# Field Sampling Plan for the National Study of Chemical Residues in Lake Fish Tissue 



Prepared for:
Leanne Stahl, National Study Manager
U.S. Environmental Protection Agency, Office of Science and Technology 1200 Pennsylvania Avenue, NW (MC 4305T)
Washington, D.C. 20460

Prepared by:
Tetra Tech, Inc.
10045 Red Run Boulevard, Suite 110
Owings Mills, MD 21117

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### 1.0 PROJECT BACKGROUND AND DESCRIPTION

### 1.1 Why conduct the National Study of Chemical Residues in Lake Fish Tissue?

The U.S. Environmental Protection Agency (USEPA) Office of Water conducted a national screening-level investigation in 1987 (USEPA 1992) to determine the prevalence of selected bioaccumulative pollutants in fish and to correlate elevated fish tissue contaminant levels with pollutant sources. Gamefish and bottom-dwelling species were collected from 388 locations across the country thought to be influenced by various point and nonpoint sources. These fish tissue samples were analyzed to determine levels of 60 target analytes, including dioxins and furans, PCBs, pesticides and herbicides, mercury, and several other organic compounds. Results of the 1987 study indicated that target analytes were present in fish tissue at many of the sampling sites, and some of the contaminants (e.g., PCBs, dieldrin, mirex, and combined chlordane) occurred at levels posing potential human health risks.

The Office of Science and Technology (OST) within the Office of Water has initiated work on the new four-year National Study of Chemical Residues in Lake Fish Tissue (or National Fish Tissue Study), which is designed to expand the scope of the 1987 study. In October 1998, USEPA convened a two-day workshop of more than 50 scientists from state, federal, and tribal agencies to obtain technical input on sampling design, target analytes, sampling methods and data management. Input from scientists at the workshop and other technical experts that participated in numerous study planning meetings was used to develop a final study design. The current study is statistically designed and will provide screening-level data on a larger suite of fish tissue contaminants from a greater number of waterbodies than were sampled in 1987.

The National Fish Tissue Study broadens the scope of the 1987 study (USEPA 1992) which focused on chemical residues in fish tissue near point source discharges. This new study will:

- provide information on the national distribution of selected persistent, bioaccumulative, and toxic (PBT) chemical residues in gamefish and bottomdwelling fish in lakes and reservoirs of the coterminous United States (excluding the Great Lakes and the Great Salt Lake),
- include lakes and reservoirs selected according to a probability design,
- involve the collection of fish from those randomly selected lakes and reservoirs over a four-year survey period (1999-2002),
- not be used to set fish consumption advisories; however, states and Native American tribes may choose to initiate a detailed fish study in a particular lake based on the screening contaminant concentrations provided by the National Fish Tissue Study, and
- include the analysis of fish tissue for PBT chemicals selected from USEPA's multimedia candidate PBT list of 451 chemicals and from a list of 130 chemicals from several contemporary fish and bioaccumulation studies. A final target analyte list of 274 PBT chemicals (including breakdown products and PCB congeners) was compiled based on input from study design workshop participants and a review team of analytical experts convened in October 1998 and March 1999, respectively.


### 1.2 What are the study objectives?

The specific objective of the National Fish Tissue Study is to estimate the national distribution of the mean levels of selected persistent, bioaccumulative, and toxic chemical residues in fish tissue from lakes and reservoirs of the continental United States.

In so doing, the study will provide the following types of information:

- information to meet objectives of the President's Clean Water Action Plan (CWAP).

CWAP Key Action \#1: USEPA and NOAA will conduct a national survey of mercury and other contaminants in fish and shellfish throughout the country, and will coordinate the effort with states and tribes to maximize geographic coverage. The shellfish survey will be based on the data obtained by NOAA's ongoing Mussel Watch Project.

- information about persistent, bioaccumulative, and toxic chemicals (PBTs) for the Agency's PBT Initiative.

The PBT Initiative seeks to identify areas of concern for human and/or ecological health. Study of fish tissue may reveal where PBTs not previously considered a problem are present at levels of concern.

- data to answer important questions concerning the national occurrence of fish tissue contamination.

What is the national extent of selected chemical contaminants in fish from lakes and reservoirs of the coterminous United States (excluding the Great Lakes)?

Are contaminant levels in fish high enough to warrant further investigation?

### 1.3 Where will field sampling occur and who will be conducting the field work?

The USEPA Office of Science and Technology will coordinate with regional staff, state resource agencies, and Native American tribes to collect fish tissue samples from randomly selected lakes and reservoirs in the continental United States. With a combined network of partners and contractors (refer to the List of Contacts on pages $v$ to xxii), USEPA anticipates the sampling of approximately 500 lakes across the country (Appendix A) during the four-year sampling duration of the study (1999-2002). The fish tissue samples will be collected based on a probability design to provide information on national distribution of the mean levels of contaminants in fish. This random selection of lakes and reservoirs is important for fulfilling the study design objectives.

### 1.4 When will field sampling occur?

Field sampling activities began in the fall of 1999, will continue in 2000 and 2001 during the summer and fall, and will conclude in the fall of 2002. Due to the effort required to initiate the study (e.g., project coordination and development of study materials such as the Quality Assurance Project Plan), fewer lakes were sampled in 1999 than will be sampled during other sampling years. Field collection effort summaries will be prepared and distributed annually to all study participants and contacts. The final study report is scheduled to be completed in 2003.

Field sampling will be conducted during the period when water and weather conditions are conducive to safe and efficient field sampling, and when the target species are most frequently harvested by anglers. For most inland freshwaters, the most desirable sampling period is from late summer to early fall, since lipid content is usually highest and water levels are usually lowest at that time. If possible, sampling should not occur during the spawning period of the particular target species being sought. With these recommendations in mind, and considering the geographic extent of the study area (i.e., range of latitudes and longitudes) the field sampling period will begin in August and last through November (and possibly into December in warmer regions). Answers to frequently asked questions regarding the annual sampling schedule and the seasonal sampling period are provided in Sections 5.1 and 5.2, respectively.

### 2.0 SAMPLING PROCESS DESIGN

### 2.1 What is a target lake?

This study defines a lake as a permanent body of water of at least one hectare (2.47 acres) in surface area with a minimum of $1,000 \mathrm{~m}^{2}$ of open (unvegetated) water and a minimum depth of one meter. The lakes in this study must also have a permanent fish population.

A critical element of the statistical survey design is the determination of the status of each lake in the sample of 900 candidate lakes (Appendix A). This means that each lake is checked to determine if it meets the definition of a lake for the study. Other situations can also occur that
will result in a lake not being sampled. It is essential that a complete record of this information be reported to the USEPA National Study Manager, since this information is required to complete the survey estimation procedures. First, the lake may be on private land and require landowner permission to visit the lake. If a landowner refuses access to a lake selected for the study, then this needs to be reported to the USEPA National Study Manager. Second, a lake may occasionally be physically inaccessible. If there are logistical or safety constraints that make a lake inaccessible, then the reason why the lake is inaccessible needs to be reported to the USEPA National Study Manager. Third, the lakes in this study must also have a permanent fish population. Examples of nonpermanent fish populations are lakes that are subject to annual fish winterkill, or are recently stocked with fingerlings. Stocked lakes with adult fish are defined as having a permanent fish population. As a contingency (i.e., if a large number of lakes are determined to be non-targets via reconnaissance), a second sample of lakes has been selected as a reserve. Information regarding lake re-selection is provided in Section 5.3.

For this study, the best way to develop an estimate of variability is to simply revisit a subset, $10 \%$ of the sites, and repeat the lake sampling procedure, compositing, and chemical analyses. Each year the National Study Manager will make arrangements with individual states to obtain the appropriate number of duplicate samples for that year (see Section 2.4)

### 2.2 What type of samples will be collected?

To meet the study objectives, the National Fish Tissue Study will include composite sampling of fish fillets for predator/gamefish species and whole fish for bottom-dwelling species from each sample lake. Five individuals per composite will be collected, all of which will be large enough to provide sufficient tissue for analysis of the group of target analytes. It has been determined that at least 560 grams of edible tissue for predators, and 560 grams of total body tissue for bottom-dwellers will be required from the composites to allow for analysis of all target analytes.

> In the event that predator species are available at a target lake but bottomdwellers are not, Sampling Teams should retain a predator composite and document on the Field Record Form that bottomdweller species were unavailable (refer to Section 5.4). The outcome of field sampling efforts will ultimately depend on the natural diversity, abundance, and availability of fish in the target lakes.

### 2.3 What is a composite sample?

Based on the recommendations of USEPA's Guidance for Assessing Chemical Contaminant Data for Use in Fish Advisories, Volume 1: Fish Sampling and Analysis, Second Edition (USEPA 1995), fish used in a composite sample should meet the following criteria:

- all be of the same species,
- satisfy any legal requirements of harvestable size or weight, or at least be of consumable size if no legal harvest requirements are in effect,
- be of similar size so that the smallest individual in a composite is no less than $75 \%$ of the total length of the largest individual,
- be collected at the same time (i.e., collected as close to the same time as possible but no more than 1 week apart) [Note: This assumes that a sampling crew was unable to collect all fish needed to prepare the composite sample on the same day. If organisms used in the same composite are collected on different days (no more than 1 week apart), individual fish will be frozen until all the fish to be included in the composite are available for delivery to the Sample Preparation Laboratory.], and
- be collected in sufficient numbers (five per composite) and of adequate size (five adult specimens that collectively will provide greater than 560 grams of edible tissue for predators, and 560 grams of total body tissue for bottom-dwellers) to allow analysis of recommended target analytes.

Answers to frequently asked questions regarding numbers of fish per composite, fish length and composite weight are provided in Sections 5.5, 5.6, and 5.7, respectively.

### 2.4 What is a duplicate sample?

Duplicate fish samples will be collected and analyzed from $10 \%$ of the lakes during the study to provide an estimate of variability. States designated by the EPA National Study Manager to collect duplicate samples can choose which lake(s) to collect duplicate samples from, and can collect the duplicate samples at the same time as the regular composite sample. At sites selected for duplicate sampling, this would involve collecting 10 predators and 10 bottom-dwelling fish to form the following 5-fish composites: one predator composite, one duplicate predator composite, one bottom-dweller composite, and one duplicate bottom-dweller composite.

### 2.5 What are the criteria for selecting target species?

According to USEPA's Guidance for Assessing Chemical Contaminant Data for Use in Fish Advisories, Volume I: Fish Sampling and Analysis, Second Edition (USEPA 1995), the primary criteria for selecting target species is that they:

- are commonly consumed in the study area,
- may potentially accumulate high concentrations of chemicals, and
- have a wide geographic distribution.

Secondarily, the target species should be:

- easy to identify,
- abundant,
- easy to capture, and
- large enough to provide adequate tissue for analysis (i.e., adult specimens that as a five-fish composite will provide at least 560 grams of edible tissue for analysis).

Two distinct ecological groups of fish, bottom-dwellers and predators, will be sampled for this study. This permits monitoring of a wide variety of habitats, feeding strategies, and physiological factors that might result in differences in bioaccumulation of contaminants.

### 2.6 Are certain species preferred as targets?

Suggested target species for this study are listed below (Table 1) in order of preference. Additional target species may be added on an as-needed basis. State personnel, with their knowledge of site-specific fisheries and human consumption patterns, will aid in the determination of the availability of target species. Every effort will be made to collect the desired species and number of fish; however, the outcome of field sampling efforts will ultimately depend on the natural diversity and abundance of fish in the study lakes.

Table 1. Recommended Target Species for Inland Freshwaters (in Order of Preference).

|  | Family name | Common name | Scientific name |
| :---: | :---: | :---: | :---: |
|  | Centrarchidae | Largemouth bass | Micropterus salmoides |
|  |  | Smallmouth bass | Micropterus dolomieu |
|  |  | Black crappie | Pomoxis nigromaculatus |
|  |  | White crappie | Pomoxis annularis |
|  | Percidae | Walleye | Stizostedion vitreum |
|  |  | Yellow perch | Perca flavescens |
|  | Percichthyidae | White bass | Morone chrysops |
|  | Esocidae | Northern pike | Esox lucius |
|  | Salmonidae | Lake trout | Salvelinus namaycush |
|  |  | Brown trout | Salmo trutta |
|  |  | Rainbow trout | Oncorhynchus mykiss |
|  |  | Brook trout | Salvelinus fontinalis |

Table 1 (continued). Recommended Target Species for Inland Freshwaters (in Order of Preference).

|  | Family name | Common name | Scientific name |
| :---: | :---: | :---: | :---: |
| Bottom-dwelling Species(in order of preference) | Cyprinidae | Common carp | Cyprinus carpio |
|  | Ictaluridae | Channel catfish | Ictalurus punctatus |
|  |  | Blue catfish | Ictalurus furcatus |
|  |  | Brown bullhead | Ameiurus nebulosus |
|  |  | Yellow bullhead | Ameiurus natalis |
|  | Catostomidae | White sucker | Catostomus commersoni |

### 3.0 FIELD SAMPLING LOGISTICS AND METHODS

### 3.1 Who will participate in the field sampling efforts?

Each Field Sampling Team is required to have the necessary knowledge and experience to perform all field activities. This includes both knowledge and experience in the collection and identification of fish, in the use of fisheries sampling gear specified for the study, and in the operation of small boats. It also includes training in project-specific sample collection and handling procedures. The field sampling crews will be primarily composed of state, tribal, and regional fisheries biologists or contracted biologists with a strong technical background in fisheries sampling activities (refer to the List of Contacts on pages v to xxii). Each Field Sampling Team should consist of (at a minimum) one experienced fisheries biologist, one field technician, and a quality control specialist, all of whom must have experience with the array of fisheries sampling gear types to be used. In some cases the fisheries biologist may serve in dual capacities, assuming responsibility for site quality control.

### 3.2 How will fish be collected?

Sampling Teams will be equipped with an array of both active and/or passive gears to ensure the collection of the desired target numbers and species of fish. Selection of the most appropriate gear type(s) for a particular target lake will be at the discretion of the experienced on-site fisheries biologist. Passive collection devices (e.g., gill nets) must be checked frequently (e.g., at least once every 24 hours) to ensure a limited time lag between fish entrapment and sample preparation/preservation.

### 3.3 What equipment will be required?

A list of equipment and expendable supplies is provided below (Table 2). Sampling Teams will be responsible for providing fisheries sampling gear and sampling vessels, and the Sample Control Center will provide sample packaging and shipping supplies.

Table 2. Equipment and Supply List for Fish Tissue Sampling.

| Provided by Field Sampling Teams | Provided by the Sample Control Center |
| :---: | :---: |
| Sampling vessel (including boat, motor, trailer, oars, gas, and all required safety equipment) ${ }^{(\mathrm{a})}$ | Ice chests and ice chest packing instruction sheet |
| Electrofishing equipment - OPTIONAL (including variable voltage pulsator unit, generator, electrodes, wiring cables, dip nets, protective gloves, protective boots, and all necessary safety equipment ${ }^{(\text {a) }}$ | Aluminum foil (solvent-rinsed and baked) |
| Nets - OPTIONAL (including trawls, seines, gill nets, fyke nets, trammel nets, hoop nets, pound nets, trap nets ${ }^{(\mathrm{a})}$ | Heavy-duty food grade polyethylene tubing |
| Angling equipment - OPTIONAL (including fishing rods, reels, line, terminal tackle, trot lines $)^{(\mathrm{a})}$ | Large plastic (composite) bags |
| Coast Guard-approved personal floatation devices | Clean nitrile gloves |
| Maps of target lakes and access routes | Field Record Forms |
| Global Positioning System (GPS) unit - OPTIONAL ${ }^{(2)}$ | Sample Identification Labels |
| pH meter (including associated calibration supplies) ${ }^{(a)}$ | Chain-of-Custody Forms |
| Livewell and/or buckets | Chain-of-Custody Labels |
| Measuring board (millimeter scale) | Dry ice |
| Knife or scissors | Adhesive (dry ice) hazmat label |
| Scientific collection permit | Preaddressed overnight courier airbill |
| Black ballpoint pens and/or waterproof markers | Preaddressed adhesive shipping label |
| Clipboard | Adhesive "perishable goods" label |
| Packing/strapping tape | Plastic cable ties |
| First aid kit and emergency telephone numbers | Plastic bubble-wrap |

(a) Selection and exact specifications at the discretion of the experienced on-site fisheries biologist.

### 3.4 Where should samples be collected in target lakes?

The field objective is for sampling teams to obtain a representative composite sample for both a predator and a bottom-dwelling species (if available) from each lake or reservoir selected for the National Fish Tissue Study. Each composite must consist of all the same species, individual fish must be of similar size, and the composite must be able to deliver 560 grams of fish tissue (fillets for predators, and whole bodies for bottom-dwellers) for chemical analysis. To obtain a representative sample of the targeted species in lakes and reservoirs, field teams will obviously have to consider factors such as available, suitable habitat, and should also consider the presence of contaminant gradients in planning sampling locations for the target lake. Ideally, the habitats suitable for target species would be determined for the lake, and up to three locations of that habitat would be randomly selected for sampling in the lake (particularly in large waterbodies). If a contamination gradient may be present in the waterbody, then three locations across the gradient should be selected for sampling. For example, in reservoirs, the three locations may be in habitat near the inflow, middle, and outflow of the reservoir. The composite is intended to estimate the mean fish tissue contaminant concentration for the lake or reservoir. Given the
diversity of lakes and reservoirs in the study, and given the multiple species that must be used, the study must rely on the local knowledge of the field teams in the selection of the representative composite sample.

### 4.0 SAMPLE HANDLING AND INTEGRITY

### 4.1 How should fish be handled in the field?

As soon as fish are obtained via active collection methods, or removed from passive collection devices, they should be identified to species. Species identification should be conducted only by experienced personnel knowledgeable of the taxonomy of fish in the waterbodies included in the fish contaminant monitoring program. Nontarget species collected by the field team should be returned to the water. Individuals of the selected target species should be rinsed in ambient water to remove any foreign material from the external surface, handled using clean nitrile gloves (provided by the Sample Control Center), and placed in clean holding containers (livewell, buckets, etc.) to prevent contamination. Each fish of the selected target species should be measured to determine total body length (mm). Maximum body length should be measured, i.e., the length from the anterior-most part of the fish to the tip of the longest caudal fin ray (when the lobes of the caudal fin are depressed dorsoventrally). When sufficient numbers of the target species have been identified to make up a suitable composite sample, the species name, specimen lengths, and all other site and sampling information should be recorded on the Field Record Form (see Section 4.6).

After initial processing to determine species and size, each of the five fish found to be suitable for the composite sample will be individually wrapped in heavy-duty aluminum foil (provided by the Sample Control Center as solvent-rinsed, oven-baked sheets). For specimens with sharp fins, spines may be broken (via gloved hands or with the use of a tool covered with the aluminum foil provided by the Sample Control Center) to prevent perforation of

Fish tissue sample collection procedures are presented in Appendix B as sequential steps, and include equipment, materials and methods required to perform field sampling activities. the wrapping materials. The broken section of the fins should be included with the fish sample. A Sample Identification Label (see Section 4.7) will be prepared for each aluminum foil-wrapped specimen. Each foil-wrapped fish will be placed into a waterproof plastic tubing that will be cut to size to fit the specimen (i.e., heavy duty food grade polyethylene tubing provided by the Sample Control Center), and each end of the tubing will be sealed with a plastic cable tie. The completed Sample Identification Label will be fastened to one of the cable ties, and the entire specimen package will be "double-bagged" (i.e., placed inside a large plastic composite bag with all the specimens of the same species from that site and sealed with another cable tie).

### 4.2 How should samples be "preserved" following collection?

Once packaged, samples should be immediately placed on dry ice for shipment. If samples will be carried back to a laboratory or other facility to be frozen before shipment, wet ice can be used to transport wrapped and bagged fish samples in the coolers to that laboratory or facility. If possible, all of the specimens in a composite sample should be kept together in the same shipping container (ice chest) for transport. Sampling Teams have the option, depending on site logistics, of:

- $\quad$ shipping the samples packed on dry ice (in sufficient quantities to keep samples frozen for up to 48 hours), via priority overnight delivery service (i.e., Federal Express), so that they arrive at the Sample Preparation Laboratory within less than 24 hours from the time of sample collection, or
- freezing the samples within 24 hours of collection (at $\leq-20^{\circ} \mathrm{C}$ ), and storing the frozen samples until shipment within approximately 1 week of sample collection (frozen samples will subsequently be packed on dry ice and

Dry ice can be obtained by contacting the Sample Control Center approximately 5 days before it will be needed in the field. Contact: Chris Moore (703) 461-2360 or Chris Maynard (703) 461-2395. Field Teams interested in obtaining dry ice from a local vendor should contact field support contractors Blaine Snyder or Mandy Richardson (410) 356-8993 to inquire about setting up an account.

The time of sample collection, and time of their relinquishment by the sample team must be recorded on the Chain-of-Custody Form (see Section 4.8).

### 4.3 Are there any special shipping requirements?

Field Sampling Teams should avoid shipping samples for weekend delivery to the Sample Preparation Laboratory unless prior plans for such a delivery have been agreed upon with the Sample Control Center.

Field sampling teams must notify the Sample Control Center (DynCorp) by telephone (Chris Moore 703/461-2360 or Chris Maynard 703/461-2395) of an incoming shipment.

Shipping container packing instructions are detailed in Appendix C. Not all Federal Express drop-off locations will accept shipments containing dry ice. Call in advance (800-Go-FedEx) to ensure that your drop-off location accepts dry ice. If you cannot locate a station in your area that accepts dry ice, simply call for a pickup (800-Go-FedEx). Federal Express will gladly pick these shipments up for you.

Federal Express Dangerous Goods personnel have approved shipment of packages for this study, providing that the packing instructions in Appendix C are followed.

### 4.4 What field paperwork is required?

Thorough documentation of all field sample collection and handling activities is necessary for proper processing in the laboratory and, ultimately, for the interpretation of study results. Field sample collection and handling will be documented in writing (for each sampling site) using the following forms and labels:

- a Field Record Form that contains information about each individual specimen and lake site (see Section 4.6),
- a Sample Identification Label that accompanies and identifies each fish specimen (see Section 4.7),
- a Chain-of-Custody Form that provides constant tracking information for all samples (see Section 4.8), and
- a Chain-of-Custody Label that seals each sample shipping container (provided by the Sample Control Center) (see Section 4.9).


### 4.5 How are samples identified on the field paperwork?

A unique tracking code (i.e., composite sample identification code) will be used to identify each composite sample. The ten-character code will include:

- state of collection (two-character abbreviation),
- year of collection (two-number abbreviation),
- lake identification number (four-digit code from Appendix A),
- composite type (one character -- $\mathrm{P}=$ predator species; $B=$ bottom-dwelling species), and
- $\quad$ sample type (one character -- $\mathrm{S}=$ standard sample; $\mathrm{D}=$ duplicate sample).


### 4.6 What information is recorded on the Field Record Form?

Detailed documentation of the samples collected in the field (for shipment to the Sample Preparation Laboratory) and information about the collection location will be recorded on a Field Record Form. One form must be completed for each sample composite. One page of the fourpage carbonless copy form will be retained by the sampler, and the other copies will be included with sample shipment to the Sample Preparation Laboratory. The latter three copies will be
placed in a waterproof plastic bag (provided by the Sample Control Center) and sealed inside the shipping container. All entries will be made in black ink and no erasures will be made. Each form will have the proper entry requirements, as demonstrated in the example below (Figure 1, page 13).

### 4.7 What information is recorded on the Sample Identification Label?

Each individual fish will be labeled by fastening a Sample Identification Label to the wrapped specimen (i.e., one end of the plastic tubing) (see Section 4.1). All sample label entries, as detailed in the example below (Figure 2, page 14), will be made with black indelible ink. The sample label will accompany each fish throughout the chain-of-custody.

### 4.8 What information is recorded on the Chain-of-Custody Form?

All samples will be transferred to the Sample Preparation Laboratory under chain of custody. The Chain-of-Custody (COC) Form acts as a record of sample shipment and a catalog of the contents of each shipment (coinciding with information on the Field Record Form). The COC forms will be produced as five-page carbonless copies with one copy retained by the sampler and four for shipment to the Sample Preparation Laboratory. The latter four copies will be placed in a waterproof plastic bag (provided by the Sample Control Center) and sealed inside the shipping container. All COC entries, as demonstrated in the example below (Figure 3, page 15), will be made in black ink.

### 4.9 What information is recorded on the Chain-of-Custody Label?

Immediately following the packing of each shipping container, each container (ice chest) will be secured with packaging tape and sealed with a Chain-of-Custody Label (provided by the Sample Control Center). The Chain-of-Custody Label must contain the signature of the sampler, and the date and time written in ink. The seal must be affixed such that the shipping container cannot be opened without breaking the seal (e.g., label adhered across the ice chest latch), so as to protect and document the integrity of the contents from field to laboratory.

### 4.10 Who should field sampling teams contact if questions arise?

Leanne Stahl, National Study Manager, can be contacted via telephone at EPA Headquarters (202/260-7055), or via email at stahl.leanne@epamail.epa.gov. The Sample Control Center (DynCorp) will be available from 7 a.m. to 6 p.m. (Eastern Time), Monday through Friday, at 703/461-2100. There will be 24-hour access to the national field support contractor (Tetra Tech, Inc.). Blaine Snyder, Tetra Tech’s Work Assignment Leader, will be available around the clock via a national toll-free number. The toll-free pager number is 1-800-722-0812. Callers can either log in a return number or leave a brief voice mail message. The pager can also accept an email message (sent to 7220812@ skytel.com) of up to 100 characters.

A complete list of National Project Management and Administration contacts, Analytical Team Members, Regional Fish Tissue Study Coordinators and Sampling Team contacts is provided on pages v through xxii.

## Figure 1. Field Record Form



Collector Name (print and sign): Bob Smith Bole Smith
Affiliation: PA Dept of Environmental Protection Phone: (717)555-1212
Address: Fulton Building, Harrisburg, PA 17120


FORM DISTRIBUTION: White -- Tetra Tech Task Leader Yellow \& Pink -- Sample Prep Lab Gold -- Sampler
EPA Sample Number (to be assigned by Prep Lab): ____ _-_

Figure 2. Sample Identification Label


Figure 3. Chain-of-Custody Form.
CHAIN-OF-CUSTODY RECORD

FORM DISTRIBUTION: WHITE - Sample Prep Lab GREEN - Tetra Tech YELLOW \& PINK - Analytical Labs GOLD - Sampler
FTetra Tech, Inc. | Biological Research Facility
reproduced electronically

### 5.0 FREQUENTLY ASKED QUESTIONS

National Fish Tissue Study orientation meetings were conducted in the ten USEPA Regions during August 1999 through June 2000 for Regional, state, and tribal study coordinators and other study participants and contacts. Orientation meeting attendees raised questions related to the study design and field logistics not specifically addressed in the study design document or the field sampling Quality Assurance Project Plan. The following sections provide responses to frequently asked questions. Further explanation on these or any additional issues is available by contacting Leanne Stahl, the USEPA National Study Manager (refer to List of Contacts, page v).

### 5.1 Can a lake be sampled in years other than when it was scheduled to be sampled?

Some flexibility may be possible regarding the sampling of lakes in years other than when they were scheduled to be sampled for the National Fish Tissue Study, particularly in cases where states will be visiting a lake (or lakes) in a different year to collect fish for their own monitoring programs. In some cases, states are on a four- or five-year fish monitoring cycle to sample their lakes, and they may not have sufficient resources to commit to collecting fish for the National Fish Tissue Study at one or more of these lakes in another year. However, since funding for this study is allocated only for fish sampling and analysis scheduled each year, changes to the schedule that result in additional lakes being sampled in a particular year could impact the capacity to analyze fish tissue for that year. States and other participants should contact the USEPA National Study Manager to discuss all proposed deviations from the sampling schedule to confirm that capacity for analyzing fish tissue will not be a problem.

### 5.2 Is the sampling period restricted to only late summer or fall?

Ideally, it would be better to collect all fish for the National Fish Tissue Study during a single or similar index period in an effort to avoid notable differences in spawning condition and active growth phases, which can influence tissue chemical concentrations. Sampling outside the August through November (and possibly December in warmer regions) period defined for the study will introduce more variability in the results. However, there may be cases where states and other study participants can only commit resources to collect fish at other times of the year, or where the late summer through fall period is not a sufficient window of time to collect fish from all lakes scheduled to be sampled. Given these constraints, states and other participants that require flexibility in the sampling period should contact the USEPA National Study Manager to discuss alternate sampling schedules. The goal remains to collect as many fish as possible during the sampling period defined for the study.

### 5.3 If necessary, how will lakes be re-selected for the study?

USEPA needs reconnaissance information from states and other participants for all sites on the original list of 900 lakes to determine if re-selecting lakes is necessary. Re-selection (or adding reserve lakes to replace those that do not meet the target lake definition or are not accessible)
will only be necessary if a large percentage of lakes are rejected within a particular size category of the original target population of 900 lakes. Lakes will be re-selected randomly on a national basis, so a lake eliminated from the original lake list in one state may be replaced by a lake of similar size in another state. A reserve list of 900 lakes is available for this process. New lakes will be added to the study lake list in increments of 20-25 lakes.

### 5.4 Should lakes with no bottom-dwelling species be sampled?

Lakes with predators, but no bottom-dwelling species, should be sampled. For these lakes, field sampling teams should collect the five-fish predator composite, complete a Field Record Form for the composite, and note on the Field Record Form that no bottom-dwelling species were available from the lake.

### 5.5 What if five fish are not available for compositing?

Field sampling teams should make every effort to consistently obtain five fish per composite, since strict adherence to the National Fish Tissue Study field sampling protocol is essential to support proper implementation of the study design. However, there may be cases where a sampling team has expended its full effort and was unable to obtain the full complement of five fish needed for a composite. In these cases, the field sampling team should document the deficiency in the number of fish on the Field Record Form and ship the available fish to the Sample Preparation Laboratory.

### 5.6 What if five fish are available, but they do not meet the 75\% length rule?

Due to the existence of size dependent correlations with body burdens for many target analytes, the $75 \%$ length rule (refer to Section 2.3) should be strictly adhered to in submitting fish for the National Fish Tissue Study. If a sampling team cannot find five fish for a composite that meet the $75 \%$ length rule, the team should retain only the fish that meet this rule, document the deficiency in the number of fish for that composite on the Field Record Form, and ship the fish to the Sample Preparation Laboratory.

### 5.7 What if the five-fish composite does not meet the 560 g weight requirement?

Adult specimens of most of the target species listed for the National Fish Tissue Study should produce at least 560 g of tissue from five fish of harvestable or consumable size. This translates to about 20 oz . of fish tissue, which would be about 4 oz . ( $1 / 4 \mathrm{lb}$.) per fish for bottom dwellers or about 2 oz . per fillet ( 2 fillets per fish) for predators. For the few exceptions where a field sampling team cannot collect fish of at least this size, the team should add another fish (or two) to the composite to provide adequate tissue to meet the 560 g requirement for analysis. Field sampling teams should enter the required field data for each fish and note the need for additional fish on the Field Record Form .

## APPENDIX A

## STATE LISTS OF RANDOMLY SELECTED TARGET LAKES

## Appendix $B$

Standard Operating Procedure:
Fish Tissue Sample Collection Procedures for a
National Study of Chemical Residues in
Lake Fish Tissue

# Fish Tissue Sample Collection Procedures for a National Study of Chemical Residues in Lake Fish Tissue 

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Scope and Applicability: This Standard Operating Procedure (SOP) must be followed by all Field Sample Collection Teams involved with the USEPA Office of Water's National Study of Chemical Residues in Lake Fish Tissue. Adherence to the SOP will ensure that field sampling activities will be performed the same way every time, i.e., are standardized, for all sampling participants.

Fish tissue sample collection procedures are presented below as sequential steps, and include specific equipment, materials, and methods required to perform field sampling activities only.

Responsibility and Personnel Qualifications: This procedure may be used by any Field Sampling Teams that have been authorized by the USEPA Project Manager or the USEPA Regional/State/ Tribal Fish Sampling Coordinators to collect fish for the National Study of Chemical Residues in Lake Fish Tissue.

References: U.S. Environmental Protection Agency (USEPA). 1995. Guidance for Assessing Chemical Contaminant Data for Use in Fish Advisories. Volume 1: Fish Sampling and Analysis. Second Edition. U.S. Environmental Protection Agency, Office of Water, Washington, D.C. EPA 823-R-95-007.
U.S. Environmental Protection Agency (USEPA). 1997. Quality Assurance Project Plan for the Cook Inlet Contaminant Study Sampling. U.S. Environmental Protection Agency, Office of Water, Office of Science and Technology, Washington, D.C.
U.S. Environmental Protection Agency (USEPA). 1997. Sampling Plan for Conducting Field Sampling and Chemical Analysis for the Cook Inlet Contaminant Study. U.S. Environmental Protection Agency, Office of Water, Office of Science and Technology, Washington, D.C.
U.S. Environmental Protection Agency (USEPA). 1999a. EPA Requirements for Quality Assurance Project Plans. U.S. Environmental Protection Agency, Quality Assurance Division, Washington, D.C. Interim Final. EPA QA/R-5.
U.S. Environmental Protection Agency (USEPA). 1999b. National Study of Chemical Residues in Lake Fish Tissue: Study Design. U.S. Environmental Protection Agency, Office of Water, Office of Science and Technology, Washington, D.C.

Precautions: Follow usual safety precautions for working in the field. Boats and/or electrofishing equipment should only be operated by qualified, experienced operators trained in their proper use. Each vessel must be equipped with the appropriate Coast Guard-required safety equipment (including personal floatation devices for each field team member). If electrofishing equipment is used for sample collection, each team member must be insulated from the water, boat, and electrodes via rubber boots and gloves.

## Equipment/Materials:

Sampling vessel (including boat, motor, trailer, oars, gas, and all required safety equipment) ${ }^{(\mathrm{a})}$
Electrofishing equipment - OPTIONAL (including variable voltage pulsator unit, generator, electrodes, wiring cables, dip nets, protective gloves, protective boots, and all necessary safety equipment) ${ }^{(\mathrm{a})}$
Nets - OPTIONAL (including trawls, seines, gill nets, fyke nets, trammel nets, hoop nets, pound nets, trap nets) ${ }^{(a)}$
Angling equipment - OPTIONAL (including fishing rods, reels, line, terminal tackle, trot lines) ${ }^{(\mathrm{a})}$
Coast Guard-approved personal floatation devices
Maps of target lakes and access routes
Global Positioning System (GPS) unit - OPTIONAL ${ }^{\text {(a) }}$
pH meter (including associated calibration supplies) ${ }^{(\mathrm{a})}$
Livewell and/or buckets
Measuring board (millimeter scale)
Ice chests ${ }^{(b)}$
Aluminum foil (solvent-rinsed and baked) ${ }^{(\text {b })}$
Heavy-duty food grade polyethylene tubing ${ }^{(b)}$
Large plastic (composite) bags ${ }^{(b)}$
Knife or scissors
Clean nitrile gloves ${ }^{(b)}$
Field Record Forms ${ }^{(b)}$
Sample Identification Labels ${ }^{(\mathrm{b})}$
Chain-of-Custody Forms ${ }^{(b)}$
Chain-of-Custody Labels ${ }^{(b)}$
Scientific collection permit
Dry Ice ${ }^{(b)}$
Black ballpoint pens and/or waterproof markers
Clipboard
Packing/strapping tape

$$
\text { Overnight courier airbills }{ }^{\text {(b) }}
$$

Plastic cable ties ${ }^{(b)}$
Plastic bubble-wrap ${ }^{(b)}$
First aid kit and emergency telephone numbers
(a) Selection and exact specifications at the discretion of the experienced on-site fisheries biologist.
(b) Provided by the sample control center.

## Procedures:

1. Identify the target lake to be sampled using the USEPA Office of Water's Target Lake List. Locate the target lake via the coordinates provided in the Target Lake List and USGS topographic maps (or equivalent maps).
2. Based on site reconnaissance, determine whether the target lake meets the definition of a suitable lake for sampling for the purposes of this study, i.e., each lake must:

- be a permanent body of water of at least one hectare in surface area,
- have a minimum of $1,000 \mathrm{~m}^{2}$ of open (unvegetated) water,
- be greater than 1 meter deep, and
- have a permanent fish population (e.g., no annual winterkill, not recently stocked with young fish).

If the target lake meets all of the above criteria, and if in the case of private property, the landowner allows access/permission to sample the lake, proceed with Step 3. If the lake does not meet the definition of a lake and/or if a private landowner denies access, record the problem and contact the USEPA Project Manager and/or the Tetra Tech Task Leader.
3. Assemble an array of both active and passive gear types, to ensure the collection of the desired target numbers and species of fish. Selection of the most appropriate gear type(s) for a particular target lake will be at the discretion of the experienced on-site fisheries biologist. Detailed procedures for use or deployment of all possible gear types are not included here. However, if passive collection devices (e.g., gill nets) are used, they must be checked frequently (e.g., several times daily if possible, but at least every 24 hours) to ensure a limited time lag between fish entrapment and sample preparation. Sampling Teams must be qualified, experienced, and/or trained on the safe and effective use of each gear type selected.
4. Sampling gear will be selected and deployed to obtain samples of both predator species and bottom-dwelling species. Suggested target species, listed in order of preference, are as follows:

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|  | Family name | Common name | Scientific name |
| :---: | :---: | :---: | :---: |
|  | Centrarchidae | Largemouth bass | Micropterus salmoides |
|  |  | Smallmouth bass | Micropterus dolomieu |
|  |  | Black crappie | Pomoxis nigromaculatus |
|  |  | White crappie | Pomoxis annularis |
|  | Percidae | Walleye | Stizostedion vitreum |
|  |  | Yellow perch | Perca flavescens |
|  | Percichthyidae | White bass | Morone chrysops |
|  | Esocidae | Northern pike | Esox lucius |
|  | Salmonidae | Lake trout | Salvelinus namaycush |
|  |  | Brown trout | Salmo trutta |
|  |  | Rainbow trout | Oncorhynchus mykiss |
|  |  | Brook trout | Salvelinus fontinalis |
| Bottom-dwelling Species(in order of preference) | Cyprinidae | Common carp | Cyprinus carpio |
|  | Ictaluridae | Channel catfish | Ictalurus punctatus |
|  |  | Blue catfish | Ictalurus furcatus |
|  |  | Brown bullhead | Ameiurus nebulosus |
|  |  | Yellow bullhead | Ameiurus natalis |
|  | Catostomidae | White sucker | Catostomus commersoni |

5. As soon as fish have been obtained via active collection methods (or removed from passive collection devices) they must be identified to species. Clean nitrile gloves must be worn during the sample handling process. Potential target species/individuals will be rinsed in ambient water to remove any foreign material from the external surface and placed in clean holding containers (e.g., livewells, buckets). Nontarget fishes or small specimens are returned to the lake.
6. One predator and one bottom-dwelling species composite will be retained from each target lake. Each composite must consist of five fish of adequate size (i.e., adult specimens that collectively will provide greater than 560 grams of edible tissue for predators and 560 grams of total body tissue for bottom dwellers) for analysis. Select fish for each composite based on the following criteria:

- all are of the same species,
- all satisfy legal requirements of harvestable size (or weight), or at least be of consumable size if no legal harvest requirements are in effect,
- all are of similar size, so that the smallest individual in a composite is no less than $75 \%$ of the total length of the largest individual, and
- all are collected at the same time, i.e., collected as close to the same time as possible, but no more than one week apart (Note: Individual fish may have to be frozen until all fish to be included in the composite are available for delivery to the sample preparation laboratory).

Accurate taxonomic identification is essential in assuring and defining the organisms that have been composited and submitted for analysis. Under no circumstances should individuals from different species be used in a single composite sample.
7. Following selection of five fish for each of the two composites that meet the above-listed criteria for compositing, measure each to determine total body length. Measure total length of each specimen in millimeters, from the anterior-most part of the fish to the tip of the longest caudal finray (when the lobes of the caudal fin are depressed dorsoventrally).
8. Record species retained, specimen length, location collected and sampling date and time on the Field Record Form (Figure 1) in black ink. Complete site location description portions of the form, and draw a simple sketch of the sampling area in the space provided. One Field Record Form will be completed for each composite collected from the target lake.
9. Assign the unique ten-character composite sample ID number to each composite as directed on the Field Record Form (Figure 1):

- state of collection (two-character abbreviation),
- year of collection (two-number abbreviation),
- lake identification number (four-digit code from Appendix A),
- composite type (one character -- $\mathrm{P}=$ predator species; $\mathrm{B}=$ bottom-dwelling species), and
- sample type (one character -- $\mathrm{S}=$ standard sample; $\mathrm{D}=$ duplicate sample).

10. Sign and date the Field Record Form.
11. Remove each fish retained for analysis from the clean holding container(s) (e.g., livewell) using clean nitrile gloves. Dispatch each fish using a clean wooden bat (or equivalent wooden device).
12. Wrap each fish in extra heavy-duty aluminum foil (provided by the sample control center as solvent-rinsed, oven-baked sheets).
13. Prepare a Sample Identification Label (Figure 2) (in black ink) for each sample, ensuring that the label information matches the information recorded on the Field Record Form.
14. Cut a length of food grade tubing (provided by sample control center) that is long enough to
contain each individual fish and to allow extra length on each end to secure with cable ties. Place each foil-wrapped specimen into the appropriate length of tubing. Seal each end of the tubing with a plastic cable tie, and attach the appropriate Sample Identification Label.
15. Double-bag each entire specimen package, that is, place inside a large plastic bag with all specimens of the same species from that site and seal with another cable tie.
16. As soon as each sample is packaged, place it immediately on dry ice for shipment. If samples will be carried back to a laboratory or other facility to be frozen before shipment, wet ice can be used to transport wrapped and bagged fish samples in the coolers to a laboratory or other interim facility.
17. If possible, keep all (five) specimens designated for a particular composite in the same shipping container (ice chest) for transport.
18. Samples may be stored on dry ice for a maximum of 24 hours. Sampling teams have the option, depending on site logistics, of:

- shipping the samples packed on dry ice in sufficient quantities to keep samples frozen for up to 48 hours, via priority overnight delivery service (e.g., Federal Express), so that they arrive at the sample preparation laboratory within less than 24 hours from the time of sample collection, or
- freezing the samples within 24 hours of collection at $\leq-20^{\circ} \mathrm{C}$, and storing the frozen samples until shipment within 1 week of sample collection (frozen samples will subsequently be packed on dry ice and shipped to the sample preparation laboratory via priority overnight delivery service).

19. Complete a Chain-of-Custody Form (Figure 3). All entries must be in black ink and coincide with specimen/sample information on the Sample Identification Labels and Field Record Forms.
20. Retain one copy of the Chain-of-Custody Form and Field Record Form, place and seal all other copies in a waterproof bag, and enclose the sealed forms in the shipping container (ice chest).
21. Pack each shipping container (completely) with dry ice, secure each container with packaging tape, and seal it (e.g., across the latch of the ice chest) with a Chain-of-Custody Label (provided by the sample control center). Include the signature of the sampler and the date/time sealed (in black ink) on each Chain-of-Custody Label.
22. Ship each container to the laboratory via priority overnight express delivery service, as directed by the USEPA Project Manager or Tetra Tech Task Leader. Monitor sample holding time, and factor time required for shipment/delivery to ensure that the preservation and holding criteria described in Step 18 have been met.

Figure 1. Field record for fish samples

## Field Record for National Study of Chemical Residues in Lake Fish Tissue



Address: $\qquad$

| Site Location <br> County: $\qquad$ <br> Latitude: $\qquad$ <br> Site Name: $\qquad$ <br> Site Description: $\qquad$ <br> Circle one: natural lake, modified natural lake, human-made reservoir, other <br> Estimated maximum lake depth $\qquad$ meters <br> $\mathrm{pH} \quad$ surface $\qquad$ mid (optional) $\qquad$ bottom (optional) $\qquad$ |  |
| :---: | :---: |
|  |  |
|  |  |
|  |  |
|  |  |
|  |  |



| Sampling Site Diagram |
| :--- |
|  |
|  |

Figure 2. Sample identification label ${ }^{(\mathrm{a})}$.

|  | Project Name |
| :--- | :--- |
| Site Identification | Date |
|  | Specimen \# |
|  | Composite Sample ID \# |

${ }^{(a)}$ See Appendix D for key to complete Sample Identification Label.

Figure 3. Chain-of-Custody Form.


## APPENDIX C

## SHIPPING CONTAINER PACKING INSTRUCTIONS



1. Laboratory Address Label: Be sure to completely tape over this label with clear tape.
2. Class 9 Dangerous Goods Label: This label will already be completed. Be sure to completely tape over this label with clear tape.
3. Perishable Goods Label: Be sure to completely tape over this label with clear tape.
4. FedEx Airbill: Remember to complete the "Date" section with the date shipped.
5. Strapping Tape: Each end of the cooler needs to be wrapped with strapping tape at least three (3) times.
6. Please make sure the cooler drain hole has been taped so that the drain hole is OPEN. If the tape has been removed please tape the plug so that the drain hole remains OPEN.

NOTE: If you have any questions regarding the packing or shipping of these samples, please contact
Chris Moore or Chris Maynard at (703) 461-2100.

## Special Instructions:

- Not all FedEx locations will accept shipments containing dry ice. Please be sure to call in advance ( 800 -Go-FedEx) to ensure that your FedEx drop-off location accepts dry ice. In the event that you cannot locate a station in your area that accepts dry ice, simply call for a pickup ( 800 -Go-FedEx) and tell them you have a shipment containing dry ice. FedEx will gladly pick these shipments up for you.
- FedEx Dangerous Goods personnel have given approval for shipment of these packages using the instructions listed above. Failure to follow these instructions could result in the package being "burmped" and therefore not reaching its destination. If you have any problems with FedEx personnel accepting your package, please contact Chris Moore or Chris Maynard at (703) 461-2100.
- Please note that the maximum amount of dry ice for this shipment is 25 kg ( 50 lbs ). If suppling your own dry ice, please make sure the weight of the dry ice does not exceed this amount. If you put less than 25 kg ( 50 lbs ) in the cooler please do not change the weight on the airbill or Class 9 label.

