CHAPTER 04 – PESTICIDES AND CHEMICAL CONTAMINANTS

SUBJECT: **CHEMOTHERAPEUTICS IN AQUACULTURE SEAFOOD COMPLIANCE PROGRAM**	IMPLEMENTATION DATE: UPON RECEIPT
DATA REPORTING	
PRODUCT CODES	PRODUCT/ASSIGNMENT CODES
See Table 1	REPORT COLLECTION AND ANALYSIS USING PAC: 04018

FIELD REPORTING REQUIREMENTS:

A. There are no hard copy reports for this Compliance Program.

B. Resources for completion of this Compliance Program can be found in the current ORA Field Workplan under PAC 04018, Chemotherapeutics in Seafood Compliance Program.

C. FACTS/OASIS reporting for domestic and import sample collections and for domestic and import sample analyses:
   1. PAC: 04018
   2. PAF: ANT

Table 1 – Product Codes Description for Different Species

INDUSTRY CODES: 16

PRODUCT CODES:

Crab	16J[][]01
Crayfish, Aquaculture Harvested Fishery/Seafood Products	16X[][]20
Crayfish (Crawfish), Fresh Water	16J[][]02
Eel	16A[][]15
Eel, Aquaculture Products	16X[][]45
Salmon, Aquaculture Harvested Fishery/Seafood Products	16X[][]03
Salmon (Humpback, Silver, King, Sockeye, Ect.)	16A[][]32
Shrimp and Prawns, Aquaculture Harvested Fishery/Seafood Products	16X[][]21
Shrimp and Prawns	16J[][]05
Tilapia, Aquaculture Harvested Fishery/Seafood Products	16X[][]06
Tilapia	16A[][]58
Trout, Aquaculture Harvested Fishery/Seafood Products	16X[][]05
Trout (Rainbow, Brook, Brown, Char, Steelhead, Ect.)	16A[][]44
Frogs, Aquaculture Harvested Fishery/Seafood Products	16X[][]42
Frog Legs, Other Aquatic Species	16M[][]01
Fish, N.E.C.	16A[][]99
Aquaculture Harvested Fishery/Seafood Products, N.E.C.	16X[][]99
Crustaceans, N.E.C.	16J[][]99
Barramundi	16A[][]85
Bass, Salt or Brackish Water	16A[][]04
Bass, Hybrid Striped, Aquaculture Harvested Fishery/Seafood Products	16X[][]01
Bream	16A[][]80
Carp	16A[][]09
Cobia	16A[][]94
Cobia, Aquaculture Harvested Fishery/Seafood Products	16X[][]47

Croaker	16A[][]13
Dace	16A[][]57
Dace, Aquaculture Harvested Fishery/Seafood Products	16X[][]44
Drum, Totoaba	16A[][]68
Grouper	16A[][]17
Lobster	16J[][]04
Langostino	16j[][]07
Milkfish	16A[][]53
Mudfish	16A[][]64
Mullet	16A[][]23
Perch Freshwater	16A[][]26
Pompano (Permit, Pompanito)	16A[][]29
Pompano, Aquaculture Harvested Fishery/Seafood Products	16X[][]07
Snapper	16A[][]37
Sturgeon (River and Sea)	16A[][]40
Sturgeon, Aquaculture Harvested Fishery/Seafood Products	16X[][]04
Turbot	16A[][]46
Yellowtail, Amberjack	16A[][]79

***For any other species, refer to Fish and Fishery products Hazard and Controls Guidance: CHAPTER 3 Potential Species-Related Hazards**

Note: 16A – Wild Caught Seafood Products

16X – Aquaculture Seafood Products

Contents

PART I – BACKGROUND

Over the past twenty years, there has been an extensive commercialization and an increased consumption rate of seafood products originating from aquaculture operations.

Aquaculture, also known as fish and shellfish farming, refers to the breeding, rearing and harvesting of aquatic food animals under environmentally controlled conditions with some form of intervention to enhance production, such as regular stocking, feeding, prevention and treatment of diseases, protection from predators, etc. Aquaculture activities are conducted in all types of water environments; freshwater, coastal and marine, inland ponds, tanks, reservoirs, rivers, lakes, estuaries, bays, fjords, and open sea.

In 2013, the world aquaculture production accounted for almost half of all fish for human food. This share is projected to rise to 62% by 2030 as catches from wild capture fisheries level off and demands for seafood constantly increases. The worldwide aquaculture production for human consumption has grown from 6.7 million metric tons in 1984 to 70.5 million metric tons ($144.4 billion value) in 2013. Fifteen main producer countries accounted for 92.7% of all farmed food fish. Asia accounted for 83% of world aquaculture production by volume and this was dominated by China with more than 62% of the global production. Other major producers are India, Vietnam, Indonesia, Bangladesh, Thailand, Chile, Egypt, Norway, Myanmar, Philippines, Brazil and Malaysia. (FAO: Fishery & Aquaculture Department in The State of World Fisheries and Aquaculture, 2014).

Aquaculture production is vulnerable to diseases and changing environmental conditions. Disease outbreaks significantly affect aquaculture production and trade. In recent years, farming areas in Asia and South America have been affected by diseases and problems from viruses, bacteria, fungi, parasites and other undiagnosed and emerging pathogens. The use of veterinary drugs in animal production is necessary to treat and control diseases; however, this use may result in the residues found in products for human consumption. The use of unapproved drugs and the misuse of FDA approved new animal drugs in aquaculture species is a concern. These uses may increase due to limited availability of treatment options, specifically for new emerging diseases. Veterinary drugs can also be administered to improve the production, e.g., to enhance feed efficiency and rate of weight gain. To protect consumers, it is important to ensure that both imported and domestic aquaculture seafood products are free from potentially harmful drug residues. Residues of animal drugs, the parent compound or its metabolite, in food may cause acute or chronic effects.

An acute response can occur from hypersensitivity or allergic reaction to the drug. For example, drug residue responses were reported in Spain, France, and China, where people became seriously ill after consumption of liver contaminated with clenbuterol residues. Other type of drugs, such as penicillin and, cephalosporins can cause allergic response at low doses. Chronic, long-term effects are difficult to detect.Effects from consumption of food contaminated with drug residues for a prolonged period of time are typically underreported to FDA or it’s difficult to identify the cause of illness. However, the long term hazards include extremely serious conditions. For example, nitrofurans and triphenylmethane dyes (malachite green, gentian violet) are considered to be carcinogenic and genotoxic, and chloramphenicol has been implicated as the causative agent in several cases of fatal aplastic anemia. Sulfonamides used at subtherapeutic and therapeutic concentrations in food-animal production revealed issues with carcinogenic and mutagenic potential and thyroid toxicity.

Antimicrobial use in aquaculture production may also contribute to microbial responses and antimicrobial resistance in bacteria that may be transferred to humans. Antibiotic resistance is a global problem found in human and animal environments inadequate oversight and inappropriate use in all areas of human and animal medicine. Microbial responses of drug residues can affect human intestinal flora diminishing the activity of intestinal bacteria. Also, antibiotic residues can develop transient resistant microorganisms and zoonotic spread of pathogens to humans. For example, the unapproved use of fluoroquinolones, such as ciprofloxacin and enrofloxacin, poses risk of antimicrobial resistance in bacteria and the related serious human health consequences as untreatable infections due to lack of drug effectiveness in treatment.

Improper use of chemical compounds in global aquaculture production is a concern of FDA, due to the significant safety impact on US consumers.

PART II - IMPLEMENTATION

1. Objective

To sample and analyze identified imported and domestic aquaculture seafood products and processed crab to determine the presence and amount of:

a) unapproved chemical compounds and,

b) unapproved new animal drugs such as antibiotics or anti-fungal agents used for therapeutic and non-therapeutic purposes.

2. Program Management Instructions

The Act and FDA regulations apply equally to aquaculture seafood products produced in this country and aquaculture seafood products produced in other countries and imported into this country for commercial distribution. This compliance program establishes the collection and testing of residues of new animal drugs and unapproved chemical compounds in samples of aquaculture seafood products and processed crabmeat. Any samples found to contain illegal residues may lead to follow-up investigations at suspected firms and regulatory action.

CFSAN/ Office of Compliance/Division of Field Programs and Guidance will issue the Sample Collection Operations Planning Effort (SCOPE) that identifies the domestic and import aquaculture seafood species prior to the start of each Fiscal Year. SCOPE is located through links on the CFSAN/Office of Compliance intranet website. Information about analyzing laboratories is provided in the SCOPE. It is imperative that the SCOPE information be followed to ensure proper sample collections.

Domestic

During sample collection of domestic product, if the investigator suspects that an unapproved new animal drug has been used on the farm or during transport, evidence of the intended use should be obtained at the point of sample collection and relayed to CVM contact. CVM may issue assignments to: i) document the use of the unapproved new animal drugs for possible enforcement action and, ii) follow-up on reports of positive drug residues.

If the presence of an unapproved drug residues or approved drug residues at or above level exceeding the established tolerance level is confirmed by the analysis of a domestic sample, the districts inform CFSAN and CVM to conduct a follow- up investigation.

Imports

Sampling of imported aquaculture seafood products are identified and directed in SCOPE.

If the presence of a drug residue is confirmed by the analysis of an import sample, the district should detain the entry and proceed with the detention and hearing process. In addition,

confirmation of the presence of a drug residue by sample analysis may result in recommendation for addition to an Import Alert for Detention Without Physical Examination (DWPE) of the combination of foreign processor and product or revision of existing Import Alert.

3. Program Interaction

- Illegal Drug Residues in Meat, Poultry, Aquacultured Seafood and other Animal Derived Foods Compliance Program, 7371.006: Follow the instructions to conduct on-farm investigations or investigations at a veterinarian or any other involved parties for drug residues from domestically produced product.

- Import Seafood Products Compliance Program, 7303.844: Use as a guide to conduct follow-up investigations at importers.

- Seafood Processor Inspection Program - Domestic and Foreign Facilities Compliance Program, 7303.842: Use for aquaculture seafood concerns other than chemotherapeutic agents.

- Aquaculture seafood products are also collected for Dioxin analysis as part of Pesticides and Industrial Chemicals in Food - Domestic and Imported Compliance Program (7304.004A). Districts may coordinate sample collections as appropriate.

PART III - INSPECTIONAL

1. Operations

Collecting Districts: Refer to ORA Field Workplan and the domestic and import Sample Collection Operation Planning Effort (SCOPE) issued by CFSAN before the start of each fiscal year for the specific numbers and the species of seafood to be collected and countries of interest. SCOPE is located at http://inside.fda.gov:9003/ProgramsInitiatives/Food/FieldPrograms/ucm272937.htm

Note: Only those products listed in the SCOPE should be collected.

A. Inspections

Resource for inspection has not been planned under this compliance program.

B. Investigations

During domestic sample collection at the aquaculture farm if the investigator suspects that use of unapproved new animal drugs on the farm or during transport, evidence of the intended use should be obtained, at the point of sample collection. The District Office should notify CVM for further follow-up investigation and a possible enforcement action.

For import samples, if a drug residue is confirmed by sample analysis, the District's Compliance Branch should detain the entry and proceed with the detention and hearing process. In addition, confirmation of the presence of a drug residue by sample analysis should result in a referral recommendation for addition to an Import Alert for Detention Without Physical Examination (DWPE) of the combination of foreign processor, sent via MARCS-CMS to DIO. DIO will forward the recommendation to CFSAN for assessment and evaluation. CFSAN will then communicate conclusion of evaluation to DIO for revision of the applicable Import Alert.

C. Sample Collections

Sampling Instructions

General:

- Samples should be collected from the largest lot size available.
- Attempt to sample lots with the smallest unit packages whenever possible.
- Raw, unprocessed, breaded, seasoned, cooked, fresh, or frozen product is acceptable for collection unless otherwise specified in the SCOPE.
  Canned product except for canned meat of crab should not be collected.
- For shipping purpose, samples of fresh seafood product can be frozen.
- Frozen, refrigerated, pasteurized, canned meat of crab (typically in pound cans), cut crab, soft shell crab, crab claws, cut crab pieces and crab legs should be

collected. Whole crab should not be collected.

- When collecting any fresh product, the investigator should coordinate with the assigned laboratory so that the applicable timeframes are met.
- Whole crayfish is acceptable for collection.
- **Do not use black or blue sharpies.** When marking aquaculture chemotherapeutic samples to avoid contamination of sample with ink of sharpie. As an alternative the Investigator can use a red pen or stick on labels.

Domestic Sample Collections

Refer to the current SCOPE for the number of domestic samples to collect of each identified product.

All products except for processed crab and crawfish **must be aquaculture raised.** Processed crab and crayfish from either aquaculture operation or wild caught can be collected.

Review the firm's inspection history or call the firm to determine if the firm does grow the desired aquaculture product. Samples should be collected as near to the point of harvest as possible and **the domestic grower should be identified in the collection report.**

If the sample is fresh, the investigator should notify the lab that fresh seafood is on hold pending completion of analysis by annotating the FDA 525 and using the appropriate sample flag on the Collection Report.

Domestic aquaculture product samples may also be collected from processors or wholesalers provided that the domestic grower can be identified and is reported in the collection report. Prior to collection, verify that the product is **domestic origin.** Do not collect DI samples to meet domestic sample collection obligations.

Import Sample Collections

Refer to the current SCOPE for the number of import samples to collect of each product and for countries of interest. Import tracker SCOPE is located http://inside.fda.gov:9003/ora/officeofregionaloperations/officeofenforcement/divisionofimportoperationsandpolicy/ucm352360.htm.

Imported seafood may not always be identified on labeling or in shipping documents as "aquaculture or farm raised." Barramundi, carp, cobia, dace, eel, frog legs, milkfish, pompano, shrimp/prawns, salmon, trout and tilapia should be assumed to be farm raised unless accompanying paperwork or labeling states “wild caught.” Collect finfish species

listed in the current FY SCOPE only when identified as farm raised. Processed crab and crayfish may be collected regardless of whether it is "wild caught" or farm-raised.

If the sample is fresh, the investigator should notify the lab that fresh seafood is on hold pending completion of analysis by annotating the FDA 525 and using the appropriate sample flag on the Collection Report. When collected as a domestic import sample, the collector must include the OASIS entry number in the remarks section of the FACTS Sample Collection Report.

Import obligations may be met by collecting Domestic Import (DI) samples only if the district can identify the foreign processor/shipper, and the country of origin.

The investigator should not target for sample collection a product from the foreign processor when the combination of foreign processor and product are already subject to DWPE due to the presence of unapproved drug residues.

Avoid collecting multiple samples from the same foreign processor in a short period of time unless otherwise instructed.

The collecting districts are encouraged to work with their Regional Activities Managers (RAMs) to adjust the May Proceed rates for those products that are required for collection by their District. The RAM will work with the Division Compliance Systems to adjust the May Proceed rates accordingly.

Sample Sizes

Domestic Samples

Finfish Salmon Tilapia Trout Arctic Char, Barramundi, Bass Striped, Cobia, Carp, Drum, Eel, Grouper, Snapper, Yellow Croaker, Pompano, Milkfish, Mudfish, Mullet, Bream, Sturgeon, Perch, Turbot, Yellowtail	Each fresh or frozen sample collected from a single lot should consist of 12 subsamples, equaling a minimum of 225 grams (0.5 lb.) per subsample, for a total weight of 2.7 kg (6.0 lbs.) of product. The sample includes the 702(b) portion.
Shrimp/Prawn	Each fresh, frozen or breaded sample collected from a single lot should consist of twelve (12) subsamples, equaling a minimum of 450 grams (1 lb.) per subsample. If the sample is whole, in-shell, a minimum of 680 grams (1.5 lbs.) per sub should be collected.
Processed Crab, Crawfish, Lobster and Langostino	Each sample should consist of twelve (12) subsamples, minimum 450 g (1 lb.) per subsample, total 5.4 kg (12.0 lbs.) of product. If the sample is whole, in-shell or is cut pieces still in shell, a minimum of 680 grams (1.5 lbs.) per sub should be collected. If the individual product package size is larger than 450 g (1 lb.) and less than or equal to 1.35 kg (3 lbs.), collect one package unit per subsample. **Note: For units larger than 3.0lb. only:** If the units must be sampled and shipped intact, collect 6 subsamples (product package units) and send to assigned laboratory. Alternatively, subsamples of at least 450 g (1 lb.) may be broken/sawed off (keep frozen) from each of 12 package units, and the twelve (12) 450 g subsamples shipped to the analyzing lab. If sampling from bulk, collect using aseptic technique (refer to IOM 4.3.6).

	If the individual product package size is less than 450 g (1 lb.), collected per subsample equals a minimum of 450g (1lb.) or 680 grams (1.5 lbs.) for whole, in-shell product. These samples should include the 702(b) reserve for domestic processed crab/crawfish/lobster/langostino.

Import and Domestic – Import Samples

Frog legs Eel, Salmon, Tilapia, Trout Arctic Char, Barramundi, Bass Hybrid Striped, Carp, Cobia, Dace, Drum, Grouper, Snapper, Yellow Croaker, Pompano, Milkfish, Mudfish, Mullet, Bream, Sturgeon, Perch, Turbot, Yellowtail	Each fresh or frozen fish sample collected from a single lot should consist of twelve (12) - 225 gram (0.5 lb.) subsamples, totaling 2.7 kg (6.0 lb.) of product. For frog legs, twelve (12) – 680 gram (1.5 lb.) subsamples, totaling ~8.2 kg (18 lbs.) of product should be collected. If the individual product package size of fish is larger than 225 grams (0.5 lb.), collect one package unit per subsample. If the individual product package is less than 225 grams (0.5lb.), collect an adequate number of package units so that the total amount collected per subsample equals a minimum of 225 grams (0.5lb.). Note: In situations where a district feels the cost of above sample sizes is prohibitive, the following alternate sampling scheme may be applied: 12 subs (equaling a minimum of 225 grams (0.5 lb.) per subsample or 680 grams (1.5 lb.) of frog legs per subsamples) for individual package size less than or equal to 3 lbs., 6 subsamples for fish less than or equal to 6.0 lbs. and 3 subsamples for fish larger than 6.0 lbs. The labs will make a single composite from equal portions of each sub received.

Shrimp/Prawn	Each fresh, frozen or breaded sample collected from a single lot should consist of twelve (12) subsamples, equaling a minimum of 450 grams (1 lb.) per subsample. If the product is whole, in-shell, a minimum of 680 g (1.5 lbs.) should be collected. If the individual product package size is larger than 450 grams (1 lb.), collect one package unit per subsample. If the individual product package is less than 450 grams (1 lb.), collect an adequate number of package units so that the amount collected per subsample equals a minimum of 450 grams (1 lb.) or 680 grams (1.5 lbs.) of whole, in-shell product. **Note: For units larger than 3.0 lb. only:** If the units must be sampled and shipped intact, collect 6 subsamples (product package units) and send to assigned laboratory. Alternatively, subsamples of at least 450 g (1 lb.) may be broken/sawed off (keep frozen) from each of 12 package units, and the twelve (12) 450 g subsamples shipped to the analyzing lab. If sampling from bulk, collect using aseptic technique (refer to IOM 4.3.6).

Processed Crab, Crawfish	Each sample should consist of twelve (12) subsamples, minimum 450 g (1 lb.) per subsample, total 5.4 kg (6.0 lbs.) of product. If the product is whole, in-shell or cut crab pieces, a minimum of 680 grams (1.5 lbs.) should be collected per sub. If the individual product package size is larger than 450 g (1 lb.) and less than or equal to 3 lbs., collect one package unit per subsample. If the unit size is less than 450 g (1 lb.), collect an adequate number of package units so that the total amount collected per subsample equals a minimum of 450 g (1 lb.) for processed product and 680 g (1.5 lbs.) for whole, in-shell product. **Note: For units larger than 3.0 lbs only**: If the units must be sampled and shipped intact, collect 6 subsamples (product package units) and send to assigned laboratory. Alternatively, subsamples of at least 450 g (1 lb.) may be broken/sawed off (keep frozen) from each of 12 package units, and the twelve (12) 450 g subsamples shipped to the analyzing lab. If sampling from bulk, collect using aseptic technique (refer to IOM 4.3.6).

Shipping Instructions

Shipping instructions to maintain the integrity of frozen and refrigerated samples and the procedure to notify the receiving laboratory can be found in IOM, Sections 4.5.3.5, 4.5.3.6, and 4.5.5.5. Samples should be packaged with the appropriate refrigerant and shipped so that they arrive at the laboratory no later than Thursday of each week.

Samples should only be shipped to the assigned laboratory designated in the current FY SCOPE.

D. Import Activities

N/A

E. Other

N/A

2. Reporting

N/A

PART IV - ANALYTICAL

1. Analyzing Laboratories

Refer to the current Lab Servicing Table for Chemotherapeutics to determine the analyzing laboratories.

For residues requiring both determinative and confirmatory methods, the laboratory performing the determinative method has the option of performing the confirmatory testing on presumptive violations or sending it to Denver Laboratory at the following address:

Denver Laboratory (DENL)

ATTN: Sample Custodian

Gianna Costo, (303) 236-3065

6th Avenue & Kipling St.

1 Denver Federal Center

Bldg. 20, Ent W10

Denver, CO 80225-0087

Email: ORA DEN LAB Supervisors

2. Analyses to be Conducted

Refer to the current FY SCOPE to determine the analyses to be conducted.

3. Methodology

A. Sample Preparation and Methods

1. General Instructions

The chemical compounds identified in the following methods to be tested in seafood species listed in this Compliance Program are the human health concern and might have the significant food safety impact on US consumers. The determinative method will identify and quantify the amount of the compound and the confirmation method will validate findings of the determinative method (if applicable). Therefore, no further analyses are required.

Report all analytical results in FACTS. Retain the remaining portion of each subsample when classified as Lab Class 3 (Adverse Finding).

2. Analytical Protocol

Prepare one composite of all subsamples, unless otherwise noted for specific drug residue testing requiring individual sub sample analysis.

The determinative method should be run on all samples. Whenever the analytical result of the residue from the determinative method is equal to or above the level

referenced below (under C. **Testing Target LEVELS (TTL)** for each drug residue, a confirmative method must be run (where applicable). The confirmative method for each residue/species combination employs mass spectrometry detection to confirm the identity of the residue

3. Sample Preparation:

Homogenize only edible portion. Thoroughly remove skin, bones, shell, any breading or seasoning before analysis. It is suggested that laboratories homogenize sample by grinding with dry ice (reference:Bunch, E.A.,Altwein, D.M., Johnson, L.E. Farley, J.R., and Hammersmith, A.A. (1995) Homogenous Sample Preparation of Raw Shrimp with the Aid of Dry Ice.*J. AOAC Int*. 78, 883-887.

Divide the prepared (processed with dry ice) composite or individual sub in half. Use half of the prepared sample for analysis and retain the other half of the prepared sample in a freezer as a reserve.

For domestic samples, retain the 702(b) portion from each of the 12 subsamples collected.

For Chloramphenicol and Nitrofurans:

1. All chloramphenicol analyses are to be run on an individual sub basis.
2. All nitrofurans analyses are to be run on an individual sub basis.

The confirmation of chloramphenicol or nitrofurans in a single subsample is sufficient to establish the residue presence.

The lab is to continue analyzing individual subsamples until either chloramphenicol or nitrofurans is confirmed in a subsample portion, or a total of 12 negative subsample portions are completed. Alternately, a two subsample composite may be prepared provided that the lab has a documented study that demonstrates adequate sensitivity and has prior approval from ORS. If the analysis of the two subs composite indicates the presence of a target analyte at 40% of the Target Testing Level (TTL), the two subs must be analyzed individually.

If 12 subsamples are collected (3 lbs. or less per unit), select at random approximately 100 grams of product (chipped from block if frozen) from each subsample. If 6 subsamples (>3 lbs. units) were collected, analyze individually each of two (2) Approximately100 g portions, taking the portions from opposite ends of the subsample.

For All Other Drug Residues*:* Prepare one composite by combining portions of all subsamples.

If 12 subsamples are collected (3 lbs. or less), select an equal representative sample, randomly or chipped from block, (~50g of product depending on individual laboratory SOPs) from each subsample.

If 6 subsamples (>3 lbs. units) were collected, select an equal representative sample from each opposite end of the subsample for frozen blocks (~50g each depending on individual laboratory SOPs). For bags of product, randomly select two equal representative samples from the bag for the composite (~50g each depending on individual laboratory SOPs).

B. Methodology

Refer to the current FY SCOPE for priority of residues to be tested for each species.

It is very important that when results are entered into FACTS, the PAF = ANT must be used.

In the following table, hyperlinks when available have been provided for methods. Where there is no hyperlink and when a lab does not have access to the referenced methods, they should contact Cai Yanxuan (Tina), ORA/ORS/FFSS at 240-402-1369 or e-mail her at Yanxuan.Cai@fda.hhs.gov for a copy of the methods.

Method LIB#	**Method Name**	**Drug Class**	**Analytes**
LIB 4600	Simultaneous Determination of **Nitrofuran** Metabolites and **Chloramphenicol** in Shrimp Using Acetonitrile Extraction and Liquid Chromatography-Tandem Mass Spectrometric Detection: Single Laboratory Validation	Nitrofuran and Chloramphenicol	Chloramphenicol, Semicarbazide (SC) 3-amino-2-oxazolidinone (AOZ) 3-amino-5-morpholinomethyl-2-oxazolidinone (AMOZ) 1-aminohydantoin (AHD)
LIB 4597	Quantitation of **Chloramphenicol** and **Nitrofuran** Metabolites in Aquaculture Products Using Microwave-Assisted Derivatization, Automated Solid-Phase Extraction and LC-MS/MS (catfish, crawfish, frog legs, crab, and shrimp)	Nitrofuran and Chloramphenicol	Chloramphenicol Semicarbazide (SC) 3-amino-2-oxazolidinone (AOZ) 3-amino-5-morpholinomethyl-2-oxazolidinone (AMOZ) 1-aminohydantoin (AHD)
J. AOAC International Vol. 98, No. 3, 2015	Determination of Triphenylmethane Dyes and Their Metabolites in Salmon, Catfish, and Shrimp by LC-MS/MS Using AOAC First Action Method 2012.25: Collaborative Study	Dyes	Malachite Green, Crystal (Gentian) Violet, And Their Metabolites

LIB 4567	Liquid Chromatography-Tandem Mass Spectrometry Method for the Determination and Confirmation Analysis of **Emamectin** Residues in Salmon Tissue	Emamectin	Emamectin
LIB 4496	Liquid Chromatography-Tandem Mass Spectrometry Method for the Confirmation and Quantitative Analysis of **Avermectin** Residues in Salmon	Avermectins	Ivermectin, Emamectin, Abamectin, Doramectin
LIB 4562/ **LIB 4614**	Analysis of sulfonamides, trimethoprim, fluoroquinolones, quinolones, triphenylmethane dyes (and their leuco metabolites) and methyltestosterone in fish and shrimp using liquid chromatography mass spectrometry	Florfenicol	Florfenicol Amine
LIB 4619/ **LIB 4626**	Development and Validation of a LC/MS/MS/MS Method for the Determination of Isoeugenol in Finfish with Rapid Extraction and Simple Off-line Derivatization	Isoeugenol	Isoeugenol
LIB 4535	Analysis of **Stilbene** Residues in Aquacultured Finfish using LC-MS/MS	Stilbenes	Diethylstilbestrol, Dienestrol, Hexestrol
LIB 4562/ **LIB 4614**	Analysis of **sulfonamides, trimethoprim, fluoroquinolones, quinolones, triphenylmethane dyes (and their leuco metabolites) and methyltestosterone** in fish and shrimp using liquid chromatography mass spectrometry	Multi Residues - fish and shrimp	Sulfacetamide Sulfachloropyridazine Sulfadiazine Sulfadimethoxine Sulfadoxine Sulfaethoxypyridazine Sulfamerazine Sulfamethazine Sulfamethoxazole Sulfamethoxypyridazine Sulfapyridine

			Sulfaquinoxaline Sulfathiazole Trimethoprim Ciprofloxacin Enrofloxacin Norfloxacin **Sarafloxacin** **Difloxacin** Flumequine Nalidixic Acid Oxolinic Acid Methyl Testosterone Malachite Green Crystal Violet Brilliant Green Leucomalachite Green Leucocrystal Violet

C. Testing Target Levels (TTL)/ Regulatory Target Level

The following values are the current testing target levels for each method supported by CVM as sufficient for detecting the presence of each residue. These levels are also considered as a regulatory action target levels.

Animal Drug Residue	**Testing Target Level (ppb)**
Chloramphenicol	0.3
Nitrofurans AOZ metabolite of Furazolidone AMOZ metabolite of Furaltadone SC metabolite of Nitrofurazone AH metabolite of Nitrofurantoin	1.0 1.0 1.0 1.0 1.0
Fluoroquinolones: Sum of Enrofloxacin and Ciprofloxacin Difloxacin Norfloxacin	 5.0 5.0 5.0

Sarafloxacin	5.0
Quinolones Oxolinic Acid Flumequine, Nalidixic Acid	 10.0 10.0 10.0
Sum of Malachite Green and Leuco Malachite Green Sum of Gentian (Crystal)Violet and Leuco Gentian Violet Brilliant Green	1.0 1.0 1.0
Sulfonamides: Sulfamerazine Sulfadiazine Sulfachloropyridazine Sulfathiazole Sulfaquinoxaline Sulfamethazine Sulfadimethoxine Sulfacetamide Sulfadoxine Sulfaethoxypyridazine Sulfamethoxypyridazine Sulfamethoxazole Sulfapyridine	 10.0 10.0 10.0 10.0 10.0 10.0 10.0 10.0 10.0 10.0 10.0 10.0 10.0
Trimethoprim	10.0
Methytestosterone	0.8
Sum of Mebendazole + Mebendazole Amine + Hydroxymebendazole	5.0
Avermectin: Ivermectin (IVR) Emamectin (EMA) Abamectin (ABA) Doramectin (DOR)	 10.0 10.0 10.0 10.0
Isoeugenol	200.0
Florfenicol Amine (residue marker)*	1000.0
Stilbenes: Diethylstilbesterol (DES) Dienesterol (DEN) Hexestrol (HEX)	 0.25 0.25 0.25
Oxytetracycline**	2000.0

* Tolerance Level of 1.0 ppm (1000.0 ppb) established for florfenicol

amine (residue marker) in aquaculture species (salmonids, and other freshwater-reared finfish) (21CFR556.283(b))

******Tolerance Level of 2.0 ppm (2000.0 ppb) established for the sum of residues of the tetracyclines including chlortetracycline, oxytetracycline, and tetracycline in finfish and lobster (21CFR556.500(b)(1))

D. Reporting

Enter all analytical results into FACTS using PAC 04018 and PAF ANT.

The following criteria should be used for classification of samples;

Lab Class "1":

- The sample contains no residue of unapproved animal drug, or
- The sample contains residues of FDA approved drug that are below the tolerance and Testing Target Levels.

Lab Class "2":

- Labs do not have to report the Lab Class 2 samples in FACTS. Samples are to be classified as Lab Class 1 or Lab Class 3.

Lab class "3":

- The reported residue concentration of unapproved drug at or above TTL (listed in Part IV, Analytical, C),
- The reported residue concentration of FDA approved drug at or above the tolerance level.

PART V - REGULATORY/ADMINISTRATIVE STRATEGY

Samples must meet the following criteria to be submitted to DIO/CFSAN:

- Sample was collected consistent with the instructions in Part III, Inspectional, A. Sample collection.
- Sample preparation and analytical methods used are only those listed under Part IV, Sample Preparation, 3 and Methodology, B.
- The presence and confirmation (if applicable) of the residue found in any sample is at a level greater than or equal to the testing target level listed in Part IV, Testing Target Levels (TTL)/Regulatory Target Level, C.

One of the goals of this program is to obtain sufficient evidence to support broad-based enforcement strategies. Districts should be aware of detention patterns that could indicate a prevalent problem with a particular product, a processor, an importer, a particular country or a specific region and notify CFSAN's Compliance Office when these situations arise. The center intends to refocus its regulatory efforts as appropriate.

Import Compliance Actions:

If the presence of a drug residue is confirmed by the analysis of an import sample, the district should refer to the appropriate Import Alert (IA) below for appropriate follow-up action, including submitting a recommendation for addition to an Import Alert red list or removal from an Import Alert green list to DIO's Import Operation and Maintenance Branch via CMS.

Unapproved Animal Drug Residues in All Aquaculture Seafood Products

Import Alert #16-124

DETENTION WITHOUT PHYSICAL EXAMINATION OF AQUACULTURE SEAFOOD PRODUCTS DUE TO UNAPPROVED DRUGS

Chloramphenicol in Crustaceans (Crab, crawfish, shrimp, lobster, langostino)

Import Alert #16-127

DETENTION WITHOUT PHYSICAL EXAMINATION OF CRUSEACEANS DUE TO CHLORAMPHENICOL

Nitrofurans in All Seafood Products

Import Alert #16-129

DETENTION WITHOUT PHYSICAL EXAMINATION OF SEAFOOD PRODUCTS DUE TO NITROFURANS

Unapproved Animal Drug Residues in Aquaculture Seafood Products (shrimp, dace, and eel) from China-countrywide

Import Alert#16-131

DETENTION WITHOUT PHYSICAL EXAMINATION OF AQUACULTURED SHRIMP, DACE, AND EEL FROM CHINA- PRESENCE OF NEW ANIMAL DRUGS AND/OR UNSAFE FOOD ADDITIVES

Unapproved Animal Drug Residues in Aquaculture Shrimp and Prawns from Peninsular Malaysia Import Alert # 16-136

DETENTION WITHOUT PHYSICAL EXAMINATION OF AQUACULTURED SHRIMP AND PRAWNS FROM PENINSULAR MALAYSIA DUE TO PRESENCE OF DRUG RESIDUES FROM UNAPPROVED ANIMAL DRUGS OR THE PRESENCE OF UNSAFE FOOD ADDITIVES

The District should refer to any additional instructions in the Regulatory

Procedures Manual, Chapter 9.

The home district should consider a follow-up HACCP Inspection, per the Import Seafood Products Compliance Program, 7303.844, at the importer whose products were found to contain illegal residues.

The foreign processing establishment that produced and offered for entry the seafood product adulterated with residues should be considered for a HACCP inspection during planning of foreign regulatory inspections.

Domestic Compliance Actions:

At this time, all positive domestic sample results should be discussed with CFSAN's Office of Compliance/Division of Enforcement who will coordinate regulatory follow-up with CVM.

If a drug residue is confirmed in domestic produced seafood, CVM will take the lead on providing direction to determine the cause of the residue. If a follow-up inspection is warranted, the Compliance Program, 7371.006, Illegal Drug Residue in Meat, Poultry, Aquacultured Seafood and other Animal Derived Proteins will be used to conduct investigations of producers and other involved parties as appropriate. All the residues of the drugs that are being tested are unapproved

new animal drugs, therefore, CVM will consider enforcement action for any violative residues when the responsibility, jurisdiction and violation are all documented.

CVM General and Regulatory Contact: Alfred Montgomery, (240) 402-6216, Alfred.Montgomery@fda.hhs.gov

PART VI REFERENCES, ATTACHMENTS, AND PROGRAM CONTACTS

1. References (Use headings 1-3 as needed)

FDA/ORA Regulatory Procedures Manual (Including Updates) Investigations Operations Manual (Most Current)

E. Attachments – N/A

F. Program Contacts

CFSAN

Program Contact:	Teja Patel	CFSAN, Office of Compliance, Division of Field Programs and Guidance, Program Assignment and Monitory Branch HFS-615, (240) 402-2339, Teja.Patel@fda.hhs.gov
Regulatory Contact:	Brandon Bridgman	CFSAN, Office of Compliance, Division of Enforcement, Food Adulteration Assessment Branch, HFS-607, (510) 337-6794, brandon.bridgman@fda.hhs.gov
Scientific Contact:	Barbara Montwill	CFSAN, Office of Food Safety, OFS, Division of Seafood Safety, HFS-325, 240-402-1426, barbara.montwill@fda.hhs.gov

CVM

Technical and Regulatory Contact:	Phillip Kijak	CVM, OR, Division of Residue Chemistry, HFV-510, (240)-402-6689, phillip.kijak@fda.hhs.gov

ORA

Import Operations:	John Sakowski	OEIO/DIO/IOMB, 301-796-8969, John.Sakowski@fda.hhs.gov
	Lisa Elrand	OEIO/DIO, HFR-PA340, 425-302-0415, Lisa.Elrand@fds.hhs.gov

Domestic Investigations:	Rina Vora	ORA, Office of Food and Feed Program Operations Branch, HFC-130 (562) 256-9292 rina.vora@fda.hhs.gov
Scientific Contact:	Connie P Drake	ORA, ORS, Division of Food and Feed Scientific Staff, HFR-SW500 (870) 370-4003 Connie.drake@fda.hhs.gov

SCIENTIFIC METHOD ANALYSIS CONTACTS:

General CVM Method

Contact:	Philip J. Kijak	CVM/OR	240-402-6689
Chloramphenicol	Brian Veach Susan Clark	ORA/ARKL ORA/DENL	870-543-4085 303-236-9629
Fluoroquinolones	Sherri Turnipseed Hui Li	ORA/DENL CVM/OR	303-236-3072 301-210-4271
Mectins/Ivermectin	Sherri Turnipssed	ORA/ADRC	303-236-3072
Malachite Green/ Gentian Violet/ Leucomalachite	Wendy Anderson Sherri Turnipseed	ORA/ADRC ORA/DENL	303-236-3037 303-236-3072
Methyltesterone	Pak Chu Susan Clark	CVM/OR ORA/DENL	240-402-5498 303-236-9629
Multi-class, Multi-reside	Mary Carson Hui Li	CVM/OR/DRC CVM/OR/DRC	240-402-6687 240-402-5496
Nitrofurans	Pak Chu Brian Veach	CVM/OR ORA/ARKL	240-402-5498 870-543-4085
Quinolones	Sherri Turnipseed	ORA/ADRC	303-236-3072

PART VII - CENTER RESPONSIBILITIES

Program Evaluation

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 and recommended program changes. The Office of Compliance will lead the effort and work in conjunction with the Office of Food Safety and CVM to prepare summary reports for this compliance program. The summaries will outline the program office's current objectives, highlight accomplishment data, and list recommendations. The reports will be made available on FDA's intranet site: http://inside.fda.gov:9003/ProgramsInitiatives/Food/FieldPrograms/ucm015369.htm.