CODEX ALIMENTARIUS COMMISSION

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World Health Organization

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JOINT FAO/WHO FOOD STANDARDS PROGRAMME

CODEX COMMITTEE ON CONTAMINANTS IN FOODS

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DISCUSSION PAPER ON AFLATOXINS IN CEREALS

In order to assist the Committee on how to proceed further with aflatoxin contamination in cereals, Codex members and Observers are invited to consider the conclusions and recommendations on page 7.

BACKGROUND

1. On the 23rd Session of the Committee on Food Additives and Contaminants (CCFAC) (1991), a maximum level (ML) of 10 µg/kg total aflatoxins (B1 + B2 + G1 + G2) was proposed for all foods. However, as there was no consensus over the issue among the country members, the development of a ML for aflatoxins in foods was discontinued and the Committee decided to discuss the issue on a commodity basis.[1]

2. At the 6th Session of the Committee on Contaminants in Foods (CCCF) (2012), the Committee agreed to the development of a discussion paper on aflatoxins in cereals through an electronic Working Group led by Brazil and co-chaired by the United States of America for consideration and discussion at the next session with the view of identifying possible actions or new work on this issue. The Committee also agreed to initiate new work on the development of an annex for the management of aflatoxins and ochratoxin A in sorghum to the Code of Practice for the Prevention and Reduction of Mycotoxin Contamination in Cereals (CAC/RCP 51-2003).[2]

3. This paper does not include data on processed food.

INTRODUCTION

4. Aflatoxins (AFs) are considered the most important group of mycotoxins in the world's food supply and are produced in nature primarily by *Aspergillus flavus* and *Aspergillus parasiticus.* AFs B1, B2, G1 and G2 are the four major naturally produced AFs, B and G referring to the blue and green fluorescence colours produced under UV light (Pitt and Hocking, 2009).

5. *A. flavus* is often found in most food produced in tropical countries, having special affinity with maize, peanuts and cottonseed. Usually, *A. flavus* produces only B aflatoxins and yet is considered the main source of AFs. *A. parasiticus* produces both B and G aflatoxins and is commonly isolated from peanuts, being quite rare to find it in other foods (Frisvad et al., 2006). Optimum conditions for AFs production by these two species is 33°C and 0.99 a_w (Sanchis and Magan, 2004). AFs could be produced by fungi either before and/or after harvesting of cereals, being influenced by several environmental factors such as temperature, relative humidity, insect damage, drought and stress condition of the plants (Miraglia et al., 2009).

TOXICOLOGICAL ASPECTS

6. At its 49th Meeting (1998), the Joint FAO/WHO Committee on Food Additives (JECFA) evaluated toxicological data on AFs (B1, B2, G1 and G2) and evaluated the human dietary exposure to AFs (FAO/WHO, 1998). The JECFA reviewed a wide range of studies, in both animals and humans, and concluded that AFs are human liver carcinogens, AFB1 being the most potent carcinogen of them. No tolerable daily intake was proposed since these compounds are genotoxic carcinogens.

7. The risks arising from the exposure to AFs were evaluated through potency estimates for human liver cancer derived from epidemiological and toxicological studies. The potency of AFs was defined by the JECFA to be 30 times higher in carriers of hepatitis B virus ($HBsAg^+$; about 0.3 cancers/year/100000 individuals) than in non-carries of hepatitis B virus ($HBsAg^-$; about 0.01 cancers/year/100000 individuals). Thus, reduction of AFs intake in populations with a high prevalence of hepatitis B carriers will have a greater impact on reducing liver cancer rates than in populations with a low prevalence of carriers.

[1] ALINORM 92/12A, para. 118.
[2] REP12/CF, para. 175.

8. At its 64th meeting, the JECFA (FAO/WHO, 2005) decided that evaluations on compounds that are both genotoxic and carcinogenic, such as AFs, should be based on the estimation of Margins of Exposure (MOEs). The MOE is defined as the ratio between a toxicological threshold (such as the BMDL[3]) and the intake. MOE lower than 10000 may indicate a public health concern (EFSA, 2005).

METHODS OF ANALYSIS

9. There is a range of methods available to analyze AFs and any one of them should provide reliable and reproducible results. Methods typically used for AFs analysis are based on three main steps: extraction, clean up and detection (Brera et al., 2008). Usually, samples are extracted with a mixture of water and organic solvents such as acetonitrile, methanol or acetone (Reiter et al., 2009). Sample clean-up uses mostly multifunctional (Fu et al., 2008; Garrido et al., 2012) or immunoaffinity columns (Daniel et al., 2011; Mazaheri, 2009; Mohammadi et al., 2012).

10. Detection and quantification methods include thin layer chromatography (TLC) (Hussain et al., 2011; Moreno et al., 2009) and high performance liquid chromatography (HPLC) with fluorescence (Almeida et al., 2012; Bansal et al., 2011; Ghali et al., 2010) or mass spectrometer detectors (Martos et al., 2010; Oueslati et al., 2012; Soleimany et al., 2012). Analysis using fluorescence detector (FD) usually needs pre/post column derivatization to enhance the fluorescent intensity of AFB1 and AFG1, thereby increasing sensitivity (Bakirdere et al., 2012). Trifluoroacetic acid (TFA) is commonly used to derivatize aflatoxin extracts before injecting in the HPLC system (Giray et al., 2007; Shah et al., 2010), while the most used post-column derivatization treatments are electrochemical using Kobracell (Almeida et al., 2012; Reiter et al., 2010) and photochemical using PHRED (Lutfullah and Hussain, 2012; Rahmani et al., 2010).

11. The use of ultraviolet detectors (UV) is less common, but still found (Binder et al., 2007; Fu et al., 2008). The enzyme-linked immunosorbent assay, ELISA, is a practical alternative for the determination of aflatoxins and has been widely used (Aydin et al., 2011; Karami-Osboo et al., 2012; Sun et al., 2011). Limits of quantification of the methods vary considerably, depending on the aflatoxin analyzed and on the method chosen, ranging from 0.01 µg/kg (HPLC-FD) (Almeida et al., 2012) to 4.0 µg/kg (TLC) (Rocha et al., 2009). LC-MS/MS methods have LOQs ranging from 0.5 µg/kg (Soleimany et al., 2012) to 2.0 µg/kg (Oueslati et al., 2012).

AGRICULTURAL, TECHNOLOGICAL AND COMMERCIAL ASPECTS

12. Complete elimination of mycotoxins from the food supply is not possible. The Code of Practice for the Prevention and Reduction of Mycotoxin Contamination in Cereals (CAC/RCP 51-2003) was established in an attempt to control and manage mycotoxins contamination worldwide. The Code states the importance of the implementation of the good agricultural practices (GAP) and the good manufacturing practices (GMP) by producers, besides indicating the adoption of a complementary management system, the Hazard Analysis Critical Control Point (HACCP) principles.

13. The Code of Practice contains recommendations to reduce the presence of mycotoxins in cereals. The recommendations include maintaining the crop on a rotation schedule, removing old seed heads and stalks that may serve as substrates for the mycotoxin-producing fungi, assuring adequate soil pH and plant nutrition to avoid plant stress, growing seed varieties developed for resistance to fungi and insect and avoiding exposure to high temperature and drought stress. At pre-harvest, the main action should be to minimize insect damage and fungal infection using insecticides and fungicides, when applicable. It is very important to harvest the grain at low moisture content, avoiding mechanical damage to the grain and contact with soil. Drying and storage facilities should be clean, dry and free of insects. Grains should be dried as soon as possible (generally to less than 15% moisture) and cleaned to remove damaged kernels.

Additional approaches to prevent aflatoxin contamination in cereal grain

14. The use of atoxigenic isolates of *A. flavus* to reduce AF concentration in the crop through the suppression of AF producers, is being considered as a promising method for AF control (Abbas, et al., 2011; Atehnkeng, et al., 2008). Probst et al. (2011) isolated atoxigenic strains of *A. flavus* from corn produced in Kenya and tested the potential to reduce AF concentrations in maize kernels co-inoculated with highly toxigenic strains. They found a reduction of up to 80.0% in AF concentration. Another approach to biological control of AF contamination in corn was evaluated by Accinelli et al. (2012). The authors applied bioplastic granules inoculated with atoxigenic strains of *A. flavus* on the soil surface of corn crops, achieving a reduction of AF contamination by 59-92%.

15. Giorni et al. (2008) tested the potential of using modified atmospheres (25.0–75.0% CO_2) to control *A. flavus* development and aflatoxin B1 production on maize grain post-harvest. The populations of *A. flavus* were significantly lower with 25 and 75% CO_2 in the atmosphere and all treatments with CO_2 were able to reduce toxin production (57.0-98.0%).

OCCURRENCE IN FOOD

16. Worldwide occurrence of AFs in cereals such as corn, rice, sorghum and wheat was evaluated from published studies related to samples collected from 2000 to 2012 and a summary of the results obtained in each study is shown in Annex 1. Data on aflatoxins in these commodities were grouped by continent and were summarized in Table 1. The means of positive samples as well as global mean, for commodities and continents, were obtained through a weighted mean of the data taken from literature. As function of the heterogeneity of published data, a few assumptions had to be made in order to permit grouping the data found in the literature.

[3] Benchmark dose, lower confidence limit.

17. Corn represented 57.2% of samples analyzed around the world, while rice, sorghum and wheat contributed about 15.0% each. Of all samples analyzed in the studies (16490), 35.9% were positive for at least one aflatoxin. Sorghum had the highest incidence of positive samples (70.7%), followed by rice (53.7%), wheat (36.3%) and corn (23.2%). Samples of sorghum also had the highest mean aflatoxin level among the grains analyzed (122.6 µg/kg). Levels of aflatoxins on the data analyzed ranged from 0.002 to 48000 µg/kg, with the highest level found in a study that also analyzed samples during aflatoxicosis outbreaks in Kenya. The upper bound of the total mean of all samples analyzed was 23.4 µg/kg (Table 1).

18. Among the 2193 contaminated corn samples analyzed in the studies (Table 1), 34.5% were from Africa. Contaminated rice, sorghum and wheat samples came mostly from Asia (78.5, 82.4 and 82.3% of the positive samples, respectively). Samples from Asia had the highest incidence of positive samples for all cereals (Figure 1). The lowest incidence of contamination was found in American samples, with no positive wheat samples reported (Figure 1).

Table 1 – Worldwide occurrence of aflatoxins in cereals.

	N[a]	Positive/analyzed samples (%)	Positive samples (µg/kg)		Total mean (µg/kg)[b]
			Mean ± SD	Range	Lower[c]–Upper[d] bound
Corn	**36**	**2193/9431 (23.2)**	**23.8 ± 39.3**	**0.01–48000**	**5.5-6.5**
Africa	13	756/2219 (34.1)	15.9 ± 11.1	0.01–48000	5.4–5.8
Americas[e]	9	494/4666 (10.6)	27.3 ± 13.6	0.1-1393	2.9–4.5
Asia	11	645/1068 (60.3)	36.3 ± 71.4	0.1-888.3	21.9–22.2
Europe[f]	5	298/1478 (20.2)	20.4[g] ± 8.9	0.01-820	2.3–2.6
Rice[h]	**26**	**1471/2738 (53.7)**	**87.9 ± 34.8**	**0.002-371.9**	**47.2–47.4**
Africa	5	45/78 (57.7)	41.1 ± 35.4	1.5-371.9	23.7–23.8
Americas	6	200/589 (34.0)	4.0 ± 17.0	0.002–158.1	1.4–1.9
Asia	13	1155/1890 (61.1)	109.1 ± 43.8	0.01–308	66.7–66.7
Europe	2	71/181 (39.2)	8.9 ± 6.2	0.05–21.4	3.5–3.9
Sorghum	**11**	**1428/2019 (70.7)**	**122.6 ± 65.1**	**0.01–263.9**	**86.7–86.9**
Africa	8	248/393 (63.1)	81.4 ± 68.0	0.34–1164	51.4–51.6
Asia	2	1176/1616 (72.8)	131.7 ± 89.8	0.01–263.9	95.8–96.0
Europe	1	4/10 (40.0)	20.0	NR	8.0–8.3
Wheat	**15**	**836/2302 (36.3)**	**18.9 ± 41.8**	**0.1–643.5**	**6.8–8.1**
Africa	6	66/206 (32.0)	10.0 ± 6.6	0.21–37.4	3.2–3.6
Americas	1	0/40 (0.0)	-	-	ND–5.0
Asia	6	688/1711 (40.2)	14.5 ± 5.6	0.1–606	5.8–7.3
Europe	3	82/345 (23.8)	62.7 ± 103.3	10.4–643.5	14.9–15.3
Total	**64**	**5928/16490 (35.9)**	**62.8 ± 43.6**	**0.002-48000**	**22.6–23.4**

[a] Number of published studies found in the literature; [b] mean of all samples; [c] samples below LOD or LOQ were considered as zero; [d] samples below LOD or LOQ were considered as 0.5 LOD or 0.5 LOQ; [e] includes monitoring data from US FDA; [f] includes monitoring data collected by EFSA (2007); [g] mean of positive samples was not available in the EFSA report; [h] mostly rice collected on the market, but some studies may include rice samples with the husk.

Africa: includes samples from Algeria, Benin, Togo, Côte d'Ivore, Egypt, Kenya, Malawi, Morocco, Nigeria, Tanzania, Tunisia, Uganda and Zambia;

Americas: includes samples from Argentina, Brazil, Canada and USA;

Asia: includes samples from China, India, Iran, Japan, Korea, Malaysia, Pakistan, Qatar and Vietnam;

Europe: includes samples from Austria, Belgium, Cyprus, Czech Republic, Denmark, Estonia, Finland, France, Germany, Greece, Hungary, Ireland, Italy, Latvia, Luxembourg, Serbia, Slovakia, Slovenia, Spain, Sweden, United Kingdom and Turkey.

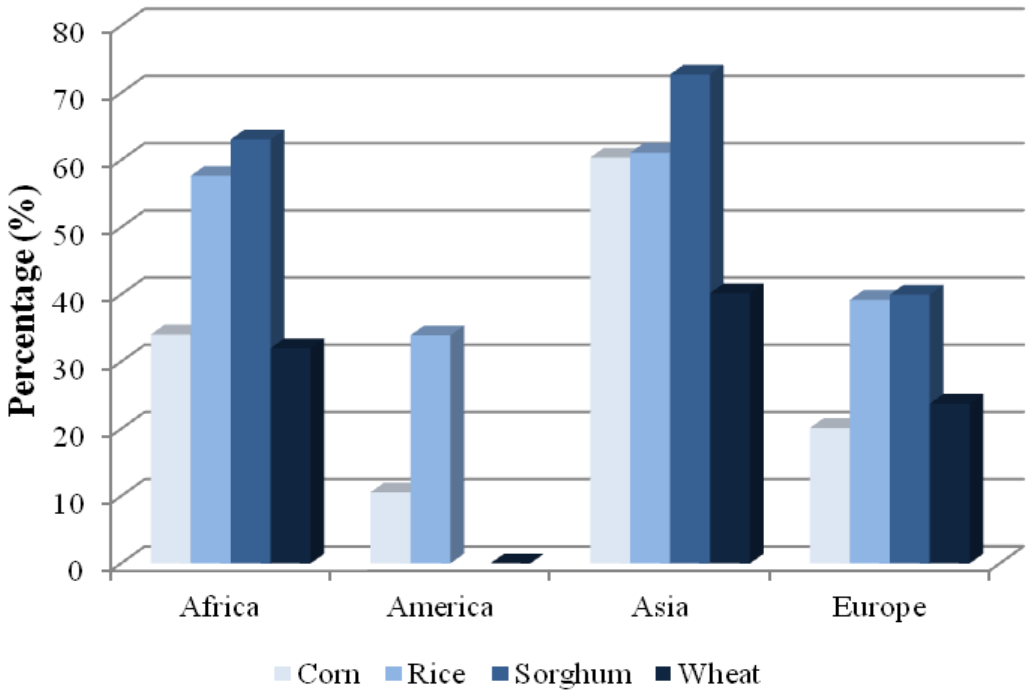

Figure 1 – Incidence of AFs in cereals samples analyzed on each continent, between 2000 and 2012.

STABILITY DURING PROCESSING

19. AFs are relatively stable compounds that are not completely destroyed by most food processes, and therefore, cereal based foods ready for consumption may still be contaminated. Sorting, cleaning, milling and thermal processing (cooking, baking, roasting, flaking, extrusion) may reduce AFs content in food products. Figure 2 illustrates the time course of AF formation and reduction in corn in reference to the Food Safety Objective (Pitt et al, 2013).

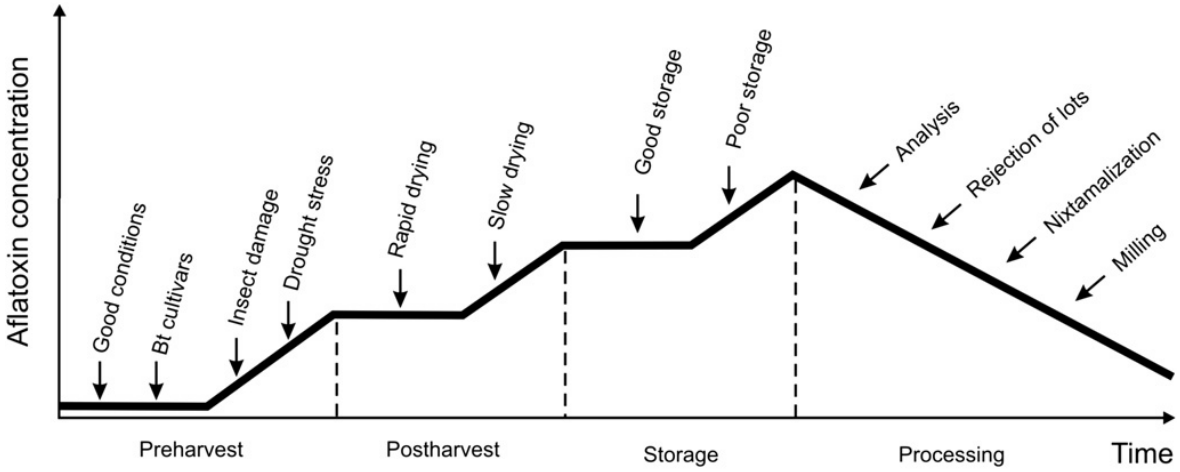

Figure 2. The time course of AF formation and reduction in maize, with reference to the Food Safety Objective (Pitt et al., 2013)

20. Sorting and cleaning usually remove the contaminated parts of the cereals, lowering AF concentration. Johansson et al. (2006) demonstrated that AFs are concentrated in the poor-quality grade components of shelled corn. About 60% of AF mass was found in damage kernels (DM), broken kernels and foreign materials (BCFM), representing only 5% of total mass. This study also found a correlation (0.964) between AF mass in the combined DM and BCFM components with AF concentration in the lot, indicating its potential value as a screening method to predict AF in a bulk lot of corn.

21. Pearson et al. (2004) tested a high-speed dual-wavelength sorter for removing corn contaminated with AFs. Reduction in AF content reached 82% in samples of yellow corn with initial AF level higher than 10 µg/kg, and 38% in samples contaminated with less than 10 µg/kg. The same approach was applied in white corn, reducing 46% of the AF content in the first sorting and 88% after a re-sorting (Pearson et al., 2010).

22. The same occurs in the milling process, where AFs may be redistributed and concentrated in certain fractions. Siwela et al. (2005) showed that AF concentration in corn meal was reduced by approximately 92% after dehulling of corn grains. During the production of polished rice (after dehulling and whitening process), AF reduction of 92-97% from the initial concentration of the raw grain was observed by Castells et al. (2007).

23. Several studies investigated the distribution of AFs during the corn wet-milling process (CRA, 2011). These studies demonstrated that AFs are mostly found into the aqueous phase of the process, due to their relatively high solubility in the water fraction. Therefore, starch, the fraction commonly used for food, is essentially aflatoxin-free.

24. The distribution of AFs in dry-milled corn fractions was evaluated by Castells et al. (2008). The authors found higher levels of AFs in the outer layers of the kernels, while processed products from the inner parts of grain, such as corn meal and flaking grits, had decreased mycotoxin levels. Pietri et al. (2009) found reductions of 8.0% (from a 5 µg/kg contaminated corn lot) and 57.0% (from a 120 µg/kg lot) of AF levels after cleaning steps. The subsequent removal of bran and germ led to a further decrease in contamination levels in the products destined for human consumption. In both papers, the most contaminated parts were those usually intended for animal feed production.

25. Hwang and Lee (2006) evaluated the reduction of AFB1 contamination in wheat after washing (10 to 30 min) and heating dry and wet wheat in an oven at various temperatures (50 to 200°C) during different periods of time (30 to 90 min). AFB1 reduction in all wheat samples was proportional to washing time (increased with longer time), ranging from 41.0 to 62.0%. AFB1 concentration decreased as temperature increased, with the most significant reduction at temperatures above 100°C. Reductions under wet heating were between 40.0 and 47.0% (100°C/30 min), up to 20% higher than what was found under dry conditions.

26. The effect of cooking (ordinary and under pressure) on AFB1 levels in polished rice was investigated by Park and Kim (2006). The ordinary process reduced AF levels by 31.0-36.0%, while in the pressure-cooked rice the reduction of AF was considerably higher (78.0-88.0%). The Ames mutagenicity test showed reductions in the aflatoxin-induced toxicity of 19.0-29.0% for ordinary-cooked rice and 68.0-78.0% for pressure cooked rice. Hussain and Luttfullah (2009) found the highest AFB1 reduction in rice cooked with excess water (87.5%), followed by ordinary (82.5%) and microwave (77.6%) cooking.

27. AF inactivation by extrusion cooking of corn flour was evaluated by Cazzaniga et al. (2001). The effects of flour moisture, extrusion temperature and sodium metabisulphite addition were evaluated. The AFB1 reduction in corn flour ranged from 10.0% to 25.0%, with the greatest reduction when the additive was used. Extrusion of rice meal showed higher reductions of AFs content, ranging from 51.0% to 95.0%, depending on the AF and extrusion conditions (initial moisture content, barrel temperature and residence time) (Castells et al., 2006).

28. The reduction of AFB1 content in the process of nixtamalization and extrusion of corn during production of tortillas was investigated by Elias-Orozco et al. (2002). The traditional nixtamalization process reduced the levels of AFB1 by 94.0% and the extrusion process by 46.0%. However, when extrusion process was combined with treatment with calcium hydroxide, reductions of AFB1 achieved 85.0%.

29. Pérez-Flores et al. (2011) evaluated the effect of microwave heating during alkaline-cooking (calcium hydroxide) of AF (B1+B2) contaminated corn during the production of tortillas. The modified tortilla-making process caused a decrease of 68.0-84.0% in AF content and, after an extract acidification (as occur during digestion), there was an increase of up to 3.0% in AF content in tortillas.

30. It is important to state that reduction of AFs levels resulting from food processing does not necessarily mean decreased toxicity of the compounds, since they may not have been destroyed, but may be bound to the food matrix or may have been changed to an unknown degradation product (Park and Kim, 2006). Thus, it is essential to perform tests to determine the toxicity and the biological activity of the remaining compounds as well as to develop analytical methodologies capable of detecting those bound and changed products.

HUMAN EXPOSURE AND RISK ASSESSMENT

31. Exposure to AFs was estimated using the total mean upper bound level of contamination on cereal grains (Table 1) and the consumption Cluster diets from GEMS/Food (WHO, 2006) (Figure 3). For each commodity, total upper mean level found in Africa was used to estimate the exposure for clusters A, C, I and J, in Americas for clusters H, K and M, in Asia for clusters G and L and in Europe for clusters B, D, E and F (Table 2). Additionally, exposure assessment was conducted using the same total upper mean level for all clusters in the calculation (Table 3). Body weight was 60 kg for all clusters except G and L (55 kg). In all scenarios, the risk arising from the exposure to AFs was characterized by calculating the margin of exposure (MOE), using a $BMDL_{10}$ of 170 ng/kg bw/day (EFSA, 2007).

32. In the first estimation (Table 2), the lowest intakes were found in Clusters H, K and M (21.8-31.0 ng/kg bw/day) and the highest were found in Clusters G and L (Asia; 511.3 and 506.6 ng/kg bw/day, respectively), mainly from the consumption of rice (about 90% of the total intake).

33. Intakes from Clusters H came mostly through the consumption of corn (72.3%). Sorghum accounted for 72% of the total intake in Cluster J and wheat for 72.2-98.0% of the total intake in Clusters B, D, E, F and M.

34. The MOE ranged from 0.3 (Cluster G and L) to 7.8 (Cluster K) and the values estimated for all Clusters indicated a health concern (MOE lower than 10000) (EFSA, 2005).

35. Using the same total mean for each crop for all clusters (Table 3), the total dietary exposure estimated to AFs ranged from 40.0 ng/kg bw/day (Cluster F) to 369.9 ng/kg bw/day (Cluster G) and the MOE from 0.5 (Cluster G and L) and 4.2 (Cluster F). Rice contributed with over 80% of AF intake in Clusters G, K and L, sorghum had the highest contribution in Cluster J (69.7%) and wheat the highest intake in Clusters B, D, E, F and M (43.5 to 73.0%).

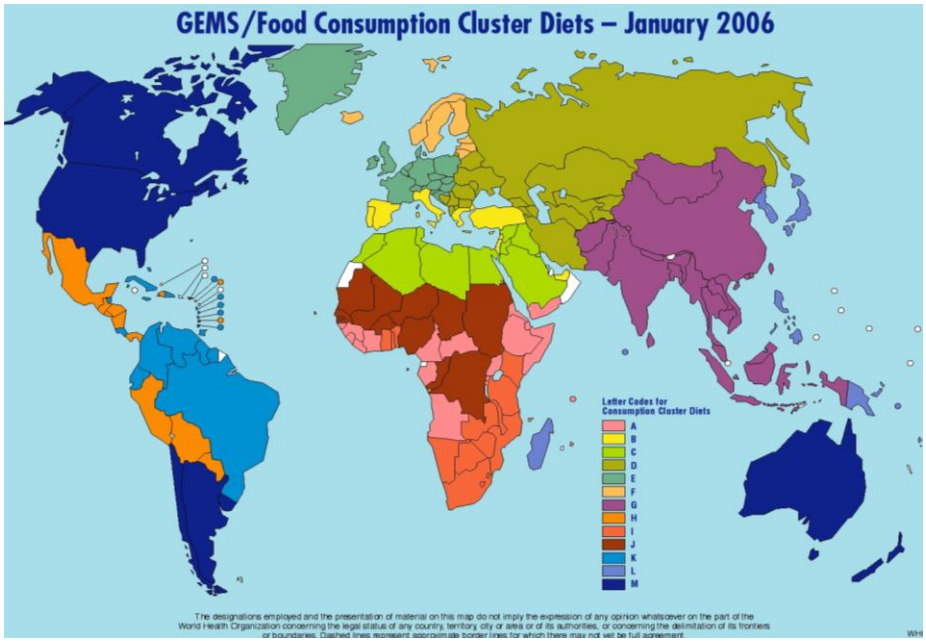

Figure 3. GEMS/Food Cluster Diets

Table 2 - AFs intake (upper bound) through the consumption of corn, rice, sorghum and wheat for each GEMS/Food Cluster (ng/kg bw/day).

	A	B	C	D	E	F	G	H	I	J	K	L	M
Corn	8.0	6.4	13.1	1.4	1.4	0.3	14.4	22.4	24.0	5.5	4.7	24.0	6.4
Rice	36.1	2.1	37.5	2.2	0.8	0.8	470.8	2.0	15.1	29.5	7.5	476.3	1.1
Sorghum	31.7	0.0	8.8	0.0	0.0	0.0	17.1	0.0	16.0	96.6	0.0	5.8	0.0
Wheat	5.3	101.1	25.6	99.5	60.3	55.1	22.9	6.6	4.1	2.5	9.5	13.7	19.5
Total	**81.1**	**109.5**	**85.0**	**103.0**	**62.5**	**56.2**	**511.3**	**31.0**	**59.1**	**134.1**	**21.8**	**505.6**	**27.0**
MOE[a]	**2.1**	**1.6**	**2.0**	**1.6**	**2.7**	**3.0**	**0.3**	**5.5**	**2.9**	**1.3**	**7.8**	**0.3**	**6.3**

[a]BMDL10=170 ng/kg bw/day (EFSA, 2007).

Table 3 – AFs intake (upper bound) through the consumption of corn, rice, sorghum and wheat for each GEMS/Food Cluster (ng/kg bw/day), using the same total mean for all Clusters.

	Af (µg/kg)[a]	A	B	C	D	E	F	G	H	I	J	K	L	M
Corn	6.5	9.0	16.1	14.7	3.4	3.6	0.8	4.2	32.3	26.9	6.2	6.8	6.9	9.3
Rice	47.4	71.9	25.0	74.7	26.2	10.0	10.0	324.8	50.8	30.0	58.7	188.3	328.6	27.3
Sorghum	86.9	53.4	0.0	14.8	0.0	0.0	0.0	15.5	28.8	26.9	162.6	0.1	5.2	4.3
Wheat	8.1	11.9	53.5	57.6	52.7	31.9	29.2	25.5	10.7	9.2	5.6	15.4	15.2	31.6
Total	**23.4**	**146.2**	**94.5**	**161.8**	**82.4**	**45.5**	**40.0**	**369.9**	**122.6**	**93.0**	**233.2**	**210.7**	**356.0**	**72.6**
MOE[a]	**-**	**1.2**	**1.8**	**1.1**	**2.1**	**3.7**	**4.2**	**0.5**	**1.4**	**1.8**	**0.7**	**0.8**	**0.5**	**2.3**

[a] samples below LOD/LOQ were considered as 0.5 LOD/LOQ; [b] BMDL10=170 ng/kg bw/day (EFSA, 2007).

RISK MANAGEMENT CONSIDERATIONS AND PUBLIC HEALTH CONCERNS

36. In the European Community, the maximum limit for AFs (AFB1+AFB2+AFG1+AFG2) is 4 µg/kg for all cereals and products derived from cereals, 10 µg/kg for corn that will be subjected to physical treatments before human consumption and for processed cereal-based foods and baby foods for infants there is only a limit of 0.1 µg/kg for AFB1 (EC, 2006). In the United States, there is a general limit for AFs of 20 µg/kg for all foods (USFDA, 2000). In Brazil, maximum limits for AFs were established for cereals and derived products (5 µg/kg, except for corn), processed cereal-based foods and baby formulas for infants (1 µg/kg) and for corn and its products (20 µg/kg) (ANVISA, 2011).

CONCLUSIONS

a) The Code of Practice for the Prevention and Reduction of Mycotoxin Contamination in Cereals, adopted by the Codex Alimentarius Commission in 2003, contains various recommendations to reduce the presence of mycotoxins in cereals. More recent research concerning the reduction of AF in a crop was conducted under laboratory conditions. However, these studies may be considered of limited application on a field situation.

b) This paper reviewed information provided in 64 published scientific papers, EFSA report and FDA report provided by the USA delegation concerning the presence of AFs in cereal grain samples (corn, rice, sorghum and wheat) from 48 countries (period of 2000 to 2012). From the 16490 samples analyzed in these studies, 35.9% contained at least one AF. Sorghum was the cereal with the highest incidence of positive samples (70.7%) and corn had the lowest incidence (23.2%). Samples coming from Asian countries had the highest incidence of positive samples for all cereals.

c) Cereal processing may reduce AFs content in the products that go to the market or used directly for human consumption. Sorting and cleaning usually remove the most contaminated parts. Higher levels of AFs are found in the outer layers of the corn kernel and processed products such as corn meal and flaking grits have decreased mycotoxin levels. Dehulling and polishing the rice may reduce AF content by over 90% and cooking the polished rice may reduce the contamination by over 30%.

d) Exposure assessments for the 13 GEMs/Food Cluster diets under different AF contamination scenarios were conducted using the data evaluated in this paper. The highest AF intakes were found in clusters G and L (Asian countries), about 90% from the consumption of rice. In all scenarios and clusters, the MOE was below 10, indicating a possible public health concern.

e) The results presented in this paper have shown that cereal grains are contaminated with AFs and that the higher exposure occurs in populations for which rice or sorghum are important component in the diet. However, these results were based on contamination data taken mostly from the literature, and to make the best use of it, some assumption had to be made due the limitation of the data provided.

f) In order to conduct a more sound evaluation of the current situation of AF contamination on cereal grains, the exposure levels and the impact to human health, it would be necessary to have raw data on cereal grains (rice, corn, sorghum, wheat, rye, oat and barley) and processed products from different parts of the world.

RECOMMENDATIONS

1. The Committee should request the JECFA to conduct an assessment on the effects of various MLs on AF exposure, and the risk from the consumption of AF contaminated cereals and cereal products.

2. Member countries are encouraged to submit raw data to allow the assessment by the JECFA on AF contamination in rice, corn, sorghum, wheat, rye, oat and barley. These data should be submitted as complete datasets with results of individual samples and not data presented in summarized or aggregate form.

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ANNEX 1

Country	Mycotoxins analyzed	Food	Positive/analyzed samples	Mean, µg/kg (range)	Method	LOD/LOQ	Reference
Algeria	AFB1	Wheat	28/45	NR [a] (0.21-37.42)	HPLC-FD	LOD=0.005	Riba et al., 2010
Argentina	AFT	Corn	264/3192 [b]	0.3–17.8 (NR-711)	TLC HPLC-FD (quantification)	LOD=0.2-0.3 LOQ=1.0	Garrido et al., 2012
Argentina	AFT	Corn	14/31	4.9–6.4 [c] (ND-22.4)	TLC	LOD=0.2-0.3 LOQ=0.4-0.5	Broggi et al., 2007
Austria	AFT	Rice	15/81	NR (0.45-11.36) [a]	HPLC-FD	LOD=0.1-0.16 LOQ=0.44-0.6	Reiter et al., 2010
Benin and Togo	AFB1	Corn	43/502	7.6–27.7 (NR)	Fluorescence densitometer	NR [d]	Egal et al., 2005
Brazil	AFT	Rice	75/166	9.09 (0.01-158.14)	IAC HPLC-FD	LOQ=0.01-0.03	Almeida et al., 2012
Brazil	AFB1 AFB2	Corn	21/200 7/200	29.12 (2.0-1393) 2.81 (5.6-55.7)	TLC	LOQ=2 LOQ=4	Rocha et al., 2009
Brazil	AFT	Rice	0/56	ND	TLC	LOD=2.5	Nunes et al., 2003
Brazil	AFB1	Rice	2/32	42.8 (11.53-74.0)	TLC	LOD=2.6	Dors et al., 2011
Brazil	AFT	Rice	1/36	1.2 (ND-1.2)	HPLC-FD	LOD=0.02–0.05 LOQ=0.05-0.16	de Carvalho et al., 2010
Brazil	AFT	Corn	24/300	23.4–40.0 [c] (ND-56.0)	TLC	LOD=4.0	Moreno et al., 2009
Brazil	AFT	Corn	7/10	1.8 (1.0-2.6)	ELISA	LOD=1.0	Oliveira et al., 2010
Canada	AFB1 AFB2	Rice	99/199 23/100	0.34–0.39 [c] (0.002-7.1) 0.08 (0.02-0.63)	IAC HPLC-FD LC-MS/MS (confirmation)	LOD=0.002 LOQ=0.05	Bansal et al., 2011
Canada	AFT	Wheat Corn	0/40 0/15	ND	LC-MS/MS	LOD=1.0-4.0	Martos et al., 2010
China	AFT	Corn	211/279	44.04 (0.2-888.3)	HPLC	NR [d]	Gao et al., 2011
China	AFB1	Corn Rice	108/108 29/29	1.3–13.5 [c] (0.4-136.8) 0.56(0.1-1.4)	ELISA IAC/HPLC–UV/FD (confirmation)	LOD=0.1	Sun et al., 2011
China	AFT	Corn	4/18	AFB1: 2.41 AFB2: 0.68 AFG1: 1.72 AFG2: 0.86 (NR)	Mycosep UPLC-UV	LOD=0.19-0.32 LOQ=0.63-1.07	Fu et al., 2008
China	AFT	Corn Rice	71/73 36/37	0.99 (NR) 0.88 (NR)	HPLC-FD	LOD=0.0074-0.1	Liu et al., 2006
Côte d'Ivoire	AFB1	Corn Rice	10/10 10/10	(<1.5-20) [a] (<1.5-10) [a]	ELISA	NR [d]	Sangare-Tigori et al., 2006
Egypt	AFT	Corn	8/80	9.85 (7.5-11.6)	TLC	NR [d]	Nogaim et al., 2011

Country	Mycotoxins analyzed	Food	Positive/analyzed samples	Mean, µg/kg (range)	Method	LOD/LOQ	Reference
India	AFB1	Sorghum	1173/1606	NR (0.01–264.0) [a]	ELISA	NR [d]	Ratnavathi et al., 2012
India	AFB1	Rice	814/1200	NR (0.1-308) [a]	ELISA	LOD=0.02	Reddy et al., 2009
India	AFB1	Wheat	664/1646	11.0-32.0 [c] (ND-606)	TLC	LOD=5,0	Toteja et al., 2006
Iran	AFT	Rice	59/71	2.09 (NR)	HPLC-FD	LOD=0.07-0.4	Mazaheri, 2009
Iran	AFB1	Corn	146/373	0.5–214.4 [c] (NR)	ELISA	LOD=1	Karami-Osboo et al., 2012
Iran	AFT	Rice	117/152	0.67 (0.15-4.27)	HPLC-FD	LOD=0.07-0.1	Mohammadi et al., 2012
Iran	AFT	Corn	17/51	22.17 [c] (0.1-316.9)	HPLC-FD	LOD=0.1	Ghiasian et al., 2011
Italy	AFT	Corn	36/36	26.3 1.7-820.0	ELISA	LOD=1.7	Covarelli et al., 2011
Japan	AFT	Corn Rice	0/10 0/53	ND	HPLC-FD	LOQ=0.1	Sugita-Konishi et al., 2006
Kenya	AFT	Corn	100/716	9.1 (1.0-48,000)	IAC Fluorometer	LOD=0.01	Daniel et al., 2011
Kenya	AFT	Corn	104/104	<20 [a] (NR)	IAC Fluorometer	NR [d]	Mwihia et al., 2008)
Kenya	AFB1	Wheat	23/50	1.7–2.2 [c] (ND-7.0)	ELISA	NR [d]	Muthomi et al., 2008
Korea	AFB1	Rice	5/88	4.8 (2.1-7.7)	ELISA HPLC-FD (confirmation)	LOD: 0.1	Park et al., 2004
Malawi	AFT	Sorghum	2/13	(1.7-3.0) [a]	Fluorometer	LOD=1.0	Matumba et al., 2011
Malaysia	AFT	Rice Wheat	11/31 2/6	1.02 (0.01-3.83) NR (0.1-5.93) [a]	IAC HPLC-FD	LOD=0.0037-0.0125	Rahmani et al., 2010
Malaysia	AFT	Rice Wheat	10/40 15/20	NR (0.15-4.42) [a] NR (0.2-3.2) [a]	UPLC-MS/MS	LOD=0.06-0.45 LOQ=0.5-1.0	Soleimany et al., 2012
Malaysia	AFB1	Wheat	3/15	1.14 (0.42-1.89)	ELISA	LOD=0.02 LOQ=4	Reddy and Baharuddin, 2010
Moroco	AFT	Wheat	0/20	ND	HPLC-FD	LOD=0.35 LOQ=0.7	Zinedine et al., 2006
Nigeria	AFT	Corn	19/103	28 (3-138)	TLC	NR [d]	Bankole and Mabekoje, 2004
Nigeria	AFT	Rice	21/21	82.5 (27.7-371.9)	HPLC/DAD	LOD=0.01-0.06	Makun et al., 2011
Nigeria	AFT	Corn Sorghum	23/23 40/40	36.0 (1.1-480.0) 8.8 (1.6-90.0)	ELISA	LOD=1.0	Bandyopadhyay et al., 2007
Nigeria	AFB1	Sorghum	93/168	199.51 (0-1164)	TLC	NR [d]	Hussaini et al., 2009

Country	Mycotoxins analyzed	Food	Positive/analyzed samples	Mean, µg/kg (range)	Method	LOD/LOQ	Reference
Nigeria	AFB1	Wheat Corn Sorghum Rice	2/11 7/18 3/10 12/20	(4.25-5.17) [c] (2.51-3.94) [c] (5.20-6.25) [c] (4.16-7.25) [c]	ELISA	NR [d]	Ayejuyo et al., 2011
Pakistan	AFB1	Corn	30/36	18.68 (ND-30.96)	HPLC-FD	NR [d]	Shah et al., 2010)
Pakistan	AFT	Rice Wheat Corn Sorghum	8/40 4/20 6/15 3/10	4.5 [c] (1.5-10.8) 6.6 (1.8-15.5) 10.4 (3.0-18.5) 5.0 (2.0-9.4)	HPLC-FD	LOD=0.5-1.0	Lutfullah and Hussain, 2012
Pakistan	AFT	Rice	28/40	4.9 (1.5-13.9)	TLC	LOD=0.5-1.0	Hussain et al., 2011
Pakistan	AFT	Corn	34/40	56.7 [c] NR	HPLC-FD	NR [d]	Ahsan et al., 2010)
Pakistan	AFT	Corn	18/65	241.0 NR	TLC	LOQ=0.5-1.0	Khatoon et al., 2012
Qatar	AFT	Rice Wheat	3/9 0/4	(0.14-0.24) [a] ND	HPLC-FD	LOD=0.1	Abdulkadar et al., 2004
Serbia	AFT	Corn Wheat Sorghum	81/443 58/304 4/10	20.0 (ND-50.0)[a]	ELISA	NR [d]	Jakic-Dimic et al., 2009
Tanzania	AFT	Corn	22/120	24.0 [e] (1.0-158.0)	IAC HPLC-FD	LOD=0.07-0.6	Kimanya et al., 2008
Tunisia	AFT	Sorghum Wheat	3/3 9/34	71.3 (27.4–116.7) 6.6 (5.2-8.7)	SLE UHPLC-MS/MS	LOD=0.4 LOQ: 1.0	Oueslati et al., 2012
Tunisia	AFT	Rice Corn Wheat Sorghum	0/11 1/17 4/46 36/49	ND 0.42 (0.15-18.6) [a] (0.4-25.8) [a]	HPLC/FD	LOD=0.02-0.05 LOQ=0.05-0.1	Ghali et al., 2010
Tunisia	AFT	Sorghum Corn Rice	13/17 9/21 2/16	22.3 (1.7-67) 7.6 (2.9-12.5) 4.7 (2-7.5)	ELISA	LOD=0.05	Ghali et al., 2008
Tunisia	AFT	Sorghum	58/93	9.9 (0.34-54.5)	HPLC-FD	LOD=0.025-0.05	Ghali et al., 2009
Turkey	AFT	Rice	56/100	NR (0.05-21.4) [a]	ELISA	LOD=0.05	Aydin et al., 2011
Turkey	AFT	Wheat	24/41	166.0 (10.4-643.5)	HPLC-FD	LOD=0.01-0.02	Giray et al., 2007
Turkey	AFT	Corn	26/26	8.2 [c] (0.01-32.3)	ELISA	NR [d]	Oruc et al., 2006
Turkey	AFT	Corn	19/30	27.8 (0.62-116.7)	HPLC-FD	NR [d]	Alptekin et al., 2009
Uganda	AFT	Corn	296/390	19.5 [c] (0-50)	IAC Fluorometer	LOD=0	Kaaya and Kyamuhangire, 2006
USA	AFT	Corn	69/72	25.6	NR	NR	Bruns et al., 2006
USA	AFT	Corn	18/18	6.5	HPLC-FD	LOD=0.5	Abbas et al., 2006
Vietnam	AFB1	Rice	35/100	3.31 (ND-29.8)	HPLC-FD	LOD=0.07 LOD=0.22	Nguyen et al., 2007
Zambia	AFT	Maize	114/114	2.7 (0.01–10.0)	ELISA	LOD=1.0	Mukanga et al., 2010

NR=not reported; ND=not detected; [a] it was used the mean of the interval; [b] samples collected in 1999 were removed; [c] weighted mean was realized with the available data; [d] it was used the LOD/LOQ of a similar methodology; [e] the median was used.

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