

## Occurrence of Fumonisin B<sub>1</sub> and Hydrolyzed Fumonisin B<sub>1</sub> in Mexican Nixtamalized Cornmeal

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Fumonisin is a group of mycotoxins produced by *Fusarium* species belonging to the Liseola section, primarily by strains of *Fusarium verticillioides*, *F. proliferatum*, and other *Fusarium* species commonly associated with corn and other cereal grains (Gelderblom *et al.* 1988). Fumonisin B<sub>1</sub> (FB<sub>1</sub>) is the most abundant toxin representing about 70% of the total fumonisins concentration in naturally contaminated foods and feeds (Ross *et al.* 1992). Fumonisin is considered cancer promotor (Gelderblom *et al.* 1992, Bacon and Nelson 1994). Fumonisin has been associated with equine leucoencephalomalacia (ELEM) and porcine pulmonary edema syndrome (Bezuidenhout *et al.* 1988, Norred and Voss 1994).

Fumonisin levels in corn-based products have also been associated with human esophageal cancer in the Republic of South Africa, where FB<sub>1</sub> concentration reached up to 11.1 mg/Kg (Sydenham *et al.* 1990). In Mexico, Rosiles *et al.* (1998) reported an outbreak of ELEM in donkeys, and fumonisins were found to be responsible. The state of Sonora, located in northwest Mexico, is one of the main agricultural areas of this country; consequently, a large number of storage facilities are distributed throughout its territory. Of the grains produced in Sonora, maize is one of the most commonly stored at farms and the occurrence of FB<sub>1</sub> was recently reported by Cortez-Rocha *et al.* (2003).

Nixtamalization is a traditional process of cooking and steeping the corn with Ca(OH)<sub>2</sub> and heat to produce masa flour. Nixtamalized cornmeal is a basic material to produce tortillas and other products, which are a basic food for Mexican people. Even though cornmeal is subjected to high temperature processes, fumonisins levels are not decreased enough and, in addition, the hydrolyzed toxin is formed (Jackson *et al.* 1999). The aim of this study was to determine the occurrence and concentration of FB<sub>1</sub> and HFB<sub>1</sub> in nixtamalized cornmeal commercialized in Hermosillo, Sonora, Mexico, which is used to make tortillas and other food products.

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## MATERIALS AND METHODS

A total of 80 nixtamalized cornmeal samples of 1 Kg were collected during May and July, 2002 from supermarkets, retail outlets, and groceries stores from 4 geographical sections of Hermosillo, Sonora, Mexico. All samples were manufactured during 2002. From May to July 2003, 71 samples were collected from all the retail outlets (71) in the city. Once the samples were received at the laboratory, the levels of total fumonisins were screened using the Fumonitest<sup>®</sup> immunoaffinity column method from VICAM (Fumonitest Manual).

High performance liquid chromatography was used to confirm FB<sub>1</sub> and HFB<sub>1</sub> levels on positive samples. The samples were extracted according to the method described by Dombrink-Kurtzman and Dvorak (1999) with slight modifications: 1) The sample and MeOH:EDTA 0.01 M solution were mixed for 3 min and centrifugated at 6000 x G during 10 min, 2) EDTA 0.01 M was used, and 3) HCl 1.0 M to adjust pH.

Following derivatization with o-phthaldehyde (OPA) according to Sydenham *et al.* (1996), the extract was analyzed using a reversed-phase HPLC/fluorescence detection system. Methanol/0.1M sodium monohydrated phosphate (70:30) solution adjusted to pH 3.3 with orthophosphoric acid was used as mobile phase, at flow rate of 1.0 ml/min. The peak areas were measured to quantify FB<sub>1</sub> and HFB<sub>1</sub>. The limit of detection of the analytical method was 0.5 µg/g for FB<sub>1</sub> and HFB<sub>1</sub>. Recoveries were determined by adding 5 ng/g FB<sub>1</sub> to 25 g of cornmeal. Average recoveries for FB<sub>1</sub> and HFB<sub>1</sub> were 107 and 81.3 %, respectively.

To determine the significance of the differences in concentrations FB<sub>1</sub> and HFB<sub>1</sub> according to geographical section of Hermosillo, origin of the sample, and kind of store from which the sample was collected, a statistical test of hypothesis for two means was performed.

## RESULTS AND DISCUSSION

Fumonisin B<sub>1</sub> (FB<sub>1</sub>) and hydrolized fumonisin B<sub>1</sub> (HFB<sub>1</sub>) were found in nixtamalized cornmeal from all the studied geographical regions of Hermosillo, Sonora. There was not significative difference in the levels of FB<sub>1</sub> and HFB<sub>1</sub> in the samples collected among these regions. FB<sub>1</sub> and HFB<sub>1</sub> were found in nixtamalized cornmeal in both years. The mean of FB<sub>1</sub> and HFB<sub>1</sub> content in samples from each year was statistically different. There were significantly more FB<sub>1</sub> and HFB<sub>1</sub> in samples collected from 2002 sampling. During sampling from May to July 2002, 62.5% of samples obtained from supermarkets (20 of 32) contained FB<sub>1</sub> and HFB<sub>1</sub>, according to the Fumonitest<sup>®</sup> and HPLC method. The concentrations ranged from 1.05 to 22.88, and 1.46 to 13.71 µg/g, of FB<sub>1</sub> and HFB<sub>1</sub>, respectively, according to HPLC method. Table 1 shows the average levels of FB<sub>1</sub> and HFB<sub>1</sub> in samples collected in 2002. All means values are higher than FDA recommendations, 2 to 4 µg/g for humans.

**Table 1.** Average levels<sup>1</sup> of fumonisin B<sub>1</sub> and hydrolyzed fumonisin B<sub>1</sub> (µg/g) in cornmeal collected during 2002.

	FB <sub>1</sub>	HFB <sub>1</sub>
Grocery stores	12.32 ± 0.76	2.57 ± 0.86
Supermarkets	10.42 ± 4.62	3.61 ± 2.40
Retail outlets	13.19 ± 4.15	3.61 ± 2.40

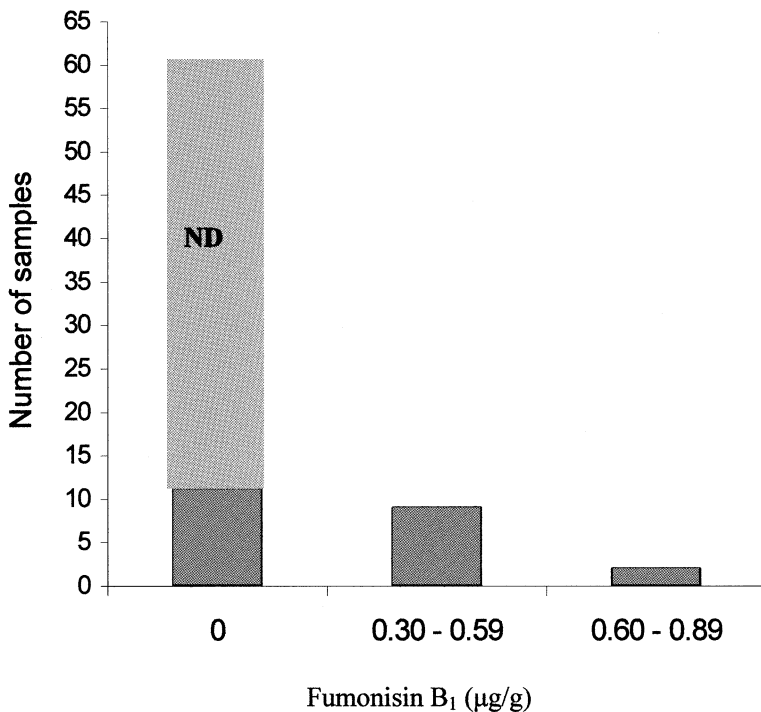
<sup>1</sup>Values are means of triplicates ± standard deviation.

Most of the samples from retail outlets (17/18) included in 2002 sampling, contained FB<sub>1</sub> and HFB<sub>1</sub> according to both methods. Levels from 3.84 to 22.57 and from 1.59 to 4.33 µg/g, of FB<sub>1</sub> and HFB<sub>1</sub>, respectively, were detected with HPLC method. All the samples from grocery stores (20/20) collected in the 2002 sampling contained FB<sub>1</sub> and HFB<sub>1</sub> according to both methods. Levels from 3.34 to 24.82 and from 1.22 to 3.67 µg/g, of FB<sub>1</sub> and HFB<sub>1</sub>, respectively, were detected with HPLC. In general, all FB<sub>1</sub> and HFB<sub>1</sub> levels detected were higher than those recommended by the FDA (U.S. FDA/CFSAN 2001).

Fumonisin B<sub>1</sub> content in samples collected during 2003 from retail outlets ranged from undetectable (< 0.3 µg/g) to 0.89 µg/g (Figure 1). FB<sub>1</sub> was not detected in fifty-two samples. Eight samples contained FB<sub>1</sub> in levels that could not be quantified with the method used. Eleven samples contained quantifiable levels of FB<sub>1</sub> ranging from 0.3 to 0.89 µg/g. In two samples the peak for HFB<sub>1</sub> was present but it could not be quantified. Probably, this finding may constitute an evidence that companies are concerned about fumonisins occurrence and attempts to maintain their levels down had been done. It was impossible to obtain information about where the corn was harvested, processing conditions, neither if corn was locally grown or was obtained from national or international suppliers.

Dombrink-Kurtzman and Dvorak (1999), reported mean levels of 0.79 µg/g of fumonisins in cornmeal dough and tortillas from Mexico. In a study by Reyes *et al.* (2001), FB<sub>1</sub> levels of 7.2, 6.3 and 4.3 µg/g were reported for corn, cornmeal dough and tortilla, respectively. FB<sub>1</sub> and HFB<sub>1</sub> mean levels obtained in our study (11.86 and 2.78 µg/g, respectively) showed higher presence of these toxins in corn products, which may suggest that levels of fumonisin were even higher than in their products.

Based on the amount of FB<sub>1</sub> and HFB<sub>1</sub> detected in commercial nixtamalized cornmeal in Sonora, fumonisin contamination may pose a risk for human health. However, an additional survey to analyze the fumonisin levels could improve the information to establish the needs for the risk management.



**Figure 1.** Number of nixtamalized cornmeal samples collected during 2003 contaminated with fumonisin B<sub>1</sub> and hydrolyzed fumonisin B<sub>1</sub> and levels detected. ND = Not detected.

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