

CHAPTER 07-Molecular Biology and Natural Toxins

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|---------------------------|---|
| SUBJECT: | Mycotoxins in Domestic and Imported Human Foods |
| IMPLEMENTATION DATE: | Upon Receipt |
| Product Codes: | See Attachment A |
| Product/Assignment Codes: | 07001-All domestic and import sample collections and analyses |

FIELD REPORTING REQUIREMENTS:

1. Reporting under PAC 07001 is required for all domestic and imported food samples collected and analyzed for mycotoxin contamination unless samples were collected during operations conducted under other compliance programs (e.g., Juice HACCP).
2. The analyzing laboratory will report analytical results into FACTS/uALIS using PAF = "MYC" and ensure that the correct Mycotoxin Code for mycotoxin analyzed is selected in the reporting system.
3. When entering information into FACTS / uALIS / Import Systems:
 - use operation code 31 to report domestic sample collection and operation code 41 to report domestic sample analysis.
 - use operation code 33 to report import sample collection and operation code 43 to report import sample analysis.
4. Refer to Part V for sample classification instructions.

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Change History

| Item | Change | Date |
|------|--|-----------|
| | <ul style="list-style-type: none">• Updated Format to reflect CP Changes• CSOs no longer need to select specific mycotoxins during sampling process• Added information on the Multi-Mycotoxin Method• Added information for additional mycotoxins of concern (Zearalenone and T-2/HT-2 toxins)• Updated Program contacts• Addition of Mycotoxin Resource Page• Updated instructions for labs to determine methodology to utilize for samples | 9/23/2024 |

PART I – BACKGROUND

1. General

Mycotoxins are toxic metabolites produced by certain fungi that can infect and proliferate on various agricultural commodities in the field and/or during storage and transit. The occurrence of these toxins on grains, nuts, and other commodities susceptible to mold infestation is influenced by environmental factors such as temperature, humidity, and extent of rainfall during the pre-harvesting, harvesting, and post-harvesting periods. Mycotoxins may exhibit various toxicological manifestations; some are teratogenic, mutagenic, and/or carcinogenic in susceptible animal species and are associated with various diseases in domestic animals, livestock, and humans in many parts of the world.

The occurrence of mycotoxins in food is not entirely avoidable; small amounts of these toxins may be legally permitted in human and animal foods. This Compliance Program (CP) solely covers human foods, and a separate CP covers animal foods (currently under development). Strategies used by the Food and Drug Administration (FDA) to minimize mycotoxins in the United States (U.S.) food supply include establishing guidelines (e.g., limits established in compliance policy guides (CPGs)) and monitoring the food supply by collecting and analyzing domestic and import foods. The data obtained over the years from FDA's monitoring programs are used to provide: (a) estimates of the incidence and levels of contamination by various mycotoxins in affected areas in the country, (b) dietary exposure data (estimates) for use in making risk assessments for specific mycotoxins, (c) background data for use in considering the establishment of guidelines for specific mycotoxins, (d) information on the co-occurrence of mycotoxins in a single commodity, (e) an estimate of the economic impact of the enforcement of regulatory guidelines on foods during a given crop year, (f) information needed to prepare answers to questions including congressional inquiries, and (g) basic information needed to support the position and recommendations of U.S. delegates participating in international meetings. The monitoring data also serves as a database describing the background distribution of various mycotoxins in domestic grains and their products in the U.S. as a function of geographic area and environmental conditions.

Advances in technology and methods of analysis now allow the FDA to examine food products for more than one mycotoxin in a single analysis. Therefore, the FDA is implementing compendial method C-003 which allows for simultaneous detection of multiple mycotoxins while preserving the ability to utilize legacy methods for individual mycotoxins covered by this CP. This new method is able to detect currently monitored mycotoxins in human food (aflatoxins, fumonisins, deoxynivalenol (vomitoxin), ochratoxin A), and new ones (zearalenone, T-2 toxin and its deacetylated analogue, HT-2 toxin). Patulin and aflatoxin M₁ are not included in C-003.

2. Specific Mycotoxins to be included in this Program

- A. Aflatoxins (AFL) - (Aflatoxin B₁, Aflatoxin B₂, Aflatoxin G₁, Aflatoxin G₂, and Aflatoxin M₁)** - are metabolic products of the molds *Aspergillus flavus* and *A. parasiticus* that may occur in food as a result of mold growth in a number of susceptible commodities, including peanuts, corn, tree nuts, and some small grains. Aflatoxin M₁ contamination of milk can occur through consumption of aflatoxin B₁-contaminated animal food by dairy cows. Aflatoxin B₁ is metabolized and excreted as aflatoxin M₁ in milk. Aflatoxins are known carcinogens in susceptible laboratory animals and in humans; therefore, the presence of aflatoxins in foods should be restricted to the lowest practical levels attainable using modern processing techniques.

The current CPG limits for aflatoxins can be found in the appropriate section of the CPG. See Part V for CPG references. Historically, aflatoxin levels in peanuts and corn have been highest in the Southeastern states. Corn from anywhere in the U.S. may be affected, however, contamination levels will depend on the growth, harvesting and storage conditions involved.

- B. Patulin (PAT)** - is a toxic substance produced by *Penicillium*, *Aspergillus*, and *Byssoschlamys* molds that may grow on apples, and may be present if rotten, moldy or damaged apples are used to make apple juice. Patulin is not destroyed by heat processing, and can occur at high levels in apple juice, including pasteurized apple juice, therefore both pasteurized and non-pasteurized single strength juice and concentrated juices are to be collected. Animal feeding studies have demonstrated that high levels of patulin in apple juice could pose a health risk if the juice is consumed over an extended period of time. In 2001, FDA established a limit for patulin in apple juice and in the apple juice component of a food that contains apple juice as an ingredient ([CPG Sec 510.150 Apple Juice, Apple Juice Concentrates, and Apple Juice Products - Adulteration with Patulin | FDA](#)).
- C. Deoxynivalenol (DON)** - commonly called vomitoxin, is a natural toxin produced by several molds of the genus *Fusarium*, especially *F. graminearum*, which is a common contaminant of several grains, including wheat, corn, barley, and rye. DON has been associated with a number of adverse health effects in humans and animals. Several adverse weather-related DON contamination episodes in the U.S. caused the FDA to issue advisory levels for human food (wheat) and animal food in 1982 and updated levels in 1993. A review of the scientific literature since 1993 revealed that higher levels of DON in food for cattle would not appear to present an animal or public health hazard, therefore, a revised advisory level for animal foods was issued in 2010 (See Part V for guidance references). FDA is continuing to study the scope and toxicological significance of the DON problem to determine if further regulatory measures are needed to control DON in human and animal food products.
- D. Fumonisin (FUM) - (Fumonisin B1, Fumonisin B2, and Fumonisin B3)** - are natural toxins produced by *Fusarium verticillioides* (previously known as *F. moniliforme*), and other

Fusarium species; these molds are common natural contaminants of corn. Fumonisin have been linked to fatalities in horses and swine. Recent studies have demonstrated the presence of fumonisins in human foods, including corn meal and breakfast cereals. Epidemiological investigations demonstrating a possible association of *F. verticillioides* with esophageal cancer and recent animal studies indicating the carcinogenicity of fumonisin B1 have highlighted the need to ensure that foods do not contain excessive amounts of fumonisins. Dry milling of whole corn kernels generally results in the production of fractions called bran, flaking grits, grits, meal, and flour. Because fumonisins are concentrated in the germ and the hull of the whole corn kernel, dry milling results in fractions with different concentrations of fumonisins. For example, dry milled fractions (except for the bran fraction) obtained from degermed corn contain lower levels of fumonisins than dry milled fractions obtained from non-degermed or partially-degermed corn. Industry information indicates that dry milling results in fumonisin-containing fractions in descending order of highest to lowest fumonisin levels: bran, flour, meal, grits, and flaking grits.

- E. Ochratoxin A (OTA)** - is a naturally occurring nephrotoxic fungal metabolite produced by certain species of the genera *Aspergillus* and *Penicillium*. It is mainly a contaminant of cereals (corn, barley, wheat, and oats), and has been found in edible animal tissues as well as in human blood sera and milk. Studies indicate that this toxin is carcinogenic in mice and rats. It is not completely destroyed during the processing and cooking of food, therefore the implication of risk to human health and safety must be considered. FDA needs up-to-date information on the incidence and levels of occurrence of this toxin in human food for use in considering any necessary regulatory control measures for this substance.
- F. T-2 and HT-2 toxins** - are type A trichothecenes produced by *Fusarium* species which invade cereal grains and produce the toxins under cool and moist conditions. Exposure to toxins may occur through surface contact or ingestion. Toxic effects in humans at levels of concern include acute gastroenteritis, high fever, hemorrhage, necrosis of muscles and skin, and possible asphyxiation. Toxic effects in animals also include disrupted ovulation and semen production.
- G. Zearalenone (ZEA)** - is an estrogenic metabolite produced by *Fusarium* species commonly found in corn, oats, wheat, sorghum, and rice. It has been linked to both reproductive and non-reproductive effects in both humans and animals.

PART II – IMPLEMENTATION

1. Objectives

- To collect and analyze domestic and import samples of various human food products to determine the occurrence and levels of aflatoxins, patulin, fumonisins, deoxynivalenol, ochratoxin A, T-2 and HT-2 toxins, and zearalenone.
- To collect monitoring and incidence data to support establishment of future limits for fumonisins, deoxynivalenol, ochratoxin A, T-2 and HT-2 toxins, and zearalenone in foods.
- To take appropriate regulatory actions against violative products when warranted.

2. Program Management Instructions

A. Planning instructions

To implement the program, Division personnel are directed to collect samples of selected foods and analyze them for mycotoxins as defined in Part I of this document. Each fiscal year, FDA's Center for Food Safety and Applied Nutrition (CFSAN), Office of Compliance issues a [Sample Collection Operation Planning Efforts \(SCOPE\)](#), which identifies the food, priority analyte, and the number of samples to be collected by each division. **NOTE:** Laboratories will determine if the sample will be analyzed by the multi-mycotoxin method or utilize Attachment A for priority analytes for the sampled product.

FDA has established advisory, guidance or action levels for some mycotoxins in certain food products. CFSAN will evaluate sample results for these mycotoxins on a case-by-case basis and may recommend follow-up, as appropriate.

B. Interactions with Other Compliance Programs

This compliance program may have some interactions with [CP 7303.847 Juice HACCP Inspection Program](#). Use the appropriate PAC when reporting sample collections under this compliance program.

The compliance program may have some interactions with the Center for Veterinary Medicine (CVM) animal food program. Please refer to C.P. 7371.000 – Comprehensive Animal Food Inspection for guidance. Before collecting animal food mycotoxin samples, contact the rTAN at: cvmanimalfoodprograms@fda.hhs.gov.

“For-cause” patulin samples or samples collected for verification during a juice HACCP inspection would be covered by CP 7303.847.

Patulin samples collected outside of a “for-cause” or verification situation during an inspection, e.g., from SCOPE, would be covered by CP 7307.001.

C. Resource Instructions

Resources for sample collections and analysis for mycotoxins in foods are provided in the [FDA Field Workplan](#).

D. Interactions with Other Federal Agencies, State and Local Counterparts, and Foreign Authorities

- i. State officials are valuable sources of information on current and potential aflatoxin problems in foods. In the past, a number of states have participated in a data exchange program with FDA. Divisions should encourage state participation via State Liaisons in this data exchange program and should coordinate activities with State officials to prevent duplication of efforts in both domestic human and animal food sampling.
- ii. Divisions will collaborate with commissioned state agencies to make them aware of the requirements of the program (in advance of the beginning of the program) and deadlines for deliverables. Divisions will offer state agencies an opportunity to assist FDA with sample collections as necessary. State laboratories may share violative results with FDA. After reviewing the results, FDA may follow-up.
- iii. USDA has a comprehensive testing program for aflatoxin in raw peanuts. USDA provides FDA with a quarterly summary of the total number of lots sampled, range of levels of total aflatoxins found, and a list of the applicants for each lot containing total aflatoxins above 15 parts per billion (ppb) (ng/g). Details on individual lots will be made available to FDA upon request (see [MOU 225-19-031](#)).
- iv. Foreign Authorities: The FDA works with foreign governments and international standard-setting bodies to harmonize food safety laws, regulations and standards based on science. Further, Divisions may review the information found at [International Cooperation on Food Safety](#).

PART III - INSPECTIONAL

1. Operations

A. Inspections

NOTE: This program does not direct inspections. However, Divisions may conduct follow up inspections, at the request of CFSAN Program Contacts, if mycotoxin amount exceeds established level of concern.

Inspectional instructions for mycotoxin inspections are contained in Section 8 of the *Guide to Inspections of Manufacturers of Miscellaneous Food Products Volume II*. This guide is available online at:

<http://www.fda.gov/ICECI/Inspections/InspectionGuides/ucm074988.htm>

B. Sample Collections

General Instructions:

- The [Sample Collection Operation Planning Effort \(SCOPE\)](#), consistent with the current [FDA Field Work Plan](#), will be issued at the beginning of each fiscal year. **Attachment A** of this compliance program list acceptable products for mycotoxin analysis. The collecting division should contact the CFSAN Compliance Program Contact early in the fiscal year if there may be difficulties in completing the workplan-directed collections. .
- Collect subsamples randomly to obtain a sample that is representative of the lot. Mycotoxin contamination can occur in localized pockets at high concentrations in foods such as unprocessed grains and nuts (see more details in Chapter 4 Sample Schedule Chart 6 of the IOM). Ensure sub-samples are not co-mingled during sampling, and sample unit integrity is maintained. For sampling bulk products, representative samples should be obtained by using a trier or other device that will provide representative portions from all sections of the container sampled. Commodities such as fruit juice, other fluid items, and mixed preparations (paste, spreads, butters) are generally homogenous and do not require any special devices for sampling.
- Do not collect more than two samples of any specific commodity from any firm per fiscal year.
- Ensure samples are handled in a manner which avoids contaminating the samples with mycotoxins of interest. If perishable samples are collected, the division may notify the

analyzing laboratory in advance to prevent sample spoilage and contamination during transportation to the laboratory.

The following information should be included in the Collection Report (C/R) for assistance in follow-up, if necessary:

- Collect pictures of the product, labeling and packaging information, such as the nutrition facts panel and serving size. For products not in retail packaging (exceptions: spices, juices, juice concentrates) attempt to collect/determine intended use.
- Document the brand name, country of origin, manufacturer, batch or lot number, and any other pertinent identifying information. For juice samples, document whether the sample is a concentrate or ready-to-drink product. If the juice sample is a concentrate, document the Brix value, if available.
- If the SCOPE assignment details a specific product and mycotoxin analyses, this information should be included in the “Remarks” and “Reason for Collection” sections of the C/R.

Domestic Foods

Collect domestic samples, as identified in the SCOPE. If divisions have reason to collect samples that are not included in the mycotoxins SCOPE, see the section below: Non-SCOPE Sample Collection.

Import Foods

Collect imported food samples, as identified in the SCOPE, at the port of entry or any location throughout the import admissibility process. If divisions have reason to collect samples that are not included in the mycotoxins SCOPE, see the section below: Non-SCOPE Sample Collection.

Non-SCOPE Sample Collection

If divisions have reason to believe that there is a mycotoxin problem with specific commodities that are not listed in the SCOPE, the division may collect these foods after consultation with the Domestic and Import Mycotoxin Monitor, Jacob Reynolds at (202) 617-5227 or email at Jacob.Reynolds@fda.hhs.gov. If the additional product collection is pre-authorized by CFSAN, the Investigator must note this in the "Remarks" section of the C/R. Otherwise, the laboratory will need to consult with CFSAN prior to beginning analyses.

NOTE: Attachment A of this compliance program lists acceptable products for mycotoxin analysis beyond what is listed in SCOPE. Additional center-issued memo(s) may be used to instruct on the collection of specific commodities.

Sample size

The most recent version of the [IOM Chapter 4 Sample Schedule Chart 6: Mycotoxin Sample Sizes](#) should be used for all mycotoxin sample collections.

For domestic samples only, a 702(b) should be collected for food samples (IOM 4.3.3.2), except where it is exempted by statute or by regulation 21 CFR 2.10(b) ([Collecting and Providing 702\(b\) Portion of FDA Official Sample](#)). If the required number of subsamples of the same lot are not available, prioritize collection of the appropriate number of sub-samples for laboratory analysis, then collect as many sub-samples as possible as a 702(b) portion. Do not collect less than the required number of sub-samples for laboratory analysis unless this is pre-approved by CFSAN Program Contacts and communicated to the laboratory by e-mail and in the “Remarks” section of the C/R.

Sample Shipping

Shipping instructions to maintain the integrity of frozen and refrigerated samples and the procedure to notify the receiving laboratory can be found in the most recent version of the [IOM Chapter 4 Section Sampling: Preparation, Handling, Shipping](#).

Samples should be packaged with the appropriate refrigerant and shipped to the laboratory no later than Thursday of each week. Unless prior arrangements are made, shipping on Fridays should be avoided as staff may not be present on the weekends to ensure proper sample receipt.

Submit samples per the [ORA Lab Servicing Table \(LST\) Dashboard](#), specifically for the mycotoxin (MYC) program.

Specific sampling instructions for certain commodities:

- i. **Corn and Corn Products-** Do not sample unpopped popcorn for aflatoxin unless there is reason to believe that contamination may be present due to late harvest or adverse environmental conditions. The characteristics of the cultivar of corn used for popping make it unlikely to be contaminated with aflatoxin.

Mycotoxin levels, in food products made from corn (grits, meal, flour, snack foods or cereals), are likely to be higher in "full fat" than in degermed products since the highest levels of mycotoxin in the kernel are associated with the germ.

- ii. **Peanuts-** The testing for mycotoxins in roasted in-shell and roasted shelled peanuts, as well as processed peanut products for consumer use, is the responsibility of FDA.

Do not sample in-shell peanuts or shelled peanuts destined for processing that is intended to remove mycotoxin contaminated nuts.

Do not sample raw peanuts (shelled or in-shell) for aflatoxins. USDA will collect and analyze samples of domestic and imported raw peanuts (shelled or in-shell) to determine if aflatoxin is less than 15 ppb (ng/g). USDA will conduct all testing of raw peanuts, domestic and imports, for aflatoxins in accordance with [FDA Memorandum of Understanding \(MOU\) with the United States Department of Agriculture \(USDA\), Agricultural Marketing Service \(AMS\) – MOU with USDA/AMS Concerning Aflatoxins in Peanuts and Peanut products, Brazil Nuts and Pistachio Nuts \(MOU-225-19-031\)](#). See also [CPG Sec 570.375 Aflatoxins in Peanuts and Peanut Products](#).

- iii. **Almonds, Brazil nuts, macadamia nuts, pecans, pistachios, walnuts, and hazelnuts** are susceptible to aflatoxin and potentially OTA contamination. FDA surveillance of these crops is necessary to assure that industry implemented quality control procedures continue to effectively prevent the marketing of mycotoxin contaminated nuts.

Do not sample raw pistachios or raw in-shell Brazil nuts for aflatoxins. USDA will test imported unroasted pistachios and imported unroasted in-shell Brazil nuts for aflatoxins in accordance with [MOU 225-19-031](#).

- iv. **Domestic Milk Products Only-** When dairy animals consume food containing high levels of aflatoxins, one of the metabolized aflatoxins (aflatoxin B₁) may be secreted into the animals' milk as aflatoxin M₁. Cattle consuming food that contains less than 20 ppb (ng/g) of total aflatoxins, however, should produce milk that complies with FDA's limit that is listed in the CPG Sec 527.400 for aflatoxin M₁ in milk.

The FDA and State animal food programs routinely collect animal food samples for mycotoxin analysis. Use the results of those sample collections to assist you in determining if a sample of milk is warranted due to the presence of mycotoxins in dairy rations.

Do not sample dairy products such as cheese or yogurt, unless there is reason to suspect they were made from milk containing levels of aflatoxin M₁ that exceed CPG limits.

- v. **Apple juice.** See Attachment A, Section 2. Collect only apple juice and concentrated apple juice, and freshly pressed apple cider. Since patulin is not destroyed by heat processing, collect both pasteurized and non-pasteurized single strength juice and concentrated juices. [CPG Sec 510.150 Apple Juice, Apple Juice Concentrates, and Apple Juice Products - Adulteration with Patulin | FDA.](#)

C. Sample Submission

Per IOM Chapter 4: Collecting Divisions are instructed to submit samples utilizing the Lab Servicing Table (LST) located here: [LST Dashboard.](#)

D. Reporting

Report resources utilized for sample collection using the following Program Assignment Codes (PACs) and Problem Area Flags (PAF):

| PAC | PAF | PAF Description |
|-------|-----|-------------------|
| 07001 | MYC | Mycotoxin in food |

FACTS shall be utilized for gathering information as part of the C/R. The C/R should include the PAF “MYC” as the basis for collection. **NOTE: A specific mycotoxin does not need to be chosen in the collection reason unless specified in the assignment or SCOPE.**

PART IV - ANALYTICAL**1. Analyzing Laboratories**

See [ORA LST Dashboard](#) for mycotoxins.

2. Analysis to be conducted:

Food samples will be analyzed for a single or multiple mycotoxins. Patulin and aflatoxin M1 will be single mycotoxin analyses. All other samples will be multi-mycotoxin analyses unless the laboratory determines that product has not been validated for the multi-mycotoxin method. The laboratory should refer to Attachment A for the appropriate single analyte priority.

A. Multi-mycotoxin analysis (12 analytes: AB₁, AB₂, AG₁, AG₂, OTA, DON, FB₁, FB₂, FB₃, T-2, HT-2, ZEA).

B. Single mycotoxin analysis:

- i. Aflatoxins (AB₁, AB₂, AG₁, AG₂)
- ii. Ochratoxin A
- iii. Fumonisin (FB₁, FB₂)
- iv. Deoxynivalenol
- v. Patulin
- vi. Aflatoxin M₁

3. Methodology**A. General Information**

- i. Prior to beginning analysis, review the C/R to verify that the collected products are listed in the SCOPE. If the product is not on the list of SCOPE requests or Attachment A, but it is pre-authorized by CFSAN, as noted in the C/R, the laboratory should proceed with the analysis. If the product is not specified in these sources, contact the general assignment or center contact for instructions.
- ii. All methods of analysis must be validated, and samples shall be defined within the scope of the selected method. When required, method extensions shall be completed prior to the submission of regulatory sample results following the FDA Foods Program Guidelines for the Validation of Chemical Methods
<https://www.fda.gov/media/81810/download>.

- iii. All potentially violative sample findings (Laboratory Class 2 or 3) will be confirmed by check analysis and the check analysis should be performed by an experienced second analyst using the same method. Confirmation of identity of toxins in violative samples must be performed by mass spectrometric analysis. Appropriate quality control measurements are required in all analyses to document method performance.
- iv. Do not use a qualitative test as it cannot provide quantitative data. CFSAN must have data for risk assessment so a quantitative method must be used. Specific methods for each mycotoxin analysis are listed in the following sections.
- v. SAFETY: Be aware of the potential hazards in the preparation of mycotoxin samples. Refer to the most recent edition of Official Methods of Analysis AOAC International, Method 977.16, for guidance.
- vi. The chart below may be used to facilitate calculations for in-shell nut samples.

| <u>Nuts</u> | <u>Meat, % by Wt.</u> |
|---------------|-----------------------|
| Almonds | 40 |
| Peanuts | 70 |
| Pecans | 50 |
| Pistachios | 50 |
| Macadamia | 29 |
| Pumpkin seeds | 74 |
| Walnuts | 50 |
| Brazil nuts | 50 |

B. Methods

- i. **Multi-mycotoxin method:** Method (C-003) can be found on the [Foods Program Compendium Analytical Laboratory Methods](#) website.
- ii. **Single mycotoxin methods:**
 - The laboratories may use an appropriate method in the current edition of the Official Method of Analysis (OMA) of the AOAC International to quantitate mycotoxins listed in section 2.B above.

- If an AOAC official method is not available, laboratories may use a validated method from the peer-reviewed scientific literature or an FDA laboratory information bulletin (LIB) to quantitate mycotoxins listed in section 2.B above. Multi-laboratory validated methods shall be considered over single laboratory validated methods.
- Prior to using an analytical method from the scientific literature or a LIB for routine use, laboratories shall conduct at minimum an appropriate single laboratory validation in accordance with the FDA Foods Program Guidelines for the Validation of Chemical Methods, <https://www.fda.gov/media/81810/download>.
- **For Patulin:** Samples of frozen concentrate should be diluted either as per recommendation for dilution or to a Brix value of 11.5 (single strength) before analysis (Federal Register 56 No. 127, pp. 30452-30466, 1991).

C. Confirmation of findings

- i. Where compendial method C-003 or any LC-MS/MS method is used, a separate confirmation of identification is not required.
- ii. Where single-analyte methods using liquid chromatography (LC)-ultraviolet (UV) or fluorescence (FLD) detection methods are used, a separate confirmation of identity using LC-MS/MS must be performed using a method that has been shown to be fit for this purpose. A worksheet of the analyses shall include the following information:
 - Sample preparation – see method of analysis for specific toxins
 - Specify LC parameters used in the analysis
 - Specify MS acquisition parameters used in the analysis
 - Conclusion - a concise statement indicating confirmation based on retention time and mass spectral comparison against a reference standard as defined by FDA Mass Spectrometry for Confirmation of the Identity of Animal Drug Residues, CVM, Guidance for Industry #118.

D. Reporting

- i. Report sample results and limit of quantitation (LOQ)/reporting limit into the FACTS/uALIS under PAC 07001 using the Problem Area Flag (PAF) MYC and the appropriate mycotoxin analyte code. Results between the limit of detection (LOD) and LOQ are indicated as trace. Results below LOD are reported as not detected. Mycotoxin findings in food are reviewed on a case-by-case basis by CFSAN. Samples

of food with any findings that represent a potential health concern, that exceed an action level or guidance level, or represent atypically high values for that food type should be classified Lab Class 2 or 3 and referred to the respective Division Compliance Branch for review and referral to CFSAN's Office of Compliance, Division of Enforcement, Food Adulteration Assessment Branch. Additional information on can be found on the Mycotoxin Resource Page. Laboratory results that represent a potential health concern should be referred to CFSAN for evaluation.

- ii. Results for additional quality control measurements (e.g., method blanks, fortified analytical portions, reference materials, etc.) are not required to be entered in the FACTS/uALIS system but the laboratory must maintain this information for potential review by CFSAN to document analytical methods performance.

- iii. Electronic worksheet packages of samples are uploaded to CMS.

PART V - REGULATORY/ADMINISTRATIVE STRATEGY**1. General Information**

- If a human food sample is found to have one or more mycotoxins at a concentration that exceeds an action level, or if there is a potential health concern, the laboratory should conduct a check analysis for the analyte(s). If the check analysis confirms the exceedance(s), the sample should be classified as Lab Class 2 or 3 and referred to CFSAN Office of Compliance, Division of Enforcement, Food Adulteration Assessment Branch via the Compliance Management System (CMS) for review.
- For imported food samples, a CMS case (Center review of detained shipment for Refusal) should be created.
- For domestic samples, a CMS Work Activity (District – Worksheet/Other Exam Review) should be created in response to sample results and the sample linked to the Work Activity.
- For tested products subject to a CPG that exceeds the action levels stated in the CPG or for products not subject to a CPG that were tested and represent a potential health concern, CFSAN concurrence is needed to support action. Please consult with the Center and review information on the Mycotoxin Resource Page.
- FDA has established action levels for some mycotoxins in certain food products. The Center will evaluate sample results for all mycotoxins on a case-by-case basis and may recommend follow-up as appropriate.

In general, refer to the relevant [Compliance Policy Guides](#) (CPGs) and [Import Alerts](#).

The following CPGs are applicable when recommending legal actions against products collected under this program:

- [Section 527.400-Whole Milk, Low Fat Milk, Skim Milk - Aflatoxin M₁](#)
- [Section 570.200-Brazil Nuts - Adulteration with Aflatoxin](#)
- [Section 570.375-Aflatoxin in Peanuts and Peanut Products](#)
- [Section 570.500-Pistachio Nuts - Aflatoxin Adulteration](#)
- [Section 555.400-Foods, Adulteration with Aflatoxin](#)
- [Section 510.150-Apple Juice, Apple Juice Concentrates, and Apple Juice Products](#)

The following guidance documents are applicable for recommending follow-up actions for specified products collected under this program:

- [Guidance for Industry: Fumonisin Levels in Human Foods and Animal Feeds | FDA](#)
- [Guidance for Industry and FDA: Advisory Levels for Deoxynivalenol \(DON\) in Finished Wheat Products for Human Consumption and Grains and Grain By-Products used for Animal Feed | FDA](#)

NOTE: Findings of T-2 toxin greater than or equal to the method LOD must be reported by the laboratory to the Federal Select Agent Program at Centers for Disease Control within seven days of identification—to include disposition details—using APHIS/CDC Form 4. Refer to 42 CFR 73.5 and <https://www.selectagents.gov/> for additional guidance.

Domestic Products Only

The Home Division must report analytical results on compliance samples that exceed CPG limits to the responsible firm and to cooperating State Liaisons within their Divisions. Initiate appropriate follow-up action consistent with CPG 527.400, for dairy products under this compliance program.

For follow-up in animal food, when milk samples exceed the CPG limits for aflatoxin M₁, Divisions must contact the CVM Regulatory Technical Assistance Network (rTAN) at cvmanimalfoodprograms@fda.hhs.gov before collecting any animal food samples.

PART VI REFERENCES, ATTACHMENTS, AND PROGRAM CONTACTS

1. References

A. U.S. Food and Drug Administration, CPG Sections:

- 675.300, Moisture Damaged Grain, (CPG Section 675.300)
(<http://www.fda.gov/ICECI/ComplianceManuals/CompliancePolicyGuidanceManual/ucm074695.htm>);
- 683.100, Action Levels for Aflatoxins in Animal Feeds, (CPG Section 683.100)
(<http://www.fda.gov/ICECI/ComplianceManuals/CompliancePolicyGuidanceManual/ucm074703.htm>)

B. DEIO Guide to Inspections of Manufacturers of Miscellaneous Food Products, Volume II, issued September 1996.

C. Guidance for Industry and FDA: Advisory Levels for Deoxynivalenol (DON) in Finished Wheat Products for Human Consumption and Grains and Grain By-Products used for Animal Feed Document issued on: June 29, 2010; Revised July 7, 2010. [Guidance for Industry and FDA: Advisory Levels for Deoxynivalenol \(DON\) in Finished Wheat Products for Human Consumption and Grains and Grain By-Products used for Animal Feed | FDA](#)

2. Attachments

A. Mycotoxin Products for Sampling and Acceptable Product Codes

3. Program Contacts

CFSAN Compliance Program Inquiries: Jacob Reynolds, CFSAN/OC/Division of Field Programs & Guidance, Program Assignment and Monitoring Branch, HFS-615, at 202-617-5227 or via email at Jacob.Reynolds@fda.hhs.gov

CFSAN Scientific Inquiries: Dr. Anthony Adeuya, CFSAN/OFS/Division of Plant Products and Beverages, Plant Products Branch, HFS-317, at 240-402-5759 or via email at Anthony.Adeuya@fda.hhs.gov

Dr. Tabitha Miller, CFSAN/OFS/Division of Plant Products and Beverages, Plant Products Branch, HFS-317, at 240-402-4552 or via email at Tabitha.Miller@fda.hhs.gov

CFSAN Analytical Inquiries: Dr. Kai Zhang, CFSAN/ORS/Division of Bioanalytical Chemistry, Bioanalytical Methods Branch, HFS-717, at 240-402-2318, or via email at Kai.Zhang@fda.hhs.gov

Dr. Christine Parker, CFSAN/ORS/Division of Bioanalytical Chemistry, HFS-717, at 240-402-2019 or via email at Christine.Parker@fda.hhs.gov

CFSAN Regulatory Inquiries: For domestic and imported food regulatory action Inquiries, Email monitored by team of DE/FAAB COs: DEChemicalContaminants@fda.hhs.gov

ORA Investigations Inquiries: **Martha (Marti) Myrick** DHAFO/HFR-SE55, at 240-402-5840 or 616-304-6283 (cell) or via email at Martha.Myrick@fda.hhs.gov

ORA Import Operations Inquiries: DIO CFSAN Liaisons. Email: oraoeiodiocfsanliaisons@fda.hhs.gov

Mycotoxin Program Lab Service Inquires: **Dr. Yanxuan (Tina) Cai**, ORA/ORS/OHAFLO, at 240-402-1369 or via email at Yanxuan.Cai@fda.hhs.gov

CVM Scientific and Analytical Inquiries: CVM Animal Food Contaminants via email at cvmanimalfoodcontaminants@fda.hhs.gov

CVM Regulatory Inquiries: CVM Regulatory Technical Assistance Network (rTAN) at cvmanialfoodprograms@fda.hhs.gov

PART VII - CENTER RESPONSIBILITIES

The CFSAN/Office of Food Safety (OFS) will provide subject matter expertise in the maintenance and evaluation of the Compliance Program and provide guidance to the Office of Compliance with regard to program priorities, relevant evaluation questions, and recommended program changes. The Office of Compliance will lead the effort and work in conjunction with the Office of Food Safety to prepare routine compliance program evaluations. Evaluation will be conducted on a periodic basis and outline the program office's current objectives, general and specific program evaluation questions, list recommendations for process improvement, and highlight data patterns and trends for better targeting and resource allocation. The Office of Compliance will make these evaluations available internally to FDA.

**ATTACHMENT A (FOR LAB USE) – PRIORITY PRODUCTS FOR SINGLE ANALYTE
ANALYSIS AND PRODUCT CODES**

FOODS FOR HUMAN USE ONLY

Domestic and Import Products

Section 1 - Aflatoxins (see section 1b below for additional samples)

| <u>PRODUCT</u> | <u>PRODUCT CODES</u> |
|---|---|
| Bakery Products | 03A to Y[] []01-1 & 99 |
| Corn, shelled, whole kernel | 02A[] []01 |
| Corn Flour (masa) or Meal (including Hominy Grits) | 02B[] []01 to 99 |
| Cereals, Corn (including Ready to Eat, Quick Cook, Instant, and Grits) | 05A or B[] []01 |
| Corn Grits, Brewers Corn Grits, Corn Flaking Grits | 05B[] []01 |
| Hominy | 24A or B[] []61 |
| Snack Foods ¹ | 07A or B[] []02 |
| Peanut Butter | 23C[] []07 |
| Peanut Products, imitation | 23N[] []01 |
| Peanuts, in shell, roasted | 23A to H[] []07 |
| Peanuts, shelled roasted | 23B to H[] []07 |
| Peanuts, toppings | 23F[] []07 |
| Peanuts, in shell, raw (follow-up samples only) | 23A[]B07 |
| Peanuts, shelled, raw (follow-up samples only) | 23B[]B07 |
| Tree Nuts: (DO NOT COLLECT CASHEW NUTS) | |
| Almonds | 23A to F[] []01 |
| Brazil Nuts | 23A to F[] []02 |
| Hazelnut | 23A to F[] []06 |
| Pecans | 23A to F[] []08 |
| Pine nuts/pinon | 23A to F[] []09 |
| Pistachios | 23A to F[] []11 |
| Walnuts | 23A to F[] []12 |
| Mixed Nuts (cashews excluded) | 23A,Y[] []99 |
| Coconut, shelled | 23A,Y[] []99 |
| Edible seeds: | |
| Pumpkin seeds | 23K[] []01 |
| Sunflower seeds | 23K[] []04 |
| Melon seeds | 23K[] []05 |
| Ginger spice | 28A to B[] []19 |
| Cayenne pepper spice (Flaked or ground) | 28A 08 and 28B[] []08 |
| Rice, cultivated, whole grain (Brown) | 02A[] []05 |
| Rice, wild, whole grain | 02A[] []06 |
| Candy with nuts &/or seeds &/or fruit &/or coconut, &/or chocolate covered nuts &/or seeds &/or fruit &/or coconut -imports | 33A to Y[] []01-14 & 99; 34A to Y[] []01-14 & 99 |

¹ Miscellaneous corn-based foods such as tacos, chips, cereals and similar products that are primarily corn are acceptable for collection. It is,

however, preferable to collect the corn ingredients that will be used in these foods.

NOTE: Nut and seed pastes and similar ethnic foods such as coated peanuts, nut &/or seed snack mixes and nut &/or seed sugar/brittle candies containing nuts &/or seeds are susceptible to aflatoxin contamination and may be sampled.

Refer to PART III, section B(iv) before collecting milk or milk products.

| PRODUCT | PRODUCT CODES |
|----------------------|-------------------------------------|
| Milk, fluid | 09C to E[] []01, 07, 09 to 11 |
| Milk, concentrated | 09C to E[] []03 |
| Milk, nonfat dried | 09C to E[] []16 |
| Yogurt | 09C to E[] []15 |
| Cheese, natural | 12A[] []01 to 61 |
| Cheese products | 12B[] []01 to 13, and 99 |
| Frozen milk products | 13A to G and Y[] []01 to 06, and 99 |

Section 1b - "SPECIAL SURVEILLANCE" PRODUCTS FOR AFLATOXIN ANALYSIS

| PRODUCT | PRODUCT CODE |
|----------------|--|
| Sesame Seed | 23K[] []02 |
| Nutmeg | 28A or B[] []30 |
| Figs/Dates | 21S or T[] []03 and 21G to J[] [];05-06 and 21H[] []16 |

Section 2 -- Patulin

| PRODUCT | PRODUCT CODES |
|---|----------------------|
| Apple Juice and Apple Juice Concentrate | 20S[] []01 |

Section 3 -- Deoxynivalenol

| PRODUCT | PRODUCT CODES |
|--|----------------------|
| Whole wheat or white flour | 02E[] []01 |
| Wheat Bran and other milled wheat products for human consumption | 02F[] []01 |
| Wheat based prepared foods: | |
| Pretzels, baked or fried | 07A or B[] []07 |
| Pasta | 04A[] []05 |
| Bread | 03A to E[] []12-13 |
| Breakfast cereal derived from wheat: | |
| Ready to Eat | 05A[] []04 |
| Quick Cook | 05B[] []04 |
| Wheat based baby food | 40M to R[] []01-99 |

Baby Wheat Cereal 40B[] []06
Section 4 -- Fumonisin

| PRODUCT | PRODUCT CODES |
|---|---|
| Popcorn (unpopped) | 02A[] []04 |
| Cleaned corn aimed for masa flour production | 02A[] []04 |
| Dry milled corn bran | 02B[] []99 |
| Refer to Part III, 4.c. regarding the need to determine the fat content before collecting these products: | |
| Corn based breakfast cereals: | |
| Ready to Eat | 05A[] []01 |
| Quick Cook | 05B[] []01 |
| Baby Corn Cereal | 40B[] []02 |
| Corn flour (includes masa flour) | 02B[] []01 |
| Corn meal | 02B[] []01 to 05, 07 thru 99 |
| Corn Hominy Grits | 02B[] []06 |
| Flaking grits | 05B[] []01 |
| Corn Grits, Brewers Grits, Enriched Corn Grits, & Corn Meal Mush | 05B[] []01 |
| Corn Based Bakery Products | 03A to E[] []04 & 99 |
| Corn Based Snack Foods | 07A to B[] []02, 07A to B[] []04; 07A to B[] []05, 33S[] []03 |

Section 5 -- Ochratoxin A

| PRODUCT | PRODUCT CODES |
|--|---|
| Barley (whole) | 02A[] []02 |
| Barley Malt | 02J[] []01 |
| Buckwheat flour | 02G[] []02 |
| Corn Meal (including hominy grits) | 02B[] []01 thru 99 |
| Oats (whole) | 02A[] []03 |
| Rye Flour | 02G[] []10 |
| Wheat kernels (whole) | 02A[] []09 |
| Whole wheat or white flour | 02E[] []01 |
| Cereals (barley) | 05A or B[] []99 |
| Cereals, (corn) (including Ready to Eat, Quick Cook, Instant, and Grits) | 05A or B[] []01 |
| Cereals (oat) | 05A or B[] []02 |
| Cereals (wheat) | 05A or B[] []04 |
| Cereals (rice) | 05A or B[] []03 |
| Coffee Beans | 31A[] []01 |
| Dried Beans/Peas | 24B[] []02 thru 99 |
| Raisins | 20B[] []10 |
| Fig, dried | 21S or T[] []03 |
| Ginger (Ground) | 28B[] []19 |
| Baby Cereals (barley, corn, oat, rice, wheat) | 40B[] []01 to 06 |
| Soya Based Baby Food Products | 40A or B[] []99, 40C[] []25, 40Y[] []99, 40N or O or P[] []02 |

**Special Note: Please use the correct product codes and collect only sample types listed

above and in the SCOPE.