.; Day, N. E.; Aramesh, B.; Mahboubi, E. Dietary Factors and Esophageal Cancer in the Caspian Littoral of Iran. *Cancer Res.* **1975**, *35*, 3493−3498.

Jaskiewicz, K.; Marasas, W. F. O.; Lazarus, C.; Beyers, A. D.; Van Helden, P. D. Association of Esophageal Cytological Abnormalities with Vitamin and Lipotrope Deficiencies in Populations at Risk for Esophageal Cancer. *Anticancer Res.* **1988**, *8*, 711−716.

Joint Iran−IARC Study Group. Esophageal Cancer Studies in the Caspian Littoral of Iran. Results of Population Studies−A Prodrome. *J. Natl. Cancer Inst.* **1977**, *59*, 1127−1138.

Joint Mycotoxin Committee. *J. AOAC Int.* **1994**, *77*, 257−258.

Kellerman, T. S.; Marasas, W. F. O.; Thiel, P. G.; Gelderblom, W. C. A.; Cawood, M.; Coetzer, J. A. W. Leukoencephalomalacia in Two Horses Induced by Oral Dosing of Fumonisin B_1. *Onderstepoort J. Vet. Res.* **1990**, *57*, 269−275.

Kmet, J.; Mahboubi, E. Esophageal Cancer in the Caspian Littoral of Iran: Initial Studies. *Science* **1972**, *175*, 846−853.

Ledoux, D. R.; Brown, T. P.; Weibking, T. S.; Rottinghaus, G. E. Fumonisin Toxicity in Broiler Chicks. *J. Vet. Diagn. Invest.* **1992**, *4*, 330−333.

Li, M. H.; Li, P.; Li, P. J. Research on Esophageal Cancer in China. *Adv. Cancer Res.* **1980**, *33*, 174−249.

Makaula, N. A.; Marasas, W. F. O.; Venter, F. S.; Badenhorst, C. J.; Bradshaw, D.; Swanevelder, S. Oesophageal and Other Cancer Patterns in Four Selected Districts of Transkei, Southern Africa. *Afr. J. Health Sci.* **1996**, *3*, 11−15.

Marasas, W. F. O. *Fusarium*. In *Foodborne Disease Handbook: Diseases Caused by Viruses, Parasites, and Fungi*; Hui, Y. H., Gorham, J. R., Murrell, K. D., Cliver, D. O., Eds.; Dekker: New York, 1994; Vol. 2, pp 521−573.

Marasas, W. F. O. Fumonisins: History, Worldwide Occurrence and Impact. In *Fumonisins in Food*; Jackson, L. S., De Vries, J. W., Bullerman, L. B., Eds.; Plenum Press: New York, 1996; pp 1−17.

Miller, M. A.; Honstead, J. P.; Lovell, R. A. Regulatory Aspects of Fumonisins with respect to Animal Feed. In *Fumonisins in Food*; Jackson, L. S., De Vries, J. W., Bullerman, L. B., Eds.; Plenum Press: New York, 1996; pp 363−368.

Musser, S. M.; Plattner, R. D. Fumonisin Composition in Cultures of *Fusarium moniliforme*, *Fusarium proliferatum* and *Fusarium nygami*. *J. Agric. Food Chem.* **1997**, *45*, 1169−1173.

NCI (National Cancer Institute). *Cancer Statistics Review 1973−1987*; National Institutes of Health Publication 90-2789; NIH: Bethesda, MD, 1990.

O'Neill, C. H.; Hodges, G. M.; Riddle, P. N.; Jordan, P. W.; Newman, R. H.; Flood, R. J.; Toulson, E. C. A Fine Fibrous Silica Contaminant of Flour in the High Oesophageal Cancer Area of North-East Iran. *Int. J. Cancer* **1980**, *26*, 617−628.

Rheeder, J. P.; Marasas, W. F. O.; Thiel, P. G.; Sydenham, E. W.; Shephard, G. S.; Van Schalkwyk, D. J. *Fusarium moniliforme* and Fumonisins in Corn in Relation to Human Esophageal Cancer in Transkei. *Phytopathology* **1992**, *82*, 353−357.

Riley, R. T.; Hinton, D. M.; Chamberlain, W. J.; Bacon, C. W.; Wang, E.; Merrill, A. H., Jr.; Voss, K. A. Dietary Fumonisin B_1 Induces Disruption of Sphingolipid Metabolism in Sprague−Dawley Rats: A New Mechanism of Nephrotoxicity. *J. Nutr.* **1994**, *124*, 594−603.

Shephard, G. S.; Thiel, P. G.; Stockenström, S.; Sydenham, E. W. Worldwide Survey of Fumonisin Contamination of Corn and Corn-based Products. *J. AOAC Int.* **1996**, *79*, 671−687.

Sydenham, E. W.; Thiel, P. G.; Marasas, W. F. O.; Shephard, G. S.; Van Schalkwyk, D. J.; Koch, K. R. Natural Occurrence of Some *Fusarium* Mycotoxins in Corn from Low and High Esophageal Cancer Prevalence Areas of the Transkei, Southern Africa. *J. Agric. Food Chem.* **1990**, *38*, 1900−1903.

Sydenham, E. W.; Shephard, G. S.; Thiel, P. G.; Marasas, W. F. O.; Stockenström, S. Fumonisin Contamination of Commercial Corn-based Human Foodstuffs. *J. Agric. Food Chem.* **1991**, *39*, 2014−2018.

Sydenham, E. W.; Shephard, G. S.; Thiel, P. G.; Stockenström, S.; Snijman, P. W.; Van Schalkwyk, D. J. Liquid Chromatographic Determination of Fumonsins B_1, B_2, and B_3 in Corn: AOAC−IUPAC Collaborative Study. *J. AOAC Int.* **1996**, *79*, 688−696.

Ueno, Y.; Iijima, K.; Wang, S.-D.; Sugiura, Y.; Sekijima, M.; Tanaka, T.; Chen, C.; Yu, S.-Z. Fumonisins as a Possible Contributory Risk Factor for Primary Liver Cancer: A 3-Year Study of Corn Harvested in Haimen, China, by HPLC and ELISA. *Food Chem. Toxicol.* **1997**, *35*, 1143−1150.

Vainio, H.; Heseltine, E.; Wilbourn, J. Report on an IARC Working Group Meeting on Some Naturally Occurring Substances. *Int. J. Cancer* **1993**, *53*, 535−537.

Weibking, T. S.; Ledoux, D. R.; Bermudez, A. J.; Turk, J. R.; Rottinghaus, G. E.; Wang, E.; Merrill, A. H., Jr. Effects of Feeding *Fusarium moniliforme* Culture Material, Containing Known Levels of Fumonisin B_1, on the Young Broiler Chick. *Poult. Sci.* **1993**, *72*, 456−466.

Xia, A. J. Carcinogenesis in the Esophagus. In *Carcinoma of the Esophagus and Gastric Cardia*; Huang, G. J., Wu, Y. K., Eds.; Springer-Verlag: Berlin, Germany, 1984; pp 52−76.

Yoshizawa, T.; Yamashita, A.; Luo, Y. Fumonisin Occurrence in Corn from High- and Low-Risk Areas for Human Esophageal Cancer in China. *Appl. Environ. Microbiol.* **1994**, 60, 1626−1629.

Zhang, H.; Nagashima, H.; Goto, T. Natural Occurrence of Mycotoxins in Corn, Samples from High and Low Risk Areas for Human Esophageal Cancer in China. *Mycotoxins* **1997**, *44*, 29−35.

Received for review November 3, 1999. Revised manuscript received February 16, 2000. Accepted February 23, 2000.

JF991196T