## CHAPTER 03 – Foodborne Biological Hazards

| SUBJECT: SAMPLING FOR FOODBORNE BIOLOGICAL HAZARDS, AND FILTH– DOMESTIC AND IMPORT | IMPLEMENTATION DATE: Upon receipt |
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| DATA REPORTING | |
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| PRODUCT CODES | PRODUCT/ASSIGNMENT CODES |
| All Food Codes<br><br>**EXCEPT:**<br>Industry 15 – Egg and Egg Products<br>Industry 16 – Fishery and Seafood Products<br>Industry 54 – Dietary Supplements<br>Industry 40 – Infant Formula (only) | REPORT PROGRAM ACTIVITIES UNDER THE FOLLOWING PACs:<br><br>03037D Domestic and Import Cheese and Cheese Products Microbiological Samples<br>03803D Domestic Human Food Microbiological Samples<br>03819C Imported Human Food Microbiological Samples<br>03050B Domestic and Import Human Food Filth Samples<br>03050E Domestic and Import Human Food Facility Exhibit Sample<br>03050N Domestic Human Food Environmental Samples<br>03S050N State Contract Domestic Human Food Environmental Samples |
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**FIELD REPORTING REQUIREMENTS:**
Report domestic sample collections and all analytical results into the Field Accomplishment and Compliance Tracking System (FACTS). Import sample collections should be reported in the System for Entry Review and Imports Operations (SERIO) and/or the Operational & Administrative System for Import Support (OASIS), as appropriate.

Note: This compliance program does not address "seafood decomposition" (fishery products), which is addressed in separate compliance programs (7303.844, 7303.842).

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## PART I – BACKGROUND

The Food and Drug Administration (FDA) enforces the Federal Food, Drug, and Cosmetic (FD&C) Act to protect public health. The FDA Food Safety Modernization Act (FSMA) was signed into law on January 4, 2011 and amended the FD&C Act. FSMA aims to ensure the U.S. food supply is safe by shifting the focus to preventing contamination of the food supply, rather than responding to it. Since the enactment of FSMA, FDA issued several large-scale risk-based microbiological sampling assignments designed with the goal of preventing foodborne contamination. For example, an assignment from 2014 to 2016 estimated the prevalence(s) of *Salmonella enterica*, *Listeria monocytogenes* and *Escherichia coli* (*E. coli*) O157:H7 in domestically produced sprouts (results are available at FDA.gov). The data from this assignment informed FDA's prioritization of short- and longer-term program initiatives.

The Sampling for Foodborne Biological Hazards (Domestic and Import) Compliance Program was developed to support FSMA implementation by providing streamlined instructions to FDA personnel for conducting activities covering domestic and imported food products such as produce, ready-to-eat (RTE) food, cheese and cheese products, and other food for foodborne biological hazards associated with public health risk. This program focuses on the analysis of (1) food for microbiological hazards such as *Listeria monocytogenes*, *Salmonella* spp., and pathogenic *Escherichia coli*, (2) environmental samples for pathogens; and (3) food for filth.

*Listeria monocytogenes* (*L. monocytogenes*) has been associated with food such as cheeses (particularly soft-ripened varieties), ice cream, raw vegetables, raw fruit, sprouts, caramel apples, fermented raw-meat sausages, raw and cooked poultry, raw meats (all types), and smoked fish. *L. monocytogenes* differs from other foodborne pathogens because it is widely distributed, adapts to diverse environmental conditions, including low pH and high salt concentrations, and is a psychrotrophic facultative anaerobe. *L. monocytogenes* is a major concern for many manufacturing industries because of its ability to grow and/or survive for long periods of time in environments having a constant source of moisture, ability to grow at low temperatures, and ability to survive prolonged periods under adverse conditions. Eating food contaminated with *L. monocytogenes* may cause listeriosis, a rare but serious illness. Listeriosis can be fatal, especially in certain high-risk groups, including the elderly and those with weakened immune systems. In pregnant women, listeriosis can cause miscarriage, stillbirth, premature labor, and serious illness or death in newborn babies. CDC estimates there are approximately 1,600 cases of listeriosis in the United States every year and the infections kill an estimated 260 people annually.

*Salmonella* is a major cause of bacterial foodborne illness in the U.S. The major reservoirs for *Salmonella* in the global food chain are raw meats, poultry, and eggs; the organism has also been isolated from fruits, vegetables, spices, and nut meats. Some *Salmonella* spp. can survive drying particularly well and are difficult to kill with heat when the *Salmonella* spp. are dehydrated or are in low moisture products such as chocolate, milk powder, spices, and peanut butter. Salmonellosis, an illness caused by *Salmonella*, has been associated with a variety of food including fresh produce, eggs, cake mixes, unpasteurized milk, cheese and cheese products, peanut butter, RTE breakfast cereals, chocolate, and salad dressings. Symptoms of salmonellosis are usually described by severe

fever, abdominal pain, nausea and sometimes vomiting. CDC estimates that *Salmonella* bacteria cause about 1.35 million infections, 26,500 hospitalizations, and 420 deaths in the United States every year. Food is the source for most of these illnesses.

*Escherichia coli* (*E. coli*) are bacteria that live in human and animal intestines. Most *E. coli* are harmless, but some are pathogenic. Shiga toxin-producing strains of *E. coli*, or STECs, have been associated with many foodborne outbreaks. Examples of food implicated included unpasteurized juices, unpasteurized soft cheeses, and raw fruits and vegetables. Symptoms of illness could be bloody diarrhea and/or kidney failure which has been reported to cause the death of young children, elderly, and those with compromised immune system. CDC estimates *E. coli* O157:H7 causes approximately 2,100 hospitalizations and about 40 deaths in the United States every year.

Environmental pathogens may cross-contaminate food in a facility. Environmental sampling is one of FDA's current strategies to determine whether an environment in a food facility contains harmful human pathogens, such as *Salmonella* spp. or *Listeria monocytogenes*. It is critical that food establishments vulnerable to pathogen harborage and cross-contamination maintain an environment that adheres to current good manufacturing practices, and as applicable and necessary, sanitation preventive controls.

The Sampling for Foodborne Biological Hazards (Domestic and Import) Compliance Program provides consolidated instructions covering sampling for biological hazards, environmental sampling (ATTACHMENT A), and filth (ATTACHMENT B) in food to enable the field to accomplish FDA's mission and to better prioritize sampling to ensure the U.S. food supply is safe and wholesome. The compliance program plays a significant role in helping reduce contaminated product from reaching consumers, understand food safety risks, and assess strategies to control those risks.

## PART II - IMPLEMENTATION

### 1. Objectives

- To collect domestic and import food samples planned for the current fiscal year.
- To accomplish the analyses of all samples collected for foodborne biological hazards.
- To accomplish the collection and analysis of samples per ATTACHMENT A – ENVIRONMENTAL SAMPLING and ATTACHMENT B – FILTH AND DECOMPOSITION.
- To prevent the introduction of contaminated food into U.S. commerce.
- To take appropriate regulatory action to remove food that violates the FD&C Act from the U.S. food supply.
- Evaluate the analytical results data to identify future trends and strategies for prevention and mitigation.

### 2. Program Management Instructions

**Inspection Priorities**

Establishment inspections (EI) are not covered under this compliance program; refer to the interacting compliance programs section below for inspectional coverage.

**Planning Instructions**

This compliance program covers sample collection and analysis of domestic and imported food products for foodborne biological hazards, environmental sampling (ATTACHMENT A), and filth (ATTACHMENT B).

- Domestic and import food sampling for biological hazards are planned each fiscal year in the Sample Collection Operation Planning Effort (SCOPE).
- Environmental sampling will be planned each fiscal year in the ORA Field Work Plan.
- If a facility is involved in ongoing compliance activities or the current inspection may be classified OAI, the Division should consult with their Compliance Branch to determine whether collection of samples for surveillance purposes is appropriate.
- For-cause product and environmental samples can be collected during inspections and investigations conducted under other compliance programs if the for-cause sampling criteria are met.

**Interactions with Other Compliance Programs**

This compliance program may have some interactions with the following compliance programs. Use the appropriate PAC when reporting sample collections under this compliance program.

- Preventive Controls and Sanitary Human Food Operations compliance program
    - Domestic coverage of incorrect application and storage of pesticides and other chemical contaminants in food processing plants, warehouses, and transport vehicles will be covered by Preventive Controls and Sanitary Human Food Operations (PCHF) compliance program during inspections.
    - Inspections at cheese and cheese product processing facilities will also be covered by the PCHF compliance program.
- National Conference on Interstate Milk Shipments (NCIMS) Milk Safety Program

- Juice HACCP compliance program
- Import Food Operations compliance program
- Domestic and Import Acidified and Low-Acid Canned Foods compliance programs
- Produce Safety compliance program (under development)

**Resource Instructions**
Divisions should coordinate resources obligations from other assignments and/or compliance programs, if possible.

**Interactions with other Federal agencies, State and Local Counterparts, and Foreign Authorities**

1. Federal Agencies
   When samples are collected during an inspection or investigation, follow the Investigations Operation Manual (IOM) subchapter 3.1.3.2 'Discussion with Federal Inspector' when federal officials from other agencies are present during FDA inspections or investigations. See IOM subchapter 3.2 'Federal Agency Interaction' for a list of Memoranda of Understanding (MOU) between the FDA and other Federal agencies that may be applicable. A complete list of MOUs may be found at FDA Memoranda of Understanding.

2. State and Local Counterparts
   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.

3. 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.

**Information relating to work planning, supervision, or other resource management that is not specified in another part of the compliance program**

Resources dedicated to this program may change on a yearly basis based on data evaluation, risk ranking, and emerging trends. Sample collection and analytical results are monitored on a regular basis to track accomplishments during the fiscal year.

## PART III - INSPECTIONAL

### 1. Operations

#### A. Inspections

Inspections are not planned under this compliance program; refer to Interactions between Compliance Programs (PART II). For-cause product and environmental samples may be collected during inspections when warranted.

#### B. Investigations

An investigation is an information-gathering activity conducted for several reasons and may lead to a situation in which sampling is recommended. See IOM subchapter 8.1.1.

#### C. Sample Collections

**General Information (domestic and import)**

Detailed instructions for sampling are contained in IOM Chapter 4 – Sampling. See also, Bacteriological Analytical Manual (BAM) online Chapter 1: Food Sampling/Preparation of Sample Homogenate. Investigators should specify the reason for sample collection and/or the microorganism for analysis.

When collecting samples from **bulk containers** investigators should provide photos of the product label in sufficient detail to permit a thorough review of all label elements. If the container is unlabeled, copies of records related to the bulk product should be collected that show in sufficient detail the information about the source of and ingredient(s) sampled. If available, photographs may be taken of the same product in labeled containers (with the same or different lot code) and submitted with a statement in the collection report that the label is representative of the sampled product.

When collecting samples of food included on the Proposed Food Traceability List (FTL), investigators should collect additional traceability records described in the Final Rule for Food Traceability, after publication, to expedite tracking of a food to mitigate foodborne illness. Note, that the additional recordkeeping requirements in the rule apply not only to food specifically listed on the proposed FTL, but also to these foods when used as food ingredients, unless otherwise exempted.

When collecting **frozen fruits and vegetables**, collect only single ingredient, finished packages, without further processing (e.g., do not collect items such as frozen berries coated with chocolate or yogurt, berry products with sugar, vegetables with cream sauce, etc.) unless otherwise directed.

When collecting **cheese and cheese products**, subsamples should consist of intact units from the same lot. Larger retail packages should not be broken or cut to obtain a subsample. Collect the intact retail unit as the subsample even if it is large. Do not collect cheese portions that were cut from larger loaves, bricks or wheels and rewrapped at the retail level, unless otherwise directed.

Refer to the current fiscal year's (FY) SCOPE for number of samples to collect. When planning sample collections under this compliance program, consider prioritizing based on seasonality of the commodity, a commodity's history of violations, a firm's history of violations, a specific region's history of violations, and emerging issues.

If Divisions identify any areas of concern other than what is described in this compliance program, they should contact the compliance program monitor to discuss possible sample collections and analysis.

The primary food and pathogens of concern covered under this compliance program are summarized in Table 1 below. For more detailed information on foodborne biological hazards and commodity/hazard pairs, see Guidance for Industry: Guide to Minimize Microbial Food Safety Hazards of Fresh-cut Fruits and Vegetables (also see, Guide to Minimize Food Safety Hazards of Fresh-cut Produce: Draft Guidance for Industry) and Draft Guidance for Industry: Hazard Analysis and Risk-Based Preventive Controls for Human Food, Appendix 1: Potential Hazards for Foods and Processes.

**Table 1. Commodities and Associated Microbiological Analytes of Concern**

| Categories | Examples of Commodities | Analytes of Concern |
|---|---|---|
| **Produce (Fruits & Vegetables)** | Raw Agricultural Commodities (RAC), e.g. :<br>• Green leafy vegetables<br>• Herbs<br>• Microgreens<br>• Sprouts<br>• Tomatoes<br>• Tree nuts and peanuts<br>• Other fresh, whole, raw produce<br>Minimally processed produce, e.g.<br>• Fresh-cut produce<br>• Fresh Green Salads<br>• Frozen or dried produce<br>• Non-LACF vacuum packed and modified atmosphere packed raw produce, such as fresh mushrooms and salads, etc. | • *Salmonella* spp.<br>• *Listeria monocytogenes* (primary concern for minimally processed produce)<br>• Shiga-toxin producing *Escherichia coli* (STEC) and enterohemorrhagic *E. coli* (EHEC)<br>• *Cyclospora cayetanensis*<br>• Hepatitis A<br>• Norovirus |
| **Ready-to-Eat** | • Prepared salads (e.g., deli salads such as macaroni and potato salads)<br>• RTE sandwiches<br>• Baked Goods, Custard or Cream-filled (Egg Containing)<br>• Ice Cream<br>• Dips and spreads | • *Salmonella* spp.<br>• *Listeria monocytogenes*<br>• EHEC/STEC<br>• *Staphylococcus aureus* (*S. aureus)*<br>• *Staphylococcus* enterotoxin |

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| **Cheese & Cheese Products (See Note below)** | Examples:<br>• Raw Milk Cheese<br>• Soft, Soft-ripened, Smeared Cheese<br>• Semi-Soft Cheese<br>• Brined Cheese<br>• Hard Cheese<br>• Extra Hard Cheese and<br>• Cheese Products (except shelf-stable)<br>• Other cheese (ex. Mexican style cheese, aged gouda, blue cheese) | • *Salmonella* spp.<br>• *Listeria monocytogenes*<br>• EHEC/STEC<br>• *S. aureus*<br>• *Staphylococcus* enterotoxin<br>• Alkaline phosphatase (The methodology for detecting the presence of alkaline phosphatase has been validated for bovine milk and bovine milk products only.) |
| **Other (processed)** | • Grains, nuts, spices, seeds (with focus on ready to eat products)<br>• Tahini and Halva Candy<br>• Dried Milk and Dried Milk Products<br>• New/novel foods (newly developed innovative food or food produced using new technologies and production processes) | **Routine:**<br>• S*almonella* spp.<br><br>***Upon Request:**<br>• *Listeria monocytogenes*<br>• EHEC/STEC<br>• *S. aureus*<br>• *Staphylococcus* enterotoxin<br>• Generic *E.coli* |
| **Other** | Other | ***Upon Request:**<br>• *Vibrio*, including *V. cholera*<br>• *Shigella*<br>• *Bacillus cereus* and *Bacillus cereus* enterotoxin<br>• *Clostridium botulinum*<br>• *Cronobacter* (other than infant formula)<br>• *Campylobacter jejuni*<br>• *Yersinia enterocolitica*<br>• Norovirus<br>• Hepatitis A Virus |

*Division should discuss these analytes with CFSAN/OC Compliance Program monitor prior to collection

**NOTE:** Cheese and cheese product samples collected under this compliance program will generally be analyzed for both microorganisms and alkaline phosphatase; filth analysis, if required, should be specifically requested in the collection report.

**Domestic Sample Collection**

Depending on the commodity, domestic samples could be collected from a variety of establishments such as packers, wholesalers, manufacturers, fresh-cut produce processing plants, distributors/warehouses, coolers, grower/shipper, and retail (except do not sample fresh berries from retail locations).

Food samples may also be collected while conducting environmental sampling during inspection under routine surveillance sampling programs such as SCOPE, under CFSAN or ORA active assignments, or as directed for compliance purposes.

Determine if 704(d) applies (Refer to IOM 4.4.10.3.64 for more information). If the investigator is informed that a sampled lot will be held pending receipt of FDA analytical results or specifically requests that FDA inform the facility of the results, the investigator should make note of the hold in the collection report by flagging the Collection Report (CR) in FACTS/MARCS with "Dealer Voluntarily Holding" as per guidance in FMD 147 and the Release of ORA Laboratory Analytical Results to the Responsible Party: Guidance for Food and Drug Administration Staff.

For domestic import samples, follow IOM 4.1.4.8 and include documentation of country of origin (e.g., label statement).

For-cause surveillance samples may be collected under this compliance program.
When samples are collected, record the reason for sample collection in the collection report (e.g., compliance program, assignment, complaint, etc.).

**Import Sample Collection**

Import samples will be collected per routine import procedures (see IOM, Chapter 6, Section 6.5 "Import Sample Collection") from standard collection sites, such as, importer's warehouses, other storage facilities, and ports of entry. Emerging issues will be addressed through CFSAN issued assignments and Import Bulletins (IBs) and may change from year to year.

Refer to Regulatory Procedures Manual (RPM) Chapter 9, Subchapter "Import Information Directives" for procedural guidelines about FDA Import Alerts (IAs) and IBs. All additional entry lines determined to be the same or similar to the sampled line item, should be forwarded to the compliance branch pending sample results as violative FDA analytical findings would provide evidence to support concurrent inclusion of these additional entry lines to the same import alert. Per 21 CFR 1.90, sampled imported products must be held intact by the owner or consignee pending the analytical results.

When selecting products for FDA sample collection, investigators should not generally collect products that are subject to DWPE per an existing IA, unless a potential concern exists with another analyte. Refer to the Compliance Management System (CMS) for relevant existing IAs and IBs. Divisions should also be aware of food products covered under Cooperative

Arrangements and should not sample products covered under Cooperative Arrangements unless otherwise instructed.

Prioritize routine sample collection based on product risk including, but not limited to, the entry's PREDICT risk score. Coverage should be consistent with the other priorities in this program.

Import juice samples should be collected under this compliance program, as there are no collections planned under the Juice HACCP Inspection Program. Emphasis should be given to unpasteurized and non-shelf-stable pasteurized ready to drink fruit and vegetable juices. Refer to IOM 4.1.4.12 Audit/Certification Sample when collecting a sample to verify analytical results provided by a certificate of analysis or private laboratory analysis that purports to show a product complies with the FD&C Act and/or regulations.

**For-Cause Sample Collection**

Inspections are not planned under this compliance program; however, if it is determined that for-cause samples will be collected during inspections under other interacting compliance programs (e.g., 7303.040 Preventive Controls and Sanitary Human Food Operations), Divisions should discuss inspection and for-cause sample collection with CFSAN/Division of Enforcement to ensure that evidence/samples obtained will support further regulatory action.

Also, refer to the following instructions:

- When collecting for-cause samples, record any suspected product abuse in the collection report; this information helps the laboratory determine if additional pathogens should be included in the microbiological analysis of the sample.

- Collect samples if Sanitary Transportation rule/controls are not in place and/or if microbial treatment to affect pathogen inactivation of the incoming product component(s) and/or of the finished product(s) is not being utilized.

- Collect **cheese and cheese product samples** when either of the following criteria has been met: (1) the firm has a previous history of unmitigated microbiological contamination in the environment and/or in finished product (i.e., follow up to illness or injury complaint, recalled/seized product, previous inspectional history, etc.), or (2) inspectional observations warrant collection of samples for microbiological analyses. If **filth** analysis is required, indicate that request in the collection report.

**Sample Size**

For general sample size information see Tables 2 and 3. The sample size does not include 702(b) portions. See IOM Chapter 4 and 6 for additional information on 702(b) portion. Do not collect 702(b) sample portions for perishable products (see 21 CFR 2.10(b)(3)) or for samples collected in import status (unless otherwise requested). Investigators should contact their supervisor or the compliance program monitor for questions about sample size and/or 702(b) portions.

In general, when collecting samples for multiple analytes (e.g., both *Salmonella* and *Listeria*), collect enough product for the intended analyses to be performed. For example, if 30 subs are needed for *Salmonella* analysis and only 10 of those subs are needed for *Listeria* analysis, please ensure that the sample consists of 30 subs total with each sub containing enough product for both analyses, as indicated in Tables 2 and 3.

There are instances, specifically for fresh produce, when a sub may consist of multiple units (whole intact single units of fruits or vegetables). An example of this would be one sub of cucumbers or onions consisting of multiple units of cucumbers or onions in one sampling container (e.g., 3 onions in one "Whirl-Pak" bag). In this case, the units may be split by the lab for multiple analyses (e.g., a portion of at least 454 grams can be analyzed for *Salmonella* and another portion of at least 454 grams can be analyzed for *Listeria*). Some units cannot be cut for analysis, for example large fruits such as cantaloupe or a whole head lettuce. These must be analyzed as whole fruits and vegetables. If this particular item is needed to be tested for multiple analytes (such as *Salmonella* and EHEC) then you will need to collect multiple units (one for each analyte). Refer to the chart below and Part IV – Analytical, Methods for Produce for guidance on collection of whole units.

**Table 2. Sample Size for *Salmonella* Analysis (Sample sizes do not include 702(b) portion.)**

| Food Category | Examples of Commodities | Minimum Number of Subs | Minimum Amount Per Sub Needed |
|---|---|---|---|
| Raw Agricultural Commodities (RAC) | Green leafy vegetables, fresh herbs, microgreens, sprouts | 30 | 100g (0.22 lb.) |
| | Tomatoes | 10 | 454g (1 lb.) |
| | Large units (e.g., melon, papaya) | 10 | 454g (1 lb.) minimum 1 whole unit |
| | Whole Fruits and Vegetables collected at retail | 10 | 454g (1 lb.) minimum 1 whole unit |
| | Whole Fruits and Vegetables collected at firms other than retail (e.g., wholesale, packer, or farm) | 30 | 454g (1 lb.) minimum 1 whole unit |
| Minimally Processed Produce | Fresh-cut produce (except very large produce) | 30 | 100g (0.22 lb.) |
| | Fresh green salads | 30* | 100g (0.22 lb.) |
| | Frozen or dried produce | 30* | 100g (0.22 lb.) |
| | Non-LACF vacuum packed and modified atmosphere packed raw produce, such as fresh mushrooms and salads, etc. | 30 | 100g (0.22 lb.) |

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| | Unpasteurized and non-shelf-stable pasteurized fruit and vegetable juices (*Import only*) | 10 | 100g (0.22 lb.) |
| Ready-to-eat (RTE) food | Prepared salads (e.g., deli salads such as macaroni and potato salads) | 10 | 100g (0.22 lb.) |
| | RTE sandwiches<br>Baked Goods<br>Custard or Cream filled (Egg containing)<br>Ice Cream | 30 | 100g (0.22 lb.) |
| | Dips and spreads | 30* | 16oz |
| Cheese and Cheese Products | For retail units (solid, shredded, grated, curds or extruded forms) weighing less than 454 g (1 lb.) | 10 | Each sub should equal at least 454 g (1 lb.). If the product unit is less than 454 g (1 lb.) collect multiple units to equal at least 454 g (1 lb.) or more. |
| | For retail units (solid, shredded, grated, curds or extruded forms) ranging in weight from 454 g (1 lb.) to less than 2.27 kg (5 lbs.) | 10 | 75g (0.16 lb.) |
| | For solid cheese (wheels, loaves, or bricks) ranging in weight from 2.27 kg (5 lbs.) to less than 4.54 kg (10 lbs.) | 2 | 375g (0.83 lb.) |
| | For solid cheese (wheels, loaves, or bricks) weighing from 4.54 kg (10 lbs.) or greater | 1 | 750g (1.65 lb.) |
| Other (processed) | Grains, nuts, spices, and seeds (with focus on ready to eat products)<br>Tahini and Halva Candy<br>Dried Milk and Dried Milk Products | 30 | 100g (0.22 lb.) |
| | New/novel foods (newly developed innovative food or food produced using new technologies and production processes) | 15 | 100g (0.22 lb.) |

*NOTE: Collect 30 sub samples at wholesale, warehouse, and packer. May collect 10 sub samples at retail if 30 subs of the same lot are not available.

**Table 3: Sample Size for Microbiological Analytes of Concern Other than *Salmonella* Analysis (Analytes of concern are listed in priority order.)**

| Food Category | Examples of Commodities | Analytes of Concern | Minimum Number of Subs | Minimum Amount per sub needed |
|---|---|---|---|---|
| Raw Agricultural Commodities (RAC) | • Fresh herbs, microgreens, sprouts<br>• Tomatoes<br>• Whole fruits and vegetables collected at retail<br>• Whole fruits and vegetables collected at firms other than retail (e.g., wholesale, packer, or farm) | *Listeria monocytogenes* | 10 | 200g (0.44 lb.) |
| | | EHEC/STEC | 10 | 50g (0.11 lb.) |
| | | *Cyclospora* (fresh produce) | 5 | 50g (0.11 lb.) |
| | | *Cyclospora* (berries) | 5 | 100g (0.22 lb.) |
| | | Hepatitis A/Norovirus | 3 | 150g (0.33 lb.) |
| | • Green leafy vegetables | *Listeria monocytogenes* | 10 | 200g (0.44 lb.) |
| | | EHEC/STEC | 10 | 400g (0.88 lb.) |
| | | *Cyclospora* | 5 | 50g (0.11 lb.) |
| | | Hepatitis A/Norovirus | 3 | 150g (0.33 lb.) |
| | • Large units (e.g., melon, papaya) | Collect at least one whole unit per sub for the desired analysis above. | | |
| Minimally Processed Produce | • Fresh-cut produce (except very large produce)<br>• Fresh green salads<br>• Frozen or dried produce<br>• Non-LACF vacuum packed and modified atmosphere packed raw produce, such as fresh mushrooms and salads, etc. | *Listeria monocytogenes* | 10 | 200g (0.44 lb.) |
| | | EHEC/STEC | 10 | 50g (0.11 lb.) |
| | | *Cyclospora* (fresh produce) | 5 | 50g (0.11 lb.) |
| | | *Cyclospora* (berries) | 5 | 100g (0.22 lb.) |
| | | Hepatitis A/Norovirus | 3 | 150g (0.33 lb.) |
| | • Unpasteurized and non-shelf-stable pasteurized fruit and vegetable juices (*Import only*)<br>• Clear liquid (i.e., bottled water, clear fruit juices) | *Listeria monocytogenes* | 10 | 200g (0.44 lb.) |
| | | EHEC/STEC | 10 | 400mL (0.88 lb.) |

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| Ready-to-eat (RTE) food | • Prepared salads (e.g., deli salads such as macaroni and potato salads)<br>• RTE sandwiches<br>• Baked goods custard or cream filled (Egg containing)<br>• Ice cream<br>• Dips and spreads | *Listeria monocytogenes* | 10 | 200g (0.44 lb.) |
| | | *Staphylococcus aureus* | 10 | 100g (0.22 lb.) |
| | | EHEC/STEC | 10 | 100g (0.22 lb.) |
| | | *Staphylococcus* Enterotoxin Testing (SET) | 10 | 50g (0.11 lb.) |
| Cheese and Cheese Products | For retail units (solid, shredded, grated, curds or extruded forms) weighing less than 454 g (1 lb.) | *Listeria monocytogenes*<br>*Staphylococcus* Enterotoxin Testing (SET)<br>EHEC/ STEC<br>*Staphylococcus aureus* | 10 | Each subsample at least 454 g (1 lb.). If the product unit is less than 454 g collect multiple units to equal at least 454 g. |
| | For retail units (solid, shredded, grated, curds or extruded forms) ranging in weight from 454 g (1 lb.) to less than 2. 27 kg (5 lbs.) | *Listeria monocytogenes*<br>*Staphylococcus* Enterotoxin Testing (SET)<br>EHEC/ STEC<br>*Staphylococcus aureus* | 10 | Each subsample at least 454 g (1 lb.) to less than 2. 27 kg (5 lbs.) |
| | For solid cheese (wheels, loaves, or bricks) ranging in weight from 2.27 kg (5 lbs.) to less than 4.54 kg (10 lbs.) | *Listeria monocytogenes* | 2 | 500g (+250g for enumeration) |
| | | EHEC/ STEC | 2 | 125g (0.27 lb.) |
| | | *Staphylococcus aureus* | 2 | 250g (0.55 lb.) |
| | | *Staphylococcus* Enterotoxin Testing (SET) | 2 | 125g (0.27 lb.) |
| | For solid cheese (wheels, loaves, or bricks) weighing from 4.54 kg (10 lbs.) or greater | *Listeria monocytogenes* | 1 | 1000g (+500g for enumeration) |
| | | EHEC/ STEC | 1 | 250g (0.55 lb.) |
| | | *Staphylococcus aureus* | 1 | 500g (1.1 lbs.) |
| | | *Staphylococcus* Enterotoxin Testing (SET) | 1 | 500g (1.1 lbs.) |

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| Other (processed) Collection and analysis for these analytes of concern should be done upon request | • Grains, nuts, spices and seeds (with focus on ready to eat products)<br>• Tahini and Halva Candy<br>• Dried Milk and Dried Milk Products<br>• New/novel foods (newly developed innovative food or food produced using new technologies and production processes) | *Listeria monocytogenes* | 10 | 200g (0.44 lb.) |
| | | EHEC/ STEC | 10 | 100g (0.22 lb.) |
| | | *Staphylococcus aureus* | 10 | 125g (0.27 lb.) |
| | | *Staphylococcus* Enterotoxin Testing (SET) | 10 | 50g (0.11 lb.) |

**Note:** Samples for multiple analytes (e.g., both *Salmonella* and *Listeria*): collect enough product for the intended analyses to be performed. For example, if 30 subs are needed for *Salmonella* analysis and only 10 of those subs are needed for *Listeria* analysis, the sample should consist of 30 subs total with each sub containing enough product for both analyses, as indicated in Tables 2 and 3.

**Sample Shipping**

Investigators should ship product samples to a servicing laboratory per the Lab Servicing Table (LST). See IOM SUBCHAPTER 4.5 - SAMPLING: PREPARATION, HANDLING, SHIPPING for detailed shipping instructions. For any technical errors or issues identified in the lab servicing dashboard, please reach out to ORALabCapacity@fda.hhs.gov.

**D. Import Activities**

Other import activities such as entry review and field/label examinations are covered under Compliance Program 7303.878 Import Food Operations.

**2. Reporting**

Report sample collections of domestic products in the Field Accomplishment and Compliance Tracking System (FACTS) and samples collected in import status in FDA Import Systems (SERIO/OASIS) using the Program Assignment Codes (PACs) and Problem Area Flags (PAF) in the table below (Table 4). If the proper PAC/PAF combinations are not available, contact the CFSAN Program Monitor.

**Table 4. Reporting PAC and PAF Combinations – Collections**

| PAC Description | PAC | PAF | PAF Description |
|---|---|---|---|
| Domestic & import cheese and cheese product microbiological samples | 03037D | MIC, NAR | Microbiological and Alkaline Phosphatase |
| Domestic human food microbiological samples | 03803D | MIC, NAR, PAR, and VIR | Microbiological |
| Imported human food microbiological samples | 03819C | MIC, NAR, PAR, and VIR | Microbiological |
| Domestic human food environmental samples | 03050N | MIC | Microbiological |

| | | | |
|---|---|---|---|
| State contract domestic human food environmental samples | 03S050N | MIC | Microbiological |
| Domestic and import human food filth samples | 03050B | CCD, FDF, FIL, NAR, and PAR | Filth |
| Domestic and import human food facility exhibit sample | 03050E | FIL, NAR | Filth |

## PART IV - ANALYTICAL

**1. Analyzing Laboratories**

Refer to the LST Dashboard for analyzing laboratory. Send to the lab that has the least amount of (%) capacity utilized at the time of shipping.

**2. Analyses to be Conducted**

Types of microbiological analyses to be performed under this compliance program are based on hazard commodity pair (refer to Table 1. Food Category and Associated Pathogen of Concern).

**3. Microbiological Methodology for all Food Products, excluding Produce**

Use the methodology, special instructions or sample preparation cited in this compliance program and/or the FDA's electronic Bacteriological Analytical Manual (eBAM).
NOTE: For any commodities NOT included in the eBAM or this compliance program, reach out to the designated CFSAN SME to determine the best way to analyze that commodity.
Methodology for CORE Assignments or Mission Critical/follow-up assignments would supersede this document.

**A. Methodology Listed by Organism:**

a) ***Escherichia coli (E. coli)***
Refer to eBAM Chapter 4, Enumeration of *Escherichia coli* and the Coliform Bacteria

b) **EHEC/STEC**
Refer to eBAM Chapter 4a, Diarrheagenic *Escherichia coli*

c) ***Listeria***
Refer to eBAM Chapter 10, Detection of *Listeria monocytogenes* in Foods and Environmental Samples, and Enumeration of *Listeria monocytogenes* in Foods

1. Rapids test kit methods validated as AOAC OMA methods can be used.
2. If *L. monocytogenes* is detected in the sample, enumeration is required when reserve remains.
3. **Compositing:** *Listeria*: Refer to the eBAM Chapter 10; only approved for Buffered *Listeria* Enrichment Broth (BLEB)
4. Enumeration guidance: see Section 5.b. below.

d) ***Salmonella***
Refer to eBAM Chapter 5, *Salmonella*

1. **Compositing**: Refer to the eBAM Chapter 1, Food Sample and Preparation of Homogenate
    a. Contact CFSAN SMEs for any additional information not covered in the eBAM.

2. VIDAS screening methods AOAC 2004.03 or AOAC 2011.03 may be utilized if the product matrix is listed in the official method validation. For unvalidated

matrices the VIDAS screening methods may be used if: in-house matrix extension studies support its use, the most current LIB matrix extension report shows method suitability, or by performing time of use routine spiking; and when authorized by local laboratory management and documented in by lab's quality system.

e) ***Staphylococcus aureus***
Refer to eBAM Chapter 12, *Staphylococcus aureus*
and Appendix 2, Most Probable Number from Serial Dilutions, (MPN dilutions).

1. If *S. aureus* counts exceed 10,000/g (MPN or CFU) analyze the sample for *Staphylococcus* enterotoxin (see Section 3f).

f) ***Staphylococcal* Enterotoxin**
Refer to eBAM Chapter 13B, *Staphylococcal* Enterotoxins Detection Methods
Other Guidance:

1. Enterotoxin testing should be performed if:
   a. Product abuse (e.g., temperature, outbreaks, etc.) is suspected **(or)**
   b. *Instructed by Compliance Program or Field Assignment to analyze the sample for *Staphylococcus*.*
   c. If viable *Staphylococcus sp.* colonies are observed to exceed 10,000/g (MPN or CFU).

2. There is zero tolerance for staphylococcal enterotoxin presence in foods. Detection of any level of staphylococcal enterotoxin is justification that the food is unfit for consumption.

3. **NOTE:** Some species of *Staphylococcus* produce an enterotoxin that is extremely heat stable and not destroyed during food processing procedures or pasteurization temperatures. Therefore, it's possible to have staphylococcal enterotoxin presence without viable *Staphylococcus aureus.* If the product is suspected to have caused staphylococcal food poisoning and undergone retort procedures, see BAM Chapter 13B for detection methods for staphylococcal enterotoxin.

4. **NOTE:** The total contents of each subsample should be retained until the original analyses are completed to ensure that enough product is available for subsequent additional and confirmation tests, if necessary.

5. **CAUTION: Staphylococcal enterotoxins are highly toxic and procedures that may create aerosols should be performed in approved biological safety cabinet (BSC**). **Staphylococcal enterotoxins (SEA, SEB, SEC, SED, and SEE) are select agents. Scientists must follow guidelines established by CDC**: https://www.selectagents.gov/faq-general.html.

g) **Other organisms (Upon special request only)**

**a.** *Vibrio*, including *V. cholera*
Refer to eBAM Chapter 9 *Vibrio*

**b.** *Shigella*
Refer to eBAM Chapter 6 *Shigella*
**c.** *Bacillus cereus* and *Bacillus cereus* enterotoxin
Refer to eBAM Chapter 14 *Bacillus cereus*
**d.** *Clostridium botulinum*
Refer to eBAM Chapter 17 *Clostridium botulinum*
**e.** *Cronobacter* (other than infant formula)
Refer to eBAM Chapter 29 *Cronobacter*
**f.** *Campylobacter jejuni*
Refer to eBAM Chapter7: *Campylobacter*
**g.** *Yersinia enterocolitica*
Refer to eBAM Chapter 8 *Yersinia entercolitica*
**h.** Norovirus
**i.** Hepatitis A

Note: Contact CFSAN SME for any additional information regarding above microorganisms

h) **Alkaline phosphatase (Cheese)**

1. **Screening**: Refer to the eBAM Chapter 27 Screening Method for Phosphatase (Residual) in Cheese Report screening results that exceed the violative threshold as >12ug/g (or applicable violative threshold for the cheese type being tested).
   **NOTE:** Do NOT perform this screening method on any *soft* cheeses containing herbs. Instead perform the method, referenced under section 2 Check Analysis below, directly on these cheeses to determine the phenol equivalent value.

   Refer to Table 1 of BAM, Chapter 27 for the maximum phenol equivalent values allowed in different types of cheese and the 21 CFR Part 133 reference.
   i. Classify alkaline phosphatase analytical results as 'Lab Class 2' and note Center review is indicated: if the cheese is listed in the **List of Standardized Cheeses with Phenol Levels from 21 CFR Part 133 (**ATTACHMENT C**)**, and the phenol equivalent value per gram is greater than the value listed for the cheese in ATTACHMENT C; or,
   ii. if the cheese is NOT listed in ATTACHMENT C, and the phenol equivalent value per gram is greater than 12 ug.

2. Check Analysis (Confirmation): **MUST be performed on ALL samples other than non-violative ones.**
   a. Method: AOAC,18th Ed., 33.7.27 AOAC Official Method 946. 03, Phosphatase (Residual) in Cheese. **NOTE:** This method is equivalent to AOAC Method 16. 275-16. 277 (13th Ed., 1980), cited in 21 CFR 133.5(c).
   b. If a laboratory chooses to use the AOAC method for the screening method, then the check analysis is for a different analyst to run the AOAC method.

**NOTE**: Any cheese sample that has been made from raw/unpasteurized milk, without proper curing or use of a process that is an acceptable alternative to pasteurization as outlined in the applicable regulation in 21 CFR Part 133, is prohibited from introduction into interstate commerce as cited in 21 CFR 1240.61(a).

3. Products to be Analyzed:
    a. Soft/semi-soft/soft-ripened cheese samples **MUST** be analyzed for alkaline phosphatase and for all the other attributes specified in Table 5 "**Compositing and Analysis of Cheese"**
    b. Soft cheeses containing herbs **MUST** be analyzed for alkaline phosphatase and for all the other attributes specified in Table 5 **"Compositing and Analysis of Cheese"**.
    c. Naturally aged (cured) cheese samples should NOT be analyzed for alkaline phosphatase but **MUST** be analyzed for all the other attributes specified in Table 5 "**Compositing and Analysis of Cheese"**. **NOTE**: A naturally aged cheese is one made from unpasteurized milk which has been cured at a temperature of not less than 35 degrees F for not less than 60 days.
    d. Other cheeses that are labeled as either pasteurized or made from dairy ingredients that have all been pasteurized **MUST** be analyzed for alkaline phosphatase and for all the other attributes specified in Table 5 "**Compositing and Analysis of Cheese".**

**B. Compositing and Analysis of Cheese**

Microbiological Analysis: Since the product will be analyzed for multiple pathogens under this program, compositing and individual subsample analysis on the same sample will be necessary. See Section 3 for methodology to utilize for cheese analysis.

**NOTE: IT IS VERY IMPORTANT TO REMOVE PORTIONS FOR MICROBIOLOGICAL ANALYSES FIRST BEFORE PERFORMING ALKALINE PHOSPHATASE ANALYSES. SPECIAL PRECAUTIONS MUST BE TAKEN TO AVOID CONTAMINATING THE PORTIONS FOR MICROBIOLOGICAL ANALYSES AS WELL AS THE PORTION FOR PHOSPHATASE ANALYSES WITH MICROBIAL PHOSPHATASE THAT MAY BE PRESENT ON THE SURFACE.**

**Table 5. Cheese, Cheese Products, and Dried Milk Products**

| Size | Analysis Indicated | # Units Collected | # Subs Analyzed | Compositing or Subsampling Information |
|---|---|---|---|---|
| | EHEC/ STEC | 10 | 10 | 25g from each sub analyzed individually |
| | *Staphylococcus aureus* | 10 | 10 | 50g from each sub analyzed individually |

| | | | | |
|---|---|---|---|---|
| 454g (1lb) to <2.27kg (<5lb) | *Staphylococcus* Enterotoxin Testing (SET) | 10 | 10* | 25g from each sub analyzed individually |
| | Alkaline Phosphatase ** | 10 | 2 | Remove 15g from each of 2 subs avoiding surface when possible and analyze individually |
| | *Listeria monocytogenes* | 10 | 10 | Examine 2 composites: Remove 50 g from 5 subs to make 2 (250g) composites for analysis |
| | *Salmonella* | 10 | 10 | Examine 2 composites: Remove 75 g from 5 subs to make 2 (375g) composites for analysis |
| 2.27kg (5lb) to >4.54kg (<10lb) | EHEC/ STEC | 2 | 10 | Take 5 core plugs (25g each) from each wheel and examine individually for a total of 10 subs |
| | *Staphylococcus aureus* | 2 | 10 | Take 5 core plugs (50g each) from each wheel and examine individually for a total of 10 subs |
| | *Staphylococcus* Enterotoxin Testing (SET) | 2 | 10* | Take 5 core plugs (25g each) from each wheel and examine individually for a total of 10 subs |
| | Alkaline Phosphatase ** | 2 | 2 | Remove a 15g core plug from each sub and analyze individually. The test portions to be analyzed are sampled by taking a portion of 1 cm thick, taken at 0.5 cm below the rind of the round side. |
| | *Listeria monocytogenes* | 2 | 2 | Examine 2 composites: Prepare 1 composite per sub by taking 5 plugs of 50g from each wheel for a 250g composite. |
| | *Salmonella* | 2 | 2 | Examine 2 composites: Prepare 1 composite per wheel by taking 187.5g (including surface of cheese) from each half of the wheel to obtain a 375g composite. Repeat this for the second wheel for a total of 2 composites. |

| | | | | |
|---|---|---|---|---|
| 4.54kg (10lb) and greater | EHEC/ STEC | 1 | 10 | Take 10 plugs (25g each) from the wheel and examine individually for a total of 10 subs |
| | *Staphylococcus aureus* | 1 | 10 | Take 10 plugs (50g each) from the wheel and examine individually for a total of 10 subs |
| | *Staphylococcus* Enterotoxin Testing (SET) | 1 | 10* | Take 10 plugs (25g each) from the wheel and examine individually for a total of 10 subs |
| | Alkaline Phosphatase ** | 1 | 2 | Remove a 15g core plug from each half of the wheel and analyze individually. The test portions to be analyzed are sampled by taking a portion of 1 cm thick, taken at 0.5 cm below the rind of the round side |
| | *Listeria monocytogenes* | 1 | 2 | Prepare 2 composites by taking 5 plugs of 50g for a 250g composite from each half of the wheel. Repeat this process to obtain the second composite. |
| | *Salmonella* | 1 | 2 | Prepare 2 composites by taking 187.5g (including both surfaces of the cheese) from each half of the wheel to obtain a 375g composite. Repeat this process to obtain the second composite. |

* If temperature abuse is suspected or each sub $>10^4$ (MPN or CFU) S. aureus

** Refer to Attachment C- List of Standardized Cheeses with Phenol Levels from 21 CFR Part 133

**4. Microbiological Methodology for Produce**

Use the following methodology for produce samples only, utilizing the previously listed methods for all other commodities.

**a) EHEC/STEC**

Refer to eBAM Chapter 4a, Diarrheagenic *Escherichia coli*

**b) *Listeria***

Refer to eBAM Chapter 10, Detection of *Listeria monocytogenes* in Foods and Environmental Samples, and Enumeration of *Listeria monocytogenes* in Foods

1. Rapids test kit methods validated as AOAC OMA methods can be used.
2. If *L. monocytogenes* is detected in the sample, enumeration is required when reserve remains EXCEPT for environmental swabs and some produce (if decaying or otherwise compromised). If you are unsure if enumeration is necessary, please contact the ORS Program Coordinator.
3. **Compositing:** *Listeria*: Refer to the eBAM Chapter 10; only approved for Buffered *Listeria* Enrichment Broth (BLEB)
4. Enumeration guidance: see Section 5.b. below.

**c) *Salmonella***

Refer to eBAM Chapter 5, *Salmonella* and CFSAN guidance for produce analysis for exceptions and/or supplements to the BAM method detailed below.

1. For "large fruit or vegetable" produce such as honeydew and Korean melon; papayas; cucumbers; avocados; apples; peaches; etc., follow the instruction in BAM Chapter 5, section C.21 for whole cantaloupe.
    a. Typically, only ten (10) subsamples are collected due to produce size. If more are collected, only analyze ten (10) subs, unless additional instruction is provided.
    b. Day 1 - individually incubate each subsample pre-enrichment.
        i. Each subsample should be either one large produce item, or when multiple units make up a subsample, enough product to weigh approximately 454 grams.
        ii. Float each subsample in 1.5 times the volume of broth as the total sub weight.
    c. Day 2 - wet composite the individual subs into two (2), five (5) sub composites.

2. For whole round tomatoes and Roma tomatoes, follow the instruction in BAM Chapter 5, section C.23 for whole tomatoes, *except blend individual subs.*
    a. Typically, only ten (10) subsamples are collected. If more are collected only analyze ten (10) subs unless additional instruction is provided.
    b. Day 1 - individually incubate subsample pre-enrichments. Each subsample should be either one large tomato, or when multiple tomatoes make up a subsample, enough product to weigh approximately 454 grams.
    c. Day 2 - wet composite the individual subs into two (2), five (5) sub composites.

3. For cherry tomatoes and husk tomatoes (tomatillos), follow the instruction in BAM Chapter 5, section C.23 for whole tomatoes.
    Either ten (10) or thirty (30) subsamples may be collected. Make two, 375-gram composites of either five (5) or fifteen (15) subs respectively, unless additional instruction is provided.

4. For fresh green leafy vegetables (lettuce, kale, spinach, etc.); herbs (basil, cilantro, parsley, etc.); and sprouts, follow the instruction in BAM Chapter 5, section C.27.
   Either ten (10) or thirty (30) subsamples may be collected. Make two (2), 375-gram composites of either five (5) or fifteen (15) subs respectively, unless additional instruction is provided.

5. For green onions or scallions; bulb onions including red, yellow, and white onions; berries including blueberries and strawberries; and peppers including both sweet and hot peppers (all varieties), follow the instruction in BAM Chapter 5, section C.7.d for fresh, frozen, or dried vegetables.
   a. For green onions or scallions, and berries (blueberries, strawberries, etc.), either ten (10) or thirty (30) subsamples may be collected.
      Day 1 - make two (2), 375-gram composites of either five (5) or fifteen (15) subs respectively, unless additional instruction is provided. Float the product in 3375 mL pre-enrichment.
   b. For larger bulb onions (red, yellow, or white), hot peppers (jalapeno, serrano, habanero, etc.), and sweet peppers, normally only ten (10) subsamples of at least 454 grams are collected.
      i. Note that bulb onions should be cut in half during sample prep.
      ii. Day 1 - individually incubate each subsample pre-enrichment.
         1. Each subsample should be enough product to weigh approximately 454 grams.
         2. Float each subsample in 1.5 times the volume of broth as the total sub weight.
      iii. Day 2 - wet composite the individual subs into two (2), five (5) sub composites.

6. **Compositing:** Refer to the eBAM Chapter 1, Food Sample and Preparation of Homogenate
   a. Wet compositing generally only applies to large fruits or vegetables or other produce where instruction is specified.
      i. On day 1, enrich and incubate ten (10) subs individually.
      ii. On day 2, prepare two (2), five (5) sub c as follows.
         1. RV: transfer 0.1 mL pre-enrichment broth, from each of 5 subsamples into 50 mL RV medium. Incubate per eBAM Chapter 5.
         2. TT: transfer 1.0 mL pre-enrichment broth, from each of 5 subsamples into 50 mL TT broth. Incubate per eBAM Chapter 5.
         3. Alternatively, if using *Salmonella* VIDAS Easy method (AOAC 2011.03): transfer 0.1 mL pre-enrichment broth, from each of 5 subsamples into 50 mL SX2 broth. Incubate per official method.

7. Matrix microbial load guidance for selective enrichment temperature selection:

a. High Microbial load matrices: Onions (whole round), cilantro, scallions (green onion), berries, melons, apples, peaches, papaya, basil, sprouts, parsley.
b. Low Microbial load matrices: peppers, (all varieties), tomatoes, limes, avocado, mangoes, cucumbers.

8. VIDAS screening methods AOAC 2004.03 or AOAC 2011.03 may be utilized if the product matrix is listed in the official method validation. For unvalidated matrices the VIDAS screening methods may be used if: in-house matrix extension studies support its use, the most current LIB matrix extension report shows method suitability, or by performing time of use routine spiking; and when authorized by local laboratory management and documented in by lab's quality system.

**d) *Cyclospora***
Refer to eBAM Chapter 19b, Detection of *Cyclospora cayetanensis* in Fresh Produce using real-time PCR.

**e) Hepatitis A and/or Norovirus**
Refer to eBAM Chapter 26 Concentration, Extraction and Detection of Enteric Viruses in Foods
- Non-acidic Produce (pH 7 or above): Concentration: Section A1: Leafy greens
- Acidic Produce (pH<7): Concentration: Section A2: Soft Fruit

**5. Positive Samples**

**a. Whole Genome Sequencing (WGS)**
All positive samples for the following pathogens must send an isolate to WGS for analysis, unless otherwise specified or requested.
*a.* *Salmonella*
b. *Listeria monocytogenes*
c. *E. coli* O157:H7 or other Shiga toxin producing *E. coli*

**b. *Listeria* enumeration:**
Cultural Enumeration Method: Perform on all *Listeria monocytogenes* positive samples with remaining product by the enumeration method in the current version of eBAM Ch. 10. Typical enumeration can be accomplished by MPN or direct plate count methodologies. For non-CORE or routine samples, enumeration analysis is performed as composite(s). This is performed by creating new composite(s) from retained subs that made the original composite(s). For CORE samples or death related samples, enumeration analysis is performed on individual subs (no compositing).

**c. Serotyping for *Salmonella*:**
1. Molecular Serotyping
    a. SeqSero: All laboratories will process their samples internally
    b. Refer to eBAM Chapter 5.
2. Isolates that Require Traditional Serotyping:

a. Laboratory should assign to DENLHAF in FACTS.
b. Refer to eBAM Chapter 5, *Salmonella*, section E.11 for instructions.

**d. Antibiotic Resistance Testing (ABR):**

1. Organisms to submit:
    a. *Salmonella* Isolates
        i. *Salmonella* isolates submitted for traditional serotyping
            1. One of each serotype from each sub/composite will be assigned to and sent to DENLHAF for ABR analysis.
            2. The analyst performing serotyping is responsible for submitting the isolate(s) to DENLHAF for ABR analysis.

        ii. *Salmonella* isolates **NOT** submitted for traditional serotyping
            1. One of each serotype from each sub/composite will be assigned to and sent directly to DENLHAF for ABR analysis.
            2. The laboratory will designate an individual responsible for submitting the isolate(s) to DENLHAF for ABR analysis.

    b. *Listeria monocytogenes* and *Escherichia coli* isolates
        1. As per assignment or otherwise requested
            a. One of each serotype from each sub/composite will be assigned to and sent directly to DENLHAF for ABR analysis.

2. How to label tubes for submission:
    a. Sample Number
    b. Composite/Sub-isolate number
    c. Date
    d. Initials
    e. Name of organism

**6. Reporting**

Refer to FMD-147: Communicating Laboratory Analytical Findings for Food Products and Environmental Samples Directive for detailed reporting instructions. Report all analytical results into the Field Accomplishment and Compliance Tracking System (FACTS) using the following Program Assignment Codes (PACs) and Problem Area Flags (PAFs):

**Table 6. Reporting PAC and PAF Combinations - Analysis**

| PAC Description | PAC | PAF | PAF Description |
|---|---|---|---|
| Domestic & import cheese and cheese product microbiological samples | 03037D | MIC, NAR | Microbiological and Alkaline Phosphatase |
| Domestic human food microbiological samples | 03803D | MIC, NAR, PAR, and VIR | Microbiological |
| Imported human food microbiological samples | 03819C | MIC, NAR, PAR, and VIR | Microbiological |
| Domestic human food environmental samples | 03050N | MIC | Microbiological |
| State contract domestic human food environmental samples | 03S050N | MIC | Microbiological |
| Domestic and import human food filth samples | 03050B | CCD, FDF, FIL, NAR, and PAR | Filth |
| Domestic and import human food facility exhibit sample | 03050E | FIL, NAR | Filth |

## PART V - REGULATORY/ADMINISTRATIVE STRATEGY

The overarching compliance goal is to prevent the introduction of adulterated product into U.S. commerce, remove adulterated product from U.S. commerce and prevent future entry of adulterated product into U.S. commerce. Use the following table as an aid when determining appropriate regulatory responses for microbiological findings on a case-by-case basis. The table is not inclusive of all FDA enforcement strategies. FDA may consider additional enforcement strategies to ensure food products do not present a risk to consumers. Any possible response depends on the firm's voluntary corrective actions, type of food processed, packed, or held in the firm, historical information related to the firm and product, and WGS genetic information related to the isolates. Enforcement action recommendations should be submitted directly to CFSAN Division of Enforcement via the Compliance Management System (CMS), and sample(s) should be linked to appropriate inspection or investigation, if performed. Contact the CFSAN Division of Enforcement point-of-contact for further direction.

**Table 7. Possible FDA Response to Microbiological Findings**

<table>
<tr><th rowspan="2">Microorganism</th><th rowspan="2">Charge(s)</th><th colspan="2">Possible FDA Response<br>*This list is not meant to be all encompassing</th></tr>
<tr><th>Domestic</th><th>Foreign</th></tr>
<tr><td>Listeria monocytogenes</td><td rowspan="4">402(a)(1)<br>402(a)(4)<br>801(a)(3)</td><td rowspan="4">• Request voluntary corrective action<br>• Voluntary recall<br>• Mandatory recall<br>• FDA issuance of a Public Warning<br>• Consider inspection of domestic firm<br>• Regulatory meeting<br>• Warning Letter<br>• Administrative detention<br>• Seizure<br>• Injunction<br>• Suspension of food facility registration</td><td rowspan="4">• Request voluntary corrective action<br>• Voluntary recall<br>• Mandatory recall<br>• FDA issuance of a Public Warning<br>• Consider inspection of foreign firm<br>• Detention/Refusal of Admission<br>• Increased Screening Criteria<br>• Import Alert<br>• Import Bulletin<br>• Warning Letter<br>• Notify FSVP importer of findings or conduct FSVP inspection</td></tr>
<tr><td>Salmonella spp.</td></tr>
<tr><td>EHEC/STEC</td></tr>
<tr><td>Staphylococcal enterotoxin or Bacillus cereus enterotoxin</td></tr>
</table>

| | | | |
|---|---|---|---|
| *Cronobacter* (*Enterobacter sakazakii*) spp if not found in infant formula | | | |
| *Clostridium botulinum* | 402(a)(1)<br>402(a)(4)<br>801(a)(3) | • Request voluntary corrective action<br>• voluntary recall<br>• Mandatory recall<br>• FDA issuance of a Public Warning<br>• Consider inspection of domestic firm<br>• Regulatory meeting<br>• Warning Letter<br>• Administrative detention<br>• Seizure<br>• Injunction<br>• Emergency permit if low acid canned food or acidified food product<br>• Suspension of food facility registration | • Request voluntary corrective action<br>• voluntary recall<br>• FDA issuance of a Public Warning<br>• Consider inspection of foreign firm<br>• Detention/Refusal of Admission<br>• Increased Screening Criteria<br>• Import Alert<br>• Import Bulletin<br>• Warning Letter<br>• Notify FSVP importer of findings or conduct FSVP inspection |
| *Vibrio*<br>Including *V. cholera*<br>*Shigella* | 402(a)(1)<br>801(a)(3) | • Request voluntary corrective action<br>• voluntary recall<br>• Mandatory recall<br>• FDA issuance of a Public Warning<br>• Consider inspection of domestic firm<br>• Regulatory meeting<br>• Warning Letter<br>• Administrative detention<br>• Seizure | • Request voluntary corrective action<br>• voluntary recall<br>• FDA issuance of a Public Warning<br>• Consider inspection of foreign firm<br>• Detention/Refusal of Admission<br>• Increased Screening Criteria<br>• Import Alert<br>• Import Bulletin<br>• Warning Letter<br>• Notify FSVP importer of findings or conduct FSVP inspection |

| | | | |
|---|---|---|---|
| *Staphylococcus aureus* (*S. aureus*) or *Bacillus cereus* greater than or equal to $10^4$ colony forming units per gram (cfu/g)<br><br>Viruses (Norovirus, Hepatitis A) | 402(a)(4)<br><br>801(a)(3) | • Request voluntary corrective action<br>• voluntary recall<br>• Mandatory recall<br>• Consider inspection of domestic firm<br>• Regulatory meeting<br>• Warning Letter<br>• Administrative detention<br>• Seizure<br>• Injunction | • Request voluntary corrective action<br>• voluntary recall<br>• Consider inspection of foreign firm<br>• Detention/Refusal of Admission<br>• Increased Screening Criteria<br>• Import Alert<br>• Import Bulletin<br>• Warning Letter<br>• Notify FSVP importer of findings or conduct FSVP inspection |
| *Cyclospora* | 402(a)(4)<br><br>801(a)(3) | • Request voluntary corrective action<br>• Request voluntary recall<br>• Consider inspection of domestic firm<br>• Regulatory meeting<br>• Warning Letter | • Request voluntary corrective action<br>• voluntary recall<br>• Consider inspection of foreign firm<br>• Detention/Refusal of Admission<br>• Increased Screening Criteria<br>• Import Alert<br>• Import Bulletin<br>• Warning Letter<br>• Notify FSVP importer of findings or conduct FSVP inspection |
| *Listeria* spp. (other than *L. monocytogenes*)<br><br>Non-pathogenic *E. coli*<br><br>**Cronobacter* (*Enterobacter* | N/A<br><br>(a)(4)- may be considered depending on a variety of factors, including | • Request voluntary corrective action<br>• Consider inspection of domestic firm | • Request voluntary corrective action<br>• Resample the shipment as resources permit<br>• Consider inspection of foreign firm<br>• Increase Screening Criteria |

| | | | |
|---|---|---|---|
| *sakazakii)* spp. if not found in infant formula | prevalence, frequency, and any other adverse observations | | • Inform the importer of record, who may not be the FSVP importer, of the significance of the findings and request that they inform the manufacturer. |

*Contact CFSAN Regulatory/Compliance contact to discuss regulatory strategy.
In addition to the above regulatory strategies, Divisions should also review Regulatory Strategy for Cheese and Cheese Products below, Preventive Controls and Sanitary Human Food Operations Compliance Program (7303.040), FY21-22 Produce Safety Assignment (DFPG #21-02), FY21 Sprout Firm Inspection and Sampling Assignment (DFPG #21-03), CFSAN Enforcement Bulletins, the Food, Color and Cosmetics Chapter 5 of the Compliance Policy Guides (CPG), and Regulatory Procedures Manual (RPM).

**Regulatory Strategy for Cheese and Cheese Products**
Divisions should review the instructions provided below, Table 8, and Attachment C to aid in determining appropriate regulatory response to microbiological contamination, filth, or alkaline phosphatase results of cheese and cheese products. Further, when pathogens are detected in a cheese or cheese product sample, Division should consider sampling of raw materials (incoming raw milk if pathogenic *E. coli* was detected in cheese), finished product cheese, and/or the environment of the manufacturing facility (if a domestic manufacturer). In addition, Division should consider interaction with Preventive Controls and Sanitary Human Food Operations compliance program to verify adequacy of sanitation.

**Direct Reference:**
The following, as specified in CPG Sec. 527.300, Dairy Products- Microbial Contamination and Alkaline Phosphatase Activity, represent criteria for direct reference seizure submission to the Division of Compliance Management and Operations (HFC-210), for direct reference import detention by the district and for direct reference submission of detention without physical examination (DWPE) to ORA, Office of Regional Operations, Division of Import Operations and Policy (HFC-170):

> Analysis of the dairy product demonstrates that one or more subsamples is positive for *Salmonella* species, *Campylobacter jejuni*, *Yersinia enterocolitica*, *Staphylococcus* enterotoxin, or *Bacillus cereus* enterotoxin.

**Domestic products:**

- When finished product samples are positive for presence of *Listeria monocytogenes,* consult with CFSAN for regulatory action instructions.
- If no INTERSTATE documentation can be associated with violative cheese samples, contact State authorities to coordinate appropriate follow-up activities. Provide assistance to the State for their follow-up activity, if requested.

**Import Products:**

- Recommendations for detention without physical examination of imported products should be referred to the Division of Import Operations, Import Compliance Branch through CMS. Enforcement action recommendations should be submitted directly to CFSAN Division of Enforcement via CMS, and sample(s) should be linked.

- Divisions may consider detention of cheese and cheese products when samples are positive for presence of human pathogens and enterotoxins as authorized (see CPG Sec. 527.300, Dairy Products - Microbial Contamination and Alkaline Phosphatase Activity).

- For imported products that are in domestic or domestic/import status, contact the CFSAN Division of Enforcement POC for further guidance when *E. coli*, Enterohemorrhagic *E. coli*, STEC or *Staphylococcus aureus* are found.

- Recommendations for detention must be accompanied by all analytical worksheets (original and check, when required) and other appropriate documentation (entry paperwork, collection report, labeling, etc.).

**Table 8: Sample Results and Possible Regulatory Responses for Cheese and Cheese Products**

| Sample Result | FD&C Act Charge | Possible FDA Response[1] |
|---|---|---|
| *Salmonella* spp.<br>*Listeria monocytogenes*<br>Enterohemorrhagic *E. coli* (O157:H7) and Shiga toxin-producing *E. coli* (Non-O157:H7)[2]<br>Staphylococcal enterotoxin<br>*Bacillus cereus* enterotoxin | DOMESTIC<br>402(a)(1)<br>402(a)(4) | • Request voluntary recall<br>• Administrative Detention<br>• Seizure<br>• Injunction<br>• Mandatory recall<br>• Suspension of Registration<br>• Environmental sampling at manufacturing site (if *Salmonella* spp. or *L. monocytogenes* is detected) |
| | IMPORT<br>402(a)(1)/ 801(a)(3)<br>402(a)(4)/ 801(a)(3) | • Detention/Refusal<br>• Addition of firm/product to applicable Import Alert(s)<br>• Consider increased screening criteria<br>• Consider inspection of foreign firm |
| *S. aureus* - Greater than or equal to 10^4 colony forming units per gram (cfu/g)<br>*B. cereus* - Greater than or equal to 10^4 colony forming units per gram (cfu/g) | DOMESTIC<br>402(a)(4) | • Warning Letter<br>• Request voluntary recall<br>• Regulatory Meeting<br>• Seizure of adulterated lot |
| | IMPORT<br>402(a)(4)/ 801(a)(3) | • Detention/Refusal<br>• Addition of firm/product to applicable Import Alert(s)<br>• Consider increased screening criteria<br>• Consider inspection of foreign firm<br>• Center supported detention/refusal |

[1] See CPG Sec. 527.300, Dairy Products- Microbial Contamination and Alkaline Phosphatase Activity

[2] Samples that are only positive for *stx1* and/or *stx2* are indicative that non-O157 STEC may be present. There are ~ 300 serotypes of STEC and not all appear to cause severe illness in humans, therefore, these isolates require further testing. Follow the procedure described in BAM Chap. 4A, Sec. R. for isolation of non-O157 STEC.

| Sample Result | FD&C Act Charge | Possible FDA Response[1] |
|---|---|---|
| Alkaline Phosphatase | DOMESTIC<br>402(a)(4) | • Determine if there is evidence that bovine milk is inadequately pasteurized or unpasteurized<br>• Request of voluntary recall Warning Letter |
| | IMPORT<br>402(a)(4)/ 801(a)(3) | • Consider Detention/refusal<br>• Consider inspection of foreign firm |
| Filth | DOMESTIC<br>402(a)(3) | • Warning Letter<br>• Request voluntary recall<br>• Seizure of adulterated lot |
| | IMPORT<br>402(a)(3)/ 801(a)(3) | • Detention/refusal<br>• Consider inspection of foreign firm |

**Alkaline phosphatase findings for both domestic and imported cheese and cheese products**: Presence of alkaline phosphatase activity at levels above those published in 21 CFR 133 indicates that some or all the milk used may not have been properly pasteurized or that it has been contaminated with raw milk; samples with such results will be categorized as 'Lab Class 2'.

Divisions should submit recommendations for regulatory action to CFSAN based on phosphatase findings using the phenol equivalent value cited in 21 CFR Part 133 (see the table below).

For standardized cheeses without a phenol equivalent value cited in 21 CFR Part 133 and non-standardized cheeses:

- a cheese sample made from PASTEURIZED milk with a phenol equivalent gram greater than 12 mg, or
- a cheese sample made from UNPASTEURIZED milk and cured (aged) properly with a phenol equivalent value per gram greater than 12 mg, or
- ANY cheese sample made from either RAW or UNPASTEURIZED milk without proper curing or use of a process that is an acceptable alternative to pasteurization as outlined in 21 CFR part 133. 21 CFR 1240.61(a) prohibits such cheeses from introduction into interstate commerce.

**NOTE:** Before recommending regulatory action, check analyses must be performed on all violative samples using the AOAC 16TH EDITION METHOD 943. 03 (33. 7.27), APHOSPHATASE (RESIDUAL) IN CHEESE (FINAL ACTION). This method is equivalent to the AOAC 13TH EDITION METHOD, APHENOL EQUIVALENT VALUE, SECTIONS 16. 275-16. 277, as cited in 21 CFR PART 133 SECTION 133. 5(c). (Current methodology for detecting the presence of alkaline phosphatase in milk and milk products has only been validated for bovine milk and bovine milk products.)

## PART VI REFERENCES, ATTACHMENTS, AND PROGRAM CONTACTS

### 1. References

A. Investigations Operations Manual (IOM)
B. Regulatory Procedures Manual (RPM)
C. Import Alerts and Bulletins (CMS; FDA.Gov)
D. Sample Collection Operation Planning Evaluation (SCOPE)
E. ORA Field Work Plan
F. Food Compliance Programs
G. CFSAN or ORA active assignments
H. CFSAN Direct Reference
I. Compliance Management System (CMS)
J. CFSAN Enforcement Bulletins
K. Lab Servicing Table (LST)
L. FDA Memoranda of Understanding
M. Cooperative Arrangements
N. Proposed Food Traceability List (FTL)
O. Draft Guidance for Industry: Standards for the Growing, Harvesting, Packing, and Holding of Produce for Human Consumption
P. Part 112 - Standards For The Growing, Harvesting, Packing, And Holding Of Produce For Human Consumption (Produce Rule)
Q. FSMA Proposed Rule for Food Traceability
R. Guidance for Industry: Guide to Minimize Microbial Food Safety Hazards of Fresh-cut Fruits and Vegetables
S. FDA's electronic Bacteriological Analytical Manual (eBAM)
T. Draft Guidance for Industry: Hazard Analysis and Risk-Based Preventive Controls for Human Food, Appendix 1: Potential Hazards for Foods and Processes
U. Resource Library
V. Raw Agricultural Commodities (RAC)
W. FMD 147 Communicating Laboratory Analytical Findings for Food Products and Environmental Samples Directive
X. Release of ORA Laboratory Analytical Results to the Responsible Party: Guidance for Food and Drug Administration Staff
Y. QMS Standard Operating Procedure (SOP) Template
Z. Inspection Guides
AA. International Cooperation on Food Safety
BB. Part 133-Cheeses and Related Cheese Products

**2. Attachments**

A. ATTACHMENT A – Environmental Sampling
B. ATTACHMENT B – Filth and Decomposition
C. ATTACHMENT C – List of Standardized Cheeses with Phenol Levels from 21 CFR Part 133

**3. Program Contacts**

Questions related to the content of Attachment A (Environmental Sampling), firm prioritization, firm replacement, RTE assessments, or enforcement actions should be directed toward the applicable contact below.

**A. CFSAN General Program Contact**

Janice King, CFSAN/OC/DFPG/Program Assignment and Monitoring Branch, Janice.King@fda.hhs.gov, 843-746-2990 ext. 16

**B. CFSAN Regulatory/Compliance Contact**

DEMicroSamplePOC@fda.hhs.gov
Contact this distribution list for regulatory follow-up on environmental or product positive findings, including requests for WGS comparison.

Leslie Hintz, CFSAN/OC/DE, Leslie.Hintz@fda.hhs.gov, 240-402-2073
Contact for other regulatory questions related to micro findings.

Robert Hatch, CFSAN/OC/DE, Robert.Hatch@fda.hhs.gov, 240-402-6081
Contact for import regulatory questions.

**C. CFSAN Office of Food Safety**

General: Donald Kautter, CFSAN/OFS/DPPB, 240-402-4072
Donald.Kautter@fda.hhs.gov

Cheese: Monica Metz, CFSAN/OFS/DDEMP, 240-402-2041
Monica.Metz@fda.hhs.gov

Filth: Monica Pava-Ripoll, CFSAN/OFS/DDEMP, 240-402-1630
Monica.Pava-Ripoll@fda.hhs.gov

RAC Produce: Mary Tijerina, CFSAN/OFS/DPS, 240-402-0405
Mary.Tijerina@fda.hhs.gov

Processed Produce: Insook Son, CFSAN/OFS/DPS, 240-402-1648
Insook.Son@fda.hhs.gov

**D. CFSAN Analytical Contacts**

Eric Brown, CFSAN/ORS (technical laboratory based WGS inquiries), Eric.Brown@fda.hhs.gov, 240-402-2020

James Pettengill, CFSAN/Biostatistics Staff (WGS data uploading inquiries), James.Pettengill@fda.hhs.gov, 240-402-1992

Thomas Hammack, CFSAN/ORS, Thomas.Hammack@fda.hhs.gov, 240-402-2010

Hua Wang, CFSAN/ORS (*Salmonella*), Hua.Wang@fda.hhs.gov, 240-402-1932

Yi Chen, CFSAN/ORS (*Listeria*), Yi.Chen@fda.hhs.gov, 240-402-2783

Julie Kase, CFSAN/ORS (STECs), Julie.Kase@fda.hhs.gov, 240-402-2923

Jacquelina Woods (Hepatitis A/Norovirus), CFSAN/OFS
Jacquelina.Woods@fda.hhs.gov, 251-406-8148

Jessica Jones (*Vibrio*), CFSAN/OFS, Jessica.Jones@fda.hhs.gov, 251-406-8136

Socrates Trujillo (*Cyclospora*), CFSAN/OARSA,
Socrates.Trujillo@fda.hhs.gov, 240-402-2037

**E. ORA/ Office of Human and Animal Food Operation (OHAFO), Division of Domestic**

Human and Animal Food Operations (DDHAFO)
Monali Yajnik, ORA/OHAFO/ DDFHAO, Monali.Yajnik@fda.hhs.gov, 240-402-1616
Martha (Marti) Myrick, Martha.Myrick@fda.hhs.gov, 240-402-5840

**F. ORA/Office of Enforcement and Import Operations (OEIO)/Division of Import Operations (DIO)**

ORA/OEIO DIO CFSAN Liaisons: ORAOEIODIOCFSANLiaisons@fda.hhs.gov

**G. ORA Office of Regulatory Science Contact**

ORA ORS OFFLO IO – Micro Mailbox (oraorsoffloiomicro@fda.hhs.gov)

## PART VII - CENTER RESPONSIBILITIES

The Office of Food Safety 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. In addition, the Office of Compliance will prepare an annual summary report of this compliance program which will be available at Compliance Program Summaries.

## ATTACHMENT A – ENVIRONMENTAL SAMPLING

**Background:**
The contamination of ready-to-eat (RTE) food by environmental pathogens (i.e. *Listeria monocytogenes* and *Salmonella*) in food processing facilities presents a significant hazard to public health. RTE food is defined in 21 CFR 117.3 as any food that is normally eaten in its raw state or any other food, including a processed food, for which it is reasonably foreseeable that the food will be eaten without further processing that would significantly minimize biological hazards. Facilities that manufacture, process, package, or hold RTE food exposed to the environment prior to packaging and the packaged food does not receive a treatment or otherwise include a control measure (such as a formulation lethal to the pathogen) that would significantly minimize the pathogen are responsible for ensuring that the hazard of environmental pathogens is controlled in their facilities through application of current good manufacturing practices (CGMPs) and/or preventive controls. For-cause and surveillance environmental sampling during inspections help to determine the prevalence of environmental pathogens in certain RTE food facilities.

When an RTE food is exposed to the environment prior to packaging and the packaged food does not receive a treatment or otherwise include a control measure that would significantly minimize the pathogen, poor implementation of CGMPs and/or preventive controls by the facility can lead to product contamination with environmental pathogens. Environmental sampling is a valuable tool to determine this risk and guide FDA's compliance action.

Environmental pathogens may establish themselves in "niche environments" and can grow to high numbers and become difficult to manage. *Listeria monocytogenes* survives and grows well in areas of the facility where there is a constant source of moisture while *Salmonella* prefers dry areas of a facility where water is periodically introduced and dries over an extended period. Moisture could be introduced into the processing environment from various sources including the manufacturing process itself, cleaning and sanitation, leaking pipes or roofs, employee or equipment movement from wet areas of the facility, and condensate. Additional information concerning environmental pathogens may be found in Background and the Control of *Listeria monocytogenes* in Ready-To-Eat Foods: Guidance for Industry (Draft Guidance).

In the past, positive environmental samples of *Listeria monocytogenes* and *Salmonella* have been analyzed using pulse-field gel electrophoresis (PFGE). Conventional approaches, like PFGE alone, lack the resolution needed for differentiating tightly linked bacteria (*i.e.*, those highly clonal isolates that comprise a common PFGE pattern). Whole genome sequencing (WGS), however, compares the 4.6 million genetic base pairs composing a *Salmonella*'s DNA, for example. This level of sensitivity and certainty associated with the resulting strain comparisons permits more precise foodborne strain cluster detection and traceback. WGS analysis of bacterial human pathogens provides high-resolution data, enabling direct links to be established between clinical isolates and food or environmental sources of bacterial contamination. WGS data can also be used to infer the evolutionary relationships (or phylogeny) within a given set of isolates as it measures each DNA position in a bacterial genome. WGS can also assist in determining if the same strain of bacteria has been present in the food facility's environment over time and may be a resident strain or if the isolates are transient strains that may be coming into the food facility's environment from other means such as a possible outside source.

**Note:** This compliance program does not address on-farm environmental sampling for covered RAC produce, which will be addressed in Produce Safety Inspection compliance program (under development).

**Objectives:**

- Perform environmental sampling as a component of inspections following Field Bulletin (FB) #30 *Instructions for Environmental Sampling* at prioritized domestic human food facilities that manufacture RTE foods that are exposed to the environment prior to packaging and will not undergo further processing or do not contain a control measure (such as a formulation lethal to the pathogen) to control pathogens.
- Perform whole-genome sequencing (WGS) for all positive isolates.
- Document positive findings and initiate compliance action as warranted.

**Program Management Instructions**:

A. Sampling Priorities

Environmental sampling should be conducted as part of inspections. Divisions may choose any facility that is recommended in the FSMA Tracker for environmental sampling and that manufactures RTE food exposed to the environment prior to packaging and will not undergo further processing or does not contain a control measure (such as a formulation lethal to the pathogen) to control pathogens.

Follow the criteria for prioritization of firms for environmental sampling:

- the facility is responsible for a Class I recall, outbreak, or lab class 3 sample of an RTE food for *Listeria monocytogenes* or *Salmonella* contamination since the previous inspection and follow-up is required to determine if the pathogen contamination originated from the facility's environment, **OR**
- the facility's previous inspection had an initial or final classification of "Official Action Indicated" (OAI) associated with **significant** CGMP and/or sanitation control observations and the Division determines that environmental sampling is required to determine if the conditions may lead to the contamination of food with environmental pathogens, **OR**
- the facility's current inspection reveals **significant** CGMP and/or sanitation control observations that may result in the contamination of RTE food with environmental pathogens, **OR**
- information is obtained from another source such as findings from FDA field assignments; federal, state, local, or tribal partners; foreign competent authorities (e.g. the rapid alert system for food and feed (RASFF) or cooperative agreement); the Reportable Food Registry (RFR); or consumer complaints that credibly indicates that the facility has **significant** CGMP and/or sanitation control deficiencies and/or RTE food is adulterated with *Listeria monocytogenes* or *Salmonella* and presents a risk to public health, **AND**
- the facility is known to manufacture RTE foods as defined in Part I Background of this attachment and the RTE foods are exposed to the environment prior to packaging and

will not undergo further processing or do not contain a control measure (such as a formulation lethal to the pathogen) to control pathogens.

Divisions may also use the above criteria to select facilities for inspection that do not appear on the list of recommended facilities in the FSMA Tracker. However, whenever such a decision is made, the Divisions may inform the compliance program monitor of this compliance program via e-mail and include the facility's name, address, and FEI. If there are any questions or if the facility does not qualify for environmental sampling under this compliance program, the compliance program monitor may contact the Division for more information.

If environmental sampling is being recommended based on observations during an ongoing inspection, the Division should determine whether the facility has an environmental monitoring program and how it is being implemented. Divisions should provide this information to the compliance program monitor. The implementation of a robust environmental monitoring program at the facility may be a factor in determining whether an environmental sampling may be approved during the inspection under this compliance program.

For-cause environmental sampling follow-up inspections may count toward environmental sampling work plan obligations if the criteria above are met. If a Division is unsure whether a facility is eligible for environmental sampling, contact the compliance program monitor.

B. Planning Instructions

Environmental sampling operations will be planned each fiscal year in the ORA Field Work Plan. Environmental sampling should not take place if a facility does not manufacture RTE foods that are exposed to the environment prior to packaging and will not undergo further processing or does not contain a control measure (such as a formulation lethal to the pathogen) to control pathogens.

C. Interactions with Other Compliance Programs

Compliance programs that cover inspections of firms with RTE food exposed to the environment with no further processing or control measures (such as a formulation lethal to the pathogen) to destroy pathogens could potentially include environmental sampling. The compliance programs listed below are those that interact with environmental sampling.

(1) Preventive Controls and Sanitary Human Food Operations (CP 7303.040)

Many inspections conducted in conjunction with environmental sampling under this compliance program will include coverage of foods subject to the CGMP & PCHF rule. Report the PAC for environmental sampling when inspections include environmental sampling.

CP 7303.040 details the criteria for performing inspections including:

- CGMP inspections (PAC 03040),
- Full scope PCHF inspections (PAC 03040F),

- PCHF follow-up inspections (PAC 03040U), and
- Modified requirements at qualified facilities inspections (PAC 03040Q).
- Human Foods Sanitary Transportation Inspections (PAC 03040T)

If an RTE food covered during an inspection is subject to 21 CFR 117 subpart C, a limited scope PCHF inspection must not be performed when performing environmental sampling operations under this compliance program.

(2) Seafood Processor Inspection Program (CPGM 7303.842)
Environmental sampling operations may be conducted during inspections of facilities covered by 21 CFR 123 *Fish and Fishery Products*.

(3) National Conference on Interstate Milk Shipments (NCIMS) MILK SAFETY PROGRAM (CPGM 7318.003)
FDA has authority to regulate the interstate commerce of dairy products under the FD&C Act as well as under the Public Health Service Act (42 U.S.C. 246 and 21 CFR 1250.26) to provide assistance to states and to advise the states on matters pertaining to the preservation and improvement of public health as it pertains to dairy products. Should a significant sanitation concern or a critical processing element be encountered and warrants for-cause environmental sampling, coordination with Liaisons and milk specialists are required prior to performing environmental sampling. Resources for environmental sample collection and analysis should be reported against PAC 03F849 (or PAC 03S849 for State inspections).

D. Resource Instructions
Divisions should coordinate resources so that environmental sampling under this Attachment A meets inspection obligations from other assignments and/or compliance programs. See Annual ORA Human Food and Animal Feed Work Plan, and ORA Laboratory Work Plan.

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

**Federal Agencies**

Follow IOM subchapter 3.1.2 *Discussion with Federal Inspector* when federal officials from other agencies are present during FDA inspections or investigations. See IOM subchapter 3.2 *Federal Agency Interaction* for a list of Memorandums of Understanding (MOUs) between the FDA and other Federal agencies that may be applicable under the environmental sampling program. A complete list of MOUs may be found at FDA Memoranda of Understanding.

**State and Local Counterparts**

Divisions will collaborate with State agencies with commissioned officials to make them aware of the requirements of this program. Divisions may offer State agencies opportunities to accompany FDA on inspections or assist as necessary.

The Division, ORA Office of Partnerships (OP), and ORA Office of Regulatory Science (ORS) will coordinate State involvement, which may be performed under contract,

cooperative agreement, partnership agreement, or other collaborative efforts. **If the Division decides to utilize state contracts, cooperative agreements, or other agreements, please maintain contact with the respective State agency and immediately obtain information such as sample number to track WGS information. The State must also be willing to work with FDA for enforcement action.**

Examples of State involvement may include the following:

- Development of a coordinated sampling work plan with FDA to mitigate sampling inconsistencies. This sampling work plan should identify the specific sampling locations, quantity of samples to collect, targeted analyses, sampling schedule, laboratory resource availability, appropriate lab methodologies, and procedures for communication of analytical results to partner agencies.
- Inclusion of key laboratory personnel in the development of the sampling work plan to facilitate laboratory coordination with field operations, provide for technical input on sampling strategies, and provide advanced notice of reagent and supply purchasing needs.
- Sharing FDA analytical results with the responsible party.

Facilities assigned under State contract may require State counterparts to revisit a facility at a later date or when scheduled RTE production occurs. If the inspection is conducted under State contract, the State should consider as appropriate their individual resources regarding washout determination. States must also report their inspections and sample collections under the PAC listed in this compliance program for state conducted environmental sampling. Samples may be analyzed by FERN labs.

**Foreign Authorities**

Foreign inspections are not planned under the environmental sampling program.

F. When to contact other offices within the FDA

For questions pertaining to environmental sampling or the determination of whether a food is considered RTE for the purpose of this compliance program, please contact the compliance program monitor.

**Inspectional**

## 1. Operations

A. Inspections

Refer to ATTACHMENT A, Implementation of this compliance program for instructions on inspection priorities.
Refer to Field Bulletin (FB) #30 *Instructions for Environmental Sampling* for detailed inspection information and areas recommended for environmental sampling.

B. Investigations

An investigation is an information-gathering activity conducted for several reasons and may lead to a situation in which inspection and environmental sampling is recommended. See IOM subchapter 8.1.1.

C. Sample Collections

**General Information**

Environmental samples must be collected according to Field Bulletin (FB) #30 *Instructions for Environmental Sampling* and IOM subchapters 4.3.6, 4.3.7.7.1, 4.3.7.7.2, and exhibit 4-20.

The investigator should evaluate the potential for environmental sample collection at the start of an inspection. To determine whether the firm is eligible for environmental sampling, consider the following criteria and refer to FB# 30 for additional information:

- The firm manufactures RTE food that is exposed to the environment prior to packaging and will not undergo further processing in the facility or does not contain a control measure (such as a formulation lethal to the pathogen) to eliminate microbiological pathogens, AND
- The firm is processing RTE food within the inspection's timeframe. Ideally, swabbing takes place during processing of RTE food.

Investigators may determine whether to swab for *Listeria* or for *Salmonella* based on the food being manufactured during the inspection and the environment in which the food is exposed. A dry processing area that occasionally becomes wet may be more suitable for *Salmonella* harborage and growth while a wet processing area may be more suitable for *Listeria* (see FB#30 for environmental pathogen targets for specific foods). Investigators may swab for another pathogen if the type of food and the conditions in the facility warrant.

Some facilities prioritized for environmental sampling may have had previous environmental sample positives. For facilities that have had previous environmental sample positives, field inspections staff should follow-up on these deficiencies and concentrate environmental sample collections in and/or near the zones where positives were found. If there are any questions regarding environmental pathogen selection for a specific food operation, field inspection staff may contact the compliance program monitor.

| Table 1 - Sampling Framework | |
|---|---|
| **Product** | Environmental swabs for *Listeria* or *Salmonella* |
| **Description** | Environmental swab subsamples |
| **Product Code** | 52Y[][]08 |
| **Sample Type** | Investigational |
| **Sample Basis** | Surveillance or compliance |
| **Collection Zones** | *Listeria*: primarily zones 1-3<br>*Salmonella*: primarily zones 2-3 |

**Sample Size**

A minimum of 50 subsamples (ideally 100 subs) should be collected for *Listeria* analysis and a minimum of 100 subs (ideally 300 subs) should be collected for *Salmonella* analysis. Investigators should determine the appropriate number of subs to collect based on facility size and number of eligible sample sites related to RTE food production.

If circumstances arise such as a small facility size or limited relevant sampling sites that lead to the conclusion that the investigator cannot collect the minimum number of subsamples (minimum of 50 for *Listeria* or a minimum of 100 for *Salmonella),* Divisions should contact the compliance program monitor. This discussion must take place prior to closing the inspection. DDHAFO will then coordinate with CFSAN to discuss the situation. Failure to collect the requisite number of swabs and/or failure to contact DDHAFO regarding limited sample sites prior to closing the inspection may result in the Division being requested to revisit the facility to collect additional swabs.

**Laboratory Selection and Sample Shipment**

Refer to the Laboratory Servicing Table (LST) to determine which lab will analyze the environmental sample. If there are any questions or concerns regarding the LST, please direct them to ORALabCapacity@fda.hhs.gov. Divisions must provide advanced notice to laboratory management (>2 days) of the upcoming swab collection to ensure the laboratories have sufficient supplies available for analysis and field inspection staff must notify the analyzing laboratory via e-mail prior to shipping samples. The e-mail must contain the sample number(s), the name of this compliance program, storage conditions, the FACTS number, and sample tracking information. Lab contact information may be found in the LST. Contact the analyzing lab directly if there are any questions related to the analysis.

Samples should be shipped by UPS "Next Day Air Early A.M." to ensure arrival at the servicing laboratory within 24 hours. Samples may be shipped Monday through Thursday. **Do not** ship samples on Friday. Samples should be stored refrigerated prior to shipment. Ship samples in an insulated transport container with frozen gel packs to keep the samples cold, but not frozen. Field inspection staff may choose to separate samples from the frozen gel packs using cardboard or other material to prevent direct contact and freezing of sample broth.

**2. Reporting**

Report environmental sample collections according to IOM subchapter 4.4.10 *Reporting Sampling Collections*.

Accomplishment hours for all environmental sample collections performed under this environmental sampling program should be entered under the PACs identified in Table 2 below.

PACs from interacting programs in Program Management Instructions of this attachment should be used as appropriate.

**Table 2: Reporting PACs and PAFs for Environmental Sampling**

| PAC Description | Reporting PACs | PAF |
|---|---|---|
| Domestic human food environmental samples | 03050N | MIC |
| State contract domestic human food environmental samples | 03S050N | MIC |

If the firm decides to voluntarily hold product that was manufactured during environmental swabbing operations, the investigator must refer to FMD 147 *Procedures for Release of Analytical Results Pursuant to Section 704(d) and Situations When Dealer is Voluntarily Holding Product.* The "Dealer Voluntarily Holding" flag should be selected and facility contact information should be included in the Remarks section. When the firm is voluntarily holding a food, the Division must notify the firm promptly of final results, positive or negative.

## Analytical

**I. Analyzing Laboratories**

Environmental sampling initial analyzing laboratories will be selected according to the LST.

*Salmonella* isolates for serotyping should be analyzed using SeqSero.
In the event an isolate requires traditional serotyping, it should be sent to:

Denver Laboratory
6th Avenue & Kipling Street
DFC Building 20, Entrance W-10
Denver, CO 80225-0087
Attention: Shauna Madson
Tel # 303-236-9631
Fax # 303-236-9675

WGS analysis will be performed at CFSAN on positive cultures ***ONLY*** if ORA/ORS labs have exceeded capacity and have discussed the workflow issues with ORA/ORS, CFSAN OC, and CFSAN/ORS.

**FDA/Center for Food Safety and Applied Nutrition**
5001 Campus Drive
HFS-711, College Park, MD 20740

Dwayne Roberson
Telephone # 240-402-3098; Fax # 301-436-2644, and
Eric Brown (Director, Division of Microbiology, CFSAN)
Telephone # 240-701-5269

## II. Analyses to be Conducted

Analysis for *Listeria monocytogenes* and *Salmonella* may be conducted for environmental sampling under this compliance program ATTACHMENT A – ENVIRONMENTAL SAMPLING. WGS must be performed on all positives. Other *Listeria spp.* may be identified during analysis, but WGS does not need to be performed in these cases unless requested.

## III. Methodology

A. *Listeria* Screening, Confirmation, and Speciation

For isolation and identification of *Listeria monocytogenes* from environmental samples, the following methods can be used:

- AOAC PTM (No. 981202) VIDAS LIS, *Listeria* in environmental samples. AOAC OMA 999.06 *Listeria monocytogenes* in Selected Foods and Environmental Surfaces
- BAM chapter 10
  Follow the sampling preparation procedures described in the method instructions.
- If AOAC methods are used, confirm culture(s) following BAM Chapter 10.
- If sample swabs are confirmed positive for *Listeria monocytogenes*, perform WGS on each swab that is confirmed positive.
- No enumeration will be performed on environmental swab/sponge samples.

Record all work done on*: L. monocytogenes* and Other *Listeria* Species on the approved harmonized worksheets. Questions regarding *Listeria* analysis may be directed to the CFSAN ORS contact in Program Contacts.

B. *Salmonella* Screening, Confirmation, and Speciation

For screening, laboratories may use AOAC OMA, 2001.09, 2004.03, and 2011.03 (validated methods by lab). If samples cannot be processed immediately, refrigerate at 4±2°C (39±2°F). Start sample analysis within 48 ±2h of collection. Samples will be considered invalid past the 48 ± 2-hour window.

For confirmation, laboratories should use methods detailed in BAM Chapter 5. If the sample is positive for *Salmonella*, prepare slants and provide hard copy information

requested under BAM online Ch.5 section E-11, *Submission of cultures for serotyping*. Positive isolate(s) should be submitted to local serotyping group for Whole Genome Sequencing and serotyping analysis. Questions regarding *Salmonella* analysis may be directed to the CFSAN POC in the Program Contacts.

C. Whole Genome Sequencing (WGS)

WGS must be performed immediately on all confirmed *Listeria monocytogenes* or *Salmonella* positives and the results must be submitted to GenomeTrakr database, the national sub-typing network for foodborne disease surveillance. When large numbers of positives are detected, the positives should be prioritized for WGS analysis in consultation with CFSAN/ORS, ORA/ORS and ORA/OHAFO POCs in Program Contacts. Prioritized positive samples should be collected from locations most likely to serve as a source of contamination of food contact surfaces and food due to proximity or due to operational activities that are most likely to result in product contamination. The remainder of the isolates should be stored frozen until further instruction.

## IV. Reporting

Analytical results (CROs (Cannot Rule Out), negatives, confirmed positive findings) will be reported in an expeditious manner after an appropriate quality assurance review has been performed by laboratory management as required by the ORA Laboratory Quality Management System to meet the requirements for laboratory accreditation of the International Organization for Standardization/ Electrotechnical Commission [ISO/IEC] 17025.

According to *FMD-147 Procedure for Release of Analytical Results Pursuant to Section 704(d) and Situations When Dealer is Voluntarily Holding Product*, the laboratories will communicate confirmed sample results to the home Division's Compliance Branch via the established email address within 24 hours of completing sample analysis and in advance of sending the FDA Form 1551, Report of Sample Analysis, to the firm.

Cannot Rule Out (CRO) results should be reported by the analyzing laboratory to the **ORA/ Office of Regulatory Science (ORS) Program Coordinator** (see Program Contacts) ***and*** to the **Division** that collected the sample. The e-mail should include as much information as necessary for Divisions to quickly identify the FEI, sample number, CRO sub numbers, and the collector's name. Laboratories should also report confirmed positive sample results by email to the CFSAN and ORA compliance program coordinators and enforcement contacts listed (see Program Contacts). Please indicate LC2 for presence of other non-pathogenic *Listeria spp.* only, and indicate LC3 for presence of pathogenic *Listeria monocytogenes* or *Salmonella* spp.

It is important to compare all isolates to historical isolates from the same processing facility, and also to clinical isolates submitted by CDC, FDA, and USDA in SRA. **If the environmental samples isolate *Listeria monocytogenes* or *Salmonella* spp. in Zone 1, 2, or 3, contact the CFSAN OC and ORA compliance program contacts** (see Program Contacts) **to request a WGS comparison be completed and discuss next steps.** No WGS comparison is requested for non-pathogenic *Listeria spp*. For FDA, the historical isolates will be identified by CFSAN contacts, pulled from the laboratory's collection, and typed/sequenced by the WGS team. The comparison review of clinical isolates to environmental sampling isolates

will be performed by CFSAN. **The Division will be notified by the CFSAN/OC/DE contacts of the WGS comparison results, including any associated clinical isolates.** Report all analytical findings into FACTS using the "MIC" PAF code. Please refer to FMD-147 for reporting analytical results when Dealers are voluntarily holding product.

**Regulatory Strategy/Administrative Strategy**

1. **Findings**
   Consideration for enforcement depends on several factors including the firm's compliance history, the significance and severity of inspection observations, the prevalence and location of pathogens in the environment, the possible route of contamination, and the adequacy and timeliness of the firm's corrective actions. Corrections should be verified and documented prior to closing the inspection. **For enforcement instructions concerning significant deviations from 21 CFR part 117 CGMP & PCHF regulation or from 21 CFR part 123 Fish and Fishery Products, see their respective compliance program guidance manuals.**

   - **Level below which no action is required**
     - No significant conditions observed
     - No environmental pathogens indicated

   - **Level at which regulatory action is warranted depending on the facility's response/corrective action, inspection history, and/or swab location**
     - Significant cGMP conditions and/or poor sanitation controls observed
     - Zones 2 - 3 environmental sample positives but a direct route of contamination to food or food-contact surfaces can't be clearly established

   - **Level at which regulatory action is expected**
     - Significant cGMP conditions and/or poor sanitation controls observed
     - Zone 1 environmental positives
     - Zone 2 and 3 environmental positives which have been observed to directly cross-contaminate with food-contact surfaces and/or finished product and may be considered a risk to public health

2. **Charges**
   Charges that may be applicable when pathogens are isolated from a production environment include:
   - An article of food is adulterated under section 402(a)(4) of the Act [21 U.S.C. 342(a)(4)] if it has been prepared, packed, or held under insanitary conditions whereby it may have become contaminated with filth, or whereby it may have been rendered injurious to health.

   Additional charges may apply depending on findings during the inspection.

For regulatory purposes, refer to the table below. This is general information; specifics of each situation will need to be considered.

**Table 3. Possible FDA Response to Environmental Findings of *Listeria monocytogenes* or *Salmonella* spp. and Adverse Conditions**

| ***Pathogen (Listeria monocytogenes* or *Salmonella* spp.*)* found in environment** | **Adverse Conditions and/or Practices (including other *Listeria* species found in the environment)** | **Charge(s)** | **Possible FDA Response *** |
|---|---|---|---|
| No | No | N/A | N/A |
| Yes | No | 402(a)(4) | • Request voluntary corrective action<br>• Regulatory meeting<br>• Warning letter<br>• Request voluntary recall |
| No | Yes | 402(a)(4) | |
| Yes | Yes | 402(a)(4) | • Request voluntary corrective action<br>• Request voluntary recall<br>• Mandatory recall<br>• Warning letter<br>• Administrative detention<br>• Seizure<br>• Injunction<br>• Suspension of food facility registration |

*Any possible response depends on a variety of factors, including the firm's voluntary corrective actions, type of food processed, packed, or held in the firm, historical information related to the firm and product, location and number of positive swabs, and WGS genetic information related to the isolates.

## 3. Actions

A. Administrative and Legal Actions for Imminent Public Health Hazards

See interacting programs (in Program Management Instructions) for additional information on administrative and legal actions. If the Division feels that administrative or legal action is warranted, management should initiate a preliminary assessment call with CFSAN Office of Compliance, Division of Enforcement (DE) compliance program contacts (see Program Contacts). State partners should be engaged to determine if enforcement actions such as embargo or permit revocation can be utilized to stop the movement of product or production while FDA is considering enforcement actions. Divisions should reach out to CFSAN OC DE as soon as this type of action is being considered. A recommendation for recall should also be considered with any imminent public health concern.

B. Compliance Activities

**CFSAN has not given Direct Reference Authority for any compliance actions related to violations resulting from environmental sampling results at this time. All possible administrative and legal regulatory actions should be discussed with State regulatory counterparts before moving forward. For compliance actions related to 21 CFR 123, please refer to** CFSAN Enforcement Bulletin No. 1**.**

If a positive environmental sample is reported to the Division by the servicing laboratory and/or egregious conditions are observed, the Division should contact the CFSAN OC DE, CFSAN OC DFPG, and ORA OHAFO DDHAFO compliance program contacts (see Program Contacts) to set up a conference to discuss possible enforcement strategies and WGS comparison review. Depending on the firm's compliance history, location of positive swab sites, and routes of contamination to RTE food, enforcement strategies including, but not limited to, regulatory meeting, warning letter, injunction, and recall of products may be considered. If the environmental isolate is virtually identical to a clinical isolate, and depending on other factors, in particular an epidemiological link to the firm's products, strong regulatory action may be warranted. FSMA enforcement tools including suspension of food facility registration could also be considered. Link Field Accomplishment Compliance Tracking System (FACTS) sample number to all Compliance Management System (CMS) work activities, actions, or recalls created under this compliance program. The Division should submit any recommendation for enforcement follow-up via CMS. Any CMS Work Activity or Action created in response to results from this compliance program should be created with a link to the related sample number or eNspect inspection ID. Submit a task to the Enforcement Contacts listed in Section VI.

Table 3 summarizes possible compliance activities based on observed conditions during an inspection and environmental sample results. This summary is a starting point and should not be the sole basis for evaluating the significance of noncompliance. Findings should be assessed on a case-by-case basis and should consider the totality of the observations. See interacting CPs referenced in this compliance program for more information concerning compliance activities associated with deviations from 21 CFR part 117 or 21 CFR part 123.

**4. Follow-Up**

Perform follow-up inspections according to the timeframes in CPGM 7303.040 *Preventive Controls and Sanitary Human Food Operations*. Prior to initiating the re-inspection, Divisions should hold an enforcement strategy discussion with CFSAN OC, ORA OHAFO DDHAFO contacts in Program Contacts of this compliance program, and state partners to discuss potential follow-up actions if the firm continues to have significant violations. If the follow-up inspection reveals that the firm continues to have conditions that are likely to lead to the adulteration of foods, the Division should initiate a call with CFSAN OC and ORA OHAFO DDHAFO contacts as soon as possible and should consider more severe enforcement actions based on these repeat offenses.

**References:**

Investigations Operations Manual (IOM)

Regulatory Procedures Manual (RPM)

Draft Guidance for Industry: Hazard Analysis and Risk-Based Preventive Controls for Human Foods

Fish and Fishery Products Hazards and Controls Guidance

21 CFR part 117 Current Good Manufacturing Practice, Hazard Analysis, and Risk-Based Preventive Controls for Human Food

21 CFR part 123 Fish and Fishery Products

Field Bulletin #30 Instructions for Environmental Sampling

CPGM 7303.040 Preventive Controls and Sanitary Human Food Operations

CPGM 7303.042 Seafood Processor Inspection Program – Domestic and Foreign Facilities

## ATTACHMENT B – FILTH AND DECOMPOSITION (MOLD/ROT)

The FD&C Act protects the public from the presence of filth, putrid or decomposed material in food products, and those products that may have been exposed to insanitary conditions that may contaminate the product with filth or render it injurious to health. The terms filth, foreign material, and extraneous material are used interchangeably by the Agency. The courts define filth in a common-sense manner; filth does not have any specialized or technical definition. Filth is any type of matter that does not belong in a food product. Examples of filth in food products include but are not limited to rodent excreta, insects, hair, bird excreta, and other extraneous materials such as metal and glass shards.

Filth can enter a product through many forms and sources and is often invisible to the consumer. Filth may be present in food naturally, or as the result of an intentional or unintentional bad practice. The identification of the type of filth can help determine how the material was introduced in the product, and whether it was a natural and unavoidable event, an accidental event, a controllable event, an unintentional event and/or a deliberate intentional event.
It is economically impractical to grow, harvest or process raw products that are totally free of non-hazardous, naturally occurring defects. Recognizing this, FDA has set maximum allowable limits for these defects (Food Defect Action Levels). Above those limits, the food is considered adulterated. Products harmful to the consumer are subject to regulatory action.

Note: This compliance program does not address "seafood decomposition" (fishery products), which is addressed in separate compliance programs (7303.844, 7303.842).

**Objectives**

- To collect and analyze samples of domestic or imported product during inspection, investigation, and/or field examination for filth and decomposition (mold/rot).

- To take appropriate regulatory action if the food samples are found to be contaminated with filth or decomposed.

**General Sampling Instruction (domestic and import)**
Divisions should consider manufacturers with a history of filth violations and foods with a history of contamination with filth which present a public health hazard, such as those listed below.

- Food products where the intended use, or further processing of which, will not remove filth elements, which can be vectors of pathogens. This includes products which will not be further processed or washed by consumers before being consumed (e.g., whole spices, dried fruit and nut products, packaged salads).
- Bakery dry mixes (e.g., biscuit, bread, cake, pancake, pastry) which are susceptible to contamination with mites that may trigger an allergic reaction when ingested.
- Products with a history of hazardous foreign objects or visually objectionable contamination. This type of filth may occur in a variety of products. This type of filth may be associated with certain manufacturing practices.

- Hazardous foreign objects include objects which are not an expected component of the product, which would cause a choking or other type of physical hazard (e.g., glass fragments, stones, metal fragments, shards of hard plastic, pit fragments in pitted olives, shell in nut products).
- Visually objectionable contaminants (e.g., whole insects, rodent droppings, cigarette butts, chewing gum).

- Foods such as whole spices and many processed fruits and vegetables susceptible to decomposition (e.g., mold/rot). Decomposition includes, but is not limited to, moldy product, rot, and other types of spoilage. Many types of decomposition are categorized as potential health hazards requiring higher priority food safety coverage. This compliance program does not address "seafood decomposition," which is addressed in separate compliance program; see Seafood Processor and Products Inspection Program (7303.842).

**Sample Collection**

Samples for filth and decomposition (mold/rot) should be collected during inspection, investigation, and/or field examination as warranted. Further:

- Samples collected for filth and decomposition analysis should be indicated in the collection report.
  - For cheese and cheese products, the laboratory will analyze the samples per the PAC, PAF, and instructions provided in the collection report.
- Collect raw materials, in-line samples, and finished product samples if contaminated raw materials were believed to be used.
  - Do not sample finished products to support contamination in the finished product if inspectional evidence indicates little likelihood of detecting this contamination in the finished product.
  - Instead, collect physical samples of contaminated raw material(s) and/or exhibits with documentation that the finished product moves interstate. Fully document the use of these contaminated raw materials in the manufacture of a specific lot or lots of finished products.
- Obtain physical samples of raw materials, in-process sub-samples, and other items such as filth exhibits showing routes of contamination to clearly show that conditions of manufacture or storage are such that there is a reasonable possibility that contamination may have occurred.
- See IOM 5.1.5.3 for domestic and 6.4 for import sample collection during field examination.

> **NOTE**: The Center will consider cases based solely on detailed observations, photographs, etc., which would convince the ordinary consumer that a condition or practice is filthy.

**Sample Size**

Refer to applicable CPGs, Import Alerts, Import Bulletins, or FDA Technical Bulletin No. 5 Macroanalytical Procedures Manual for instructions. In the absence of specific instruction, collect ten (10), 2 lb. subsamples at random. If the Division's investigations branch has contacted the

laboratory or ORA/ORS Filth Program Coordinator and established that a smaller sample size would suffice for regulatory analysis, then the established smaller sample size may be collected.

For Cheese and Cheese Products
See Tables 2 and 3 of this compliance program; remaining portions are to be used for filth analysis when necessary.

**Sample Shipment**
Refer to the current LST Dashboard for appropriate analyzing laboratory.

Refer to IOM Section 4.5.3.5 for sample handling of frozen samples or Section 4.5.3.6 for refrigerated samples as appropriate.

**Analytical Methods**
**Sample Preparation and Instructions for Filth and Decomposition (Mold/Rot) Analysis**

Refer to applicable CPGs, Import Alerts and Bulletins, Laboratory Information Bulletins (LIBs), FDA Technical Bulletin No. 1 *Principles of Food Analysis for Filth, Decomposition and Foreign Matter*, FDA Technical Bulletin No. 5 *Macroanalytical Procedures Manual*, ORA Laboratory Manual Volume IV Section 4 - Microanalytical & Filth Analysis, and AOAC, most current Ed., for filth and decomposition instruction.

**Cheese and Cheese Products - Sample Preparation Instructions for Filth Analysis**
Filth Analysis will only be performed upon request. If filth analysis is not specifically requested in the collection report, the laboratory should not perform filth analysis.

Subsamples 2.27 kg (5 ) or greater
Remove six (6) - 227 g (8 oz.) subsamples from the unit of cheese. For samples of shredded, grated or extruded forms of cheese, remove the same amount. Each subsample should be examined individually and not composited.

Consumer size units, 227 g (8 oz.)
Randomly select six (6) subsamples from the remaining portions of the ten (10) subsamples used for microbial and alkaline phosphatase analysis. Remove 227 g (8 oz.) from each of the six (6) subsamples for analysis. For samples of shredded, grated or extruded forms of cheese, remove the same amount. Each subsample should be examined individually and not composited.

Analytical Guidance: Filth, Mold and Foreign Objects: Microscopic and Macroscopic.
AOAC Official Method 994.05 Light Filth in Cheeses.

If not applicable, then use:
AOAC Official Method 960.49 Filth in Dairy Products.

**Reporting**
Report all analytical results for filth and decomposition (mold/rot) using the following PAC and PAF:

PAC: 03050B

PAF: CCD, FDF, FIL, NAR, PAR

**Regulatory Strategy**
Apply the appropriate CPG criteria (Chapter 5 - Food, Colors, and Cosmetics | FDA). In those instances when specific criteria for regulatory action are not available, submit analytical findings to CFSAN, Division of Enforcement Contact (see Compliance Program contacts section). Enforcement action recommendations should be submitted directly to CFSAN Division of Enforcement via the Compliance Management System (CMS), and sample(s) should be linked.

## ATTACHMENT C –List of Standardized Cheeses with Phenol Levels from 21 CFR Part 133

| Cheese Type | Fg phenol/ g cheese | CFR Reference |
|---|---|---|
| Brick | 20 | 133. 108(a)(2) |
| Cheddar | 12 | 133. 113(a)(2) |
| Colby | 12 | 133. 118(c)(2) |
| Cook cheese, koch kaese | 12 | 133. 127(a)(2) |
| Washed curd and soaked curd | 12 | 133. 136(a)(2) |
| Edam | 12 | 133. 138(a)(2) |
| Granular and stirred curd | 12 | 133. 144(a)(2) |
| Gruyere | 12 | 133. 149(a)(2) |
| Hard | 12 | 133. 150(c)(2) |
| Limburger | 16 | 133. 152(a)(2) |
| Monterey & monterey jack | 12 | 133. 153(a)(2) |
| Mozzarella & scamorza | 12 | 133. 155(a)(2) |
| Low moisture mozzarella & scamorza | 12 | 133. 156(a)(2) |
| Muenster & munster | 12 | 133. 160(a)(2) |
| Pasteurized process | 12 | 133. 169(a)(2) |
| Pasteurized process cheese food | 12 | 133. 173(a)(2) |
| Pasteurized neufchatel cheese spread with other foods | 12 | 133. 178(a)(2) |
| Pasteurized process cheese spread | 12 | 133. 179(a)(2) |
| Provolone | 12 | 133. 181(a)(2) |
| Samsoe | 12 | 133. 185(a)(2) |
| Semisoft | 20 | 133. 187(c)(2) |
| Semisoft part-skim | 20 | 133. 188(c)(2) |
| Spiced | 12 | 133. 190(a)(2) |
| Swiss & emmentaler | 12 | 133. 195(a)(2) |