Georgia Department of Natural Resources

Processing and Handling of Tissue Samples for Fish Consumption Guidelines

Environmental Protection Division

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SUMMARY

Composite samples will be submitted for processing and analysis to either the Georgia DNR EPD Laboratory, or under a contract with the University of Georgia Cooperative Extension Service, Agricultural and Environmental Services Laboratories. Call Clete Barton, Tommy Miklos (North GA), or Sarah Dubose (South GA) with the Watershed Planning and Monitoring Program if you have any questions, require assistance, or are ready for samples to be picked up.

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- Tommy Miklos: (404) 651-8602; <u>thomas.miklos@dnr.ga.gov</u>
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Standard collections for fish consumption guidelines (FCG) are comprised of 3 composites of each species, where each composite contains 5 individual fish of that species. A sample size of fifteen fish has been found necessary to obtain a representative contaminant level to account for natural variations within a population. Composite sample analysis allows us to assess this number of fish and minimize the laboratory analytical support costs.

Complete sample collections increase the statistical and analytical certainty of the average concentration of toxic substances in a population of any species assessed. Complete collections also provide a more sound and better representative basis of data upon which to establish consumption recommendations allowing the public to make better choices about the fish they consume. This becomes important as new data is collected on areas that have previously been tested. These new data may replace or be combined with historical data, and decisions on issuing new updated guidelines on each area will be easier to deal with having a full fifteen fish sample. Secondarily, analysis of contaminant trends in species (increasing, decreasing or staying the same), may be made if sufficient data exist.

On occasion fish may be analyzed whole. Fish that will be analyzed whole should be wrapped in aluminum foil and placed individually in a plastic bag with the fish sample tag inside and visible within the bag with the fish.

FIELD PROCEDURE FOR COLLECTING AND PREPARING FISH SAMPLES FOR ANALYSIS

1. SAMPLING PLAN: Each year, EPD will work with WRD and CRD to develop an annual FCG Sampling Plan to identify and prioritize sites and fish species to be sampled. These sites will be distributed throughout the state and will be in large public lakes over 500 acres, popular public fish lakes and ponds less than 500 acres, rivers, and estuarine sounds. The fish species chosen will be game fish commonly eaten. For freshwater systems, three fish species will be chosen to ensure a representative contaminant sample for the entire water column; bottom feeders (e.g. catfish species), mid-depth feeders (e.g. largemouth bass) and top feeders (e.g. sunfish species). For estuarine waters, the fish species will

- 2. SAMPLE COLLECTION: Three composites of five individuals of similar size, for a total of 15 fish of each fish species requested. Fish may be held on wet ice for up to 24 hours or frozen for up to 30 days before sample preparation. Most freshwater fish species are collected in the late summer through the fall as during this period fish tend to have the least variable lipid content, and organic contaminants residues will be more consistent. However, some migratory species may be seasonal visitors to target waters during other periods, and collection times may need to be adjusted. Adjustments to collection season may also be necessary to avoid collections immediately following spawning for those species that do not spawn in the spring, such as occurs with several estuarine/marine species.
- **3. COMPOSITE SAMPLE SIZE:** There should be five fish in each composite. As a practical matter, the number of fish in the composite is somewhat flexible as long as the size criterion is met (smallest is 75% the length of the largest) and all fish in a composite are of the same species. If extremely small, more than five fish may be required to obtain the minimum sample weight of 100 grams.
- 4. **COMPOSITE SAMPLE PROCESSING:** Stainless steel filleting knives and impervious boards are suggested. The workstation should be arranged to provide a clean workspace that can be renewed between each composite. To prevent cross contamination, aluminum foil or plastic back bench cover should be replaced between each composite. Knives, scalers, catfish nippers and cutting boards should be cleaned between processing composite samples.
 - a. Each species from a station should be segregated and arranged into composites. This should be done by first weighing and measuring each fish and recording this information on a field sheet and/or individual fish tag. For blue crabs, the carapace width is recorded rather than the length.
 - b. After grouping into composite samples of similar size, an individual/composite number can be assigned and added to the fish tag.
 - c. Samples will be three composites of edible flesh (filets, except for small sunfish), skin on and scales off, including the belly flap with the rib cage removed. Catfish and gar will be skin off. Each composite sample will be made up of similar size (length) fish. The smallest fish in a composite will be at least 75% of the total length or carapace width with crabs of the largest. Fish that are to be analyzed as individuals follow the same protocol. Tissues other than muscle may be required for analysis to answer additional special questions. Specific procedures will be established for these fish.
 - i. **Fish 3 pounds or smaller:** Scaled with skin on (except catfish, and gar). Entire filet, belly flap included with rib cage removed.
 - ii. **Fish larger than 3 pounds:** Scaled with skin on (except catfish and gar). Partial filet taking two hand size portions, removed from the dorso-lateral

area, one from each side of the fish, belly flap included with rib cage removed.

- iii. **Catfish and Gar:** Catfish filets as described in i. and ii. above, but with skin removed. Gar filets from above abdominal cavity only and with skin removed.
- iv. Sunfish: Scaled with skin on, head off, gutted, belly on, and left on backbone. This is typically how consumers prepare and cook sunfish. Many of these fish harvested are 10 inches or less in length and this allowance is made because of the small muscle mass produced in a filet and because of the impracticality of filleting such a small fish. Large crappie may be filleted.
- v. Very Small Fish (≤5 inches): Sometimes special collections are requested in areas where fish kills have been recent or repetitive and species present are in young age classes or quite small. To conduct analyses for all 43 analytes, a certain minimum sample amount is required (100-150 grams), which a 5-fish composite will not provide. In these cases, collection of 10 fish per composite may be necessary, as long as this does not overly stress the site by over collection. Additionally, because of the small size, filleting each fish is impractical. Prepare fish by scaling if possible, gutting and cutting head off; and cutting fins off.
- vi. **Crabs, Shrimp, Oysters or other shellfish:** Adult Blue Crabs, minimum size approximately 5-6". Three composites per site.. Minimum of five crabs per composite (up to 10 may be required to obtain a minimum sample weight of 100 grams), with the smallest within 75% of largest (carapace width, spine to spine). Adult crabs, "Lump and Claw meat (minus carapace, legs and viscera). Shrimp, 300-400 grams (10-13 ounces) each composite. Three composites per site. Head off and peeled, Oysters (or other bivalve), minimum of a pint per composite, 3 composites per site, similar shell length and legal. Shucked with shell liquid included.
- vii. Other edible organ or tissues: Roe from some species is considered a delicacy and is consumed. When roe is prepared, it should be wrapped separately in aluminum foil and included with the filets of the female that it originated from. Striped mullet is one species where the roe is commonly consumed. Additionally, striped mullet have a gizzard (specialized muscle in the alimentary tract used to grind plant matter), which is also frequently consumed.

5. SAMPLE PREPARATION:

a. Each fish tissue sample should be wrapped separately in heavy duty aluminum foil, shiny side out. The sample plus the tag should be placed in a resealable plastic bag. The tag should also be attached to the aluminum foil with clear packing tape. Please make sure tag is visible through the bag containing samples.

- b. Once all five fish for a composite have been processed, wrapped in aluminum foil, and bagged, place them all in a larger plastic bag. This constitutes the composite sample. All the fish in this bag will be ground up together in the lab and analyzed.
- c. After sample preparation, samples should be frozen and stored frozen until preparation for analyses in the laboratory begins. The plastic ties and tags maintain sample integrity from start to finish. The aluminum foil keeps the filets protected. The small (sandwich size) plastic bags keep the two filets and tag together. The large (gallon size) plastic bag keeps composite fish together. It is most important to stress personal attention to anything that might be transferred to the sample by the operator through touching foreign objects and then the sample. Work should be conducted in a clean area, and the bench cover should be changed, and utensils should be cleaned between composites to prevent cross contamination.
- **6. PAPERWORK:** A second individual should perform all record keeping, sample identification and logging duties.
 - a. **INDIVIDUAL FISH SAMPLE TAG:** This tag is attached to each individual fish. It contains the date, collector, waterbody, site location, fish species, length, weight, fish id, composite number, and any additional notes. A computer-generated page of blank tags is attached. However, you can request this computer file, which may be printed on laser jet using Rite-in-Rain [®] paper.
 - b. **FIELD & LAB DATA SHEET:** This is the sheet on which the lab identification number will be recorded when the samples reach the lab. A copy is attached. When you have your collections done, please fax a copy of these sheets to Clete Barton or Tommy Miklos at (404) 651-8455 and retain a digital copy of the lab sheet for your files.
 - c. **CHAIN OF CUSTODY FORM:** This document identifies the people responsible for the samples from collection through storage and transport all the way to the lab. A copy is attached but we typically fill them out here prior to making the sample pickup. The original chain of custody should be handed over to EPD when they pick up the samples. You should retain a digital copy of the chain of custody for your files.

LABORATORY ANALYTICAL DATA QUALITY OBJECTIVES (DQOS)

As defined by EPA, Data or Method Quality Objectives (DQO or MQO) provide a basis whereby the level of data quality needed is established and may be evaluated with respect to its intended use. Data Quality Objectives are qualitative and quantitative statements derived from the outputs of each step of the DQO process. The EPD Laboratory has established DQOs for the analysis of metals, mercury, PCBs and organochlorine pesticides in biological tissues. The current Georgia EPD Laboratory MQO/DQOs are appended to this plan of study. When assessing data for deriving consumption recommendations, the reporting limit for a parameter should be below the any restrictive triggering value. When the Science Advisory Committee made the recommendations for the Georgia risk-based program, minimum quantitative limits were established for the 43 contaminants identified for analysis in the fish tissue projects. These are presented in the following table. The alpha- and gamma-chlordane isomers were

requested by the Hazardous Waste Branch as additional parameters.

PARAMETERS AND REQUESTED MINIMUM QUANTITATIVE LIMITS FOR FISH TISSUE SAMPLES FOR FISH CONSUMPTION GUIDELINES

Metals: (mg/kg wet weight)	Pesticides/PCBs: (mg/kg wet weight)
Antimony: 1.0	Aldrin: 0.01
Arsenic: 0.06	a-BHC: 0.01
Beryllium: 1.0	b-BHC: 0.01
Cadmium: 1.0	d-BHC: 0.01
Chromium, Total: 1.0	g-BHC (Lindane) : 0.01
Copper: 1.0	Chlordane (total, alpha-& gamma-chlordane): 0.03
Lead: 1.0	4,4-DDD: 0.01
Mercury: 0.01	4,4-DDE: 0.01
Nickel: 1.0	4,4-DDT: 0.01
Selenium: 1.0	Dieldrin: 0.01
Silver: 1.0	Endosulfan I: 0.02
Thallium: 1.0	Endosulfan II: 0.03
Zinc: 1.0	Endosulfan Sulfate: 0.05
	Endrin: 0.01
	Endrin Aldehyde: 0.05
	Heptachlor: 0.01
	Heptachlor Epoxide: 0.01
	Toxaphene: 0.1
	PCB-1016: 0.03
	PCB-1221: 0.03
	PCB-1232: 0.03
	PCB-1242: 0.03
	PCB-1248: 0.03
	PCB-1260: 0.03
	Methoxychlor: 0.05
	HCB: 0.01
	Mirex: 0.1
	Pentachloroanisole: 0.01
	Chlorpyrifos: 0.01
	Total Lipid (%): 0.1%

APPENDIX

1. GADNR FISH/SEAFOOD FIELD DATA AND LAB PROCESSING SHEET FOR GENERAL CONTAMINANTS

2. FISH TISSUE CHAIN OF CUSTODY FOR GENERAL CONTAMINANTS PROJECT

3. SAMPLE TAGS FOR GENERAL CONTAMINANTS

GADNR FISH/SEAFOOD FIELD DATA AND LAB PROCESSING SHEET FISH CONSUMPTION GUIDANCE GENERAL CONTAMINANTS ANALYSIS

WATERBODY NAME:	LOCATION:
COLLECTORS:	SPECIES:
DATE:	TIME:
LATITUDE (DD.DDD):	LONGITUDE (DD.DDD):

COMPOSITE #1				Weight of Aliquot for		
Total Length (mm)	Fish #	Total Weight (g)	EPD FIELD I.D. #	Composite (g)	Lab #	Archive #
	1					
	2					
	3					
	4					
	5					

COMPOSITE #2			Weight of Aliquot for			
Total Length (mm)	Fish #	Total Weight (g)	EPD FIELD I.D. #	Composite (g)	Lab #	Archive #
	1					
	2					
	3					
	4					
	5					

COMPOSITE #3				Weight of Aliquot for		
Total Length (mm)	Fish #	Total Weight (g)	EPD FIELD I.D. #	Composite (g)	Lab #	Archive #
	1					
	2					
	3					
	4					
	5					

FISH TISSUE CHAIN OF CUSTODY FOR CONTAMINANTS PROJECT GEORGIA DEPARTMENT OF NATURAL RESOURCES

Item #	# Composites or (Other)	EPD Field ID #	WRD/CRD Region	Species	Waterbody	Location
1						
2						
3						
4						
5						
6						
7						
8						
9						
10						
11						
12						

Chain-of-Custody Receipt and Delivery Signature Record

Item #	Date	Relinquished By	Received By	Date	Relinquished By	Received By

Date:	Collector(s):		
Water Body:			
Site Location:			
Species:	Length (mm):	Weight (grams):	
Fish #:	/ Composite #	Note:	

Date:	Collector(s):		
Water Body:			
Site Location:			
Site Location.			
Species:	Length (mm):	Weight (grams):	
Fish #:	/ Composite #	Note:	

Date:	Collector(s):		
Water Body:			
Site Location:			
Species:	Length (mm):	Weight (grams):	
Fish #:	/ Composite #	Note:	

Date:	Collector(s):		
Water Body:			
Site Location:			
Species:	Length (mm):	Weight (grams):	
Fish #:	/ Composite #	Note:	

Date:	Collector(s):		
Water Body:			
Site Location:			
Species:	Length (mm):	Weight (grams):	
Fish #:	/ Composite #	Note:	

Date:	Collector(s):		
Water Body:			
Site Location:			
Species:	Length (mm):	Weight (grams):	
Fish #:	/ Composite #	Note:	