Estimation of dietary intakes of fumonisins B1 and B2 from conventional and organic corn

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Abstract

The dietary intakes of fumonisins from 60 samples of conventional and organic corn were assessed. A 13.3% of the conventional corn samples contained fumonisin B1 and B2 at mean levels of 43 and 22 ng/g, respectively, while 10% of the organic corn samples contained fumonisins at somewhat lower levels of 35 ng/g (FB1) and 19 ng/g (FB2). Overall, the fumonisin levels in the corn samples were much lower than the maximum level of 2000 ng/g (as the sum of FB1 and FB2) proposed for unprocessed maize in a recent EU regulation. The fumonisins present in conventional and organic maize are estimated to contribute with very low percentages of 0.21% and 0.17%, respectively, to the level considered at risk for human health. Based on the data exposed in this paper, the farming system is probably not of decisive importance for the final contamination of agricultural products with these mycotoxins.

Keywords: Fumonisins; Organic corn; Daily intakes

1. Introduction

In the temperate climatic conditions prevailing in Europe, Fusarium fungi are important in the cereal food chain and can reduce crop yields and contaminate grain with mycotoxins (SCOOP, 2003). In Aragón (Spain), maize diseases caused by Fusarium have been reported since the early 1980’s (Palazón & Palazón, 1982). Members of the Gibberella fujikuroi complex (species Fusarium verticilloides, synonym of F. moniliforme) are generally regarded as the most important colonizers of cereal grains with the ability to produce the fumonisin mycotoxins (Ariño & Bullerman, 1994; Shephard, Thiel, Stockenström, & Sydenham, 1996; Soriano & Dragacci, 2004a). Fumonisins have been shown to be involved in leukoencephalomalacia in equine species, associated with pulmonary oedema in pigs, and implicated in oesophageal cancer in humans (Visconti, Marasas, Miller, & Riley, 1999). Fumonisin B1 is possibly carcinogenic to humans (group 2B) by the International Agency for Research on Cancer (IARC, 2002). The Scientific Committee for Food of the European Commission (SCF, 2003) has evaluated fumonisins and established a provisional maximum tolerable daily intake (PMTDI) of 2 μg/kg body weight/day for the total of fumonisins B1, B2 and B3, alone or in combination. Also, the European Union recently regulated fumonisins (as the sum of FB1 and FB2) in maize-based products and unprocessed maize, so that if no specific level is fixed before 1 October 2007, maximum levels from 200 to 2000 μg/kg will apply thereafter (Commission Regulation No 856/2005).

With the steady and fast growth of the European and US markets for organic foods there is a strong need for quality control, including safety evaluation of the products (Dimitri & Oberholtzer, 2006). Organic products of plant origin are grown without the aid of chemical-synthetic pesticides and largely without the use of readily soluble
mineral fertilizers within a diverse range of crop rotation and extensive soil tillage. Proponents of conventional farming claim that products from organic farming may pose a higher risk due to the presence of mycotoxins. This has not however been confirmed in a literature review carried out by the Food and Agriculture Organization of the United Nations (FAO, 2000). In a recent study, organic farming systems showed lower rates of Fusarium ear blight infection and lower mycotoxin contamination in winter wheat (Triticum aestivum) than conventional farming systems (Birzele, Meier, Hindorf, Krämer, & Dehne, 2002). Occasionally, higher mycotoxins concentrations have been measured in organic foods than in their conventional counterparts. For instance, in a recent comparison between conventional and organic French foods, organic wheat and barley were more highly contaminated by deoxynivalenol than conventional ones, but the differences were not significant (Malamaret, Parent-Massin, Hardy, & Verger, 2002). In summary, fungal attack and mycotoxin contamination in organically and conventionally grown produce is still an extremely controversial issue (Magkos, Arvaniti, & Zampelas, 2006).

Regarding fumonisins, the Rapid Alert System for Food and Feed of the European Union (RASSF, 2006) has reported several alert notifications in conventional and organic corn-based commodities in the last two years, which is a cause of concern for consumers and regulatory authorities. Also, a few research papers have compared the levels of fumonisins in conventional and organic corn-based foods (Cirillo, Ritiieni, Visone, & Cocchi, 2003; Paepens et al., 2005). From such results no generalizations can be drawn except to reinforce the demand for stringent controls and further studies including organic products. The principles for the prevention and reduction of fumonisins is based on the work by Bennett and Richard (1994). Briefly, 25 g of ground corn are extracted with 100 ml acetonitrile–water (1:1), filtered through Whatman No. 1 and the pH adjusted to 6–7. The cleanup was carried out with Multisep 211 Fum columns (Romer Labs, Union, MO) and the fumonisin-containing eluate was derivatized with NDA reagent (2,3-Naphthalene dicarboxaldehyde). Samples were injected into the LC-FLD system, a Kontron Model 322 pump, a Model 360 autosampler, and an SFM 25 fluorescence detector at 420 nm (excitation) and 500 nm (emission).

The analytical method was validated in-house with respect to precision and recovery. Blank corn samples were spiked with FB1 and FB2 at a level of 250 ng/g by pipetting an aliquot of standard solution onto the milled blank corn and allowing it to dry overnight (12 h). The average recoveries and relative standard deviations (RSDr, repeatability) obtained by the described method for FB1 and FB2 were 95.0% (RSD 4%, n = 3) and 85.0% (RSD 4%, n = 3), respectively. The performance characteristics for the analytes FB1 and FB2 were within the acceptable margins indicated in the Commission Directive 2005/38/EC (2005). The study of sensitivity indicated that the limit of quantification (LOQ) for FB1 and FB2 was 25 ng/g. The linear range was from LOQ to 25xLOQ.

2. Material and methods

2.1. Origin of corn samples

Thirty samples of corn grain were selected from farms using conventional cultivation methods, and another 30 samples were collected from organic farms. These corn samples from various harvesting years (2001 to 2003) were collected 1–4 weeks after harvest, October to November, at farms of Aragon (Spain). The objective was to obtain pairs of samples (conventional and organic) from farms at neighbouring sites. These sites differed in agricultural practices, fertilizers and pesticide usage, but environmental factors such as climate and soil conditions were as comparable as possible. In the conventional farms, the maize seeds had been treated with fungicides (approved treatments included himexazol 70% at 2 kg/Tm and maneb 40% at 3 L/Tm) and insecticides and herbicides were sprayed as needed during the vegetation period. Also, mineral fertilization of soil was used in the conventional farms. In the organic farms, by contrast, no pesticides and fungicides were used, and agronomic techniques included crop rotation with legumes and alfalfa plus fertilization with compost. In addition, tilling the soil between crop applications is indispensable in organic farming as a weed control technique.

To prepare representative samples, up to ten subsamples of 250 g were drawn from each lot of cereal, the subsamples were aggregated and a single composite sample of 0.50 kg was taken to the laboratory. The samples were stored at −21 °C until analyzed for mycotoxins.

2.2. Analysis of fumonisins in corn

The technique for extraction and determination of fumonisins is based on the work by Bennett and Richard (1994). Briefly, 25 g of ground corn are extracted with 100 ml acetonitrile–water (1:1), filtered through Whatman No. 1 and the pH adjusted to 6–7. The cleanup was carried out with Multisep 211 Fum columns (Romer Labs, Union, MO) and the fumonisin-containing eluate was derivatized with NDA reagent (2,3-Naphthalene dicarboxaldehyde). Samples were injected into the LC-FLD system, a Kontron Model 322 pump, a Model 360 autosampler, and an SFM 25 fluorescence detector at 420 nm (excitation) and 500 nm (emission).

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2.3. Statistical analyses

The results from fumonisin analyses obtained by the LC-FLD method were subjected to statistical analyses (ANOVA test) according to Sachs (1978). The incidences of samples containing fumonisins B1 and B2 (% positives) were expressed as the percentage of samples containing levels above LOQ (25 ng/g). The mean was calculated using one-half the LOQ for results lower than the LOQ. A probability value of 0.05 has been used to determine the statistical significance. Calculations were performed on StatView™ SE + Graphics (Abacus Concepts, Inc. 1988, Berkeley, CA) for Macintosh personal computers.
3. Results and discussion

Fumonisin B1 was detected above the LOQ (25 ng/g) in four samples of conventional corn (13.3%), and in three samples of organic corn (10.0%), whereas fumonisin B2 occurred in 10% out of 30 samples of conventional corn, and in 6.7% out of 30 samples of organic corn. The levels of fumonisins B1 and B2 in conventional corn samples amounted to 43 and 22 ng/g, respectively, while organic corn samples showed somewhat lower rates of contamination, B1 35 ng/g, and B2 19 ng/g (Table 1). Considering the positive samples, the levels of fumonisins in conventional corn ranged from 127 to 354 ng/g (FB1) and 93 to 120 ng/g (FB2), while in organic corn ranged from 147 to 359 ng/g (FB1) and 60 to 153 ng/g (FB2). Overall, the fumonisin levels in the corn samples were very low, much lower than the maximum level of 2000 µg/kg (as the sum of FB1 and FB2) proposed for unprocessed maize in a recent EU regulation (Commission Regulation No 856/2005).

Several surveillance studies of fumonisin B1 in Spanish market corn-based foods (Sanchis et al., 1994; Vellutti, Marin, Sanchis, & Ramos, 2001) and in corn-based feeds (Sanchis et al., 1995; Castella, Bragulat, & Cabañes, 1999) have been published, but organic corn was not included. Regarding market corn-based foods for human consumption (corn flour, sweet corn, corn snacks, corn flakes, popcorn and toasted corn), the occurrence of fumonisins was 16% out of 50 samples (Sanchis et al., 1994) and 23% out of 228 samples (Vellutti et al., 2001), with calculated mean concentrations of 23.1 and 31.4 ng/g, respectively (since authors originally reported results for positive samples, the total mean was re-calculated using one-half the LOQ for negative samples for a meaningful comparison). The prevalence is slightly higher but the fumonisin contents are somewhat lower than those reported in the present study, which may be probably due to different environmental conditions in the sampling years. Therefore, the adoption of a periodic analysis of the fumonisin levels in maize grain is recommended together with a control system of risk factors through the use of Good Agricultural Practices and Good Manufacturing Practices for the prevention and reduction of Fusarium toxins (European Commission, 2005).

In our study, the small differences in fumonisin concentrations between conventional and organic corn samples were not statistically significant (p > 0.05), indicating that the farming system is probably not of decisive importance for the final contamination of agricultural products with these mycotoxins. Similarly, the agricultural practice did not have any significant effect on the fumonisin concentrations found in conventional and organic corn samples from the Belgian market (Paepens et al., 2005). A study of the Italian market (Cirillo et al., 2003) reported that the highest median concentration of fumonisin B1 (345 ng/g) occurred in conventional maize-based foods (p > 0.05) while that of fumonisin B2 (210 ng/g) appeared in organic maize-based foods (p < 0.05). Other mycotoxins have been also subjected to comparison between conventional and organic production systems. Thus, deoxynivalenol concentrations of conventionally grown wheat were found to be significantly higher than in organic wheat grown in Thuringia, Germany (Döll, Valenta, Dänicke, & Flachowsky, 2002). In another study, the levels of ochratoxin A in organically grown rye were higher than in conventionally grown based on multi-year mean contents, though ochratoxin A levels in conventional and organic Danish wheat were very similar (Jorgensen & Jacobsen, 2002). Finally, ochratoxin A occurrence in cereal grains from conventional and organic Polish farms was almost the same (Czerwiecki, Czajkowska, & Witkowska-Gwiazdowska, 2002).

The stability and persistence of fumonisins through food processing has been reviewed by the European Mycotoxin Awareness Network (EMAN, 2006). Dry milling of maize results in distribution of fumonisins into different milled fractions so that concentrations in the bran, for example, may be higher than in the original whole maize grains. In experimental wet milling, fumonisin has been detected in steep water, gluten, fibre and germ, but not in the starch. Fumonisins are quite stable and are not destroyed by moderate heat (Castelo, Sumner, & Bullerman, 1998), however, an 80% reduction by heating at higher temperatures has been reported (Visconti et al., 1999). However, caution is required in assessing risk as it has been reported that breakdown products such as ‘hydrolysed fumonisin’ may be formed and these may be almost as toxic as the parent compound (EMAN, 2006).

Even though there are only 30 occurrence data for each type of corn, an estimation of dietary exposure to fumonisin B1 and B2 from conventional and organic corn was carried out (Table 2). In Spain, an average adult (70 kg) consumes around 1.6 kg of corn per year (4.4 g/day), according to the latest Food Supply and Commodity Balance Data (FAO, 2003). The mean fumonisin content of conventional corn measured in this study was 65.1 ng/g (as the sum of FB1 and FB2) proposed for unprocessed maize in a recent EU regulation (Commission Regulation No 856/2005).
Then, the estimated daily intake of fumonisins was 4.1 ng/kg body weight/day, which is considerably lower (0.21%) than the PMTDI of 2 μg/kg body weight/day established by the Scientific Committee for Food of the European Commission (SCF, 2003) (Table 2). Similarly, considering the mean fumonisin content of organic corn (54.1 ng/g), the estimated daily intake of 3.4 ng/kg body weight/day only amounted to 0.17% of the PMTDI. Since there is no data available on the consumption of organic corn, the same intake of 4.4 g/day applied for conventional maize has been used for this comparison. Furthermore, for exposure assessment, it would be important to bear in mind that fumonisins can be reduced from the first post-harvest treatment, from sieve process, to any process involving water, temperature, humidity and time.

Based on the limited data exposed in this paper, the fumonisins present in conventional and organic maize are estimated to contribute with very low percentages of 0.21% and 0.17%, respectively, to the level considered at risk for human health. These estimated intakes are in the lower range of those reported in the recent Scientific Cooperation Task on *Fusarium* toxins in food and assessment of dietary intake by the population of EU Member States (SCOOP, 2003). This report indicated that 46% and 42% of cereal-based food samples contained fumonisins B₁ and B₂, respectively, and the range of average dietary intakes of the sum of FB₁ and FB₂ for adults was between 0.1% and 1.4% (expressed as percent of PMTDI). However, higher intakes up to 22.3% PMTDI were noted for young children. It is well known that certain population groups are exposed to higher intakes of fumonisins, such as people with gluten intolerance, celiac or Dühring’s disease, vegetarians, infants and alcohol consumers (Soriano & Dragacci, 2004b). However, in a quantitative risk assessment for fumonisins in US corn, current dietary levels of fumonisins would not result in renal lesions even at upper levels of exposure in high maize eaters (Humphreys, Carrington, & Bolger, 2001).

In the neighbouring Portugal, fumonisins were detected in 45% of 31 samples of corn and corn-based foods at mean concentrations ranging from 64 to 995 ng/g (Lino, Silva, Pena, & Silva, 2006). According to data from Portuguese Food Balance, maize consumption in 2003 reached 27.9 g per person per day, and the authors calculated that the estimated daily intake of fumonisins was 0.14 μg/kg body weight (7% PMTDI).

The results of the present study indicated that, as far as fumonisins are concerned, there is no scientifically tenable evidence that the differences observed between conventional and organic foodstuffs would lead to any objectively measurable effect on consumer health.

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### References


