

# **The New Zealand Mycotoxin Surveillance Programme**

## **Fumonisin in Maize-Based Products and Wine**

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**Cover Image:** Photomicrograph of *Fusarium verticillioides*.

**Credit:** Centers for Disease Control and Prevention(CDC)/ Dr. Libero Ajello

## Scientific Interpretative Summary

This SIS is prepared by New Zealand Food Safety (NZFS) risk assessors to provide context to the following report for NZFS risk managers and external readers

### The New Zealand Mycotoxin Surveillance Programme

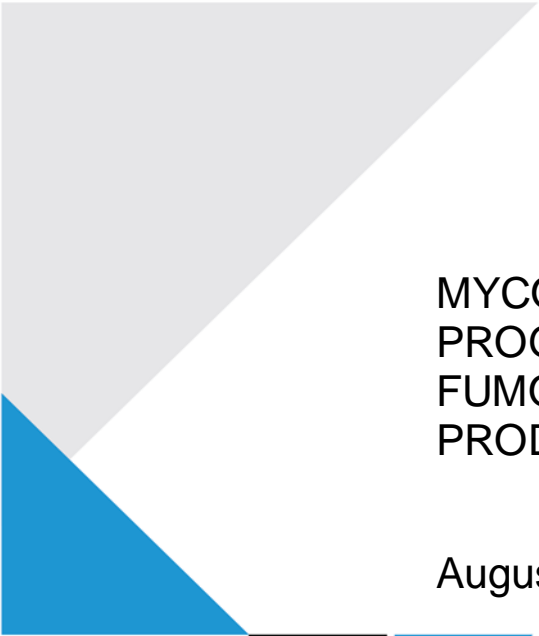
#### Fumonisin in Maize-Based Products and Wine

The risk of mycotoxins in New Zealand foods has been evaluated in NZFS's mycotoxin risk profiles. In the 2014 mycotoxin risk profile Fumonisin were listed as a high priority for which to generate New Zealand occurrence data. Fumonisin are reported to occur in maize-based foods and recent studies have also quantified levels in wines.

Fumonisin (FB<sub>1</sub>, FB<sub>2</sub> and FB<sub>3</sub>) were analysed in 80 maize-based foods and 20 wine samples. Any fumonisin result over 50 µg/kg was also tested for co-occurrence of aflatoxin. Analytical methodology for the survey is well detailed and the results can be viewed with strong confidence as being accurate for current occurrence levels.

Fumonisin were detected in 51 out of the 80 maize-based foods, being most prevalent in the maize-based foods with little secondary processing, such as maize-meal and polenta. Fumonisin were not detected in red wine. In addition none of the samples with greater than 50 µg/kg fumonisin had detectable aflatoxin. Ratios of the three fumonisin B forms are consistent with the literature and levels in general fall within, and in many cases to the lower end, of international ranges.

New Zealand origin and manufactured foods appear low in fumonisin mycotoxins by international standards.



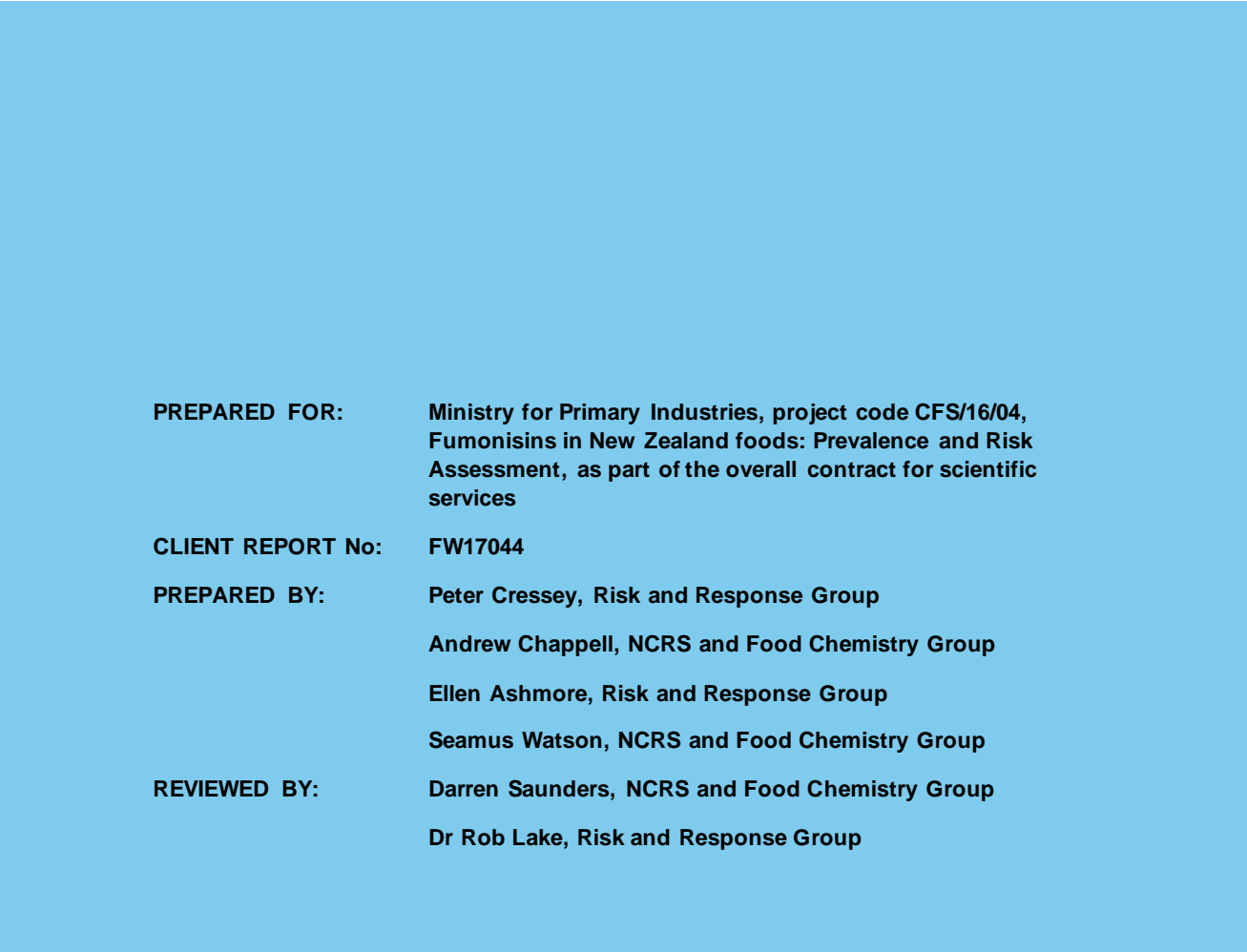
**MYCOTOXIN SURVEILLANCE  
PROGRAMME 2016-2017:  
FUMONISINS IN MAIZE-BASED  
PRODUCTS AND WINE**

August 2017



**≡/S/R**

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# EXECUTIVE SUMMARY

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The Mycotoxin Surveillance Programme (MSP) involves investigation of food safety issues associated with mycotoxins in the New Zealand food supply, as identified in risk profiling exercises carried out in 2005-2006 and 2013-2014. During 2016-2017, the MSP involved analysis of maize-based foods and red wine for the presence of fumonisin mycotoxins.

Fumonisin are mycotoxins produced predominantly by *Fusarium verticillioides* (*F. moniliforme*), *F. proliferatum* and *F. fujikuroi*. These fungal species are endemic in maize worldwide, but are rarely found in other crops. It has recently been discovered that *Aspergillus niger* strains are also capable of producing fumonisin mycotoxins. This discovery had led to the detection of fumonisins in foods not previously considered as vehicles for these toxins, such as fruits and fruit products.

Fumonisin were detected in approximately two-thirds of maize-based foods analysed in the current survey, but were not detected in any red wine samples ( $n = 20$ ). The highest prevalence of fumonisin contamination (100%) and the highest concentrations of total fumonisins (up to 1400  $\mu\text{g}/\text{kg}$ ) were detected in samples of cornmeal or polenta. This is consistent with the minimally processed nature of these foods.

The ratios of the three fumonisins determined ( $\text{FB}_1$ ,  $\text{FB}_2$  and  $\text{FB}_3$ ) were consistent with ratios reported previously by the Joint FAO/WHO Expert Committee on Food Additives. The relative concentrations of the three fumonisins were in the order  $\text{FB}_1 \gg \text{FB}_2 > \text{FB}_3$ , with  $\text{FB}_1$  accounting for about three-quarters of the total fumonisins detected.

All samples containing  $>50 \mu\text{g}/\text{kg}$  total fumonisins were also analysed for co-occurrence of aflatoxins. However, aflatoxins were not detected in any samples. It should be noted that co-exposure to fumonisins and aflatoxins from the diet is more likely to be of toxicological interest than co-occurrence in individual foods, as the toxicological effects of these toxins are not dependent on the particular food they contaminate.

Concentrations of fumonisins detected in foods available in New Zealand were within the range of results reported internationally or, for some food types, towards the lower end of the reported range. For some food types, New Zealand manufactured products showed clearly lower levels (prevalence and concentration) of fumonisin contamination than imported products. However, New Zealand manufactured maize-based products usually contain a mixture of locally-grown and imported maize.

While the fumonisin content of foods is not regulated under the Australia New Zealand Food Standards Code, fumonisin concentrations found in the current survey would have been compliant with international maximum limits, with the exception of one imported corn meal sample, containing  $>1400 \mu\text{g}/\text{kg}$  total fumonisins.

# 1. INTRODUCTION

---

The Mycotoxin Surveillance Programme (MSP) involves investigation of food safety issues associated with mycotoxins in the New Zealand food supply.

As with other activities of the Ministry for Primary Industries (MPI), activities in this area are directed on the basis of risk. The risk profile of mycotoxins in the New Zealand food supply and its update (Cressey and Thomson, 2006; Cressey, 2014a) are viewed as starting points for this process. The risk profile identified a number of issues to be investigated or clarified.

Efforts in previous years have focussed on determination of aflatoxins in a range of foods (Cressey and Jones, 2008; 2009; 2010), culminating in a dietary exposure assessment (Cressey, 2011); and analysis of ochratoxin A (OTA) in dried fruits and spices (Cressey and Jones, 2009) and cereal products, coffee, wine and beer (Cressey and Jones, 2011) and trichothecene mycotoxins in cereal products (Cressey *et al.*, 2014), culminating in exposure assessments for those two classes of mycotoxins (Cressey, 2014b).

These studies concluded assessment of the priority topics identified in the 2006 risk profile. Due to the shortage of New Zealand specific prevalence data, fumonisins were identified in the 2014 risk profile as the topic for the Mycotoxin Surveillance Programme in 2016-2017.

## 1.1 FUMONISINS

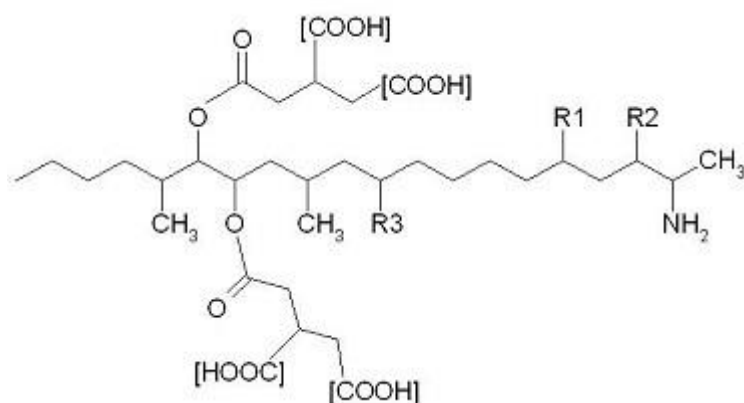
Fumonisin are mycotoxins produced predominantly by *Fusarium verticillioides* (*F. moniliforme*), *F. proliferatum* and *F. fujikuroi*, as well as some less common *Fusarium* species, for example *F. anthophilum*, *F. dlamini*, *F. napiforme* and *F. thapsinum* (Rheeder *et al.*, 2002; Suga *et al.*, 2014). These fungal species are endemic in maize worldwide, but are rarely found in other crops (Pitt and Tomaska, 2001). The fungi grow optimally at about 25°C and will grow at temperatures up to 32-37°C (Pitt and Tomaska, 2001).

It has recently been discovered that *Aspergillus niger* strains are also capable of producing fumonisin mycotoxins (Frisvad *et al.*, 2007; Scott, 2012). This discovery had led to the detection of fumonisins in foods not previously considered as vehicles for these toxins. *A. niger* produces predominantly FB<sub>2</sub> and FB<sub>4</sub> (Mogensen *et al.*, 2010).

### 1.1.1 Structure and nomenclature

Fumonisin consist of a 20 carbon aliphatic chain with 2 ester-linked hydrophilic polyol side chains. Structural details for FB<sub>1</sub> – FB<sub>4</sub> are shown in Figure 1.

**Figure 1. Structure of major fumonisins**



Fumonisin B<sub>1</sub>: R<sub>1</sub>= OH; R<sub>2</sub>= OH; R<sub>3</sub>= OH; Fumonisin B<sub>2</sub>: R<sub>1</sub>= H; R<sub>2</sub>= OH; R<sub>3</sub>= OH;  
Fumonisin B<sub>3</sub>: R<sub>1</sub>= OH; R<sub>2</sub>= OH; R<sub>3</sub>= H; Fumonisin B<sub>4</sub>: R<sub>1</sub>= H; R<sub>2</sub>= OH; R<sub>3</sub>= H

Fumonisin C<sub>1</sub>-C<sub>4</sub> are identical to the corresponding B fumonisin, except the aliphatic carbon chain does not have the terminal methyl (CH<sub>3</sub>) group on the right hand end, adjacent to R<sub>2</sub> and the amino (NH<sub>2</sub>) group (Soriano and Dragacci, 2004). Concentrations of B and C series fumonisins in maize have been shown to be correlated, but with the C series fumonisin present at concentrations 5% or less of the corresponding B series fumonisin (Shephard *et al.*, 2011).

### 1.1.2 Occurrence

*F. verticillioides* and *F. proliferatum* are amongst the most common fungi associated with maize worldwide and can be recovered from most maize kernels, even when the kernels appear healthy (WHO, 2000). *F. verticillioides* is considered to be the major cause of *Fusarium* kernel rot in maize, a significant plant disease occurring in warm, dry weather (JECFA, 2001). *Fusarium* kernel rot, and associated fumonisin synthesis, is also strongly associated with insect damage of kernels, as this provides an entry point for the fungus (WHO, 2000). Thin kernel pericarp (greater susceptibility to insect injury), propensity to kernel splitting, and previous infection with other *Fusarium* species, such as *F. graminearum*, all increase the risk of *F. verticillioides* infection and fumonisin formation (WHO, 2000).

*F. verticillioides* is widespread in the tropics and humid temperate zones, but is uncommon in cooler temperate zones (Pitt and Hocking, 1997). Surveys of *Fusarium* species in New Zealand maize are supportive of this observation, as the fungus is only rarely isolated (Hussein *et al.*, 2002; Sayer, 1991; Sayer and Lauren, 1991). *F. proliferatum* has not been reported in New Zealand maize.

Although fumonisin-producing fungi appear to be relatively rare in New Zealand, FB<sub>1</sub> was reported in New Zealand pasture grasses, at concentrations in the range 1-9 mg/kg (Mirocha *et al.*, 1992). The pastures analysed were associated with an idiopathic<sup>1</sup> disease of wapiti (elk) and wapiti hybrids grazed on the pastures.

<sup>1</sup> Relating to or denoting any disease or condition which arises spontaneously or for which the cause is unknown

While the vast majority of data available relate to fumonisins in maize and foods derived from maize, other foods have occasionally been analysed for fumonisins. While fumonisins have been detected in an increasingly wide range of foods, the concentrations found are generally much lower than those found in maize and/or the food is not consumed in large quantities. Two points are worth noting:

- Although concentrations of fumonisins found in wheat are mostly much lower than those found in maize, high concentrations (>1000 µg/kg) have occasionally been reported; and
- The discovery that *Aspergillus niger* is able to produce fumonisins has led to detection of these toxins in a range of fruits and fruit-based foods, such as wine (Al-Taher *et al.*, 2013; Logrieco *et al.*, 2010; Mogensen *et al.*, 2010; Tamura *et al.*, 2012).

### 1.1.3 Co-occurrence of fumonisins and aflatoxins

Interactions between aflatoxin B<sub>1</sub> (AFB<sub>1</sub>), a compound with known genotoxic and hepatocarcinogenic properties, and fumonisins, which have the potential to induce regenerative cell proliferation in the liver, have been identified as being of concern (JECFA, 2012).

The evidence for toxicological consequences following co-exposure to aflatoxins and fumonisins was reviewed at the 83<sup>rd</sup> meeting of the Joint FAO/WHO Expert Committee on Food Additives (JECFA) (2017). It was concluded that, although evidence in laboratory animals has suggested an additive or synergistic effect of fumonisin and aflatoxin co-exposure in the development of preneoplastic lesions or hepatocellular carcinoma, currently no data are available on such effects in humans. Co-exposure to fumonisins and aflatoxins has been implicated in childhood stunting, however, two prospective epidemiological studies did not provide support for this hypothesis. JECFA concluded that there are few data available to support co-exposure as a contributing factor in human disease. However, the mechanistic concerns expressed at the 74<sup>th</sup> JECFA meeting were reiterated.



## 2. MATERIALS AND METHODS

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### 2.1 FOODS SAMPLED

#### 2.1.1 Cereals and cereal products

Available studies have consistently shown that cereals, particularly maize and maize products, are the major dietary sources of fumonisins. Most studies suggest that maize and maize products account for at least 70% of dietary fumonisin exposure. With respect to cereal consumption, New Zealand is predominantly a wheat-consuming culture, with an increasing amount of rice consumed.

Wheat consumed in New Zealand is either from domestic production or imported, predominantly from Australia. There is some evidence to suggest that fumonisin contamination of wheat products consumed in New Zealand is unlikely:

- *F. verticillioides* is widespread in the tropics and humid temperate zones, but is uncommon in cooler temperate zones (Pitt and Hocking, 1997). Surveys of *Fusarium* species in New Zealand maize and wheat are supportive of this observation, as the fungus has not been isolated (Hussein *et al.*, 2002; Sayer, 1991; Sayer and Lauren, 1991). *F. proliferatum* has very rarely been reported in New Zealand maize and has not been reported in New Zealand wheat.
- The 23rd Australian Total Diet Study included analysis of baked beans, breakfast cereals, meat pie and frozen sweet corn kernels for FB<sub>1</sub> and FB<sub>2</sub> (Food Standards Australia New Zealand, 2011). Fumonisins were not detected in any food sample analysed. However, the analytical method and the limit of detection (LOD) were not reported.

While this evidence is by no means compelling, it suggests that wheat should not be a primary focus of the current survey.

Based on food consumption information from the 2002 Children's National Nutrition Survey (Ministry of Health, 2003) and the 2008-2009 Adult Nutrition Survey (University of Otago and Ministry of Health, 2011) the following maize-based food were identified as the most frequently consumed in New Zealand:

- Maize-based breakfast cereals (e.g. cornflakes)
- 'Mexican-style' maize products (e.g. corn chips, tortillas)
- Extruded maize flour snacks
- Popcorn

In addition, it is proposed that two minimally processed maize products (maize meal and polenta) be included as 'worst-case' scenarios.

#### 2.1.2 Fruit products

Recent studies have reported detection of fumonisins, particularly FB<sub>2</sub> in dried fruit, including figs (Heperkan *et al.*, 2012; Karbancioglu-Güler and Heperkan, 2009; Kosoglu *et al.*, 2011), dried vine fruit (Knudsen *et al.*, 2010; Mikušová *et al.*, 2013; Perrone *et al.*, 2013; Varga *et al.*, 2010), and wine (Al-Taher *et al.*, 2013; Logrieco *et al.*, 2010; Mogensen *et al.*, 2010; Tamura *et al.*, 2012). Of these products, wine is of the greatest economic relevance to New Zealand. There is some evidence to suggest that FB<sub>2</sub> is more prevalent in red wines, than white wines.

### 2.1.3 Sample numbers

The project included analysis of 100 analytical samples. This was considered to be an appropriate number for consideration of fumonisins, given that New Zealanders are not major maize consumers. On this basis, the following sample disposition was adopted:

- Maize-based breakfast cereals 20 samples
- 'Mexican-style' maize products 20 samples
- Extruded maize flour snacks 15 samples
- Maize meal/polenta 15 samples
- Popcorn 10 samples
- Wine, red 20 samples

### 2.1.4 Sampling

All food samples were obtained from supermarkets or speciality food retail outlets in Christchurch during March and April 2017. The primary focus was on sampling as wide a range of brands as possible, rather than on weighting sampling to match market share.

Mycotoxins may be distributed non-uniformly in food commodities. While retail foods are likely to have been homogenised to some extent, purchasing aimed to collect a minimum of 400-500 g of sample.<sup>2</sup> For products that were sold in units weighing less than 400 g each, multiple retail units with the same batch number and use by/best before date were collected to make up an analytical sample of sufficient size.

Samples were stored at room temperature and were homogenised prior to analysis.

### 2.1.5 Fumonisins analysed

The majority of the toxicological data available for fumonisins relates to fumonisin B<sub>1</sub> (FB<sub>1</sub>). A Provisional Maximum Tolerable Daily Intake (PMTDI) has been derived by JECFA for FB<sub>1</sub>, FB<sub>2</sub> and FB<sub>3</sub>, 'singly or in combination' (JECFA, 2012). The sum of the concentrations of FB<sub>1</sub>, FB<sub>2</sub> and FB<sub>3</sub> is referred to as total fumonisins, with FB<sub>1</sub> usually constituting about 70% of total fumonisins. Analyses of fumonisins sometimes include FB<sub>4</sub> or the hydroxylated forms of the major fumonisins. However, to date, these fumonisin forms have not been included in toxicological assessments of fumonisins and no analytical standards are available.

There is also evidence that fumonisins may become bound to other macromolecules in the food matrix during even quite mild food processing (Dall'Asta *et al.*, 2009). It was suggested that the binding is associative, rather than covalent and fumonisins can be released by digestion. There has been no suggestion that the adducts formed have toxicological implications, but may lead to underestimation of the contamination level of commodities, particularly maize. To date, assessment of free and bound forms of fumonisins has not been included in food surveys.

On the basis of this information, the current study determined FB<sub>1</sub>, FB<sub>2</sub> and FB<sub>3</sub> in all samples.

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<sup>2</sup> <https://www.food.gov.uk/sites/default/files/multimedia/pdfs/mycotoxinsguidance.pdf> Accessed 31 July 2017

## 2.1.6 Aflatoxin analyses

Studies on the co-occurrence of fumonisins and aflatoxins in foods have usually detected co-occurrence in a relative small proportion of samples (Ali *et al.*, 1998; Cano-Sancho *et al.*, 2012b; García-Moraleja *et al.*, 2015; Kimanya *et al.*, 2014; Kpodo *et al.*, 2000; Magoha *et al.*, 2016; Park *et al.*, 2002; Sangare-Tigori *et al.*, 2006; Sun *et al.*, 2011; Vargas *et al.*, 2001; Wang *et al.*, 1995; Warth *et al.*, 2012). Aflatoxin concentrations in samples containing both classes of mycotoxins are usually 100-1000-fold lower than the concentration of fumonisins. Consequently, it was decided to only carry out aflatoxin analyses on samples containing >50 µg/kg total fumonisins.

## 2.1.7 Analytical methods

### *Fumonisin*s

Solid samples (25 g homogenised) were extracted with acetonitrile/water (50:50, 100 mL), filtered, pH adjusted (pH 6-9) and the extract cleaned-up using Romer Multisep®211 Fum fumonisin columns.<sup>3</sup> Wine samples (25 mL) were diluted to 100 mL with acetonitrile/water and cleaned-up as for extracts of solid samples. Separation and quantification of fumonisins by liquid chromatography-tandem mass spectrometry (LC-MS/MS) was carried out based on an Agilent application note (Agilent Technologies, 2010).

In addition, <sup>13</sup>C-analogues of FB<sub>1</sub>, FB<sub>2</sub> and FB<sub>3</sub> were added to all sample extracts to allow correction for matrix effects. It is worth noting that, while a number of studies have employed <sup>13</sup>C-analogues in fumonisin analyses (Bryla *et al.*, 2016; Gazzotti *et al.*, 2011; Liu *et al.*, 2015), use of the full range of <sup>13</sup>C-analogues is less common (Bryla *et al.*, 2016), with other studies only using the <sup>13</sup>C-analogue of FB<sub>1</sub>.

The mass spectrometer source was used in electrospray ionisation (ESI) mode, with a capillary voltage of 2500 V, corona current of 0 µA and charging voltage of 2000 V. The gas flow rate was 5 L/minute, with a gas temperature of 325°C, nebuliser pressure of 60 psi and a vapouriser temperature of 200°C.

Mass spectrometric parameters used in the current study are given in Table 1. The mass spectrometer was operated in positive ion mode, with a dwell time of 100 ms for all analytes.

Table 1. Mass spectrometric parameters used for the analysis of fumonisins

Compound	Precursor Ion (m/z)	Product Ion (m/z)	Resolution	Fragmentor voltage (V)	Collision Energy (V)
<sup>13</sup> C-FB <sub>1</sub>	756.5	374	Widest/Wide	160	40
<sup>13</sup> C-FB <sub>1</sub>	756.5	356	Widest/Wide	160	44
<sup>13</sup> C-FB <sub>2</sub> & <sup>13</sup> C-FB <sub>3</sub>	740.6	358	Wide/Unit	135	35
<sup>13</sup> C-FB <sub>2</sub> & <sup>13</sup> C-FB <sub>3</sub>	740.6	340	Wide/Unit	135	35
FB <sub>1</sub>	722	352	Widest/Wide	150	35
FB <sub>1</sub>	722	334	Widest/Wide	150	40
FB <sub>2</sub> & FB <sub>3</sub>	706	336	Widest/Wide	135	38
FB <sub>2</sub> & FB <sub>3</sub>	706	318	Widest/Wide	135	38

<sup>3</sup> <https://shop.romerlabs.com/en/MycoSep-MultiSep/MultiSep/MultiSep-211-Fum> Accessed 27 June 2017

Precursor and product ions are consistent with those used in other studies (Bryla *et al.*, 2016; Cendoya *et al.*, 2014; Gazzotti *et al.*, 2011; Liu *et al.*, 2015).

### Aflatoxins

Aflatoxins (B<sub>1</sub>, B<sub>2</sub>, G<sub>1</sub> and G<sub>2</sub>) were analysed by LC-MS/MS using a method based on Agilent application notes (Agilent Technologies, 2007; 2008; 2011).

The mass spectrometer source was used in multimode ionisation (MMI) mode, with a capillary voltage of 2500 V, corona current of 0  $\mu$ A and charging voltage of 2000 V. The gas flow rate was 6 L/minute, with a gas temperature of 300°C, nebuliser pressure of 60 psi and a vapouriser temperature of 200°C.

The mass spectrometer was operated in positive ion mode, with a dwell time of 200 ms for all analytes. Mass spectrometric parameters for aflatoxins are summarised in Table 2.

**Table 2. Mass spectrometric parameters used for the analysis of aflatoxins**

Compound	Precursor Ion (m/z)	Product Ion (m/z)	Resolution	Fragmentor voltage (V)	Collision Energy (V)
AB <sub>1</sub>	313	241	Wide/Wide	120	30
AB <sub>2</sub>	315	259	Wide/Wide	120	30
AG <sub>1</sub>	329	243	Wide/Wide	120	30
AG <sub>2</sub>	331	245	Wide/Wide	120	30

## 2.1.8 Analytical quality control

### Fumonisin

#### Accuracy – spike recovery

The performance of the analytical method was assessed by adding ('spiking') known amounts of fumonisin to samples covering all matrix types and then analysing the samples to determine what proportion of the added fumonisin is recovered by the analytical method. Table 3 summarises spike recoveries for matrices included in the current study.

**Table 3. Spike recovery for fumonisins from survey matrices**

Sample type	Spike concentration ( $\mu$ g/kg)	Recovery (%)		
		FB <sub>1</sub>	FB <sub>2</sub>	FB <sub>3</sub>
Maize-based breakfast cereals	15	105.7	64.8	88.7
Maize-based breakfast cereals	15	87.8	61.5	81.4
Maize-based breakfast cereals	30	93.7	71.1	79.9
Maize-based breakfast cereals	300	116.0	106.5	107.8
Mexican-style maize product	15	96.3	78.4	82.7
Mexican-style maize product	15	89.4	104.4	94.2
Extruded maize flour snacks	30	76.8	66.4	76.4
Maize meal/polenta	300	116.9	95.4	95.9
Maize meal/polenta	1000	110.8	90.3	84.5
Maize meal/polenta	300	79.3	78.2	77.7
Maize meal/polenta	1000	73.7	71.4	68.8
Popcorn	30	78.8	61.9	77.7
Wine, red	15	76.7	79.7	74.8
<b>Mean</b>		<b>92.5</b>	<b>79.2</b>	<b>83.9</b>

The European Commission has specified acceptable recovery ranges for regulatory analysis of some mycotoxins (European Commission, 2006b). For FB<sub>1</sub> and FB<sub>2</sub> at concentrations less

than 500 µg/kg acceptable recovery was defined as 60-120%, while at spike concentrations greater than 500 µg/kg the acceptable range is 70-110%. The spike recoveries reported in Table 3 are all within these acceptable ranges. It has been assumed that the European recovery acceptance ranges for FB<sub>1</sub> and FB<sub>2</sub> are applicable to FB<sub>3</sub>.

#### Accuracy – quality control materials

A quality control material for fumonisins in corn (low level) was included in analytical batches.<sup>4</sup> Table 4 summarises the expected concentrations of FB<sub>1</sub>, FB<sub>2</sub> and FB<sub>3</sub> in this material and the concentrations determined by ESR.

**Table 4. Summary of results of analysis of quality control material for fumonisins in corn quality control material**

<b>Fumonisin</b>	<b>Expected result (µg/kg)<sup>a</sup></b>	<b>Number of analyses</b>	<b>Determined, mean (range) µg/kg</b>	<b>Determined as proportion of expected (%)</b>
FB <sub>1</sub>	667 ± 78	6	443 (413-463)	66 (62-69)
FB <sub>2</sub>	156 ± 21	6	131 (119-151)	84 (76-97)
FB <sub>3</sub>	89 ± 22	6	68 (62-79)	76 (70-89)

<sup>a</sup> [https://www.romerlabs.com/fileadmin/user\\_upload/romerlabs/Documents/PDF\\_Files/Biopure\\_Products.pdf](https://www.romerlabs.com/fileadmin/user_upload/romerlabs/Documents/PDF_Files/Biopure_Products.pdf)

It should be noted that the quality control material is not a certified reference material and the expected concentrations are those found in multiple analyses by Romer’s own laboratories. On this basis the performance against the expected results were assessed using the European Commission criteria for acceptable recovery ranges for regulatory analysis (see above). Results for FB<sub>2</sub> and FB<sub>3</sub> were well within the acceptable range for values below 500 µg/kg (60-120% recovery). The results for FB<sub>1</sub> sit on the boundary in two respects; the expected concentration (667 µg/kg) is near the point where the European Commission criteria changes from 60-120% to 70-110%, with the determinations also intermediate between the respective lower limits. Given the good recoveries achieved for matrix spikes (Table 2), it was considered that the method used in the current study had been demonstrated to show an acceptable degree of accuracy.

#### Sensitivity - Limits of detection and quantification

The limit of detection (LOD) was calculated from the lowest concentration standard that was significantly different to the response baseline (United States Food and Drug Administration, 1996). This is defined as a signal where the signal to noise ratio (s/n) is greater than three. For the majority of analytical runs this resulted in detection limits for each fumonisin of 0.7-1.7 µg/kg. The limit of quantification (LOQ) was taken as three times the LOD and in most cases was in the range 2.0-5.0 µg/kg.

#### Precision – coefficient of variation

Coefficients of variation (CVs), based on a method standard deviation derived from duplicate analyses of naturally contaminated samples were determined for FB<sub>1</sub>, FB<sub>2</sub> and FB<sub>3</sub> (IANZ, 2004). The respective CVs were 8.5, 5.3 and 6.9%, respectively.

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<sup>4</sup>

[https://www.romerlabs.com/fileadmin/user\\_upload/romerlabs/Documents/PDF\\_Files/Biopure\\_Products.pdf](https://www.romerlabs.com/fileadmin/user_upload/romerlabs/Documents/PDF_Files/Biopure_Products.pdf) Accessed 12 July 2017

The European Commission has specified acceptable CV or relative standard deviation (RSD) ranges for regulatory analysis of some mycotoxins (European Commission, 2006b). For FB<sub>1</sub> and FB<sub>2</sub> at concentrations less than 500 µg/kg acceptable CVs for analysis in a single laboratory were defined as ≤30%, while at concentrations greater than 500 µg/kg the acceptable range is ≤20%. CVs determined in the current study are all within these acceptable ranges. It has been assumed that the European CV acceptance ranges for FB<sub>1</sub> and FB<sub>2</sub> are applicable to FB<sub>3</sub>.

### *Aflatoxins*

#### Accuracy – spike recovery

The European Commission has specified acceptable recovery ranges for regulatory analysis of some mycotoxins (European Commission, 2006b). For individual aflatoxins at concentrations greater than 10 µg/kg recommended recoveries were defined as 80-110%. The spike recoveries reported in Table 5 are mostly within these recommended ranges. The most obvious exception is AFG<sub>2</sub>, for which three of the five recoveries were above the recommended range. However, given that AFB<sub>1</sub> is the predominant aflatoxin detected in food, the spike recoveries were considered to be acceptable.

**Table 5. Spike recovery for aflatoxins from survey matrices**

Sample type	Spike concentration (µg/kg)	Recovery (%)			
		AFB <sub>1</sub>	AFB <sub>2</sub>	AFG <sub>1</sub>	AFG <sub>2</sub>
Mexican-style maize product	15	90	85	95	111
Extruded maize flour snacks	15	101	95	103	124
Maize meal/polenta	15	112	105	115	121
Popcorn	15	100	96	107	128
Popcorn	15	77	85	93	94
<b>Mean</b>		<b>96</b>	<b>93</b>	<b>103</b>	<b>116</b>

#### Sensitivity - Limits of detection and quantification

The LOD and LOQ were calculated using the signal to noise (s/n) ratio of low concentration standards (0.5-5.0 µg/kg) to determine a concentration equivalent to s/n = 3 (LOD) and s/n = 10 (LOQ) (United States Food and Drug Administration, 1996). The rounded LOD and LOQ for each aflatoxin are 0.2 and 0.6 µg/kg.

Defining LODs and LOQs for total aflatoxins creates a dilemma. If each aflatoxin has an LOD of 0.2 µg/kg, then the sum of four aflatoxins should be 0.8 µg/kg. However, in the calculation of total aflatoxins, analytical results below the LOD are usually assigned a zero concentration. On this basis, the LOD and LOQ for total aflatoxins will be the same as the LOD and LOQ for each of the individual aflatoxins.

#### Precision – coefficient of variation

Coefficients of variation (CVs) are usually calculated from duplicate analyses of naturally contaminated samples. However, in the current study none of the analysed samples contained detectable concentrations of aflatoxins. Therefore, there is no basis for calculating a study-specific CV. Previous aflatoxin studies under the Mycotoxin Surveillance Programme have reported CVs in the range 3.3-6.2% for individual aflatoxins (Cressey and Jones, 2009; 2010).

## 3. RESULTS AND DISCUSSION

### 3.1 SUMMARY OF RESULTS

Table 6 summarises the results for FB<sub>1</sub>, FB<sub>2</sub>, FB<sub>3</sub> and total fumonisins for the foods analysed in the current survey. Individual results are summarised in Appendix 1.

**Table 6. FB<sub>1</sub>, FB<sub>2</sub>, FB<sub>3</sub> and total fumonisin content of maize-based foods and red wine in New Zealand**

Food	N	N>LOD (percent)	Fumonisin concentration, mean (range), µg/kg <sup>a</sup>			
			FB <sub>1</sub>	FB <sub>2</sub>	FB <sub>3</sub>	Total
Maize-based breakfast cereals	20	5 (25)	5 (ND-71)	0.4 (ND-9)	0.6 (ND-12)	6 (ND-92)
'Mexican-style' maize products	20	17 (85)	113 (ND-588)	26 (ND-133)	15 (ND-92)	154 (ND-813)
Extruded maize flour snacks	15	9 (60)	28 (ND-187)	5 (ND-49)	3 (ND-22)	35 (ND-258)
Maize meal/polenta	15	15 (100)	245 (3-992)	55 (ND-285)	35 (ND-158)	335 (ND-1435)
Popcorn	10	6 (60)	32 (ND-214)	3 (ND-20)	2 (ND-7)	37 (ND-242)
<b>Total maize-based foods</b>	<b>80</b>	<b>51 (64)</b>	<b>85 (ND-992)</b>	<b>18 (ND-285)</b>	<b>11 (ND-158)</b>	<b>114 (ND-1435)</b>
Wine, red	20	0 (0)	0 (ND-ND)	0 (ND-ND)	0 (ND-ND)	0 (ND-ND)

N: number of samples      ND: Not detected

N>LOD: number of samples with detectable fumonisin concentrations

<sup>a</sup> Mean values are the means of all analysed samples, assigning results below the LOD a value of zero

As might be expected the highest mean fumonisin concentrations were detected in the least processed maize products; maize meal/polenta. The lowest estimates of prevalence of detectable fumonisins were found in food types that were either highly processed (extruded maize flour snacks) or contained significant amounts of non-maize ingredients (some maize-based breakfast cereals).

Fumonisin was detected in approximately two-thirds of maize-based foods.

Fumonisin was not detected in red wine samples. This finding is discussed in greater detail in section 3.2.6.

#### 3.1.1 Ratio of fumonisins

The three major fumonisins occur in foods in reasonably constant proportions (JECFA, 2012). JECFA analysed available data on fumonisins in maize and found mean contributions of FB<sub>1</sub>, FB<sub>2</sub> and FB<sub>3</sub> to total fumonisins of 72.7, 19.3 and 8.0%, respectively.

Table 7 includes a summary of the proportions of the three fumonisins in the maize-based food categories included in the current survey and across all food categories. Proportions were only calculated for samples in which all three fumonisins were detected.

**Table 7. Proportions of FB<sub>1</sub>, FB<sub>2</sub> and FB<sub>3</sub> in maize-based foods in New Zealand**

Food	Fumonisin proportion, mean (range), % <sup>a</sup>		
	FB <sub>1</sub>	FB <sub>2</sub>	FB <sub>3</sub>
Maize-based breakfast cereals	77.8	9.7	12.5
'Mexican-style' maize products	75.0 (68.8-82.7)	16.1 (11.8-20.5)	8.9 (5.4-11.3)
Extruded maize flour snacks	78.5 (72.6-83.7)	12.6 (8.1-19.0)	8.9 (8.2-9.8)
Maize meal/polenta	74.6 (69.1-79.3)	14.7 (7.8-19.9)	10.7 (6.3-13.4)
Popcorn	84.7 (81.3-88.5)	8.4 (6.7-10.2)	6.9 (3.1-9.0)
<b>Total</b>	<b>76.3 (68.8-88.5)</b>	<b>14.2 (6.7-20.5)</b>	<b>9.5 (3.1-13.4)</b>

<sup>a</sup> Proportions were only calculated for samples in which all three fumonisins were detected

The relative order of magnitude for the percent of each fumonisin is consistent with the JECFA data, the mean proportions determined by JECFA fall within the range of values found in this study.

### 3.1.2 Co-occurrence of fumonisins and aflatoxins

All samples in the current survey found to contain greater than 50 µg/kg total fumonisins were analysed for the presence of aflatoxins (B<sub>1</sub>, B<sub>2</sub>, G<sub>1</sub> and G<sub>2</sub>). Aflatoxins were not detected in any samples analysed.

While a number of studies have reported on the co-occurrence of fumonisins and aflatoxins in individual foods (Ali *et al.*, 1998; Cano-Sancho *et al.*, 2012b; Kpodo *et al.*, 2000; Park *et al.*, 2002; Sangare-Tigori *et al.*, 2006; Sun *et al.*, 2011; Vargas *et al.*, 2001; Wang *et al.*, 1995), a recent JECFA evaluation placed greater emphasis on co-exposure to fumonisins and aflatoxins (JECFA, 2017). Previous work suggested that New Zealanders have low dietary exposure to aflatoxins (Cressey and Reeve, 2013).

## 3.2 FUMONISINS IN INDIVIDUAL FOOD TYPES

As no previous surveys of fumonisins in foods available to New Zealand consumers have been carried out, discussions in the following sections will focus on comparing the results of the current survey to surveys of similar foods performed internationally.

Results of a small survey of New Zealand “corn meal” were cited in a review of worldwide data (Shephard *et al.*, 1996). The twelve samples analysed were actually whole kernel corn and fumonisins were not detected, although the LOD was 50 µg/kg; about 50 times higher than in the current study. More recently, 50 samples, mostly of imported corn (maize) products, have been analysed for fumonisins. Only one sample was found to contain FB<sub>1</sub> (D Lauren, personal communication, 1998). However, comparisons between these results and those of the current study should only be carried out with caution, due to the large differences in the sensitivity of the methods used.

Corn-based breakfast cereals and corn chips sold in New Zealand have previously been reported to contain mostly imported maize with lower levels of New Zealand product (D Lauren, personal communication, 1998). However, it is uncertain whether this remains the case.

### 3.2.1 Maize-based breakfast cereals

Fumonisins were only detected in 25% of maize-based breakfast cereals. Of the two types of breakfast cereals examined (corn flakes and muesli), fumonisins were only detected in corn flakes. However, this may be a simple dilution effect, as muesli contains a range of non-maize ingredients. All corn flake samples found to contain fumonisins were manufactured overseas; four from Australia and one from the USA.

Fumoinisin concentrations detected in corn flakes in the current study ( $n = 12$ ; mean, all samples = 10 µg/kg; mean, positive samples only = 24 µg/kg; maximum = 92 µg/kg) were near to bottom of the range seen in international studies (see Appendix 2). Very similar levels of fumoinisin contamination in corn flakes have been reported in Argentina (Solovey *et al.*, 1999), Japan (Aoyama *et al.*, 2010) and Spain (D’Arco *et al.*, 2009).

### 3.2.2 ‘Mexican-style’ maize products

Fumoinisins were detected in the majority (85%) of Mexican-style maize products analysed in the current survey. The only samples in this category that did not contain detectable concentrations of fumoinisins were two samples of corn chips manufactured in New Zealand from a mix of local and imported ingredients and one sample of corn chips, whose country of origin was not reported. There was no clear geographical trend in the fumoinisin content of Mexican-style maize products, although four of five products with total fumoinisin concentrations greater than 300 µg/kg were from Australia.



Concentrations of fumonisins in Mexican-style maize products determined in the current survey were within the range of concentrations found in international studies (see Appendix 2).

### 3.2.3 Extruded maize flour snacks

Fumonisin were detected in just over half of the extruded maize flour snacks analysed. There was a clear difference in the fumonisin content of domestically produced product ( $n = 10$ ; total fumonisins ND-11  $\mu\text{g}/\text{kg}$ ) and imported products ( $n = 5$ ; total fumonisins 18-258  $\mu\text{g}/\text{kg}$ ). All of the New Zealand products reported that manufacture was from a mixture of local and imported materials and it is not possible to say whether these results indicate a low fumonisin content in New Zealand-grown maize, sourcing of low-fumonisin overseas maize, or a mixture of both.

The fumonisin content of this product type observed in the current study is within the range of concentrations reported internationally (see Appendix 2).

### 3.2.4 Maize meal/polenta

Fumonisin were detected in all samples of maize meal and polenta analysed in the current survey. This is not surprising as these products are the least processed maize products included in the current survey and the concentrations in these products should be similar to concentrations in whole unprocessed maize kernels. Products labelled as being produced in New Zealand generally contained lower total fumonisin concentrations (range 5-71  $\mu\text{g}/\text{kg}$ ) than products from Italy (range 57-1062  $\mu\text{g}/\text{kg}$ ), Argentina (194  $\mu\text{g}/\text{kg}$ ), Australia (424  $\mu\text{g}/\text{kg}$ ) or the USA (1435  $\mu\text{g}/\text{kg}$ ). Labelling suggests that the New Zealand products represent a mixture of imported and locally-produced ingredients.

While some studies have reported substantially higher fumonisin concentrations in maize meal (Bittencourt *et al.*, 2005; Savi *et al.*, 2016), the concentrations found in the current survey are consistent with the majority of international surveys (see Appendix 2).

### 3.2.5 Popcorn

Fumonisin were detected in 60% of popcorn samples analysed. The highest total fumonisin concentration observed (242  $\mu\text{g}/\text{kg}$ ) was in a sample originating from the USA. However, only one of the 10 samples was from the USA and caution should be exercised in drawing any conclusions from this observation.

Popcorn samples appear to have a higher proportion of total fumonisins present in the form of FB<sub>1</sub> (mean = 84.7%) than other food types. Popping corn is a subspecies of maize, *Zea mays everta*, however, it is uncertain whether genetic differences in the maize may have contributed to differences in the fumonisin contamination profile.

Levels of fumonisin contamination in popcorn found in the current survey are generally lower than those reported in international studies (see Appendix 2).

### 3.2.6 Red wine

Fumonisin were not detected in any of the red wine samples analysed as part of the current survey, which included a mixture of New Zealand ( $n = 4$ ) and imported wine, predominantly from Australia ( $n = 15$ ). It should be noted that studies that have detected fumonisins in wine have usually detected them at concentration of 1  $\mu\text{g}/\text{kg}$  or less (Logrieco *et al.*, 2010; Tamura *et al.*, 2012). The LOD for wine in the current study was 1.6-1.7  $\mu\text{g}/\text{kg}$ . While concentrations of fumonisins greater than the LOD in the current study have been reported (Logrieco *et al.*, 2010; Mogensen *et al.*, 2010), such concentrations appear to be relatively uncommon. Logrieco *et al.* (2010) reported 1/51 Italian red wines containing greater than 1

µg/L of FB<sub>2</sub>, while a Japanese study detected only trace amounts (<1.0 µg/L) of FB<sub>1</sub> or FB<sub>2</sub> in 5/14 red wines (Tamura *et al.*, 2012).

### 3.3 REGULATORY LIMITS FOR FUMONISINS

The Joint Australia New Zealand Food Standards Code does not specify maximum limits (MLs) for fumonisins in any food.<sup>5</sup>

In 2003, worldwide regulations for mycotoxins were reviewed (Van Egmond and Jonker, 2004). The results of this review are summarised in Table 8. It should be noted that the limits reported here include a mixture of regulatory maximum permitted limits and guideline levels.

**Table 8. Regulatory limits for fumonisins in various countries (food regulations only)**

Country	Commodity description	Fumonisin	Regulatory limit (µg/kg)
Bulgaria	Maize and processed products thereof	FB <sub>1</sub> + FB <sub>2</sub>	1000
Cuba	Maize, rice	FB <sub>1</sub>	1000
France	Cereals and cereal products	FB <sub>1</sub>	1000 (Target) 3000 (Maximum)
Iran	Maize	FB <sub>1</sub> + FB <sub>2</sub>	1000
Singapore	Corn and corn products	FB <sub>1</sub>	Not given
Switzerland	Maize	FB <sub>1</sub> + FB <sub>2</sub>	1000
Taiwan	Maize products	FB <sub>1</sub>	Not stated
USA	Degermed, dry milled corn products	FB <sub>1</sub> + FB <sub>2</sub> + FB <sub>3</sub>	2000 (Guide)

*From van Egmond and Jonker (2003)*

In 2006, the European Commission established MLs for fumonisins (sum of FB<sub>1</sub> and FB<sub>2</sub> in µg/kg) in maize and maize products (Table 9) (European Commission, 2006a).

**Table 9. Regulatory limits for fumonisins in the European Union (2006a)**

Commodity description	Regulatory limit (µg/kg)
Unprocessed maize	2000
Maize flour, maize meal, maize grits, maize germ and refined maize oil	1000
Maize based foods for direct human consumption, excluding foods listed in other categories	400
Processed maize-based foods and baby foods for infants and young children	200

European MLs for fumonisins were amended in 2007 (Table 10) (European Commission, 2007).

**Table 10. Regulatory limits for fumonisins in the European Union (2007)**

Commodity description	Regulatory limit (µg/kg)
Unprocessed maize, with the exception of unprocessed maize intended to be processed by wet milling	4000
Maize intended for direct human consumption, maize-based foods for direct human consumption, with the exception of foodstuffs listed in other categories	1000
Maize-based breakfast cereals and maize-based snacks	800
Processed maize-based foods and baby foods for infants and young children	200
Milling fractions of maize with particle size > 500 micron falling within CN code 1103 13 or 1103 20 40 and other maize milling products with particle size > 500 micron not used for direct human consumption falling within CN code 1904 10 10	1400

<sup>5</sup> <https://www.legislation.gov.au/Details/F2016C00167> Accessed 12 April 2017

Commodity description	Regulatory limit ( $\mu\text{g}/\text{kg}$ )
Milling fractions of maize with particle size $\leq 500$ micron falling within CN code 1102 20 and other maize milling products with particle size $\leq 500$ micron not used for direct human consumption falling within CN code 1904 10 10	2000

CN = Combined Nomenclature

The Codex Committee on Contaminants in Foods (CCCF) proposed MLs for fumonisins in unprocessed maize of 5000  $\mu\text{g}/\text{kg}$  and 2000  $\mu\text{g}/\text{kg}$  for maize flour/meal (Codex Committee on Contaminants in Foods, 2013). However, delegations from some countries, where maize is a dietary staple, expressed reservations on these MLs. They expressed a view that the setting of limits to cover both countries where maize was a staple and countries where it was a minor dietary component was not possible.

CCCF amended the MLs at their 8<sup>th</sup> meeting, reaching a compromise position of MLs of 4000  $\mu\text{g}/\text{kg}$  for raw maize grain and 2000  $\mu\text{g}/\text{kg}$  for maize flour and maize meal (Codex Committee on Contaminants in Foods, 2014). These MLs were forwarded to the Committee for adoption and are now included in the General Standard (Codex Alimentarius Commission, 2016).

The desirability of country-specific MLs and the tension between food safety and food security in countries where maize is a staple have been discussed in a recent scientific paper (Shephard *et al.*, 2013).

### 3.3.1 Compliance of foods from the current survey with EU MLs

The EU has the strictest reported MLs for fumonisins, with the 2007 amendment specifying a ML of 1000  $\mu\text{g}/\text{kg}$  for maize-based foods for direct human consumption and 800  $\mu\text{g}/\text{kg}$  for maize-based breakfast cereals and maize-based snacks. In the EU, fumonisins are defined as the sum of FB<sub>1</sub> and FB<sub>2</sub>. Of the 80 maize-based foods analysed in the current survey only one sample of imported corn meal contained fumonisins, as defined in the EU, at a concentration greater than 1000  $\mu\text{g}/\text{kg}$  (1277  $\mu\text{g}/\text{kg}$ ).

## 4. CONCLUSIONS

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Fumonisin was detected in approximately two-thirds of maize-based foods analysed in the current survey, but was not detected in any red wine samples ( $n = 20$ ). The highest prevalence of fumonisin contamination (100%) and the highest concentrations of total fumonisins (up to 1400  $\mu\text{g}/\text{kg}$ ) were detected in samples of cornmeal or polenta. This is consistent with the minimally processed nature of these foods.

The ratios of the three fumonisins determined ( $\text{FB}_1$ ,  $\text{FB}_2$  and  $\text{FB}_3$ ) were consistent with ratios reported previously by JECFA. The relative concentrations of the three fumonisins were in the order  $\text{FB}_1 \gg \text{FB}_2 > \text{FB}_3$ , with  $\text{FB}_1$  accounting for about three-quarters of the total fumonisins detected.

All samples containing  $>50 \mu\text{g}/\text{kg}$  total fumonisins ( $n = 28$ ) were also analysed for co-occurrence of aflatoxins. However, aflatoxins were not detected in any samples. It should be noted that co-exposure to fumonisins and aflatoxins from the diet is more likely to be of toxicological interest than co-occurrence in individual foods, as the toxicological effects of these toxins are not dependent on the particular food they contaminate.

Concentrations of fumonisins detected in foods available in New Zealand were within the range of results reported internationally or, for some food types, towards the lower end of the reported range. For some food types, New Zealand manufactured products showed clearly lower levels (prevalence and concentration) of fumonisin contamination than imported products. However, New Zealand manufactured maize-based products usually contain a mixture of locally-grown and imported maize.

While the fumonisin content of foods is not regulated under the Australia New Zealand Food Standards Code, fumonisin concentrations found in the current survey would have been compliant with international maximum limits, with the exception of one imported corn meal sample, containing  $>1400 \mu\text{g}/\text{kg}$  total fumonisins.

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# APPENDIX 1 INDIVIDUAL RESULTS ON FUMONISINS IN MAIZE PRODUCTS AND RED WINE

Results for individual samples analysed in the current project are summarised in Table 11.

**Table 11. Fumonisin concentrations in individual samples and aflatoxin concentrations in selected samples**

Sample	Food Group	Product	Country of Origin	Fumonisin concentration ( $\mu\text{g/kg}$ ) <sup>a</sup>				Aflatoxin (total) concentration ( $\mu\text{g/kg}$ )
				FB <sub>1</sub>	FB <sub>2</sub>	FB <sub>3</sub>	Total <sup>b</sup>	
1	Maize-based breakfast cereals	Corn Flakes	Made in Australia from local and imported ingredients	71	9	12	92	ND
2	Maize-based breakfast cereals	Corn Flakes	Made in New Zealand from local and imported ingredients	ND	ND	ND	ND	
3	Maize-based breakfast cereals	Corn Flakes	Made in New Zealand from local and imported ingredients	ND	ND	ND	ND	
4	Maize-based breakfast cereals	Corn Flakes	Made in New Zealand from local and imported ingredients	ND	ND	ND	ND	
5	Maize-based breakfast cereals	Corn Flakes	New Zealand	ND	ND	ND	ND	
6	Maize-based breakfast cereals	Corn Flakes	Made in New Zealand from local and imported ingredients	ND	ND	ND	ND	
7	Maize-based breakfast cereals	Muesli	Made in New Zealand from local and imported ingredients	ND	ND	ND	ND	
8	Maize-based breakfast cereals	Muesli	Made in New Zealand from local and imported ingredients	ND	ND	ND	ND	
9	Maize-based breakfast cereals	Corn Flakes	Australia	8	ND	ND	8	
10	Maize-based breakfast cereals	Corn Flakes	Australia	12	ND	ND	12	
11	Maize-based breakfast cereals	Corn Flakes	Australia	(4)	ND	ND	(4)	
12	Maize-based breakfast cereals	Corn Flakes	Made in New Zealand from local and imported ingredients	ND	ND	ND	ND	
13	Maize-based breakfast cereals	Corn Flakes	USA	(4)	ND	ND	(4)	
14	Maize-based breakfast cereals	Muesli	Made in New Zealand from local and imported ingredients	ND	ND	ND	ND	
15	Maize-based breakfast cereals	Corn Flakes	Made in New Zealand from local and imported ingredients	ND	ND	ND	ND	

Sample	Food Group	Product	Country of Origin	Fumonisin concentration ( $\mu\text{g}/\text{kg}$ ) <sup>a</sup>				Aflatoxin (total) concentration ( $\mu\text{g}/\text{kg}$ )
				FB <sub>1</sub>	FB <sub>2</sub>	FB <sub>3</sub>	Total <sup>b</sup>	
16	Maize-based breakfast cereals	Muesli	Made in New Zealand from local and imported ingredients	ND	ND	ND	ND	
17	Maize-based breakfast cereals	Muesli	Made in New Zealand from local and imported ingredients	ND	ND	ND	ND	
18	Maize-based breakfast cereals	Cereal Flakes	Made in New Zealand from local and imported ingredients	ND	ND	ND	ND	
19	Maize-based breakfast cereals	Muesli	Made in New Zealand from local and imported ingredients	ND	ND	ND	ND	
20	Maize-based breakfast cereals	Muesli	Made in New Zealand from local and imported ingredients	ND	ND	ND	ND	
21	Corn-based Mexican style products	Corn chips	Not stated	ND	ND	ND	ND	
22	Corn-based Mexican style products	Corn chips	Made in New Zealand from local and imported ingredients	120	28	(12)	160	ND
23	Corn-based Mexican style products	Corn chips	Made in New Zealand from local and imported ingredients	131	25	18	174	ND
24	Corn-based Mexican style products	Corn chips	Australia	19	5	ND	24	
25	Corn-based Mexican style products	Corn chips	Made in New Zealand from local and imported ingredients	ND	ND	ND	ND	
26	Corn-based Mexican style products	Corn chips	Made in New Zealand from local and imported ingredients	ND	ND	ND	ND	
27	Corn-based Mexican style products	Tortilla chips	Made in Australia from local and imported ingredients	48	(10)	ND	61	ND
28	Corn-based Mexican style products	Tortilla chips	USA	13	(2)	(2)	17	
29	Corn-based Mexican style products	Corn mini wraps	Australia	300	89	47	435	ND
30	Corn-based Mexican style products	Tortillas	New Zealand	277	76	32	384	ND
31	Corn-based Mexican style products	Tortillas	New Zealand	49	8	6	64	ND
32	Corn-based Mexican style products	Tortillas	Made in Australia from local and imported ingredients	588	133	92	813	ND
33	Corn-based Mexican style products	Corn Tapas	Made in New Zealand from local and imported ingredients	135	29	20	183	ND
34	Corn-based Mexican style products	Corn chips	New Zealand	4	ND	ND	4	
35	Corn-based Mexican style products	Corn chips	New Zealand	6	ND	ND	6	
36	Corn-based Mexican style products	Corn chips	Malaysia	7	ND	ND	7	

Sample	Food Group	Product	Country of Origin	Fumonisin concentration ( $\mu\text{g}/\text{kg}$ ) <sup>a</sup>				Aflatoxin (total) concentration ( $\mu\text{g}/\text{kg}$ )
				FB <sub>1</sub>	FB <sub>2</sub>	FB <sub>3</sub>	Total <sup>b</sup>	
37	Corn-based Mexican style products	Tortilla chips	USA	16	(2)	ND	18	
38	Corn-based Mexican style products	Taco Shells	Made in Australia from local and imported ingredients	264	53	31	348	ND
39	Corn-based Mexican style products	Taco Shells	Made in Australia from local and imported ingredients	242	58	27	326	ND
40	Corn-based Mexican style products	Taco Shells	Spain	41	6	(3)	50	
41	Extruded maize flour snacks	Corn Crispbread	Made in Australia from local and imported ingredients	44	7	6	56	ND
42	Extruded maize flour snacks	Corn Crispbread	Australia	96	9	9	115	ND
43	Extruded maize flour snacks	Corn Crispbread	Made in Australia from local and imported ingredients	51	7	6	65	ND
44	Extruded maize flour snacks	Cereal chips	Made in New Zealand from local and imported ingredients	(2)	ND	ND	(2)	
45	Extruded maize flour snacks	Cereal chips	Greece	187	49	22	258	ND
46	Extruded maize flour snacks	Shaped product	Made in New Zealand from local and imported ingredients	11	ND	ND	11	
47	Extruded maize flour snacks	Shaped product	Made in New Zealand from local and imported ingredients	(1)	ND	ND	ND	
48	Extruded maize flour snacks	Shaped product	Made in New Zealand from local and imported ingredients	ND	ND	ND	ND	
49	Extruded maize flour snacks	Shaped product	Made in New Zealand from local and imported ingredients	ND	ND	ND	ND	
50	Extruded maize flour snacks	Shaped product	Made in New Zealand from local and imported ingredients	ND	ND	ND	ND	
51	Extruded maize flour snacks	Shaped product	Australia	18	ND	ND	18	
52	Extruded maize flour snacks	Shaped product	Made in New Zealand from local and imported ingredients	ND	ND	ND	ND	
53	Extruded maize flour snacks	Shaped product	Made in New Zealand from local and imported ingredients	ND	ND	ND	ND	
54	Extruded maize flour snacks	Shaped product	Made in New Zealand from local and imported ingredients	ND	ND	ND	ND	
55	Extruded maize flour snacks	Cereal chips	Made in New Zealand from local and imported ingredients	7	ND	ND	7	
56	Popcorn	Popcorn	USA	214	20	7	242	ND
57	Popcorn	Popcorn	Spain	ND	ND	ND	ND	
58	Popcorn	Popcorn	Produced in New Zealand from local and imported ingredients	ND	ND	ND	ND	

Sample	Food Group	Product	Country of Origin	Fumonisin concentration ( $\mu\text{g}/\text{kg}$ ) <sup>a</sup>				Aflatoxin (total) concentration ( $\mu\text{g}/\text{kg}$ )
				FB <sub>1</sub>	FB <sub>2</sub>	FB <sub>3</sub>	Total <sup>b</sup>	
59	Popcorn	Popcorn	New Zealand Made	ND	ND	ND	ND	
60	Popcorn	Popcorn	New Zealand	69	5	7	81	ND
61	Popcorn	Popcorn	New Zealand	14	ND	ND	14	
62	Popcorn	Popcorn	Hungary	5	ND	ND	5	
63	Popcorn	Popcorn	New Zealand	2	ND	ND	2	
64	Popcorn	Popcorn	Made in New Zealand from local and imported ingredients	22	(3)	(2)	28	
65	Popcorn	Popcorn	Made in New Zealand from local and imported ingredients	ND	ND	ND	ND	
66	Maize meal/polenta	Cornmeal Flour	New Zealand	5	ND	ND	5	
67	Maize meal/polenta	Polenta	Italy	133	17	19	169	ND
68	Maize meal/polenta	Polenta	Italy	257	53	44	355	ND
69	Maize meal/polenta	Commeal	Italy	225	28	31	284	ND
70	Maize meal/polenta	Polenta	Italy	45	(5)	8	57	ND
71	Maize meal/polenta	Cornmeal	Italy	436	85	68	589	ND
72	Maize meal/polenta	Corn Masa Mix	Packaged in New Zealand from imported ingredients	53	10	8	71	ND
73	Maize meal/polenta	Polenta	New Zealand	7	ND	ND	7	
74	Maize meal/polenta	Cornmeal	USA	992	285	158	1435	ND
75	Maize meal/polenta	Corn Flour	Australia	316	74	35	424	ND
76	Maize meal/polenta	Polenta	Argentina	146	35	12	194	ND
77	Maize meal/polenta	Polenta	Italy	779	182	101	1062	ND
78	Maize meal/polenta	Polenta	Italy	242	48	37	327	ND
79	Maize meal/polenta	Cornmeal flour	Made in New Zealand from a local ingredient	34	9	5	48	
80	Maize meal/polenta	Polenta	Not stated	(3)	ND	ND	(3)	
81	Red wine	Wine, Merlot	New Zealand	ND	ND	ND	ND	
82	Red wine	Wine, Shiraz	Australia	ND	ND	ND	ND	



Sample	Food Group	Product	Country of Origin	Fumonisin concentration ( $\mu\text{g}/\text{kg}$ ) <sup>a</sup>				Aflatoxin (total) concentration ( $\mu\text{g}/\text{kg}$ )
				FB <sub>1</sub>	FB <sub>2</sub>	FB <sub>3</sub>	Total <sup>b</sup>	
83	Red wine	Wine, Shiraz	Australia	ND	ND	ND	ND	
84	Red wine	Wine, Cabernet Sauvignon	Australia	ND	ND	ND	ND	
85	Red wine	Wine, Pinot Noir	Australia	ND	ND	ND	ND	
86	Red wine	Wine, Merlot	Australia	ND	ND	ND	ND	
87	Red wine	Wine, Shiraz	Australia	ND	ND	ND	ND	
88	Red wine	Wine, Pinot Noir	Australia	ND	ND	ND	ND	
89	Red wine	Wine, Cabernet Sauvignon	Australia	ND	ND	ND	ND	
90	Red wine	Wine, Cabernet Sauvignon	Australia	ND	ND	ND	ND	
91	Red wine	Wine, Merlot	Australia	ND	ND	ND	ND	
92	Red wine	Wine, Merlot	Australia	ND	ND	ND	ND	
93	Red wine	Wine, Shiraz	Australia	ND	ND	ND	ND	
94	Red wine	Wine, Cabernet Sauvignon	Australia	ND	ND	ND	ND	
95	Red wine	Wine, Cabernet Sauvignon	Australia	ND	ND	ND	ND	
96	Red wine	Wine, Shiraz	Australia	ND	ND	ND	ND	
97	Red wine	Wine, Pinot Noir	New Zealand	ND	ND	ND	ND	
98	Red wine	Wine, Merlot Cabernet Sauvignon	New Zealand	ND	ND	ND	ND	
99	Red wine	Wine, Pinot Noir	New Zealand	ND	ND	ND	ND	
100	Red wine	Wine, Pinot Noir	France	ND	ND	ND	ND	

ND: not detected

<sup>a</sup> Figures in parentheses were between the method LOD and LOQ and should be regarded as indicative. These indicative figures were used in the calculation of total fumonisins

<sup>b</sup> Total fumonisins were calculated as the sum of the concentrations of FB<sub>1</sub>, FB<sub>2</sub> and FB<sub>3</sub>. Concentrations below the LOD were considered to be zero for calculating total fumonisins. Due to rounding total fumonisins as they appear in this report may not always match the sum of the three individual fumonisins

# APPENDIX 2 OVERSEAS STUDIES ON FUMONISINS IN CEREAL PRODUCTS AND WINE

Table 12 summarised data from overseas studies containing information comparable to that in the current study. To this end, studies that determined fumonisins in raw, unprocessed maize have not been summarised here, as the current study only considered processed consumer retail foods.

**Table 12. Overseas studies on the fumonisin content of maize products and wine**

Country	Year	Number of samples, positive/ total (percent)	Results, mean (range) (µg/kg) <sup>a</sup>	Reference
<b>Cornflakes</b>				
Argentina	1997	FB <sub>1</sub> 16/17 (94) FB <sub>2</sub> 0/17 (0) FB <sub>3</sub> 0/17 (0)	11 (2-38) - -	(Solovey <i>et al.</i> , 1999)
Belgium	2003-2004	FB <sub>1</sub> 24/39 (62) FB <sub>2</sub> 25/39 (64) FB <sub>3</sub> 23/39 (59)	78 (<LOQ-292) 11 (<LOQ-26) 18 (<LOQ-49)	(De Smet <i>et al.</i> , 2009)
Brazil	2003-2005	FB <sub>1</sub> + FB <sub>2</sub> Federal District 8/20 (40) Total Brazil 20/35 (57)	127 (ND-906) 505	(Caldas and Silva, 2007)
Brazil	NS	3/6 (50)	170	(Bordin <i>et al.</i> , 2015)
Brazil	2007-2010	FB <sub>1</sub> 9/11 (82) FB <sub>2</sub> 6/11 (55)	341 (120-840) 87 (50-151)	(Martins <i>et al.</i> , 2012)
Brazil	2013	FB <sub>1</sub> 14/25 (56) FB <sub>2</sub> 8/25 (32)	78 (16-317) 85 (33-286)	(Savi <i>et al.</i> , 2016)
Poland	2013	9/19 (47)	Total (free) 85 (13-248) Total (free and bound) 204 (26-745)	(Bryla <i>et al.</i> , 2016)
China (Shandong province)	NS	FB <sub>1</sub> 14/14 (100) FB <sub>2</sub> 13/14 (93) FB <sub>3</sub> 13/14 (93)	104 (1-171) 14 (ND-26) 17 (ND-32)	(Li <i>et al.</i> , 2015)
Denmark	1996	FB <sub>1</sub> 6/10 (60) FB <sub>2</sub> 2/10 (20)	110 (5-1030) 25 (4-243)	(Petersen and Thorup, 2001)
France	2009-2012	Organic FB <sub>1</sub> 4/37 (11) FB <sub>1</sub> 4/37 (11) Conventional FB <sub>1</sub> 0/96 (0) FB <sub>2</sub> 0/96 (0)	(140-190) (130-170) - -	(Rubert <i>et al.</i> , 2013)
Germany	1999-2001	117/205 (57)	Total 37 (<5-600)	(Zimmer <i>et al.</i> , 2008)
Italy	2007	Organic 0/1 (0) Conventional 0/10 (0)	-	(D'Arco <i>et al.</i> , 2009)
Japan	2004-2007	FB <sub>1</sub> 15/81 (19) FB <sub>2</sub> 0/81 (0) FB <sub>3</sub> 0/81 (0)	25 (max = 59)	(Aoyama <i>et al.</i> , 2010)
Spain	2007	Organic 1/1 (100)  Conventional 2/11 (18)	FB <sub>1</sub> 3 FB <sub>2</sub> ND FB <sub>3</sub> ND FB <sub>1</sub> 18 (5-30) FB <sub>2</sub> 4 (2-9) FB <sub>3</sub> 4	(D'Arco <i>et al.</i> , 2009)
Spain (Catalonia)	2008-2009	FB <sub>1</sub> + FB <sub>2</sub> 28/72 (39)	79 (max = 140)	(Cano-Sancho <i>et al.</i> , 2012a)
Spain	2009-2012	FB <sub>1</sub> 10/85 (12) FB <sub>2</sub> 10/85 (12)	(190-205) (170-200)	(Rubert <i>et al.</i> , 2013)
USA	NS	NS	(ND-88)	(Castelo <i>et al.</i> , 1998)
<b>'Mexican-style' maize products</b>				
Canada	NS	Tortilla chips 2/2 (100)	FB <sub>1</sub> + FB <sub>2</sub> 315 (178-451)	(De Girolamo <i>et al.</i> , 2011)
Denmark	1996	Taco shells FB <sub>1</sub> 2/2 (100) FB <sub>2</sub> 2/2 (100)	16 (13-19) 7 (7-7)	(Petersen and Thorup, 2001)
Europe	NS	Tortilla chips 1/7 (14)	FB <sub>1</sub> + FB <sub>2</sub> 960	(De Girolamo <i>et al.</i> , 2011)

Country	Year	Number of samples, positive/ total (percent)	Results, mean (range) (µg/kg) <sup>a</sup>	Reference
Mexico	NS	Masa flour 17/18 (94)	FB <sub>1</sub> + FB <sub>2</sub> 937 (48-1800)	(De Girolamo <i>et al.</i> , 2011)
UK	NS	Tortilla, taco, enchilada 6/20 (30)	Total (10-31)	(MAFF, 1995)
USA	NS	Tortilla chips, n = 19	(ND-1565)	(Castelo <i>et al.</i> , 1998)
USA	NS	Tortillas 51/53 (96) Masa flour 8/8 (100)	187 (12-750) 262 (63-689)	(Stack, 1998)
USA	NS	Masa flour 7/7 (100) Tortilla chips 4/4 (100)	FB <sub>1</sub> + FB <sub>2</sub> 389 (191-762) FB <sub>1</sub> + FB <sub>2</sub> 82 (32-115)	(De Girolamo <i>et al.</i> , 2011)
<b>Popcorn</b>				
Brazil	2003-2005	Federal District 22/24 (92) Total Brazil 36/46 (78)	664 (ND-2100) 1180	(Caldas and Silva, 2007)
Brazil	NS	11/15 (73)	540	(Bordin <i>et al.</i> , 2015)
Brazil	2007-2010	FB <sub>1</sub> 15/17 (88) FB <sub>2</sub> 10/17 (59)	338 (89-1170) 108 (57-211)	(Martins <i>et al.</i> , 2012)
Brazil	2013	FB <sub>1</sub> 4/10 (40) FB <sub>2</sub> 1/10 (10)	86 (44-180) 35	(Savi <i>et al.</i> , 2016)
Denmark	1996	FB <sub>1</sub> 4/9 (44) FB <sub>2</sub> 1/9 (11)	54 (1-474) 59	(Petersen and Thorup, 2001)
Japan	2004-2007	FB <sub>1</sub> 49/57 (86) FB <sub>2</sub> 44/57 (77) FB <sub>3</sub> 40/57 (70)	68 (max = 354) 18 (max = 94) 12 (max = 64)	(Aoyama <i>et al.</i> , 2010)
UK	NS	Ready-made 0/9 (0) Popping corn 6/13 (46)	(ND) Total (14-784)	(MAFF, 1995)
USA	NS	NS	(ND)	(Castelo <i>et al.</i> , 1998)
<b>Maize snacks</b>				
Brazil	2003-2005	FB <sub>1</sub> + FB <sub>2</sub> Federal District 17/20 (85)	178 (20-555)	(Caldas and Silva, 2007)
Denmark	1996	FB <sub>1</sub> 6/10 (60) FB <sub>2</sub> 3/10 (30)	16 (2-65) 2 (4-8)	(Petersen and Thorup, 2001)
Poland	2013	9/14 (64)	Total (free) 1006 (13-3300) Total (free and bound) 1650 (27-7330)	(Bryla <i>et al.</i> , 2016)
Japan	2004-2007	FB <sub>1</sub> 41/50 (82) FB <sub>2</sub> 40/50 (80) FB <sub>3</sub> 36/50 (72)	113 (max = 1670) 32 (max = 600) 20 (max = 280)	(Aoyama <i>et al.</i> , 2010)
Spain (Catalonia)	2008-2009	FB <sub>1</sub> + FB <sub>2</sub> 44/72 (61)	119 (max = 476)	(Cano-Sancho <i>et al.</i> , 2012a)
UK	NS	31/40 (78)	(11-220)	(MAFF, 1995)
<b>Cornmeal/polenta</b>				
Argentina	1997	Cornmeal FB <sub>1</sub> 19/21 (90) FB <sub>2</sub> 12/21 (57) FB <sub>3</sub> 11/21 (52)	503 (60-2900) 133 (61-1090) 78 (18-1020)	(Solovey <i>et al.</i> , 1999)
Brazil	2000	Commeal FB <sub>1</sub> 30/30 (100) FB <sub>2</sub> 30/30 (100) Flour FB <sub>1</sub> 30/30 (100) FB <sub>2</sub> 30/30 (100)	5200 (1100-15300) 1000 (230-3900) 2110 (470-7200) 670 (120-1800)	(Bittencourt <i>et al.</i> , 2005)
Brazil	2003-2005	Commeal FB <sub>1</sub> + FB <sub>2</sub> Federal District 73/73 (100) Total Brazil 255/259 (98)	1610 (270-6170) 3320	(Caldas and Silva, 2007)
Brazil	NS	Cornmeal 13/18 (72) Polenta 3/7 (43)	420 210	(Bordin <i>et al.</i> , 2015)
Brazil	2007-2010	Cornmeal FB <sub>1</sub> 28/19 (97) FB <sub>2</sub> 19/29 (66)	494 (81-2320) 158 (45-333)	(Martins <i>et al.</i> , 2012)
Brazil	2013	Commeal FB <sub>1</sub> 15/15 (100) FB <sub>2</sub> 14/15 (93) Flour FB <sub>1</sub> 8/15 (53) FB <sub>2</sub> 3/15 (20)	1300 (75-5400) 650 (52-1480) 415 (15-1540) 223 (32-352)	(Savi <i>et al.</i> , 2016)
Denmark	1996	Polenta FB <sub>1</sub> 1/1 (100) FB <sub>2</sub> 1/1 (100)	84 22	(Petersen and Thorup, 2001)
Germany	1999-2001	46/62 (74)	Total 307 (<5-4800)	(Zimmer <i>et al.</i> , 2008)
Italy	2007	Flour Organic 1/1 (100)  Flour Conventional 2/3 (67)	FB <sub>1</sub> 81 FB <sub>2</sub> 53 FB <sub>3</sub> 24 FB <sub>1</sub> 199 (163-235) FB <sub>2</sub> 133 (79-187) FB <sub>3</sub> 63 (55-70)	(D'Arco <i>et al.</i> , 2009)
Poland	2013	Flour 12/20 (60)	430 (13-1700)	(Bryla <i>et al.</i> , 2016)
China	NS	Commeal FB <sub>1</sub> 47/47 (100) FB <sub>2</sub> 47/47 (100)	452 (15-5050) 96 (1-1350)	(Li <i>et al.</i> , 2015)

Country	Year	Number of samples, positive/ total (percent)	Results, mean (range) (µg/kg) <sup>a</sup>	Reference
		FB <sub>3</sub> 46/47 (98)	81 (ND-712)	
Spain	2007	Flour 5/7 (71)	FB <sub>1</sub> 18 (3-45) FB <sub>2</sub> 5 (2-9) FB <sub>3</sub> 4	(D'Arco <i>et al.</i> , 2009)
Tanzania	2011-2012	Flour FB <sub>1</sub> 20/67 (30) FB <sub>1</sub> + FB <sub>2</sub> 21/67 (31)	(53-970) (48-1220)	(Magoha <i>et al.</i> , 2016)
UK	NS	Polenta 16/20 (80)	Total (16-2120)	(MAFF, 1995)
USA	NS	Commeal NS	(<75-5920)	(Castelo <i>et al.</i> , 1998)
<b>Wine</b>				
Denmark	NS	FB <sub>2</sub> 18/77 (23)	(1-25)	(Mogensen <i>et al.</i> , 2010)
Italy	NS	Red wine FB <sub>2</sub> 9/51 (18)	(0.5-2.4)	(Logrieco <i>et al.</i> , 2010)
Japan	2010	Red wine FB <sub>1</sub> 4/14 (29) FB <sub>2</sub> 2/14 (14) FB <sub>3</sub> 1/14 (7) White wine FB <sub>1</sub> 2/13 (15) FB <sub>2</sub> 0/13 (0) FB <sub>3</sub> 0/13 (0)	(<1.0-<1.0) (<1.0-<1.0) <1.0 (<1.0-<1.0) - -	(Tamura <i>et al.</i> , 2012)

NS: Not stated ND: Not detected

<sup>a</sup> FB<sub>1</sub> unless otherwise stated. 'Total' refers to the sum of FB<sub>1</sub>, FB<sub>2</sub> and FB<sub>3</sub>



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