Occurrence of deoxynivalenol and nivalenol in Spanish corn-based food products

María-Ángeles Castillo*, Rosa Montes, Adriana Navarro, Ramón Segarra, Gonzalo Cuesta, and Enrique Hernández.

Departamento de Biotecnología, Escuela Técnica Superior de Ingenieros Agrónomos, Universidad Politécnica de Valencia, Camino de Vera, 14, 46022 Valencia, Spain.

Running title: Mycotoxins in Spanish corn-based foods

* Corresponding author: María-Ángeles Castillo, Departamento de Biotecnología, Escuela Técnica Superior de Ingenieros Agrónomos, Universidad Politécnica de Valencia, Camino de Vera 14, 46022- Valencia, Spain
Fax: 34-963877429. Tel.: + 34-963877423.

E-mail address: mcastill@btc.upv.es
Abstract

The aim of the present work was to evaluate the occurrence of trichothecenes toxins, deoxynivalenol (DON) and nivalenol (NIV), in samples of corn-based foods (breakfast cereals and snacks) consumed by the Spanish population. A total of 175 commercially available samples were randomly collected during 2005. Trichothecenes were determined by gas chromatography-electron capture detector. The estimated limit of quantification was 25.4 µg/kg for DON and 15.9 µg/kg for NIV. DON was detected in 22 of the 55 samples of breakfast cereals, in 13 of the 57 samples of baked corn snacks and in 12 of the 63 samples of fried corn snacks. NIV was detected in 6 samples of breakfast cereals and 1 sample of snacks. Based on total of samples, the median concentrations of DON and NIV found were 53.9 and 60.2 µg/kg, respectively. The influence of different factors, such as the presence of additional ingredients and the type of commercial brand on the toxin incidence and content levels were also studied. The values of both mycotoxin intake found in this study are lower than the proposed Tolerable Daily Intake for the respective toxin (1 and 0.7 µg/kg bw/day for DON and NIV, respectively).

Keywords: deoxynivalenol; nivalenol; trichothecenes; breakfast cereals; snacks
1. Introduction

Deoxynivalenol (DON) and nivalenol (NIV) are type-B trichothecene mycotoxins, that is, secondary metabolites produced by several fungal genera, most notably *Fusarium*, which is known to attack various cereals. Surveys have shown that DON occurs frequently in grains like wheat, barley, and maize, and that it is also the most common toxin, occurring concomitantly with 3-acetyldeoxynivalenol, 15-acetyldeoxynivalenol and NIV (Tanaka et al., 1990; Trucksess et al., 1995; Placinta et al., 1999). Trichothecenes cause a wide range of toxic effects in animal and humans such as feed refusal, vomiting, diarrhea, hemorrhage, anemia and immunosuppression (Hussein and Brasel, 2001).

As the incidence of *Fusarium* toxins has been reported for cereals on a global scale (Placinta et al., 1999; JEFCA 2000, 2001; Schollenberger et al., 2007), regular contamination can be expected for grain-based foods as the food processing of cereal crops does not completely eliminate mycotoxins (Scott, 1991; Hazel and Patel, 2004; Cetin and Bullerman, 2006). This has been confirmed for a variety of foodstuffs, including breakfast foods, snack foods, bread, pasta, etc. (FSA, 2003, 2005; Schollenberger et al., 2005a,b; Samar et al., 2007). As a result, humans are exposed to mycotoxins in their diet. It is clear that the presence of mycotoxins in the human diet, and especially in the diet of vulnerable populations like children, is a matter of concern. In recent years the EU’s Scientific Committee on Food (SCF) has evaluated the *Fusarium* toxins DON (SCF, 1999), NIV (SCF, 2000), and a group of trichothecenes (SCF, 2002). Likewise, the SCF established a tolerable daily intake (TDI) per kg of bodyweight and day of 1 and 0.7 µg/kg for DON and NIV, respectively. In 2005, the
European Commission (EC) set maximum levels of DON and zearalenone, which were applied from July 2006 (EC, 2005a). No limits are established for NIV.

Despite corn being one of the more susceptible substrates to this type of contamination, there is a little information available on the occurrence of trichothecenes in corn-derived foods in Spain (Cerveró et al., 2007). The purpose of this study was to examine the occurrence and concentration levels of DON and NIV in Spanish corn-based foods, specifically breakfast cereals and snacks. Although there are currently no regulatory limits for NIV, their co-occurrence with DON and its toxicity justifies their inclusion in our studies. Other aims of this study were to estimate the daily intake of DON and NIV, and to assess the real contribution of cereal-derived foods to the TDI proposed by the SCF in the Spanish population.

2. Material and methods

2.1. Reagents

The trichothecene standards DON and NIV, lindane (Riedel-de-Haën®) (internal standard) and derivatization reagent, Tri-Sil TBT (Supelco), a mixture of N-trimethylsilylimidazole–N,O-bis(trimethysilyl) acetamide–trimethylchlorosilane (3:3:2) were purchased from Sigma-Aldrich (Madrid, Spain). Deionized water was purified with a Millipore Milli-Q Plus system (Millipore, Billerica, MA, USA). Potassium dihydrogen phosphate (KH$_2$PO$_4$) and sodium hydroxide (NaOH), used to prepare phosphate buffer, were purchased from Panreac (Barcelona, Spain). All solvents (acetonitrile and hexane) were analytical grade and purchased from J.T. Baker (Deventer, Holland). Stock and working standards of DON and NIV were prepared by
appropriate dilution in acetonitrile to assess the linearity of method and spiked samples. All stock solutions were stored at – 20 ºC when not in use. The internal standard was dissolved in hexane at 1 mg/l for GC-ECD measurements. The Mycosep™ 227 columns were purchased from Romer Labs, Inc., USA.

The standards of DON and NIV were purchased from Sigma-Aldrich as pure mycotoxins. Krska et al. (2004) calculated the purity of commercially available DON from Sigma, resulting in 496%, and they concluded that these products can be considered sufficiently pure for routine analysis of these mycotoxins in food and feed. So we have used the standards as they were provided.

2.2. Samples

A total of 175 packaged samples of commercial corn foods were randomly collected during the first 8 months of 2005 from supermarkets and retail outlets located in the city of Valencia (Spain). A wide range of brands were covered to ensure that the survey was representative of the range of products available to consumers in Spain. At least 0.5 kg was collected for each commercial sample, which was finely ground for 3 min using an Osterizer mill (Oster Co., USA) and was stored at -20 ºC until the moment of the analysis, according to Cirillo et al. (2003a,b).

The following food items were collected: corn-based breakfast cereals (n = 55), baked corn snacks (n = 57) and fried corn snacks (n = 63). The sample collection was divided to convenience of discussion into different groups on the basis of possible ingredients and of the commercial brand type. The possible additional ingredients were chocolate, butter, honey, wheat, oat, and aromatic flavours such as cheese, jam and barbecue flavour.
2.3. Apparatus

Mycotoxin analysis was performed using an Agilent 6890N gas chromatograph, equipped with a $^{63}$Ni ECD (Agilent Technologies, Waldbrum, Germany). A fused-silica capillary column HP-5 [5% methyl phenyl siloxane column (30 m x 0.32 mm i.d., 0.25 μm film thickness, Agilent Technologies)] was used. The carrier gas was nitrogen at a flow of 1.8 ml/min. The temperatures of splitless injection port and ECD were 250 °C and 300 °C, respectively. The injection volume was 2 μl. The column temperature program, following an initial period of 80 °C for 1 min, was 30 °C/min to 140 °C, 5 °C/min to 280 °C and hold 31 min. Signals were processed by HP GC ChemStation software Version A.09.01 [1206] (Agilent Technologies, 1990-2001).

2.4. Extraction and clean-up

The mycotoxins were extracted and purified as described previously (Eskola et al., 2001) with few modifications. A 25 g subsample was extracted with 100 ml of acetonitrile:water (84:16 v/v) into a 500 ml Erlenmeyer flask. The flask was shaken with a rotary shaker at 170 rpm for 80 min at room temperature, and filtered through a filter paper (Whatman grade 2V) from Whatman (Maidstone, UK). The filtrate was defatted with n-hexane (2 x 20 ml). A total of 8 ml of defatted extract was purified through MycoSep 227 column, according to the instructions of the manufacturer. The purified extract that passed through the column (~ 4-5 ml) was collected, and the procedure was repeated by eluting the column with 8 ml of acetonitrile:water (84:16 v/v).
v/v). The combined fractions were evaporated to dryness under a gentle stream of nitrogen.

2.5. Derivatization

A 50 µl volume of the derivatization mixture (TMSI-BSA-TCMS, 3:3:2) was placed into a vial containing the dry residue. The mixture was allowed to react for 30 min at 80 ºC. After cooling, the derivatized sample was diluted to 480 µl with hexane and mixed thoroughly on vortex for 30 s. The hexane was then washed with 1 ml of phosphate buffer (0.1 M, pH 7.2) by mixing for about 30 s and, finally 20 µl of internal standard (1 mg/l) was added and shaken on vortex. The two layers were allowed to separate. The upper hexane layer with the trimethylsilyl derivatives was transferred to a vial for the gas chromatographic analysis.

2.6. Recovery assays, detection and quantification limits

For recovery studies, mycotoxin-free samples were artificially fortified at three levels of each mycotoxin (50, 500, and 1000 µg/kg of DON, and 300, 500, and 2000 µg/kg of NIV), as follows. Twenty-five grams of ground sample, previously defrosted, were soaked in 0.5 ml of acetonitrile solution containing a suitable amount of each mycotoxin. Then the sample was submitted to dissolvent evaporation for 2 h at room temperature. Finally, the spiked sample was extracted and analyzed by GC as described above. All tests were done in triplicate using different matrices.

The detection and quantification limits for both mycotoxins were assessed at a signal to noise ratio of 3:1 and of 6:1, respectively.
2.7. Statistical analyses

The results from mycotoxin analyses were subjected to statistical analysis using STAGRAPhICS PLUS software (Statistical Graphics Corp. version 5.1) and SPSS (version 14.0.1 for Windows, SPSS Inc., Chicago). The toxin content data were checked for normal distribution (Shapiro-Wilks test) and variance homogeneity (Cochran test). As the data did not fulfill these conditions, it was subsequently subjected to the Kruskal-Wallis test. When comparing the toxin levels in the different sample groups, the Mann-Whitney U-test applied to ranks was used to determine the statistical significance of the differences. The toxin incidence data were also analyzed in order to establish a significant association between different sample groups and the presence of mycotoxin (Fisher’s test). A probability value of 0.05 was used to determine the statistical significance.

3. Results and discussion

3.1. Analytical quality control

Mycotoxins were quantified by an internal calibration procedure. Calibration curves with internal standard were linear from 0.05 to 5 mg/l for DON and NIV (regression coefficients $r = 0.9993$ and $r = 0.9990$ for DON and NIV, respectively). The estimated limits of detection and quantification were 14.4 and 25.4 µg/kg, respectively, for DON, 9.6 and 15.9 µg/kg for NIV.
The recoveries of DON and NIV in different samples are summarized in Table 1. The recoveries of mycotoxins at the lower spiking level varied between 102.7% and 107.3% for DON and 93.0% to 113.2% for NIV. At the higher spiking level, the recoveries varied between 70.5% and 97.0% for DON and 77.9% and 94.1% for NIV. The lowest recoveries corresponded to fried corn snack samples. These differences could be attributable to the differences of matrix as described by Jestoi et al. (2004) and it is possible that specific extraction analysis protocols should be used for each combination of mycotoxin and food. The RSDr (%) of the mean recoveries for DON ranged from 2.2% at the higher spiking level to 12.8% at the lower spiking level, whereas these values for NIV varied between 0.3% and 19.5%, both at the lower spiking level. The recovery and RSDr obtained for DON are in line with the legislation levels for the DON determination methods (EC, 2005b) thus the method is acceptable according to EU criteria. Specific measures for NIV have not been considered by legislation because a certain degree of co-occurrence with DON is generally observed.

3.2. Occurrence of DON and NIV in corn-based foods

The results obtained from the analysis of DON and NIV in the samples are presented in Table 2 where the predominant mycotoxin for all analyzed samples was DON.

The incidence of DON and NIV in all the samples was 26.8% and 4%, respectively, while the median content of the positive samples was 53.9 and 60.2 µg/kg. The occurrence of DON varied between 25.5%, 27.6% and 46.8% for fried snacks, baked snacks and breakfast cereals, respectively. The incidence of DON was significantly different in breakfast cereals ($P = 0.026$). With regard to NIV occurrence, 3.4% of the
breakfast cereals and 0.6% of the baked snack samples were contaminated by this mycotoxin, while no fried snack samples was contaminated with NIV. The statistical analyses showed a significant dependence between the type of food and the presence of NIV at the 99% confidence level ($P = 0.006$).

The DON contamination levels varied from 30.1 to 121.1 µg/kg in breakfast cereals, from 36.4 to 131.7 µg/kg in baked snacks, and from 26.1 to 80.4 µg/kg in fried snacks. No significant differences were found among the median DON contents for all food items ($P > 0.05$). The highest level of DON was found among the baked snack samples (131.7 µg/kg) but no samples exceeded the legally established DON limit (500 µg/kg) (EC, 2005a).

The NIV contamination levels fluctuated between 51.1 and 106.5 µg/kg in breakfast cereals. Only one sample of baked snacks was contaminated with NIV (55.7 µg/kg). No significant differences were found among the median NIV contents for all positive food items ($P > 0.05$). The results show the natural co-occurrence of both toxins in two samples at concentrations of 64.9 µg/kg of DON and 55.7 µg/kg of NIV for a baked snack sample, and of 35.6 and 106.5 µg/kg, respectively, for a breakfast cereal sample.

Comparison between Spanish surveillance and literature data indicates that the incidence of DON contamination in Spanish corn-based foods is lower than in similar foods analyzed in Italy (64% of breakfast cereals and 93% of maize-based foodstuffs) (Cirillo et al., 2003a,b), Germany (67% of breakfast cereals) (Schollenberger et al., 2005b), and the United Kingdom (35% of breakfast cereals and 83.3% of snacks) (FSA, 2005). Differences between these data and the results of the present study may be attributed, among others, to a different origin of basing corn; it is well known that cereal infection with *Fusarium* and toxin production depend strongly on environmental conditions (damp climate, cool temperatures). *Fusarium* and trichothecene
contamination is more likely under the wet and cold weather conditions of Northern and Central European regions. This would explain the relative low percentage of DON/NIV positive samples. Moisture levels during harvest, transporting and storing the grain, and differences in food production methods (Larsen et al., 2004; Cavaliere et al., 2005) are also believed to be contributing factors. Conversely however, Spanish corn-based contamination is greater than that reported by Milanez et al. (2006) in Brazil, where only one out of 78 samples of corn-based products (cornflakes, corn grits) was found to present traces of DON and NIV. This may attribute either to a different origin of corn, as mentioned above, or to the high detection and quantification limits estimated by the authors (40 and 170 µg/kg for DON and 40 and 200 µg/kg for NIV).

With regard to the median mycotoxin levels detected in this study, they were similar to both those found in Italy by Cirillo et al. (2003a,b) and in Germany by Schollenberger et al. (2005b), but lower than those registered in samples surveyed in the United Kingdom (FSA, 2005). The results from a previous study on the occurrence of *Fusarium* toxins in 25 samples of corn-based foods marketed in Spain (Cerveró et al., 2007) showed a higher incidence of deoxynivalenol (68%) and mean content (91 µg/kg) of DON. This discrepancy between these data and present results may partly be attributed to the limited number of samples analyzed in the former study.

According to the information on the package, samples have been classified into two groups to associate mycotoxin incidence and content with the presence of additional ingredients: (1) samples with additional ingredients (92 samples), (2) samples without additional ingredients (83 samples). The percentage of positive samples and the content of toxins are listed in Table 3. The occurrence of DON and NIV was similar for both groups of samples, and the Fisher exact test showed no association between the variables considered ($P = 0.225$ for DON; $P = 0.182$ for NIV). The median content of
DON was significantly higher in samples without additional ingredients (63.7 µg/kg) ($P = 0.004$), while no significant differences were found for the median NIV content, although the highest levels of mycotoxins were detected in samples with additional ingredients. Moreover, fifty per cent of samples containing another type of cereal (wheat or oats) presented detectable levels of DON. The highest level of NIV was detected in one sample containing oats. It was clear from this work that contamination by DON and NIV is not only exclusive of corn, but also of oat and wheat contents as some authors have reported for these ingredients (Müller et al., 1997, 1998, 2001; Langseth and Rundberget 1999; Schollenberger et al., 2002, 2005a,b).

In the same way, samples were classified into two groups to compare mycotoxin occurrence and content according to the commercial brand: (1) private label (52 samples), and (2) company brand (123 samples). The results of the occurrence and levels of toxins are shown in Table 3. Approximately 37% of private label brand samples and 22.7% of company brand samples were positive for DON. With regard to NIV occurrence, 7.7% of the private brands and 2.4% of the company brands were contaminated with this mycotoxin. The statistical study showed a weak association between DON occurrence and type of commercial brand ($P = 0.047$) while no significant association was found for NIV. The statistical comparison between the mycotoxin level for both sample groups indicated no significant differences between the levels of both mycotoxins ($P = 0.259$). Although private labels have been seen as low-priced and low-quality products in a social context, the results obtained herein indicate that the quality of both types of commercial brands, in terms of mycotoxin content, is approximately the same. In recent years, a significant increase in private label brands has been observed, and companies have started using them to market higher quality
products. Nowadys in Europe, private label goods account for around 45% of the products sold in supermarkets.

In order to assess the public health risk of consuming DON and NIV in the Spanish population, the exposure of consumers to these toxins can be compared to safety guidelines, such as the Tolerable Daily Intake (TDI). Table 4 shows the calculated human DON and NIV intake from the mean mycotoxin levels found in the samples analyzed in this study. The relationship between each mycotoxin intake and the TDI levels proposed by the SCF of the European Union (1µg/kg bw/day for DON and 0.7 µg/kg bw/day – temporary TDI for NIV), has been expressed as a percentage. The mycotoxin intake values found in this study are less than those proposed in the TDI for the respective toxin, and they represented a fraction which does not exceed 7.1% for adults. However, these percentages increase to 12% for DON and 15.7% for NIV for risk groups, such as children. The results of a large-scale European study on the occurrence of *Fusarium* toxins and dietary intake in the European population (EC, 2003) demonstrated that while the dietary intakes of DON and NIV were often less than the TDI’s for the respective toxin for the entire population and adults, higher intakes values were observed for infants and children.

Overall, although the mycotoxin levels found in this survey were low, it is important, however, to bear in mind that cereals are only one of the many possible sources of these mycotoxins for humans, specially children or young people. Moreover, *Fusarium* contamination in grains may differ among the years of harvest (Langseth and Elen, 1997; Müller et al., 1997, 2001; Scott, 1997). Thus, different levels of toxin contamination can also be expected for cereal-based foods.

Although longer extensive studies including more *Fusarium* toxins are advisable, the results of the present study provide evidence of the presence of these mycotoxins in
the corn-based foods marketed in Spain. This study confirms the importance of continued surveillance of mycotoxin occurrence in cereal-processed foods in Spain, especially if they are mainly consumed by children and young people. A considerable contribution of these products to *Fusarium* toxin intake of the Spanish consumers may be assumed.

**Acknowledgment**

This work has been financed by the Spanish Comisión Interministerial de Ciencia y Tecnología (Project Ref. AGL2004-07549-C05-04) whose support is gratefully acknowledged.

**References**


Cirillo, T., Ritieni, A., Galvano, F., Amodio-Cocchieri, R., 2003a. Natural co-
occurrence of deoxynivalenol and fumonisins B1 and B2 in Italian marketed
foodstuffs. Food Additives and Contaminants 20, 566 – 571.
Cirillo, T., Ritieni, A., Visone, M., Amodio-Cocchieri, R., 2003b. Evaluation of
conventional and organic Italian foodstuffs for deoxynivalenol and fumonisins B1
A study of the suitability of gas chromatography-electron capture detection for the
analysis of deoxynivalenol in cereals. Mycotoxin Research 16, 73 – 90.
and assessment of dietary intake by the population of EU member states, Report on
Tasks for Scientific Cooperation (SCOOP) 3.2.10 European Commission, Brussels.
laying down the sampling methods of analysis for the official control of the levels
of Fusarium toxins in foodstuffs. Official Journal of European Union L 143, 18 –
26.
Survey Information Sheet 35/03.
Information Sheet 72/05.


http://www.inchem.org/documents/jecfa/jecmono/v47je01.htm


http://europa.eu.int/comm/food/fs/sc/scf/out44_en.pdf

http://europa.eu.int/comm/food/fs/sc/scf/out74_en.pdf


**Table 1**

Relation of analyzed samples according to different categories

<table>
<thead>
<tr>
<th></th>
<th>Commercial brand</th>
<th>Additional Ingredients&lt;sup&gt;a&lt;/sup&gt;</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Company brand</td>
<td>Private label</td>
<td>With</td>
</tr>
<tr>
<td>Breakfast cereals</td>
<td>25</td>
<td>30</td>
<td>30</td>
</tr>
<tr>
<td>Baked corn snacks</td>
<td>49</td>
<td>8</td>
<td>29</td>
</tr>
<tr>
<td>Fried corn snacks</td>
<td>49</td>
<td>14</td>
<td>33</td>
</tr>
</tbody>
</table>

<sup>a</sup>: Samples with or without another ingredients such as chocolate, butter, honey, wheat, oat, and aromatic flavours.
Table 2

Recoveries and relative standard deviations (%) for trichothecenes in spiked samples\textsuperscript{a}

<table>
<thead>
<tr>
<th>Spiking levels (μg/kg)</th>
<th>Breakfast cereals</th>
<th>Baked corn snacks</th>
<th>Fried corn snack</th>
</tr>
</thead>
<tbody>
<tr>
<td>1000</td>
<td>94.7 ± 10.7</td>
<td>97.0 ± 12.8</td>
<td>70.5 ± 7.3</td>
</tr>
<tr>
<td>DON</td>
<td>95.2 ± 4.2</td>
<td>100.2 ± 6.6</td>
<td>94.9 ± 4.9</td>
</tr>
<tr>
<td>50</td>
<td>107.3 ± 2.2</td>
<td>106.7 ± 8.8</td>
<td>102.7 ± 2.3</td>
</tr>
<tr>
<td>2000</td>
<td>94.1 ± 8.5</td>
<td>93.2 ± 18.7</td>
<td>77.9 ± 4.7</td>
</tr>
<tr>
<td>NIV</td>
<td>96.8 ± 4.1</td>
<td>105.4 ± 9.7</td>
<td>106.2 ± 9.7</td>
</tr>
<tr>
<td>300</td>
<td>93.0 ± 7.8</td>
<td>113.2 ± 19.5</td>
<td>101.6 ± 0.3</td>
</tr>
</tbody>
</table>

\textsuperscript{a}Number of samples = 3

DON, deoxynivalenol; NIV, nivalenol
Table 3
Natural occurrence of mycotoxins in corn-based foods

<table>
<thead>
<tr>
<th>Mycotoxin</th>
<th>Samples positive/total (%)</th>
<th>Range</th>
<th>Median</th>
</tr>
</thead>
<tbody>
<tr>
<td>Breakfast cereals</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>DON</td>
<td>22/55 (40.0)</td>
<td>30.1 – 121.1</td>
<td>44.5</td>
</tr>
<tr>
<td>NIV</td>
<td>6/55 (11.0)</td>
<td>51.1 – 106.5</td>
<td>67.8</td>
</tr>
<tr>
<td>Baked snacks</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>DON</td>
<td>13/57 (22.8)</td>
<td>36.4 – 131.7</td>
<td>62.5</td>
</tr>
<tr>
<td>NIV</td>
<td>1/57 (1.7)</td>
<td>55.7</td>
<td>55.7</td>
</tr>
<tr>
<td>Fried snacks</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>DON</td>
<td>12/63 (18.2)</td>
<td>26.1 – 80.4</td>
<td>55.5</td>
</tr>
<tr>
<td>NIV</td>
<td>0/63</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Total samples</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>DON</td>
<td>47/175 (26.8)</td>
<td>26.1 – 131.7</td>
<td>53.9</td>
</tr>
<tr>
<td>NIV</td>
<td>7/175 (4.0)</td>
<td>51.1 – 106.5</td>
<td>60.2</td>
</tr>
</tbody>
</table>
Table 4

Natural occurrence of mycotoxins in corn-based foods according to their composition

<table>
<thead>
<tr>
<th>Mycotoxin</th>
<th>Samples positive/total (%)</th>
<th>Range</th>
<th>Median</th>
</tr>
</thead>
<tbody>
<tr>
<td>With additional ingredients</td>
<td>DON</td>
<td>22/92 (23.9)</td>
<td>26.4 – 131.7</td>
</tr>
<tr>
<td></td>
<td>NIV</td>
<td>2/92 (2.2)</td>
<td>51.0 – 106.5</td>
</tr>
<tr>
<td>Without additional ingredients</td>
<td>DON</td>
<td>25/83 (30.1)</td>
<td>34.8 – 105.7</td>
</tr>
<tr>
<td></td>
<td>NIV</td>
<td>5/83 (6.0)</td>
<td>53.9 – 90.2</td>
</tr>
</tbody>
</table>

DON, deoxynivalenol; NIV, nivalenol
Table 5

Natural occurrence of mycotoxins in corn-based foods according to the type of commercial brand

<table>
<thead>
<tr>
<th>Mycotoxin</th>
<th>Samples positive/total (%)</th>
<th>Range</th>
<th>Median</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Company brand</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>DON</td>
<td>28/123 (22.7)</td>
<td>26.1 – 131.7</td>
<td>59.7</td>
</tr>
<tr>
<td>NIV</td>
<td>3/123 (2.4)</td>
<td>53.9 – 106.5</td>
<td>55.7</td>
</tr>
<tr>
<td><strong>Private label brand</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>DON</td>
<td>19/52 (36.5)</td>
<td>30.1 – 121.1</td>
<td>46.7</td>
</tr>
<tr>
<td>NIV</td>
<td>4/52 (7.7)</td>
<td>51.0 – 90.2</td>
<td>67.8</td>
</tr>
</tbody>
</table>

DON, deoxynivalenol; NIV, nivalenol
Table 6

Estimated DON and NIV daily intake (μg/kg bw/day) and the percentage that it represents of the proposed tolerable daily intake (TDI) by the Scientific Committee on Food of European Union (SCF)

<table>
<thead>
<tr>
<th></th>
<th>DON</th>
<th></th>
<th>NIV</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean level (μg/kg)</td>
<td>Intake (μg/kg bw/day)</td>
<td>SCF (%)</td>
<td>Mean level (μg/kg)</td>
</tr>
<tr>
<td>Breakfast cereals a Adults c</td>
<td>53.3</td>
<td>0.03</td>
<td>3.0</td>
<td>72.8</td>
</tr>
<tr>
<td>Breakfast cereals a Children d</td>
<td>0.06</td>
<td>6.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Corn snacks b Adults</td>
<td>61.9</td>
<td>0.05</td>
<td>5.0</td>
<td>55.7</td>
</tr>
<tr>
<td>Corn snacks b Children</td>
<td>0.12</td>
<td>12.0</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

DON, deoxynivalenol; NIV, nivalenol

a Recommended intake indicated on the label of the package (30 g) is assumed.
b A mean weight by package of 50 g is assumed.
c A body weight (bw) of 60 kg is assumed for adults.
d A body weight (bw) of 25 kg is assumed for children.