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FOOD AND AGRICULTURE ORGANIZATION OF THE UNITED NATIONS

WORLD HEALTH ORGANIZATION

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DISCUSSION PAPER ON OCHRATOXIN A IN COFFEE

Governments and international organizations are invited to submit comments on the following subject matters no later than 1 March 2007, preferably in electronic format, for the attention of Ms. Tanja Åkesson, the Netherlands Secretariat of the Codex Committee on Contaminants in Foods, Fax No.:+31 70 3786141; E-mail:info@codexalimentarius.nl with a copy to the Secretary, Codex Alimentarius Commission, Joint FAO/WHO Food Standards Programme, Viale delle Terme di Caracalla, 00153 Rome, Italy (Fax +39.06.5705.4593; E-mail: Codex@fao.org).

BACKGROUND

1. The Codex Committee on Food Additives and Contaminants, at its 38th Session, agreed to establish an electronic working group led by Brazil to prepare a discussion paper on the occurrence of ochratoxin A (OTA) in coffee, for consideration at the First Session of the Codex Committee on Contaminants in Foods (see ALINORM 06/29/12 para.145). Brazil, Canada, European Community, France, Ghana, Indonesia, Switzerland, the United Kingdom, Uganda and the IFT participated in the electronic working group and prepared this document.

INTRODUCTION

2. OTA is a mycotoxin that can be found in different sources, as cereals, wine, grape juice, dried vine fruit, beer, coffee, cocoa and spices. Cereals and cereal based products (flour, bran, breakfast cereals, bread, pasta, biscuits, cereal bars and others) represent the main source of dietary exposure, both for adults and children. For this last group, very vulnerable due to their food consumption in relation to their body weight, grape juice and raisins can also be an additional source of exposure. Besides the previously mentioned products, wine, beer, cocoa and coffee can be other sources of exposure (CX/FAC 06/38/26).

3. OTA is produced in food by the following fungi: Penicillium verrucosum and Aspergillus species, such as A. ochraceus, A. westerdijkiae, A. carbonarius and A. niger. They occupy various ecological niches, affect several commodities, and have different frequency of occurrence in different geographical regions (WHO, 2002).

4. A study carried out in Brazil investigated the distribution of OTA producing fungi and their ability to produce the toxin in 872 isolates. The most common species found was Aspergillus niger (549 isolates), but only 3% of the isolates produced OTA. A. ochraceus was also commonly found (269 isolates), with 75% being able to produce OTA. A. carbonarius was found (54 isolates) in only one region, which has a warm climate, and only in beans from the drying yard or from storage. However 77% were able to produce OTA (PITT, 2001).

5. There are two main species of coffee, with several varieties, responsible for the world production and trade of the product: Coffea arabica (arabica coffee), that can be grown from an altitude of 600-2000m and average temperature between 18^{o} – 22.5^{o}C, in the wet tropics and Coffea canephora (robusta coffee), that can be grown at an altitude below 600m and average temperature between 22^{o} 26^{o}C, also in the wet tropics.

6. According to FAOSTAT (2006) coffee is one of the most important and valuable commodities produced in 78 countries around the world, by 20 to 25 million families (most of them small farmers). It represents, for many developing countries, the major share of their total exports. Nineteen of these countries were responsible for 90.08% of the total world production (ANNEX). From the estimated US$55 billion year for the total retail market, exporting countries are expected to receive around 15% of it.

7. This discussion paper considers different aspects related to coffee contamination with OTA: toxicological evaluation, sampling and analytical methods, data of occurrence, estimated intake, and measures for the prevention and reduction of OTA contamination in coffee.

CHEMICAL STRUCTURE

8. OTA consists of a polyketide-derived dihydroisocoumarin moiety linked through the 12-carboxy group to phenylalanine. Due to its chemical structure (Figure 1), it is soluble in most organic solvents such as alcohols, ketones, benzene, and chloroform, but it is not very soluble in water and it is insoluble in petroleum ethers and saturated hydrocarbons. It degrades in an alkaline medium. OTA is also stable to the level of heat utilized in ordinary cooking, being necessary temperatures above 250^{o}C for several minutes to reduce its concentration. It is detectable by blue-green fluorescence in ultraviolet light.

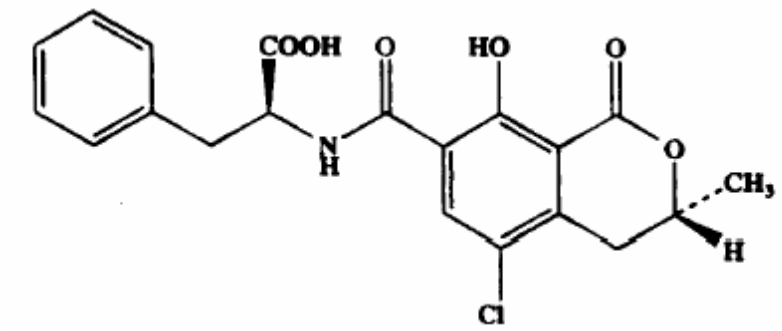

Figure 1. Chemical structure of ochratoxin A.

TOXICOLOGICAL EVALUATION

9. The toxicity of OTA has been reviewed by the International Agency for Research on Cancer (IARC), that has classified OTA as a possible human carcinogen (group 2B), and by the Joint FAO/WHO Expert Committee on Food Additives (JECFA).

10. The kidney is considered the most affected organ by OTA, whose nephrotoxic and carcinogenic properties have been the major focus of the safety evaluation performed by scientific bodies. Furthermore, OTA has also teratogenic, immunotoxic and possibly neurotoxic properties.

11. The JECFA considered at its 56^{th} meeting in February 2001 that the new data raised further questions about the mechanisms by which OTA causes nephrotoxicity and renal carcinogenicity and the interdependence of these effects. The mechanism by which OTA causes carcinogenicity is unknown, although both genotoxic and non-genotoxic modes of action have been proposed. JECFA noted that studies to solve these issues are in progress and recommended reviewing the results when they become available. JECFA retained the previously established Provisional Tolerable Weekly Intake (PTWI) of 100ng/kg of body weight, pending the results of these studies (WHO, 2002).

12. The EFSA Scientific Panel on Contaminants in the Food Chain established 120 ng/kg b.w. as the Tolerable Weekly Intake (TWI) for OTA (EFSA, 2006).

13. In the priority list of food additives, contaminants and naturally occurring toxicants proposed by CCFAC for evaluation by JECFA, OTA is listed as high priority, addressing toxicological re-evaluation, exposure assessment (special consideration to developing countries) and effects of processing on residual levels in food (ALINORM 05/28/12).

METHODS OF ANALYSIS AND SAMPLING

Sampling

14. The random nature of fungal contamination of raw materials (such as coffee) and thus the uneven distribution of subsequent OTA contamination means that sampling is a major issue (EFSA, 2002).

15. The total variance can be estimated and portioned into sampling, sample preparation and analytical variances. The sample, sub sample size, particle size and type of mill influence the uncertainty associated with the measure of the true level of total contamination.

16. These relationships can be expressed by using the estimated variance for sample size, sub sample size and number of analyses for specific OTA concentration, as described below:

    - Sampling variance:
    $S^2(s) = (1/ns)\ 1.35C^{1.09}$
    Where: ns = sample size (kg)
    C = estimate of OTA concentration measured in µg/Kg

    - Sub sampling variance:
    $S^2\ (ss) = (25/nss)\ 0.27C^{1.46}$
    Where: nss = sub sampling size (g)

    - Effect of change of the number of aliquots:
    $S^2\ (a) = (1/na)\ 0.01\ C^{1.61}$
    Where: na = number of aliquots quantified

    The total variance estimated:
    $S^2(t) = S^2\ (s) + S^2\ (ss) + S^2\ (a)$ (Vargas et al., 2004)

17. According to Vargas (2005), the lognormal theoretical distribution should be selected to model the sample OTA test results because it gave the best fit.

18. It is not possible to design the sampling method without determining the acceptable risk to consumers and producers and the maximum residue level.

Analytical Methods

19. Several analytical methods for the determination of OTA in cereals (maize, barley, wheat and rye) and derived products (wheat bran and whole meal products), and beverages (wine, beer and coffee) have been formally validated in collaborative studies.

20. A criteria-based approach, whereby a set of performance criteria is established with which the analytical method used should comply, is appropriate. The criteria-based approach has the advantage that, by avoiding setting down specific details of the method used, developments in methodology can be exploited without having to reconsider or modify the specified method. The performance criteria established for methods should include all the parameters that need to be addressed by each laboratory such as the detection limit, repeatability coefficient of variation, reproducibility coefficient of variation, and the percent recovery necessary for various statutory limits (Table 1). Utilizing this approach, laboratories would be free to use the analytical method most appropriate for their facilities. Analytical methods internationally recognized may be used. The methods are regularly monitored and improved depending upon technology (CX/FAC 06/38/18).

21. Performance Criteria for Methods of Analysis established by EC are shown in Table 1

Table 1- Performance criteria for **OTA** (EC Nº 401/2006)

Level µg/kg	RSD_r	RSD_R	Recovery
< 1	≤ 40	≤ 60	50 to 120
1-10	≤ 20	≤ 30	70 to 110

Precision RSD_r may be calculated as 0.66 times the precision RSD_R at the concentration of interest.

- The detection limits of the methods used are not stated as the precision values as given at the concentrations of interest;
- The precision values are calculated using the Horwitz equation, i.e.:

$RSD_R = 2^{(1-0.5 \log C)}$

Where :

- RSD_R is the relative standard deviation calculated from results generated under reproducibility conditions [(sR/ *X*) x 100]
- C is the concentration ratio (i.e. 1= 100g/100g; 0.001 = 1,000 mg/kg)

OCCURRENCE OF OTA IN COFFEE

22. Surveys all over the world have confirmed the presence of OTA in commercial raw, roasted and soluble coffee. Extensive sampling of raw coffee from all origins and both types of coffee (Arabica, Robusta) has shown that OTA contamination may be more frequent in some areas, but that no producing country is entirely free from contamination (Taniwaki, 2006).

23. The natural occurrence of OTA in green and roasted coffee beans has been reported in the following studies. Additional information, including the geographical origin of analysed samples, are shown at Tables 2, 3 and 4 (Taniwaki, 2006).

Green coffee beans

24. OTA as contaminant in green coffee beans was first reported by Levi et al. (1974) at levels ranging from 20 to 360 µg/Kg in 22 of 335 samples, with detection limit of 20 µg/kg.

25. Later on, revising several data from coffee companies, Levi (1980) reported that OTA was not found in 502 commercial green coffee shipments entering in the port of Trieste (Italy). On the other hand, in the USA, OTA was detected in 2 of 201 green coffee samples, at levels of 24 and 96 µg/kg.

26. OTA concentration ranging from <10 to 200 µg/kg was found in 9 of 31 green coffee samples by Norton et al. (1982).

27. Cantafora et al. (1983) reported OTA in 9 of 40 commercial green coffee samples at levels of 0.5 to 23 µg/kg and Tsubouchi et al. (1985) reported levels from 9.9 to 46.0 µg/kg in 4 of 22 samples.

28. Micco et al. (1989) found OTA levels from 0.2 to 15.0 µg/kg in 17 of 29 green coffee samples. Studer-Rohr et al.(1995) detected it in 13 of 25 samples at levels ranging from 1.2 to 56.0 µg/kg.

29. Data from MAFF (1996) reported OTA occurrence in 110 of 291 green coffee samples of *Coffea arabica* and *C. canephora*, imported by the UK from 27 different countries. The highest levels in *C. arabica* and *C. canephora* were 9 and 27.3 µg/kg, respectively.

30. OTA was detected at levels ranging from 0.1 to 17.4 µg/kg (Nakajima et al., 1997) and from 0.1 to 4.6 µg/Kg (Trucksess et al., 1999).

31. Romani et al. (2000) showed 106 of 162 green coffee samples OTA positive ranging from 0.1 to 48 µg/kg.

32. Leoni et al. (2001) detected OTA in 27 of 132 green coffee samples, collected at sale points, with levels from 0.7 to 47.8 µg/kg.

33. Data collected from EU Member States on the OTA occurrence in 1704 green coffee samples showed 36% of positive samples and the mean level was 3.6 µg/kg. (Miraglia and Brera, 2002).

34. Taniwaki et al. (2003) reported that the average OTA content in 135 samples of mature cherries from trees, overripe cherries from trees, overripe cherries from the ground, drying yard and storage was 0.1, < 0.2, 1.6, 2.1 and 3.3 µg/kg, respectively. Although OTA levels varied widely, only 9 of 135 samples exceeded 5 µg/kg, with 1 sample of poor quality coffee exceeding 100 µg/kg.

35. Batista et al. (2003) reported that 22% of the 40 green coffee samples were contaminated with OTA at levels from 0.47 to 4.82 µg/kg with an average contamination of 2.45 µg/kg.

36. Martins et al. (2003) analysed 60 green coffee samples, from which 20 (33.3%) were contaminated with levels from 0.2 to 7.3 µg/kg, with an average contamination of 2.38 µg/kg.

37. Yani (2004) reported OTA contamination in green coffee beans collected from farmers, district and regency sectors in Indonesia. Twelve (40%), 8 (53.3%), and 5 (33%) out of 30, 45, and 15 samples, were contaminated with OTA at levels from 0.092 to 3.736 µg/kg (average of 0.70 µg/kg); 0.083 – 0.751 µg/kg (average of 0.30 µg/kg) and 0.162 – 1.027 µg/kg (average of 0.38 µg/kg) at farmers, collectors district and regency, respectively.

38. Gollücke et al. (2004) reported OTA in 37 green coffee samples at levels from <0.16 to 6.24 µg/kg (average of 3.20 µg/kg). Five samples were separated for sound and defective beans. Sound beans showed levels from 0.22 to 0.80 µg/kg (average 0.46 µg/kg) and defective beans from 0.42 to 17.46 µg/kg (average 4.52 µg/kg).

39. Pardo et al. (2004) detected OTA in all 57 green coffee samples from different origins. The average contamination was 6.7 µg/kg, ranging from 1.3 to 31.5 µg/kg. OTA levels in Arabica and Robusta coffee samples were not significantly different.

40. Moraes et al. (2006) analysed 30 green coffee samples and found OTA levels ranging from <1 to 133.7 µg/kg, with an average contamination of 14.7 µg/kg.

Table 2. Incidence of ochratoxin A (OTA) in green coffee worldwide.

Origin	Nº positive/ Nº samples	Range of OTA (µg/kg)	Coffee type	Reference
Angola	0/4	< 20[a]	N.S.[b]	Levi et al. (1974)
Brazil	3/7	Trace – 360	"	"
Colombia	17/139	Trace – 50	"	"
Cameroon	0/1	< 20[a]	"	"
Ivory Coast	1/12	Trace	"	"
Uganda	1/ 2	Trace	"	"
Unknown	7/102	Trace	"	"
Unknown	0/502	N.D.[c]	"	Levi (1980)
Unknown	2/201	N.D.[c] – 96	"	"
Brazil	10/14	0.2 – 3.7	Arabica	Micco et al. (1989)
Cameroon	3/3	Traces – 2.2	Robusta	"
Colombia	1/ 2	3.3	Arabica	"
Costa Rica	1/ 2	Traces	Arabica	"
Ivory Coast	1/2	1.3	Robusta	"
Kenya	0/2	< 0.01[a]	Arabica	"
México	1/ 2	1.4	Arabica	"
Zaire	2/2	8.4 – 15.0	Robusta	"
Brazil	3/5	2.0 – 7.4	N.S.[b]	Studer-Rhor et al. (1995)
Colombia	3/5	1.2 – 9.8	"	"
Central America	0/1	< 0.5[a]	N.S.[b]	Studer-Rhor et al. (1995)
Costa Rica	0/1	< 0.5[a]	"	"
Guatemala	0/1	< 0.5[a]	"	"
Ivory Coast	2/2	9.9 – 56.0	"	"

[a] Corresponds to the detection limit of the method; [b] Not Specified; [c] Not Detected (limit not specified).

Table 2. Incidence of ochratoxin A (OTA) in green coffee worldwide.

Origin	Nº positive/ Nº samples	Range of OTA (µg/kg)	Coffee type	Reference
Kenya	0/3	< 0.5[a]	"	"
New Guinea	0/1	< 0.5[a]	"	"
Tanzania	1/1	2.2	"	"
Zaire	1/1	17.3	"	"
Unknown	2/4	2.2 – 11.8	"	"
America, Africa, Papua New Guinea	31/153	0.2 – 9.0	Arabica	MAFF, 1996
America, Africa, Asia	55/75	0.2 – 27.3	Robusta	"
Unknown	24/63	0.2 – 7.7	N.S.[b]	"
Yemen	7/10	0.7 – 17.4	Arabica	Nakajima et al. (1997)
Tanzania	5/9	0.1 – 7.2	Arabica	"
Indonesia	2/9	0.2 – 1.0	Robusta	"
Ethiopia	0/1	< 0.1[a]	Arabica	"
Central America	0/6	< 0.1[a]	Arabica	"
South America	0/12	< 0.1[a]	Arabica	"
East Africa	42/33	0.2 – 62.0	N.S.[b]	Heilmann et al. (1999)
West Africa	9/9	0.3 – 5.0	"	"
Asia	20/29	0.2 – 4.9	"	"
Central America	6/15	0.2 – 0.8	"	"
South America	5/17	0.2 – 1.0	"	"
South America	9/19	0.1 – 4.9	N.S.[b]	Trucksess et al. (1999)
Africa	76/84	0.5 – 48.0	N.S.[b]	Romani et al. (2000)
Latin America	19/60	0.1 – 7.7	"	"
Asia	11/18	0.2 – 4.9	"	"
Brazil	27/132	0.7 – 47.8	Arabica	Leoni et al. (2001)
Unknown	374/1704	0.2 – 80.0	N.S.[b]	EU (2002)
Brazil	9/135	< 0.2 – 100	Arabica	Taniwaki et al. (2003)
Brazil	5/40	0.4 – 4.82	Arabica	Batista et al. (2003)
Brazil	20/60	0.2 – 7.3	Arabica	Martins et al. (2003)
Brazil	22/54	0.3 – 160	Arabica	Moraes & Luchese (2003)
Indonesia	25/60	N.D.[c] – 3.7	Robusta	Yani, 2004
Brazil	17/37	0.2 – 6.2	Arabica	Gollücke et al. (2004)
Africa	12/12	2.4 – 23.3	Robusta	Pardo et al. (2004)
America	31/31	1.3 – 27.7	Arabica	"
Asia	14/14	1.6 – 31.5	Arabica and Robusta	"
Brazil	15/30	< 1 – 133.7	Arabica	Moraes et al. (2006)

[a] Corresponds to the detection limit of the method; [b] Not Specified; [c] Not Detected (limit not specified).

Roasted and Soluble Coffee

41. The presence of OTA in commercial roasted coffee beans was reported for the first time in five of 68 samples at levels from 3.2 to 17.0 µg/kg by Tsubouchi et al. (1985), as a result of the introduction of a HPLC method, for the determination of OTA in coffee beans and coffee products.

42. OTA was detected in 16 of 40 analyzed coffee brews, prepared from roasted coffee samples. The contamination levels ranged from 1.0 to 7.8 µg/kg (Studer-Rohr et al., 1994a, 1994b). In these studies, a partial destruction of OTA was obtained after roasting.

43. The presence of OTA in 20 out of 30 commercial roasted coffee samples was described by Koch et al. (1996) at levels ranging from 0.3 to 7.5 µg/kg.

44. Pittet et al. (1996) surveyed 116 soluble coffee samples from various countries and different manufacturers. The contamination levels ranged from <0.2 to 15.9 µg/kg. The highest levels of OTA were detected among soluble coffee samples adulterated with coffee husks and/or coffee parchments (mean contamination level: 5.9 µg/kg). By comparison, OTA concentrations in pure soluble coffee samples were significantly lower, with a mean contamination level of 1.1 µg/kg.

45. Patel et al. (1997) detected OTA in 17 out of 20 roasted coffee samples ranging from 0.2 to 2.1 µg/kg.

46. Trucksess et al. (1999) detected OTA in 9 from 13 roasted ground coffee samples in the USA. The contamination levels ranged from 0.1 to 1.2 µg/kg and the average was 0.41 µg/kg.

47. Van der Stegen et al. (1997) analysed 633 samples of coffee products. The levels of OTA in roasted coffee ranged from <0.5 to 8.2 µg/kg, with an average of 0.8 µg/kg. From 149 soluble coffee samples only four exceeded 10 µg/kg, with an average of 1.3 µg/kg.

48. OTA was detected by Jorgensen (1998) in all of 11 roasted coffee beans samples at levels ranging from 0.1 to 3.20 µg/kg, with an average of 0.51 µg/kg.

49. The occurrence of OTA was detected by Prado et al. (2000) in soluble coffee and roasted ground coffee samples. Samples of soluble and roasted ground coffee showed levels ranging from 0.31 to 1.78 µg/kg (mean of 0.73 µg/kg) and 0.99 to 5.87 µg/kg (mean of 1.75 µg/Kg), respectively.

50. In a study performed by Fazenkas et al. (2002), in 50 commercial coffee samples, 66% were contaminated with OTA ranging from 0.17 to 1.3 µg/kg, with an average of 0.57 µg/kg.

51. Lin et al. (2005) analysed 51 coffee samples detecting OTA in 13 (25%) of them, with the contamination ranging from <0.1 to 0.5 µg/kg.

52. Data collected from EU Member States on the OTA occurrence in 1184 processed coffee samples showed 46% of positive samples and the mean level was 1.1 µg/kg. (Miraglia and Brera, 2002).

53. Moraes et al. (2006) analysed 33 market samples of roasted coffee, including low cost brands and found OTA ranging from < 1 to 13 µg/kg with an average of 1.49 µg/kg.

Table 3. Incidence of ochratoxin A (OTA) in commercial roasted coffee worldwide.

Retail country	**Nº positive/ Nº samples**	**Range of OTA (µg/kg)**	**Reference**
Japan	5/68	3.2 – 17.0	Tsubouchi et al. (1988)
United Kingdom	17/20	0.2 – 2.1	Patel et al. (1997)
Europe	?/484	< 0.5[a] – 8.2	Van der Stegen et al. (1997)
Denmark	11/11	0.1 – 3.2	Jorgensen (1998)
Spain	29/29	0.22 – 5.64	Burdespal e Legarda (1998)
United States	9/13	0.1 – 1.2	Trucksess et al. (1999)
Brazil	23/34	0.3 – 6.5	Leoni et al. (2000)
Brazil	41/47	0.99 – 5.87	Prado et al. (2000)
Germany	22/67	0.3 – 3.3	Wolff (2000)
Germany	273/490	0.21 – 12.1	Otteneder & Majerus (2001)
Canada	42/71	0.1 – 2.3	Lombaert et al. (2002)
Hungary	33/50	0.17 – 1.3	Fazekas et al. (2002)
Brazil	17/33	< 1 – 13.2	Moraes et al. (2006)

[a] Corresponds to the detection limit of the method.

Table 4. Incidence of ochratoxin A (OTA) in commercial soluble coffee worldwide.

Retail country	**Nº positive/ Nº samples**	**Range of OTA (µg/kg)**	**Reference**
Austrália	7/22	0.2 – 4.0	Pittet et al. (1996)
United States	3/6	1.5 – 2.1	“
Germany	5/9	0.3 – 2.2	“
United Kingdom	64/80	0.1 – 8.0	Patel at al. (1997)
Europe	?/149	< 0.5[a] – 27.2	Van der Stegen et al. (1997)
Spain	9/9	0.19 – 1.08	Burdaspal e Legarda (1998)
Brazil	8/10	0.31 – 1.78	Prado et al. (2000)
Brazil	16/16	0.5 – 5.1	Leoni et al. (2000)
Germany	23/52	0.3 – 9.5	Wolff (2000)
Germany	12/41	0.28 – 4.8	Otteneder & Majerus (2001)
Canada	20/30	0.1 – 3.1	Lombaert et al. (2002)

[a] Corresponds to the detection limit of the method.

FACTORS AFFECTING THE PRESENCE OF OTA IN COFFEE

54. Since the presence of OTA in coffee beans is a result of contamination by a few fungi species, mainly *A. ochraceus, A. westerdijkiae*, *A.niger* and *A. carbonarius* (Urbano et al., 2001a; Taniwaki et al., 2003; Batista et al., 2003; Suarez-Quiroz et al., 2004), practices that restrict the fungi development throughout the production chain must be adopted to avoid OTA and preserve the final quality of coffee.

55. Mycological analyses of cherry beans collected from trees have not demonstrated the presence of these ochratoxigenic fungi, indicating that OTA contamination in green coffee is a post-harvesting problem. The main sources of these fungi seem to come from soil, equipment and drying yard surfaces (Taniwaki et al., 2003).

56. When dealing with over ripeness, fruits generally dry on the tree and then fall down. If these beans remain for a long period on the soil, an increase of infection with ochratoxigenic species may occur. If infected beans are mixed with healthy ones, fungi contamination will spread (Taniwaki et al., 2003).

57. The influence of the harvest procedure, fruit ripening and drying process on risk contamination was evaluated. It was concluded that gleaning coffee and coffee dried directly on bare ground were the highest sources of contamination (Moraes and Luchese, 2003).

58. Moisture content and water activity (a_w) are the most important factors that influence fungal growth. To avoid the development of toxigenic fungi in coffee, the water activity should be kept under control from post-harvesting to final processing (Palacios-Cabrera et al, 2004).

59. Drying coffee beans to 11-12% moisture content, which correspond to aw of 0.60, avoid fungal growth and subsequently OTA production. Laboratory studies have shown that the limiting aw for growth of *A. ochraceus* and *A. niger* are 0.79 and 0.77, respectively (Palacios-Cabrera et al, 2004).

60. Different climates and production systems imply different risk for the development of OTA-producing fungi. In shaded plantations, the soil remains relatively moist even if there is a dry season. In some regions the harvest period (typically stretching over three months) coincides with a rainy season or humid conditions. These scenarios are of the highest risk for fallen coffee fruit to become grossly contaminated. In unshaded production systems, where the harvest is conducted in a dry season, the risk is reduced. (FAO, 2005).

61. Coffee roasting may remove a very significant percentage of OTA, as shown in Table 5. However, conflicting data with respect to the influence of roasting, grinding and beverage preparation on the residual level of OTA are found in the literature.

Table 5. Effect of roasting on ochratoxin A (OTA) reduction.

N°. of samples	Toxin origin	Roasting condition	% of reduction	References
4	Inoculation[a]	200°C/10-20 min	0 –12	Tsubouchi et al. (1988)
2	Natural[b]	5 – 6 min/dark roasting	90 – 100	Micco et al. (1989)
3	Natural[b]	252°C/100-190 seg	14 – 62	Studer-Rohr et al. (1995)
2	Inoculation[a]	252°C/100-190 seg	2 – 28	“
6	Natural[b]	223°C / 14 min	84	Blanc et al. (1998)
3	Inoculation[a]	200ºC/10 min (medium roasting)	22.5	Urbano et al. (2001b)
3	“	200ºC/15 min (medium roasting)	48.1	“
3	“	210ºC/10 min (medium dark)	39.2	“
3	“	210ºC/15 min (medium dark)	65.6	“
3	“	220ºC/10 min (dark)	88.4	“
3	“	220ºC/15 min (dark)	93.9	“

[a] Coffee beans inoculated with toxigenic spores of *Aspergillus ochraceus*; [b] Naturally contaminated beans.SOURCE: (Taniwaki, 2006).

DIETARY EXPOSURE

62. Exposure to mycotoxins has been associated with the observation of adverse effects in humans and livestock. Health concerns related to dietary exposure to mycotoxins depend on: the levels of mycotoxins in food as consumed, the amount of food consumed, body weight and physiological state of the individual, and the bioavailability and toxicity of the compound to humans. Other dietary factors may increase or decrease the toxicity (Kuiper-Goodman, 1994).

63. In its OTA evaluation in 2001, JECFA calculated the human exposure to OTA from different food sources. The approach followed resulted in a mean total intake of OTA of about 45 ng/kg bw per week, assuming a body weight of 60 kg. Cereals and wine contributed about 25 and 10 ng/kg of b.w per week, respectively, to mean intake, whereas grape juice and coffee each contributed 2-3 ng/kg of b.w per week. Other food products (dried fruits, beer, tea, milk, cocoa, poultry and pulses) contributed less than 1 ng/kg b.w per week (WHO, 2002).

64. In the evaluation of OTA exposure, showed in the position paper prepared by Sweden for the 31st session of the CCFAC, the mean values used for coffee were those from countries with high consumption rate. In this calculation, coffee represented 12% of the total intake and 8.6 or 3.6% of the Tolerable Daily Intake (TDI) established by the Nordic group or JECFA, respectively (CX/FAC99/14).

65. In 2002, an assessment of OTA dietary intake by the population of EU was published. Coffee represented 10% of the total intake, while cereals and cereal products contributed the most (50 %) to the mean European total human OTA exposure. For the overall population, the OTA intake from coffee ranged from 0.06 to 0.42 ng/kg b.w/day. In most countries no marked differences were found in dietary intake values among groups of population (Miraglia and Brera, 2002).

66. A total diet study performed in France showed that the estimated average OTA intake of the French population was 2.2 ng/kg b.w./day for adults aged 15 or more, 4.1 ng/kg b.w./day for children aged 3 to 14. The 95th percentile exposure was 3.6 ng/kg b.w./day for adults and 7.8 ng/kg b.w./day for children. The food group that contributed most (>70 %) to the exposure for both population groups was cereals and cereal products. Grape-based products (raisins, table grapes, juice and wine), coffee, nuts and oilseeds contributed less than 5 % to the total exposure.

PREVENTION OF OTA IN COFFEE

67. Some research projects have been carried out to identify the factors related to the formation of OTA in coffee. The most recent and broad initiative came from the governments of producing countries, with collaboration of the FAO, the International Coffee Organization (ICO) and the European Coffee Industry, resulting in the project "Enhancement of Coffee quality through the Prevention of Mould Formation".

68. Good practices can be utilized at all stages of coffee production to reduce OTA contamination, as follows:

69. During harvesting, ground beneath the coffee plants should be covered using clean tarpaulin or plastic canvas during harvesting time, in order to avoid contact of the cherries just picked from the plant with the soil, foreign material and cherries already fallen during the crop season. The last ones may be highly contaminated with fungal spores, posing a high OTA risk. Also overripe cherries should not be mixed with the ones just picked from the plant.

70. After harvesting fresh cherries should be processed as fast as possible, either for dry or wet processing, preferentially on the same day.

71. The processing facilities should be located in a dry area, with equipment and facilities maintained permanently well clean. By products (husk, pulp) obtained during processing, must be disposed in a separated area and composted before its utilization in the orchard.

72. Undesirable or risk material, such as husks, floaters, un-hulled cherries or mouldy beans, should be separated from good quality cherries.

73. Water utilized in wet processing must have good quality.

74. Equipment utilized must be well cleaned after use.

75. Drying must occur as fast as possible, to avoid fungi growth and OTA production. Cherries and beans must: be dried on clean surfaces; disposed in drying layers with maximum of 4 cm of thickness; protected to avoid re-wetting and be permanently moved to allow uniform drying, in order to reach a maximum safe moisture content of 12.5%.

76. For storage and transportation purposes only clean bags should be used for dried coffee.

77. Bags must be transported and loaded/unloaded only under dry weather or protected environment to prevent re-wetting.

78. Warehouses must be: protected against rain, well ventilated, isolated from humidity on ground floor, walls and ceiling. Dry green coffee must be stored in clean bags or containers; be disposed on pallets, above ground level, and distant from walls.

CONCLUSIONS & RECOMMENDATIONS:

79. The present Discussion Paper on OTA in coffee leads to the following broad conclusions and recommendations for consideration at the 1st Session of the Codex Committee on Contaminants in Foods:

I It is recommended that a Codex Code of Practice for the Prevention and Reduction of OTA in Coffee should be established based on FAO Guidelines for the Prevention of Mould Formation in Coffee.

II To ensure that all climatic and agricultural conditions are considered, it is appropriate that all coffee producing countries take part on the elaboration of the Codex Code of Practice draft.

III The necessity of setting a maximum level for OTA in coffee should be assessed after the Code of Practice has been developed and it should consider:

- There is a significant difference between the level of OTA contamination in green, roasted and soluble coffee.
- There are significant variations on the OTA reduction, depending on the technological process to which coffee is submitted.
- It is necessary to have reliable data on wide world exposure and occurrence after the Code of Practice has been implemented.

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ANNEX

Main Green Coffee Producing Countries(years 2000 to 2004) (source: FAOSTAT)

Green Coffee, production (Mt)	**2000**	**2001**	**2002**	**2003**	**2004**	**Total**
Brazil	1,903,562	1,819,569	2,649,610	1,996,850	2,475,780	10,845,371
Viet Nam	802,500	840,600	699,500	793,700	834,600	3,970,900
Colombia	636,000	656,160	690,840	694,080	663,660	3,340,740
Indonesia	625,009	575,160	698,589	702,274	702,274	3,303,306
México	338,170	302,996	313,027	310,861	310,861	1,575,915
Índia	292,000	301,000	301,000	275,000	275,000	1,444,000
Guatemala	312,060	275,700	221,820	244,200	216,600	1,270,380
Ethiopia	229,980	228,000	225,360	221,580	259,980	1,164,900
Côte d'Ivoire	336,273	209,000	182,001	140,027	159,769	1,027,070
Honduras	193,309	205,545	182,160	152,040	178,140	911,194
Uganda	143,475	197,410	189,000	150,871	186,000	866,756
Peru	158,283	159,936	178,285	169,548	176,137	842,189
Costa Rica	161,395	150,289	140,874	132,259	126,000	710,817
Ecuador	138,030	164,790	79,149	82,720	83,000	547,689
Philippines	107,557	112,271	107,080	106,388	100,911	534,207
El Salvador	114,087	112,201	91,513	91,513	78,510	487,824
Venezuela	78,440	91,877	76,946	64,265	65,559	377,087
Papua New Guinea	83,000	62,500	66,000	69,000	60,000	340,500
Nicaragua	82,206	66,799	60,235	59,659	70,909	339,808

According to FAOSTAT data, there are 78 green coffee producers, considering the years 2000 to 2004.
From these 78, a number of 19 producers were responsible for 90,08% of the total green coffee world production. The remaining 59 países, named below, were responsible for the other 9,92% of the total green coffee world production.

- **Africa** - S.Tome and Príncipe, Gabon, Benin, Comoros, Angola, Republic of Congo, Ghana, Mozambique, Liberia, Nigeria, Equatorial Guinea, Zimbabwe, Zambia, Malawi, Central African Republic, Togo, Sierra Leone, Guinea, Rwanda, Burundi, Cameroon, Tanzania, Democratic Republic of Congo, Madagascar, Kenya
- **America** - Suriname, Guadeloupe, Martinique, Belize, Guyana, St.Vincent/Grenadines, Trinidad Tobago, Dominica, Jamaica, Paraguay, United States of America, Puerto Rico, Panama, Cuba, Bolivia, Haiti, Dominican Republic
- **Asia** - Nepal, Cambodia, Myanmar, SriLanka, Yemen, Timor-Leste, China, Malaysia, Laos, Thailand
- **Oceania** – Cook Islands, Samoa, Vanuatu, Tonga, Fiji Islands, French Polynesia, New Caledonia