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NOTES

Occurrence of Fumonisins B_1 and B_2 in Corn-Based Products from the Spanish Market

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The natural occurrence of fumonisins B_1 and B_2, the incidence of *Fusarium* organisms, and the capacity of *Fusarium* isolates to produce fumonisins were investigated with 50 corn-based samples from Spain destined for human consumption. Eight samples (16%) were found to be contaminated with fumonisins. The levels of contamination were very low, with a mean of 80 ng/g.

Investigations carried out in the last decade showed that members of the genus *Fusarium* have the capacity to produce a high number of mycotoxins. Recently, the fumonisins, a new class of mycotoxins, were identified and characterized (1, 2, 4). To date, six different fumonisins have been identified. Of them, fumonisin B_1 (FB_1), FB_2, and FB_3 are the major toxins produced in fungal cultures or present in naturally contaminated corn samples, while the other three, FB_4, FA_1, and FA_2, are produced only in minor amounts (2).

Production of these mycotoxins seems to be limited to some *Fusarium* species. *F. moniliforme* and *F. proliferatum* are the main species producing high yields of fumonisins (7, 14).

Fumonisins are implicated in leukoencephalomalacia in equine species (14, 16) and seem to be associated with pulmonary edema syndrome in swine (7) and esophageal cancer in humans (3, 13). So, humans and animals are susceptible to the toxic effects of these mycotoxins. Because of their recent isolation and characterization, there have been very few studies of their world distribution; however, their occurrence in South Africa, the United States, Switzerland, Brazil, and some Asian countries (5, 6, 9–12, 16) has been studied.

There is, however, no information available regarding the natural occurrence of these mycotoxins in Spain. This paper describes the first study carried out in Spain of the fumonisin incidence in corn destined for human consumption.

Corn-based samples. Samples were taken directly by staff from analytical laboratories (between January and May 1993) as single random purchases from the retail market in Lleida and Valencia, Spain. The samples were usually sent to the laboratory as soon as they were collected and were tested on arrival. Otherwise they were stored at −20°C to arrest any fumonisin formation up to the time of analysis. The distribution of the samples is given in Table 1.

Reference standards. FB_1 and FB_2 standards were purchased from the Division of Food Science and Technology, Commonwealth Scientific and Industrial Research Organization, Pretoria, South Africa.

FB_1 and FB_2 analysis. The samples were analyzed by a modification of the Shephard method (8). Fifty grams of each sample was blended with 50 ml of ethyl acetate and filtered. The solid part was blended with 100 ml of methanol-water (3:1) and filtered. The filtrate was centrifuged at 613 × *g* at 4°C for 10 min, and the eluate was recovered. A 10-ml aliquot was applied to an Analytichem Bond-Elut Sax cartridge (Varian) which had been conditioned with 5 ml of methanol followed by 5 ml of methanol-water (3:1). Subsequently, the cartridge was washed successively with 8 ml of methanol-water (3:1) and 3 ml of methanol, and the toxins were eluted with 14 ml of 0.5% acetic acid in methanol. The eluate was evaporated to dryness under vacuum, redissolved in 2 ml of methanol, and reevaporated to dryness under vacuum. The residue was dissolved in 0.2 μl of methanol. Two hundred microliters of *o*-phthaldialdehyde reagent, prepared according to the method of Shephard et al. (8), was added to a 50-μl sample solution, of which 20 μl was injected into a high-performance liquid chromatography system between 1 and 2 min after derivatization. The eluant was methanol–0.1 M potassium dihydrogen phosphate (4:1) adjusted to pH 3.3 with *ortho*-phosphoric acid. The flow rate was 1.5 ml/min.

The ranges and means of fumonisin concentrations together with the number of samples found to be positive for each fumonisin are given in Table 1. The distribution of fumonisin

TABLE 1. Incidence and levels of fumonisins in corn-based products from Spain

Product	Toxin	Incidence (no. positive[a]/total)	Range of toxin amt (ng/g)	Mean toxin amt (ng/g)
Corn grits	FB_1	3/15	0–90	60
	FB_2	0/15	ND[b]	
Corn flakes	FB_1	2/12	0–100	60
	FB_2	0/12	ND	
Snacks	FB_1	2/11	0–200	130
	FB_2	0/11	ND	
Toasted corn	FB_1	0/9	ND	
	FB_2	0/9	ND	
Corn flour	FB_1	1/3	0–70	70
	FB_2	0/3	ND	

[a] Contamination above 50 ng/g.
[b] ND, not detected.

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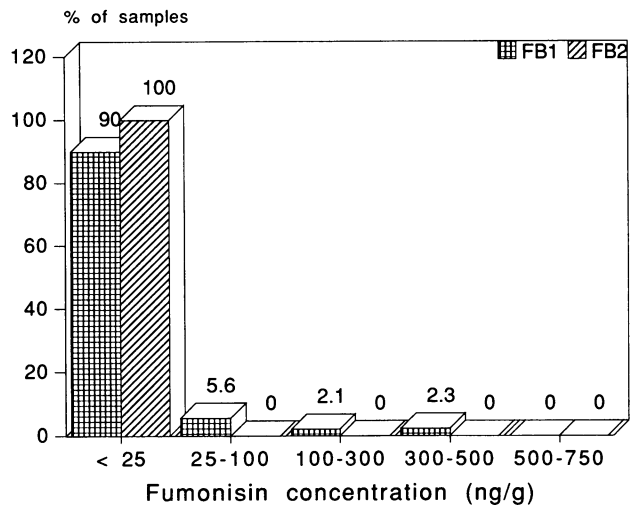

FIG. 1. Fumonisin concentrations in corn-based products from Spain.

concentrations in commercial corn product samples is given in Fig. 1.

Eight samples (16%) of 50 were found to be contaminated with FB_1 in concentrations ranging from 50 to 200 ng/g (mean, 80 ng/g). Of these eight positive samples, none contained any detectable levels of FB_2. The highest frequency of positive samples was found in corn grits (20%), followed by snacks (18%) and corn flakes (17%). Of three corn flour samples, one was found to be contaminated. None of the toasted corn samples contained fumonisins. The levels of contamination in the samples analyzed were similar to those obtained by Pittet et al. (6) in samples from the Swiss market.

There were no differences in fumonisin content among the kinds of samples and between the samples collected in Lleida and Valencia. As expected, FB_1 was always the major fumonisin in positive samples.

We think that it is necessary to have more information about the fumonisin incidence in other areas in order to know the extent to which this mycotoxin is a problem in Spain. Furthermore, it will be interesting to know the ecological factors that influence the accumulation of fumonisins in the grains in order to prevent and control it.

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