codex alimentarius commission

FOOD AND AGRICULTURE ORGANIZATION OF THE UNITED NATIONS

WORLD HEALTH ORGANIZATION

JOINT OFFICE: Viale delle Terme di Caracalla 00100 ROME Tel: 39 06 57051 www.codexalimentarius.net Email: codex@fao.org Facsimile: 39 06 5705 4593

Agenda Item 16(j) **CX/FAC 03/`35**

November 2002

JOINT FAO/WHO FOOD STANDARDS PROGRAMME

CODEX COMMITTEE ON FOOD ADDITIVES AND CONTAMINANTS
Thirty-fifth Session
Arusha, Tanzania, 17 - 21 March 2003

DISCUSSION PAPER ON DEOXYNIVALENOL

Governments and international organizations wishing to submit comments on the following subject matter are invited to do so **no later than 31 December 2002** as follows: Netherlands Codex Contact Point, Ministry of Agriculture, Nature Management and Fisheries, P.O. Box 20401, 2500 E.K., The Hague, The Netherlands (Telefax: +31.70.378.6141; E-mail: info@codexalimentarius.nl, with a copy to the Secretary, Codex Alimentarius Commission, Joint FAO/WHO Food Standards Programme, FAO, Viale delle Terme di Caracalla, 00100 Rome, Italy (Telefax: +39.06.5705.4593; E-mail: Codex@fao.org).

1. Comments on the text are requested, and more information is requested on the following specific issues:
- information on the occurrence of deoxynivalenol (DON) in cereals
- information on existing legal limits or guideline levels or action levels in foodstuffs and feedingstuffs
– information on the influence of processing
– information on decontamination, sorting etc. to lower the level of DON in a lot
– information on trade disruption (rejected lots)

2. Comments on the proposed maximum levels are requested.

Background

3. The 33rd session of the Codex Committee on Food Additives and Contaminants in 2001 agreed that a drafting group under the direction of Belgium and with assistance of Canada, Denmark, Germany, the Netherlands, Switzerland, the United States and the EC, would elaborate a Discussion Paper on Deoxynivalenol for consideration at its next Session (ALINORM 01/12A para. 197, ref 1).

4. The 34th session of the Codex Committee on Food Additives and Contaminants in 2002 agreed that the drafting group would revise the Discussion Paper on Deoxynivalenol while introducing, if possible, a proposal for a maximum limit for DON in cereals, for circulation, comment and further consideration at its next meeting (ALINORM 03/12 para 163, ref 2).

5. In circular letter CL 2002/10-FAC of April 2002 (ref 2), Codex Alimentarius requested additional information and data on the occurrence of DON in cereals, as well as results of any studies on the effects of processing, to send to the secretariat no later than 1 September 2002. No answers were received.

6. DON or vomitoxin belongs to the group of mycotoxins called trichothecenes and is produced by certain *Fusarium* species, especially *F. graminearum* and *F. culmorum* which are pathogens of several cereal grains. Trichothecenes are sesquiterpenoid compounds. DON is water-soluble and chemically very stable.

7. DON is primarily found as a contaminant of cereals and cereal products.

8. The European Mycotoxin Awareness Network (EMAN, ref 3) provides information on all areas of mycotoxins. It contains fact/information sheets, biannual newsletters, online training courses, lists of references and useful links.

Screening and analytical methods

9. A common misconception has existed in the grain trade that percentage *Fusarium*-damaged kernels can be used as a reliable basis for calculating DON concentration once the ratio of DON concentration to percent *Fusarium*-damaged kernels is known. Canadian studies showed that the potential for error is very large and that the relationships between DON concentration and percent *Fusarium*-damaged kernels are neither strong enough nor robust enough to predict DON in individual samples with a high degree of accuracy and precision. Head blight is not only caused by toxin producing *Fusarium* species (especially *F. graminearum*, and with a more limited toxin production *F. avenaceum*, and *F. poae*) but also by the non-toxin producer *Microdochium nivale* (formerly *Fusarium nivale*). Therefore on years when the predominant fungi responsible for head blight is *Microdochium nivale*, levels of DON and incidence of contamination are likely to be lower than those which could be suspected from the degree of head blight damage. More important, *Fusarium*-damaged kernels are not normally found in DON contaminated (1-5 ppm) Scandinavian wheat naturally and artificially infected with *Fusarium culmorum*. Looking at damaged kernels can thus not be used to predict DON.

10. The use of appropriate sampling techniques for obtaining a representative sample from a bulk lot of a commodity for DON analysis is a major factor for ensuring that the analytical test results will give a reliable estimate of the true concentration of DON in that lot. Other factors that may contribute to variations in analytical test results among samples from a given lot include errors associated with the sample preparation phase and the analytical procedure used.

11. Promising results are obtained with near-infrared spectroscopy for rapid DON testing (ref 4-5). Correlations ranging from 0.70 to 0.93 between DON levels predicted by near-infrared spectroscopy on both ground samples and whole kernel samples, to DON levels determined by GC technique for various hard and soft wheats, were reported. DON concentrations predicted by near-infrared testing of ground samples were in closer agreement with levels determined by GC-MS than for near-infrared testing of whole kernel samples. A fluorescence polarization immunoassay method was recently developed for DON in wheat. The method is easy to use and useful for screening of DON in wheat, however, it is not suitable for testing of maize (ref 6). A sensitive rapid ELISA test for DON in agricultural commodities has been developed that can detect levels down to 20 µg/kg and can quantitate DON in samples within 6 minutes (ref 7).

12. Analytical methods that are available for DON include GC-ECD, GC-MS, LC-UV, LC-fluorescence, LC-MS, TLC, ELISA and immunoaffinity column-fluorescence. Critical evaluations of chromatographic methods currently available for the analysis of DON in cereals were recently published (refs 5, 8-10). The results from a recent international interlaboratory study of an analytical method for DON and zearalenone in agricultural commodities revealed the desirability for further improvements in analytical procedures for these toxins in order to obtain more accurate measurements (ref 11). DON reference standards are often purchased as crystalline material or thin film. Therefore, before their use as calibrants, they are prepared gravimetrically in an organic solvent and stored in a freezer. The stability of DON in various organic solvents was recently studied, and it was observed that acetonitrile was the most suitable solvent for long-term storage of DON as a reference standard (ref 12). The analysis of trichothecenes including DON by gaschromatography have been recently studied in an EU-project "Intercomparison of trichothecene analysis and feasibility to produce certified calibrants and reference material" within the Standard, Measurement and Testing Programme. A high both between- and within-laboratory variation were found in the intercomparison studies. Several causes for the variation were identified. A matrix effect on the trichothecene response was the most prominent (ref 13-14).

13. Wide availability of reference standards and regular international comparative studies are needed to ensure improved internal and external quality assurances. The Institute for Reference Materials and

Measurements, Joint Research Centre of the European Commission, makes available BCR reference materials of DON in maize flour and wheat flour (BCR = European Community Bureau of Reference Materials, ref 15). The Food Analysis Performance and Assessment Scheme (FAPAS) from the UK included DON in wheat flour for 2001-2002 for testing laboratory proficiency. FAPAS has test material for DON in wheat which is available for use by laboratories for quality assurance purposes.

14. Methods of analysis for the determination of DON and other trichothecenes in cereals are under discussion for standardisation within the European Committee for Standardisation (CEN). Gas-chromatographic with flame ionisation detection (FID) and electron capture detection (ECD) and HPLC with UV detection methods have been studied. One HPLC-method with charcoal and immuno-affinity column clean-up has been collaboratively studied with relatively good results. No method has, however, yet been found robust enough for standardisation.

Occurrence

15. Surveys have shown that DON occurs frequently in grains such as wheat, barley, and maize and also in oats, rice, rye, sorghum, and triticale. The types of wheat affected by DON include both winter and spring varieties and hard and soft cultivars. Other trichothecenes and zearalenone do occur concomitantly with DON, but DON is usually the most predominantly occurring toxin.

16. *Fusarium* species can produce DON in the field and also during storage if the moisture content of grain kernels is high.

17. Local temperatures, rainfall and humidity are major factors for infections that occur at the time of flowering. The timing of rainfall, rather than the amount, is the most critical factor.

18. The analysis of wheat flours collected from commercial sources in Germany revealed that the median DON content was significantly higher for wheat flour originating from wheat of conventional as compared to organic production: 295 µg/kg and 120 µg/kg respectively (ref 16).

19. DON is primarily located in the outer parts of the cereals kernels which is consistent with the result of microscopical analyses which showed a preferred occurrence of *Fusarium* hyphae in aleurone and pericarp tissues (ref 16).

20. JECFA (ref 17) has assessed the levels and patterns of contamination of food commodities by DON on the basis of data received from Argentina, Brazil, Canada, China, Finland, Germany, Italy, the Netherlands, Norway, Sweden, the United Kingdom, Uruguay, and the USA and from data from the literature. DON was found to be a frequent contaminant of cereal grains such as wheat (11444 samples, 57% positive), maize (5349 samples, 41% positive), oats (834 samples, 68% positive), barley (1662 samples, 59% positive), rye (295 samples, 49% positive) and rice (154 samples, 27% positive). It was also detected in buckwheat, popcorn, sorghum, triticale and in some processed food products such as wheat flour, bread, breakfast cereals, noodles, baby and infant foods, and cooked pancakes, and also in malt and beer. The mean concentrations in data sets in which samples containing DON were found were 4-9000 µg/kg for barley, 3-3700 µg/kg for maize, 4-760 µg/kg for oats, 6-5100 µg/kg for rice, 13-240 µg/kg for rye, and 1-5700 µg/kg for wheat. A recent UK study revealed a very low incidence of DON in rice at retail outlets: 99 samples <10 µg/kg, one sample with 12 µg/kg (ref 18).

21. The following data are an example of wide annual variation and show that DON is present in finished products. In the Netherlands (ref 19) about 1200 samples of wheat and wheat containing products were sampled and analysed for DON between 1998 and September 2001, as part of the monitoring programme of the Inspectorate for Health Protection. Samples included products such as wheat, breakfast cereals, bread, pasta, wheat flour, and many other foodstuffs. The data show higher DON levels in wheat products prepared from wheat harvested in the 'wet' year 1998, compared to products from wheat harvested in the 'dry' years 1999 and 2000. In wheat harvested in 1998 a mean level of DON of 446 µg/kg was found (n= 216), whereas in wheat harvested in 1999 and 2000 the levels were 161 µg/kg (n=281) and 168 µg/kg (n=87) respectively. In bread, biscuit and crackers the mean level was found to be 220µg/kg in the 1998 harvest samples, and 118 µg/kg (17 samples) and 65 (µg/kg 22 samples) in the 1999 and 2000 harvest samples respectively. In baby

and toddler food (mainly (whole wheat) breakfast cereals) the DON levels were 949 µg/kg (28 samples) in the 1998 harvest samples, and 71 µg/kg (16 samples) and 140 µg/kg (5 samples) in the 1999 and 2000 harvest samples respectively.

22. Not only mean levels are interesting, but also knowledge of the distribution and especially knowledge on the incidence of high concentrations is important to take measures. As an example, the percentages of samples of wheat containing more than 250, 500, 750, 1000 and 1250 µg/kg, respectively, were for the wet harvest year 1998 (n=158) 66%, 41%, 26%, 19% and 15%, respectively. The figures for 2000 (n=602) were 35%, 18%, 10%, 5% and 2%, respectively (ref 19).

23. In Germany, several studies together containing 128 samples of the cereal based infant foods showed 92% of the samples had a DON level below 100 µg/kg. The mean level was 33 µg/kg.

24. Carry-over of DON to food products of animal origin does not appear of concern because animals refuse feed when the mycotoxin is present at high concentrations, and DON undergoes rapid metabolism and elimination in livestock species (JECFA 2001, ref 17).

25. The EC is undertaking a scientific co-operation task to gather all available data on occurrence of DON and other *Fusarium* mycotoxins in foodstuffs in the EC and to make dietary intake estimates. It is expected that the results of this study will become available in spring 2003 (ref 20).

Prevention

26. Document CX/FAC 02/21 on the draft Code of practice for the prevention of mycotoxin contamination in cereals, including annexes on ochratoxin A, zearalenone and fumonisin and including a new annex on trichothecenes has been discussed at the 34th session of the Codex Committee on Food additives and Contaminants in 2002 and sent to step 5 (para 125 and 177, ref 2).

27. Fungicides and insecticides may influence the presence of DON, but fungicides are mainly developed to control pathogenic fungi and are rarely if ever specifically targeted to control toxigenic fungi. One should be careful not to selectively remove dominant pathogenic but non-toxigenic species, allowing more active colonisation by toxigenic species such as *Fusarium graminearum* (ref 21). The timing and application rate are also important for optimum control. More information on the relationship between the use of fungicides and the occurrence of *Fusarium*-toxins can be found in the literature (ref 22-26).

28. Research on prevention is progressing (ref 27). Identification of antagonists may result in patenting of biocontrol agents. Research is going on about the efficacy of anti-oxidants and essential oils.

29. Agricultural practices such as crop rotation, plowing under or removing old seed heads, stalks and other debris will reduce the availability of crop residues in the soil and at the soil surface that can serve as nutrients for the saprophytic *Fusarium* species and hence will aid in the control of *Fusarium* head blight and DON contamination. It was observed that the incidence and severity of *Fusarium* head blight were greatest when wheat followed corn and least when wheat followed non-cereals in a crop rotation system (ref 28).

30. Another important means of preventing the contamination of wheat with DON is the use of wheat cultivars which are highly resistant to *Fusarium,* as well as reducing plant stress and appropriate drying after harvest.

31. The presence of DON cannot be completely avoided at the moment with the preventive measures and techniques currently available. However, the draft Code of Practice for the Prevention of Mycotoxin contamination in Cereals, which includes an annex on trichothecenes, should improve the situation. A Hazard Analysis Critical Control Point (HACCP) system should, when properly implemented, result in a reduction of the levels of mycotoxins, including *Fusarium*-toxins (ref 29-32).

Decontamination and effects of processing

32. DON is considered to be a stable compound, both during storage/milling and the processing/cooking of food. Therefore it can occur in foods prepared from DON-contaminated grains.

33. Post harvest safety management of wheat infected with *Fusarium* is an extremely important aspect of safety assurance. While strategic options are greatly limited by practical considerations, they involve developing ways to reduce, eliminate and control mycotoxin concentrations in commercial shipments and end-products.

34. Physical procedures for removing DON from contaminated grains, including cleaning, washing, dehulling, and milling have been successful to varying extents.The effectiveness of these procedures depend on the distribution of the toxin throughout the kernels as well as the level of contamination (ref 33-34). The effectiveness of various physical and chemical procedures for reducing DON contamination in grains and processed products was recently reviewed (ref 35). Simple washing procedures resulted in 65% to 69% reductions of DON in contaminated barley and corn. Washing might be a useful treatment to use prior to wet milling or ethanol fermentation, otherwise the cost of drying grains would be prohibitive (ref 36).

35. It was found that when infected wheat was simply separated into fractions of various size with laboratory sieves, DON was concentrated in the smaller fractions. The larger fractions had low levels of DON (ref 37). Conventional wheat cleaning equipment has been used to separate *Fusarium*-damaged kernels with varying degrees of success (ref 38). Specific gravity tables also appear to be effective. It was found that DON was heavily concentrated in gravity table fractions of lowest density (ref 39). The most dense fractions had much less DON than corresponding unfractionated wheat. Removing the least dense fraction also improved the milling properties of the remaining wheat. About 25% of DON contamination can be removed by cleaning and polishing whole kernels (ref 40). Bran obtained after polishing barley tends to contain higher concentrations of deoxynivelenol. (ref 36).

36. Dry milling is a process for separating grain components by grinding wheat into various particle sizes, the whitest flour having the lowest particle size. The degree of toxin contamination tended to be lower with decreasing ash content of wheat flour, i.e. the DON content tended to be lower in white compared to whole-grain flour. These results confirm the view of a localisation of *Fusarium* toxins primarily in the outer parts of grain kernels. The contents of DON in wheat flour can be reduced by 10-85% in comparison to the whole-grain (ref 16). Typically, the toxin remains in wheat flour at levels ranging from 60-80% of toxin level from the starting wheat (ref 40).

37. Wet-milling is a major process for obtaining starch for human consumption. When DON contaminated corn was processed through a commercial wet-milling process, high levels of DON were found in concentrated steep liquor fractions, low levels in germ, fiber and gluten fractions, and very low levels(close to detection limits) found in the starch fraction (ref 41).

38. Baking does not destroy or significantly reduce levels of DON (ref 40). DON reduction occurs during the fermentation step of bread making in an Argentinian process (ref 42). Similar results were obtained in a study involving different types of products in a low-technology bakery; a significant reduction was noted in the DON levels in the dough and final baked products analysed as a consequence of the bread making process (ref 43). Yeast doughnuts were shown to have higher concentrations of DON than in the flour used (ref 36). In studies involving ground maize, it has been found that moisture, especially when added as a basic solution and combined with heat, will give a useful reduction in the DON content of naturally contaminated ground maize (ref 44). A combination of heat and treatment with lime water in the process of making tortillas, reduced DON by 72% to 82% in two corn samples (cited by ref 36).

39. Because of its heat stability, DON content does not change significantly during extrusion. Cooking of spaghetti and noodles prepared from wheat reduced the level of DON to 53% (ref 45).

40. During the germination process of barley, 77% of DON can be destroyed in five days (ref 40). Nevertheless, malt may also contain more DON than the unmalted barley (ref 36). DON survives the brewing process and has been found in beers from various countries (ref 46).

41. Research is ongoing to evaluate the efficacy of physical adsorbents for decontamination of DON-contaminated grain intended for feed (ref 3, 27). Other decontamination procedures are also investigated. No commercial methods are currently available for the complete removal of DON from contaminated grains.

Toxicology

42. JECFA performed a risk assessment of DON in 2001 (ref 17). Available toxicological data did not suggest that DON presents a carcinogenic hazard. In animals, decreased feed consumption, diarrhoea and vomiting have been observed as acute effects. JECFA recognised that DON can lead to outbreaks of acute illness in humans. However, the available data did not permit setting an acute reference dose (level below which no acute effects would be expected to occur).

43. Reduced growth and suppression of the host resistance to infection with *Listeria monocytogenes* and *Salmonella enteritidis* have been seen as short or long term effects. JECFA established a provisional maximum tolerable daily intake (PMTDI) of 1 µg/kg body weight and concluded that intake at this level would not result in effects of DON on the immune system, growth, or reproduction. As the trichothecenes have similar toxic properties, albeit with different potencies, JECFA recommended that toxic equivalency factors relative to DON be developed for the other trichothecenes commonly occurring in cereal grains, if sufficient data become available.

44. The European Scientific Committee on Food expressed its opinion on DON on 2 December 1999 (ref 47). The general toxicity and the immunotoxicity of DON are considered to be the critical effects. A temporary TDI (tTDI) of 1 µg/kg bw was derived based on a chronic dietary study with mice (safety factor of 100). The tTDI was made temporary because the Committee wanted to discuss later the question whether a group TDI should be assigned for several trichothecenes. The Committee has now assessed trichothecenes as a group and concluded that the available data did not support the establishing of a group TDI nor the establishment of toxic equivalency factors relative to DON. A full TDI for DON at 1 µg/kg bw/day was established (ref 48).

Exposure and risk characterisation

45. From the submitted occurrence data, JECFA estimated the dietary intake of DON on the basis of the single weighted mean concentrations for each commodity and the GEMS/Food regional diets. However, it should be noted that there was incomplete coverage for regions outside of the European region (the GEMS/Food European region includes North America), and that data from the European region was used to estimate concentrations in other regions. This use of European data could have lead to either an over- or under-estimate of exposure in regions other than the European region. The mean intake estimates (ranging from 0.77 to 2.4 µg/kg of body weight a day) values exceeded the PMTDI for four of the five regional diets. The Committee noted that there was considerable uncertainty in the intake estimates. However, the exceedance of the PMTDI by mean intakes in 4 of 5 regions suggests that it is quite likely that the PMTDI is exceeded by a substantial percentage of the world's population. It should be noted that possible reductions in levels of DON as a result of processing were not taken into consideration in this assessment.

46. The European Scientific Committee in 1999 observed that the intake of DON from cereals and beer in the Scandinavian countries and from cereals in the Netherlands has been estimated to be in the order of the tTDI (ref 47, 49-50).

47. The Swiss Federal Office of Public Health estimated in 1997 the <u>mean</u> intake of DON in Switzerland: Adults: ≤ 170 ng/kg bw/day and young children: ≤ 800 ng/kg bw/day.

48. In the Netherlands, both the National Institute for Public Health and the Environment (RIVM) and the Dutch Health Council identified children as the population group most at risk to exceed the TDI (refs 19, 51). Of the 1-year old children, 80% exceeded the TDI and 20% exceeded 2xTDI. The intake of the 95 percentile of 1-year old children was 3 µg/kg bw. For both adults and children bread is the most important food group contributing to the intake. For 1-year old children specific infant food also contributed significantly.

49. In the middle of China (Henin province), where a human intoxication episode caused by red mold occurred, DON was the predominant toxin detected abundantly and frequently at a level of up to 14 000 µg/kg (mean 2850 µg/kg) in 30 of 31 (97%) wheat samples taken from the 1998 Puyang crop (ref. 52).

Maximum levels in food

50. The EC recently recommended an action level of 500 µg/kg for cereal foodstuffs as consumed and other cereal products at retail stage, and an action level of 750 µg/kg for flour used as raw material in food products. Such actions levels have been in use in *e.g.* the Netherlands since 2000. For some time, Austria has had a guideline level of 750 µg/kg for durum wheat and 500 µg/kg for wheat and rye. Germany recently notified a draft legislation with the following maximum levels: cleaned cereals for direct human consumption, cereal based products and pasta, not including durum wheat and durum wheat based products (because their database on durum wheat was considered to be insufficient): 500 µg/kg; bread, biscuits and pastries: 350 µg/kg; cereals based foodstuffs for infants and young children: 100 µg/kg (ref 53).

51. Switzerland adopted a guideline level of 1000 µg/kg for DON in cereals in March 1998. This guideline level is valid for cereal products and for cereals as sold to the consumer, but does not apply to raw cereals.

52. Canada has a guideline level of 2000 µg/kg for DON in uncleaned soft wheat, corresponding to 1200 µg/kg in the flour portion (for the manufacture of non-staple foods such as cakes, cookies, biscuits). With respect to uncleaned soft wheat intended for use in infant foods, the guideline is 1000 µg/kg corresponding to 600 µg/kg in the flour portion. Since levels of DON in hard wheat, the major Canadian wheat, are generally low, no guidelines have been established for DON for this type of wheat, nor for other grains.

53. The US has a guideline value of 1000 µg/kg for finished wheat products.

54. Russia has a maximum level of 1000 µg/kg for DON in cereals (wheat of hard and strong types), flour and wheat bran (ref 54).

55. In China, the regulatory limit for DON in cereals for human consumption is 1000 µg/kg (ref 52).

Maximum levels in feed

56. DON undergoes rapid metabolism and elimination in livestock species and is not known to occur at significant levels in foodstuffs of animal origin. Maximum levels in feed are therefore not needed to protect public health, but might be useful to protect animal health.

57. Sweden applies guideline levels of 4000 µg/kg for DON in cereal products used as feed material for animal feedingstuffs, 400 µg/kg in feedingstuffs for pigs and 2000 µg/kg in feedingstuffs for cattle and poultry.

58. Belgium uses a guideline level of 5000 µg/kg for DON in cereals and cereal by-products used as raw materials for animal feedingstuffs.

59. Austria recommended the following guideline levels: 500 µg/kg in feedingstuffs for pigs, 1,000 for µg/kg for feed for beef cattle and for feed for laying hens and breeding poultry, and 1,500 µg/kg for feed for fattening poultry.

60. Germany has orientation values for DON of 1000 µg/kg for pig feed, 2,000 µg/kg for calve feed, 5,000 µg/kg for bovine and poultry feed.

61. In the Netherlands, the following action levels are applied. For cereals: for pigs, for laying hens, for calf and dairy cattle: 5,000 µg/kg, for other cattle and poultry: 10,000 µg/kg. For compound feed: for pigs: 1,000 µg/kg, for calf and dairy cattle: 2,000 µg/kg, for laying hens: 3,000 µg/kg, for other cattle and poultry: 5,000 µg/kg.

62. The US has the following guideline levels for DON in feed for various animal species: 10000 µg/kg for DON in grains and grain by-products destined for ruminating beef and feedlot cattle older than 4 months and for chickens, with the added recommendation that these ingredients not exceed 50% of the diet of cattle or chickens; 5000 µg/kg for DON in grains and grain by-products destined for swine with the added recommendation that these ingredients not exceed 20% of their diet; and 5000 µg/kg for DON in grains and grain by-products destined for all other animals with the added recommendation that these ingredients not exceed 40% of their diet.

63. Canada recommends a maximum level of 5,000 µg/kg for DON in feedingstuffs for cattle and poultry and a maximum of 1,000 µg/kg for DON in feedingstuffs for swine and young calves and lactating dairy animals (ref 54).

Trade disruption

64. In view of the worldwide contamination of cereals with DON, considering the fact that cereals are important in international trade and that different countries apply different rejection levels, it is expected that DON levels in commodities involved in international trade are a matter of concern.

65. Economic losses due to DON can be high. Nearly 40% of the 1990 crop in western and central New York was not sold because of this contamination (ref 55).

Other legitimate factors

66. Foodstuffs containing wheat are staple foods and are a good source of a number of essential nutrients. The Dutch Health Council therefore advises against measures to decrease wheat consumption as a way to reduce DON exposure (ref 19).

Conclusions and recommendations

67. The appropriateness of setting of maximum levels of DON in foodstuffs derived from cereals should be discussed in CCFAC, as a result of the JECFA evaluation indicating that the TDI is exceeded in four of the five regional diets. Prevention of contamination is currently not sufficiently achievable, although the draft Code of Practice for the Prevention of Mycotoxin Contamination in Cereals, which includes an annex on trichothecenes, should improve the situation. However, the setting and implementation of maximum levels in conjunction with good agricultural practices, should contribute to the reduction of mean DON levels by preventing the marketing and consumption of highly contaminated foodstuffs.

68. In case CCFAC opts for the setting of maximum levels for DON complementary to the development of the Code of Practice, CCFAC should identify the food groups for which the setting of maximum levels could be appropriate, e.g. for (raw) cereals and cereal products and for cereal based infant foods. It can be discussed whether maximum levels should be set for the consumer products, as these have a direct impact on the exposure, and/or for raw cereals, important in international trade.

69. Setting maximum levels for consumer products only, leaves more possibilities to efforts of sorting etc. in years with high levels of contamination, and might therefore better ensure the food supply and free trade whilst protecting public health. Setting harmonised maximum levels for raw cereals provides clear guidance and transparency for the international trade. However, it is clear that the setting of a maximum level for raw cereals only, taking into account the achievability also in years with high levels of contamination, will not necessarily sufficiently protect the consumer. Therefore the appropriateness of setting in Codex Alimentarius of maximum levels for all cereal-based foodstuffs should be discussed. Finally, the maximum levels have to be based on the ALARA principle. The year to year variation has to be taken into account in order not to endanger the food supply during years with high levels of DON contamination. On the other hand, it has to be considered that cereals can be used for multiple purposes, so that cereals exceeding the maximum levels for food can be redirected to other appropriate uses and therefore not necessarily result in a complete loss of the affected cereals.

70. Taking into account the above mentioned considerations, the following maximum levels for DON are proposed for discussion:

a) raw cereal grains, to be subjected to sorting or other physical treatment (e.g. starch production) before human consumption or use as an ingredient in foodstuffs (after which the DON levels should comply with the other relevant maximum level): 2000 µg/kg

b) all products derived from cereals (e.g. flour, processed cereal products) including cereal grains intended for direct human consumption, except infant food: 500 µg/kg

c) cereal-based infant food: 100 µg/kg

Mixing of lots with the aim to decrease the contamination level below the maximum level should not be allowed. The <u>mean</u> level of DON in all cereal products consumed during a relatively long period of time (chronic risk) should remain significantly below the maximum level of 500 µg/kg, to ensure that public health is sufficiently protected.
For cereal based infant food, a lower level is appropriate as infants are the most vulnerable group (growth retardation as critical toxicological effect). This low level is achievable by carefully selecting the cereals to be used for the manufacturing of infant food.
It should be determined whether these proposals correspond to the ALARA level considering all scientific data and all legitimate factors. Data are needed from different geographical areas and from several years.

71. CCFAC could discuss the development of a harmonised sampling plan for testing DON in cereals. Additional data would be welcome on the distribution of DON in a contaminated cereal grain lot.

References

1. ALINORM 01/12A Report of the 33rd session of the Codex Committee on Food Additives and Contaminants, 12-16 March 2001. http://www.codexalimentarius.net/
2. ALINORM 03/12. Report of the 34th session of the Codex Committee on Food Additives and Contaminants, 11-15 March 2002 http://www.codexalimentarius.net/
3. http://www.mycotoxins.org/ European Mycotoxin Awareness Network
4. Williams *et al.* Near-infrared prediction of deoxynivalenol in wheat, pages 9-11 in: Proc. 1996 Regional *Fusarium*/scab forum. R. Clear, ed. Canadian Grain Commission, Winnipeg, MB., 1996.
5. Van Osenbruggen, WA, and Pettersson, H. Analysis of relevant *Fusarium* mycotoxins in cereals-state of the art. In: Scholten et al (eds) Food safety of cereals: a chain-wide approach to reduce *Fusarium* Mycotoxins. European Commission, pp 41-49, 2002.
6. Maragos, C.M., and Platner, R.D. Rapid fluorescence polarization immunoassay for the mycotoxin deoxynivalenol in wheat. J. Agric. Food Chem. 50:1827-1832, 2002.
7. Abouzied, M. A very sensitive rapid ELISA test for the detection and quantitation of the trichothecene mycotoxin deoxynivalenol (DON). In: Proceedings of the X International IUPAC symposium on mycotoxins and phycotoxins, 21-25, Sao Paulo, 2000.
8. Mateo, J.J., *et al.* Critical study of and improvements in chromatographic methods for the analysis of type B-trichothecenes. J Chromatography A 918:99-112, 2001.
9. Krska, R., *et al*., The state-of-the art in the analysis of type–A and –B trichothecene mycotoxins in cereals. Fresenius J. Analytical Chemistry 371: 285-289, 2001.
10. Lombaert, G.A. Methods for the determination of deoxynivalenol and other trichothecenes in foods. Adv. Exp. Med. Biol. 504:141-153, 2002.
11. Josephs, R. D., *et al.* International interlaboratory study for the determination of the *Fusarium* mycotoxins zearalenone and deoxynivalenol in agricultural commodities. Food Addit. Contam. 18(5): 417-430, 2001.
12. Widestrand, J. and Pettersson, H. Effect of time, temperature and solvent on the stability of T-2 toxin, deoxynivalenol and nivalenol calibrants. Food Addit. Contam. 18(11):987-992,2001.
13. Pettersson, H., and Langseth, W. Intercomparison of trichothecene analysis and feasibility to produce certified calibrants and reference material. Final report I. Method Studies. BCR Information, Project Report EUR 20285/1 EN, pp 1-82, 2002.
14. Pettersson, H., and Langseth, W. Intercomparison of trichothecene analysis and feasibility to produce certified calibrants and reference material. Final report I. Homogeneity and stability studies, final intercomparison. BCR Information. EU Project Report EUR 20285/2 EN, pp1-145, 2002.
15. http://www.irmm.jrc.be/mrm.html
16. Schollenberger, M., *et al.*, *Fusarium* toxins in wheat collected in an area in southwest Germany. Int. J. Food Microbiol., 72:85-89, 2002.
17. JECFA, fifty sixth meeting, February 2001 http://www.who.int/pcs/jecfa/summaries.htm
18. Food Survey Information Sheet 22/02, 2002. http://www.foodstandards.gov.uk/multimedia/pdfs/22rice.pdf
19. Health Council of the Netherlands, Deoxynivalenol (DON). The Hague: Health Council of the Netherlands, 2001; publication no. 2001/23.
20. Commission Decision 2002/916/EC: inventory and distribution of tasks to be undertaken within the framework of co-operation by Member States in the scientific examination of questions relating to food. Official Journal of the European Communities, L319, page 28, 23.11.2002. http://europa.eu.int/eur-lex/
21. Opinion on the relationship between the use of plant protection products on food plants and the occurrence of mycotoxins in foods, adopted on 24 September 1999 by the Scientific Committee on Plants of the European Commission. http://europa.eu.int/comm/food/fs/sc/scp/out56_en.html
22. D'Mello, J.P.F., *et al.* Pesticides use and mycotoxin production in *Fusarium* and *Aspergillus* phytogens. Eur. J. Plant Pathol., 104:741-751, 1998.
23. Gareis, M. and Ceynowa, J. Influence of fungicide Matador (tebuconazole/triadimenol) on mycotoxin production by *Fusarium culmorum*. Z. Lebensm. Unsters. Forsch., 198:244-248, 1994.
24. Miedaner, T. and Reinsbrecht, C. Trichothecene content of rye and wheat genotypes inoculated with a deoxynivalenol and nivalenol-producing isolate of *Fusarium culmorum*. J. Phytopathol., 149:245-251, 2001.

25. Jennings, P., *et al*. Overview of *Fusarium* ear blight in the UK. Effect of fungicide treatment on disease control and mycotoxin production. In: Proceedings of the Brighton crop Protection Conference: Pests and Diseases, Farnham, UK. BCPC Publications, Volume 2, pp 707-712, 2000.
26. Jennings, P. Control of the fungus through the use of pesticides. In: Scholten et al (eds) Food Safety of cereals: a chain-wide approach to reduce *Fusarium* Mycotoxins. European Commission, pp 22-24, 2002.
27. http://www.mycotoxin-prevention.com/
28. Dill-Macky, R. and Jones, R.K. The effect of previous crop residues and tillage on *Fusarium* head blight of wheat. Plant Disease 84:71-76. 2000.
29. FAO/IAEA. Manual on the application of the HACCP system in mycotoxin prevention and control. Food Nutrition Paper No. 73, Food and Nutrition Division, FAO, Rome, 2001.
30. Wareing, P. The application of the hazard analysis critical control point (HACCP) approach to the control of mycotoxins in foods and feeds. Post Harvest News & Information, 10(3):29N-33N, 1999.
31. Park *et al*. Minimising risks posed by mycotoxins utilising the HACCP concept. Third joint FAO/WHO/UNEP International Conference on Mycotoxins, Tunis, 3-6 March 1999.
32. Halacker, R. Mycotoxins and the HACCP concept. Brauwelt, 40:1820-1825, 1998.
33. Charmley, L.L., and Prelusky, D.B. Decontamination of *Fusarium* mycotoxins. IN: Miller, J.D, Trenholm, H.L.(Eds.) Mycotoxins in Grain. Compounds Other Than Aflatoxin. Eagen Press, St. Paul MN, pp 421-435,1994.
34. Pomeranz *et al.*, in: Advances in Cereal Science and Technology, Pomeranz (ed), AACC, St Paul, USA, Vol. X, pp 373-433, 1991
35. Trigo-Stockli, D.M. Effect of processing on deoxynivalenol and other trichothecenes. Adv. Exp. Med. Biol. 504:181-188, 2002.
36. Visconti, A. and De Girolamo, A. *Fusarium* mycotoxins in cereals: storage, processing and decontamination. In: Food Safety of cereals: a chain-wide approach to reduce *Fusarium* mycotoxins. Scholten et al. European Commission. 2002.
37. Chelkowski and Perkowski. Mycotoxins in cereal grains, 15, Distribution of deoxynivalenol in naturally contaminated wheat kernels. Mycotoxin Res. 8: 27-30, 1992.
38. Pomeranz *et al*., *Fusarium* head blight (scab) in cereal grains, pages 373-433 in: Advances in Cereal Science and Technology, 1990, Vol. X, Y. Pomeranz, ed. Am. Assoc. Cereal Chemists, St. Paul, MN.
39. Tkachuk *et al*. Removal by specific gravity table of tombstone kernels and associated trichothecenes from wheat infected with *Fusarium* head blight, 1991, Cereal Chem., 68: 428-431.
40. Bennett, G.A., and Richard, J.L. Influence of processing on *Fusarium* mycotoxins in contaminated grains. Food Technology, 50(5):235-239, 1996.
41. Lauren, D.R. and Ringrose, M.A. Determination of the fate of three *Fusarium* mycotoxins through wet-milling of maize using an improved HPLC analytical technique. Food Addit. Contam. 14(5):435-443,1997.
42. Samar, M.M., *et al.* Effects of fermentation on naturally occurring deoxynivalenol (DON) in Argentinean bread processing technology. Food Add. Contam., 18 (11): 1004-1010, 2001.
43. Neira, M.S., *et al.* The effects of bakery processing on natural deoxynivalenol contamination. Int. J. Food Microbiol. 37(1):21-25,1997.
44. Lauren, D.R., and Smith, W.A. Stability of the *Fusarium* mycotoxins nivalenol, deoxynivalenol and zearalenone in ground maize under typical cooking environments. Food Addit. Contam. 18(11): 1011-1016, 2001.
45. Nowicki, T.W. *et al.* Retention of the *Fusarium* mycotoxin deoxynivalenol in wheat during processing and cooking of spaghetti and noodles. J. Cereal Sci., 8:189-202 (1988).
46. Scott, P.M. Mycotoxins transmitted into beer from contaminated grains during brewing. J AOAC Intl. 79(4): 875-882, 1996.
47. Opinion on *Fusarium* Toxins. Part 1: Deoxynivalenol (DON), expressed on 2 December 1999 by the Scientific Committee on Food of the European Commission. http://europa.eu.int/comm/food/fs/sc/scf/out44_en.html
48. Opinion on *Fusarium* toxins. Part 6: Group evaluation of T-2 toxin, HT-2 toxin, nivalenol and deoxynivalenol, adopted on 26 February 2002 by the Scientific Committee on Food of the European Commission. http://europa.eu.int/comm/food/fs/sc/scf/out123_en.pdf
49. Eriksen, G.S. and Alexander, J. (eds.) *Fusarium* toxins in cereals – a risk assessment. Nordic Council of Ministers. TemaNord 502, Copenhagen, 1998.
50. Pieters *et al.* Deoxynivalenol. Derivation of concentration limits in wheat and wheat containing products. RIVM Report 388802018, The Netherlands, 1999.

51. Risk assessment of deoxynivalenol in food. An assessment of exposure and effects in the Netherlands. RIVM report 388802022. 2001. Pieters et al.
52. Li, F-Q *et al*., *Fusarium* toxins in wheat from an area in Henan Province, PR China, with a previous human red mold intoxication episode. Food Add. Contam., 19(2):163-167, 2002.
53. Notification 2002/138/D http://europa.eu.int/comm/enterprise/tris/
54. FAO. Worldwide regulations for mycotoxins 1995. A compendium. Food and Nutrition paper 64. 1997.
55. Visconti, A. Poblems associated with *Fusarium* mycotoxins in cereals. 2002. http://www.mycotochain.org