Mercury in Fish Tissue –
A Statewide Assessment of Rivers

Quality Assurance Project Plan (QAPP)
Version 1.2, October 28, 2008

Summer 2008

Prepared by Don A. Essig and Jason Pappani, IDEQ

Acknowledgements

This document is the product of many hands and minds other than the primary authors. The quality assurance project plans for Brownlee Reservoir, written by Hawk Stone, and Lake Lowell, written by Lauri Monnot, provided the template and foundation for this plan. Without their earlier efforts we would have wasted considerable time "reinventing the wheel". Tony Olsen of EPA's Corvallis Lab generated the random selection of rivers sites for this project, and we could not have proceeded without his help. Finally, several people reviewed various earlier versions of this QAPP, including Mary Anne Kosterman, Richard Lee, Amanda Fawley, Michael McIntyre, and Don Bledose. Without their help the imperfections would be more numerous and obvious. Thank you all for your kind help.

This document follows the guidelines and format recommended by *Guidance for Quality Assurance Project Plans* (USEPA 2002a). Much of the text here is taken directly from that publication. It also borrows from EPA's *Guidance for Assessing Chemical Contaminant Data for Use in Fish Advisories* (USEPA 2000), Idaho DEQ's *Implementation Guidance for the Idaho Mercury Water Quality Criteria* (DEQ 2005a), and the Idaho Fish Consumption Advisory Program's *Protocol* (IDHW 2006).

GROUP A: PROJECT MANAGEMENT

A1 – Approval Sheet

Don A. Essig
Project Manager
Idaho Department of Environmental Quality
State Office

Don A. Essig 10-28-08

Michael McIntyre
Surface Water Quality Manager
Idaho Department of Environmental Quality
State Office

Michael McIntyre 10/28/08

Donald Bledsoe
Quality Director
Idaho Department of Environmental Quality
State Administration Office

DCRBIC 28OCT08

A2 - Table of Contents

This plan covers the entire project from design, through implementation, to assessment. These elements are presented in that order and have been arranged into four general groups:

A3. Distribution List

Don Bledsoe, Quality Director, DEQ Administration

Marti Bridges, TMDL Program Manager, DEQ Surface Water Program

Barry Burnell, Water Quality Division Administrator

Amanda Fawley, Brooks Rand LLC

Xin Dai, Statistician, DEQ Technical Services Division

Michael Edmondson, 303(D) and 305(B) Program Manager, DEQ Surface Water Program

Don Essig, Water Quality Standards Coordinator, DEQ Surface Water Program

Jeffery Fromm, Environmental Toxicologist, DEQ Technical Services Division

Richard Lee, DEQ Technical Services Division

Michael McIntyre, DEQ Surface Water Manager, DEQ Surface Water Program

Jason Pappani, Monitoring and Assessment Coordinator, DEQ Surface Water Program

Jim Vannoy, Environmental Education and Assessment Manager, IDHW

Wally Baker, Idaho Bureau of Laboratories, IDHW

Shanda McGraw, EcoAnalysts, Inc.

A4. Project/Task Organization

DEQ's Surface Water Program will oversee the project. The Surface Water Program is responsible for: 1) hiring, training and supervising a DEQ seasonal crew on field procedures including collection of fish, macroinvertebrates, and waters samples and proper labeling, preservation and shipping; 2) coordinating with Brooks Rand LLC on sample shipping and reporting of analytical results; and 3) compilation and final reporting of field and laboratory results.

Michael McIntyre is Manager of DEQ's Surface Water Programs and is responsible for overall direction of this project.

Brooks Rand LLC is the contract laboratory for mercury, arsenic, and selenium analysis of water and fish tissue. They will: 1) process and prepare fish tissue for analysis; 2) perform chemical analysis fish and water samples, including field quality control samples; and 3) report results, including associated laboratory QC summaries, to the DEQ project manager.

The Idaho Bureau of Laboratories will provide sample processing and analysis of surface water samples for nutrients, total suspended solids, specific conductance, suspended sediment, and chlorophyll-a.

EcoAnalysts, Inc is the contract laboratory for macroinvertebrate sample processing, sorting, and identification. They will be responsible for following DEQ's QC protocols for macroinvertebrate identification and reporting results to DEQ.

Don Essig of DEQ is the writer of this plan and overall project manager. He will ensure day-to-day coordination with contract laboratories, DEQ technical services staff working on the project, and will produce the final report on fish contaminant levels and associated water chemistry.

Jason Pappani of DEQ is manager of field operations and will ensure sampling is conducted according to this plan and following established DEQ protocols, and will produce the final report on biological sampling and associated water chemistry.

Richard Lee of DEQ's Technical Services will provide technical support for sample tracking and maintaining records field data, shipping, and chain-of-custody paperwork.

Xin Dai will be the project quality assurance officer and will be responsible for reviewing data against the data quality objectives in this QAPP and reporting her findings to the project manager.

Don Bledsoe is DEQ's quality assurance director, and is responsible for review of this plan. He will contribute a quality assurance summary to the final report.

A5. Problem Definition/Background

Interest in mercury contamination of Idaho fish has been rising since 2003 when DEQ was petitioned to adopt a methylmercury fish tissue criterion. In April 2005 Idaho adopted a fish tissue methylmercury criterion to protect individuals that may eat fish from Idaho surface waters (IDAPA 58.0102.210). This criterion of 0.3 milligrams methylmercury per kilogram (300 ng/g) of fresh weight fish is based on protecting a person weighing 70 kilograms (155lbs) who eats on average of 17.5 grams of fish per day—about one 8-ounce meal every other week over their lifetime.

Methylmercury is a very toxic form of mercury that readily biomagnifies, increasing greatly in concentration in aquatic food chains. This often culminates with mercury in varieties of fish sought for sport in concentrations that pose a human health concern. Presently there are eight lakes and reservoirs and two streams across the state of Idaho with fish consumption advisories for mercury – advice to the public, typically young children and pregnant women, to limit their number of meals of caught fish so as to protect their health. There have also been two Total Maximum Daily Loads (TMDLs) prepared in Idaho addressing mercury contamination – Jordan Creek and Salmon Falls Reservoir.

In 2006 DEQ undertook probabilistic sampling of rivers across the state. That effort resulted in biological, habitat and water quality data from 25 sites, and fish tissue data from15 sites. The work completed in 2006 was the first step in a two-phase sample design with the goal of

providing a statewide assessment of Idaho's major rivers, with the second phase to be completed in 2008 (see Appendix A).

In 2007 DEQ conducted probabilistic sampling of lakes and reservoirs over 50 acres in size across Idaho, obtaining fish tissue contaminant information from 50 lakes and 89 fish tissue composite samples (Essig and Kosterman 2008).

A6. Project Purpose/Task Description

The present project plan is to complete the second phase of the Idaho Major River Survey sample design (see Appendix A), This will require obtaining biological, habitat, water chemistry, and fish tissue data from 25 randomly selected sites, and only fish tissue and water from 10 additional sites (35 total fish tissue and water collections to complement the 2006 effort), giving us a total of 50 probabilistic sites from which to base statistical estimates of the condition of Idaho's major rivers, and the proportion of Idaho's major rivers that meet or exceed certain criteria

Biological, water chemistry, and habitat data will provide DEQ with the necessary data for assessing the ecological condition of Idaho's major rivers (appendix A). Using the probabilistic survey design, DEQ will be able to estimate statistically the condition of Idaho's major rivers. In addition, fish tissue data will allow DEQ to make a statement about the percentage of rivers in Idaho with methylmercury concentrations in the flesh of commonly fished species greater than Idaho's methylmercury fish tissue criterion.

Although this criterion is for methylmercury fish tissue samples will be analyzed for total mercury since it has been established that the majority of total mercury in fish tissue is in the form of methylmercury (90% or more, EPA 2001a; Larosa and Allen Gil 1995); thus it is conservative to assume that all mercury in fish tissue is methylmercury. Therefore, results for total mercury concentrations will be used for comparison to Idaho's methylmercury criterion. This will provide an overall picture of risk to the fishing public from mercury contamination in Idaho's rivers. It will not however provide site-specific information about all rivers, or risks due to consumption of species not sampled.

Because much effort is involved in obtaining fish, in addition to mercury fish tissue samples will also be analyzed for total selenium, total arsenic, and inorganic arsenic. This will provide a more robust assessment of human health risks from fish tissue consumption. We will also collect water samples for analysis of arsenic (total & inorganic), total mercury, and selenium. This will help answer question about bioaccumulation of these contaminants. In addition the total mercury water data along with fish tissue data will add to the growing set of data testing Idaho's assertion that its fish tissue criterion is more protective of aquatic life (requires lower ambient mercury concentrations) than EPA's 1994 total mercury criterion for aquatic life protection.

Field operations will begin in late June 2008 and conclude by October 2008. Laboratory analysis will occur concurrently, but with a two week to one month delay before results are available. We will attempt to collect two game species from each waterbody but expect some waters to support

only one species in sufficient numbers and size to be harvestable. Fish tissue will be sub-sampled using plugs from one fillet from each fish, composited by species for analysis. Thus, aside from field duplicates, there will be one result for each analyte per species per water body. Water samples will be grab samples from a well mixed (turbulent) portion of the stream flow. Clean hands / dirty hands procedures will be used for collection of water samples to be analyzed for total mercury.

A final report summarizing field activities and results will be completed by March 2009.

A7. Quality Objectives and Criteria

DEQ believes that consistency between monitoring plans is important, therefore quality objectives closely follow the methods and criteria used in the 2007 Mercury in Fish Tissue – A Statewide Assessment of Lakes and Reservoirs QAPP (Essig and Kosterman 2007), and the Beneficial Use Reconnaissance Program Field Manual for Rivers (DEQ 2006), the Beneficial Use Reconnaissance Program Field Manual for Streams (DEQ 2007) and the 2005 Quality Assurance Project Plan: Beneficial Use Reconnaissance Program (DEQ 2005b).

The following sections describe particular goals for data quality.

A7.1 Precision/Duplicate Samples

Precision refers to the measure of agreement among repeated measurements of the same sample under similar or identical conditions. It gives information about the reproducibility of results and is determined by the generation and analysis of duplicate samples. Precision is expressed as the Relative Percent Difference (RPD) between duplicate samples or analyses and will be calculated using the following equation:

$$RPD = \frac{2\,|(C_s - C_d)|}{C_s + C_d} \times 100\%$$

Where:
C_s = the sample result, and
C_d = the duplicate sample result

There will be three kinds of duplicate used in this study: field, composite, and analytical. Composite duplicates apply only to fish samples and so for water samples there will be only field and analytical duplicates. A field duplicate is collection of a second sample from the same location at the same time. For fish this means the same site (reach of river) on the same day. Fish tissue composite duplicates consist of a second set of subsamples from a set of fillets, ground into a composite puree. Analytical duplicates are a repeated analysis of the same water or fish tissue composite by the laboratory. For tissue samples this involves a duplicate digestion. Each duplicate type provides information on reproducibility of results at different stages in the sampling, processing and analysis sequence.

Field duplicates will be collected at the rate of at least ten percent but not less than once a week for water samples. Fish duplicates will be driven by the availability of fish and the ten percent rate may not be attained. Fish tissue composite duplicates will be created at the rate of ten percent of the number of field sample delivered to the laboratory for analysis. Finally, the laboratory will analyze in duplicate ten percent of the samples they run. This may include samples from other projects as is appropriate to batching of samples for analysis.

Variability in results can increase at each step in handling of samples and is cumulative in the chain from analysis to sample collection. Our precision goals reflect this. The laboratory precision objective is an RPD no more than 30% between duplicate analyses for fish tissue and 25% for water samples. Duplicate laboratory analyses exceeding this objective will trigger an assessment of quality control and re-analysis of the samples in question. The composite precision objective is an RPD no more than 40%. If this goal is not met sample results will be flagged and considered for re-compositing from archived samples. Field duplicate precision will be reported as information on data quality to be considered in interpreting results. Table 1 summarizes these objectives for water and fish tissue.

Table 1: Summary of Precision Data Quality Objectives for Chemical Analysis of Water and Fish Tissue

Duplicate Type	Sample Matrix	Precision Goal
Analytical	Water	RPD <25%
	Fish tissue	RPD <30%
Processing	Water	not applicable
	Fish tissue	RPD <40%
Field	Water & Fish tissue	report as information

Precision of macroinvertebrate and habitat field data is achieved through extensive crew training and oversight, and through strict adherence to established DEQ protocols (see DEQ 2006 and DEQ 2007). There will be no field duplicates for macroinvertebrates or habitat.

Taxonomic precision for macroinvertebrate identification is provided by the contract laboratory. These measures include verification of sub-sampling and sorting precision. The contractor will perform QA/QC on the subsampling of at least 10% of all samples. Samples are to be combined in the Caton tray and the appropriate number of grid squares selected at random in order to obtain a minimum of 500 individuals for identification. Following this, another qualified employee of the contractor must examine all the material from the selected squares and check for invertebrates that were missed. At least 95% of all the invertebrates in the selected squares must have been removed for identification. If less than 95% of the sample has been picked, the sample and all the material from the grid squares not selected must be placed back into the Caton tray and redistributed for a new random subsample to be taken. The new subsample must be

rechecked before identifications can occur on the macroinvertebrates that were selected.

A7.2 Accuracy

For chemical measurements, accuracy is measured by analyzing materials of known concentration and tells us how true a result an analytical method gives. The ratio of the measured concentration to the actual or true value is expressed as percent recovery (measured/true x 100 = % Rcv). Recovery can be less than 100% (low bias) or greater than 100% (high bias). With samples, like fish tissue, that involve digestion in preparation for analysis, accuracy is determined by sample preparation as well as the analytical technique.

Accuracy of water sample analysis is usually determined from analysis of spiked samples, where a known quantity of analyte is added to an actual field sample. This is known as a matrix spike. For matrices other than water accuracy is usually determined from the analysis of standard or certified reference materials (SRM or CRM). Reference materials are samples of a matrix (e.g., animal tissue) similar to that being analyzed and of a known or, through round-robin analysis, agreed upon true concentration. A CRM is available for fish tissue (DORM-2 dogfish muscle), for total arsenic (As), total selenium (Se), and total mercury (Hg). No CRM value is available for inorganic arsenic; matrix spikes will be used.

While a CRM provides a check on loss of analyte in laboratory sample preparation and digestion of samples, often a critical step in overall analyte recovery, it cannot account loss or gain in analyte that may occur elsewhere in the sample handling chain. For this reason, overall method accuracy can not be measured in this study.

The laboratory will employ CRM digests to assess recovery of analytes in the laboratory. Recovery goals vary by analyte and matrix (Table 2). For samples batches in which recovery objectives are not met, the laboratory will contact the project manger promptly and discuss whether the results can be flagged and accepted or the samples rerun (re-digested and/or re-analyzed).

Table 2: Summary of Accuracy Data Quality Objectives

Matrix	Analyte	CRM/Spike	% Recovery
Fish Tissue	Total Mercury	CRM	75 to 125%
	Total Arsenic	CRM	75 to 125%
	Inorganic Arsenic	Spike	75 to 125%
	Total Selenium	CRM	70 to 130%
Water	Total Mercury	Spike	75-125%
	Total Arsenic	Spike	75-125%
	Inorganic Arsenic	Spike	65-135%
	Total Selenium	Spike	75-125%

The inadvertent addition of analyte to a sample through handling is known as contamination and causes a high bias in the samples. Contamination may come from sample contact with collection equipment, containers, exposure to the atmosphere, e.g. dust, fumes, even mercury vapor in the breath of the person conducting the sampling. Ease and degree of contamination depends on how little analyte is already present in the sample. Mercury is extremely low in most water samples. Contamination of fish tissue is difficult, but also hard to ascertain. Care in handling to avoid contamination of all samples is prudent.

Blanks will be used to check on the possible contamination (analyte gain) in sample collection and processing. For water a blank is a sample of deionized water carried to the field and handled as an ambient sample. For tissue samples a blank is a sample of deionized water processed as a fish tissue sample after the processing equipment has been cleaned. All blanks are of a water matrix.

Table 3: Summary of Blank Contamination Data Quality Objectives

Matrix	Analyte/Method	Blank Type	Acceptable Level[a]
Water	Mercury	Processing (fish) Field (water)	< 200 ng/L (< 0.2 ng/g) < 5 ng/L
	Total Arsenic	Processing (fish) Field (water)	< 200 μg/L (< 0.2 μg/g) < 0.3 μg/L
	Inorganic Arsenic	Processing (fish) Field (water)	< 10 μg/L (< 0.01 μg/g) < 0.05 μg/L
	Selenium	Processing (fish) Field (water)	< 100 μg/L (< 0.1 μg/g) < 1.0 μg/L

[a] Values in () expressed as equivalent tissue concentrations. Note change in units.

Accuracy of macroinvertebrate data is achieved by ensuring accurate identification of macroinvertebrates. The contractor will perform QA/QC on the identification of at least 10% of all samples. Once a taxonomist has completed the identification and enumeration of all the macroinvertebrates in a subsample, the subsample must be repackaged, and then another qualified taxonomist employed by the contractor will re-identify and re-enumerate the subsample independently of the first taxonomist. Once this has been completed for a site, the contractor must perform a percent similarity calculation. The percent similarity must be 95% or greater. Before further samples are processed, the taxonomists must confer to reconcile any discrepancies. For any specimens that are unknown or in question, the results will be reported at the next higher taxonomic level for that group, and the specimen will be sent to an expert in that taxonomic group for identification at the expense of the contractor.

A7.3 Data Representativeness

Representativeness expresses how accurately the sample results represent a characteristic of the population. It is best achieved by careful selection of sampling locations, following sample collection procedures, and obtaining a sufficient number of samples. Thorough documentation of sample site selection will allow an assessment of representativeness after field operations have ended.

Water samples will be collected from a well mixed portion of the river flow (e.g. riffle) in or near the thalweg.

For fish, DEQ's implementation guidance and EPA protocol prescribe that a minimum of 10 fish from the highest trophic level should be sampled per water body. We will aim to meet this minimum but may be unable to due to scarcity of fish. A sample of fish for analysis will consist of a composite of up to ten fish of a species from one site. Use of composite tissue samples averages out fish to fish variation in contaminant levels and provides an estimate of the exposure likely to result from consumption of a particular fish species caught from that site over time.

A7.4 Data Comparability

Comparability is a measure of the confidence with which one data set can be compared to another.

Water samples will be preserved and analyzed using standard methods. Clean hands / dirty hands procedures will be implemented for collection of samples to be analyzed for their total mercury content.

Species and age (size) of fish are known to greatly affect mercury bioaccumulation. Therefore the target species and size of fish will be restricted to reduce this variability, see section B1.3. Furthermore, lengths and weights of each fish will be recorded.

Fish tissue and water samples will be analyzed using EPA standard methodology. All practical safeguards will be implemented to avoid mercury contamination during sample collection and processing. These precautions are detailed in sections B.2 and B.3.

A7.5 Data Completeness

Completeness is the difference between the quantity of data obtained and the quantity expected. With careful adherence to the project plan, it is expected that all data collected will be usable. However, due to unforeseen circumstances some results may be lost due to equipment failure, environmental conditions or logistical constraints.

For this study a complete data set is initially defined as 100% of the target number of sites sampled, 100 % of the sites sampled for water chemistry and 75% of the target number of fish samples. The latter allows for the possible rejection of individual samples in tissue processing (see B2.4), and the expectation that we will not be able collect the planned two species of fish from all sites.

Since laboratory analysis requires only a small fraction of the tissue collected, the remaining tissue homogenate will be archived one year in the event repeat analysis is needed. In addition, only one fillet per fish will be homogenized. The second fillet from each fish will also be archived for one year. With these safeguards we expect to eventually get useable analytical results for all fish samples collected.

With the randomized sampling and summer-long sampling season, sampling will continue until 35 rivers (25 for biomonitoring & fish, plus 10 more for fish tissue only) are monitored. To reach the data completeness objective of 75 % of the target number of samples for fish, we need to collect 53 fish samples (ten fish each). This works out to two species per site from 18 rivers and one from the other 17 sites.

If the analytical data completeness objective is not met, the project manager and project personnel will confer to consider whether repeat analysis must occur or the data quality objective for completeness can be relaxed. Any deviations from protocol will be carefully documented to enable the project manager to decide whether data will be discarded. All deviations from the plan and procedures will be noted in field notebooks, sample collection field sheets, processing logs, or laboratory logs as appropriate. Each note of deviation will be initialed and dated by the person making the entry. In addition the QAO will be notified and will address the consequence of these deviations in their final QA/QC project summary.

A8. Special Training/Certification

At least one person on the fish collection crew shall receive instruction on fish handling and identification (Section B2.2). The individual in charge of fish handling samples should be familiar with fish filleting and will take precautions such as cleaning the filleting surface and tools between species. Similarly, at least two persons shall be trained in clean hands / dirty hands procedure for water sample collection. All crew members will be trained in proper execution of DEQ's field methods (DEQ 2006, DEQ 2007).

All field participants shall be familiar with boating safety, and will have attended training seminars and field exercises. Electro-fishing from a boat is a hazardous activity, and all

participants shall be fully briefed on proper procedure. Additional safety and operation training will be provided, should funds be available.

Records of training certificates and professional qualifications will be examined prior to assignment of project tasks. Copies of training records shall be retained with other project records generated as a result of implementation of this QAPP.

A9. QAPP Revision, Documents and Records

This QAPP may be revised upon approval of the project management team identified in the Approval Sheet (section A1). Revisions may be made to improve or address QA/QC problems that arise over the course of the study or otherwise improve or further project objectives based on knowledge gain during project execution.

The most current version of the QAPP will be distributed to project personnel as soon as it is available. Before any action is taken under this plan, it will be confirmed that all personnel have read the plan. Where possible, this document will be distributed electronically. New versions will replace prior versions.

All paperwork created during this project will be collated into a 'project file'. This paperwork could include:

- Completed field forms (see Appendix A),
- Sample processing logs (see Appendix C),
- Field notebook with all deviations from protocol and other pertinent information noted,
- Calibration logs for any equipment used, and
- Site photographs (electronic photos will be included on compact disc).

A final report will be prepared by Don A. Essig and made available to all on the distribution list. It will summarize the field activities, provide results, and evaluate the overall success of monitoring. The report will be available by March 2009.

The laboratory will report results to Don A. Essig, in electronic format. This will include both a PDF of laboratory data reports, and an Excel spreadsheet summarizing analytical results.

Compact discs will be used to store all electronic information associated with this project. The project file will be kept at DEQ's State Office for at least five (5) years.

GROUP B: DATA GENERATION AND ACQUISITION

The elements in this group address all aspects of project design and implementation. Implementation of these elements ensure that appropriate methods for sampling, measurement and analysis, data collection or generation, data handling, and QC activities are employed and are properly documented.

B1. Sampling Process Design (Experimental Design)

This element describes the project's data collection or research experimental design.

B1.1 Sampling Locations

A random probability design is employed in this study (See Appendix A). The chosen target population is Idaho's Major Rivers, as defined by DEQ (see Appendix A). Tony Olsen of EPA's Corvallis Laboratory provided a draw of 50 waters from this sampling frame as the primary set of waters to be sampled, with 25 to be sampled in 2006, and an additional 25 to be sampled in 2008.

We have already screened this list in the office and know that not all the primary waters are suitable for sampling due to being impounded or inaccessible. Replacement sites were taken from an 'over-sample' of 200%, or 100 additional rivers randomly drawn with the primary sites (See Figure 1). As primary sites are eliminated as unsuitable, replacement sites from the overdraw list are taken in the order given so as to maintain a statistically valid random sample.

A river sample reach is defined as 40 times the general wetted width with a minimum reach length of 500 m and maximum reach length of 1000 m. The site coordinates are located in the middle of the sample reach and this point is known as the "x-site". The sample reach is comprised of 6 equidistant cross-channel transects for habitat and biological sampling. Electrofishing will occur throughout the sample reach.

In the event electrofishing the reach does not yield ten fish per species (up to 2 species) electrofishing will continue downstream until the takeout or 10 fish per species. Fishing beyond the bottom of the reach is only for purpose of obtaining fish for tissue analysis and not for fish community description. The location and time at which electrofishing is ended will be recorded. See the Beneficial Use Reconnaissance Program Field Manual for Rivers for detailed description of field methods (DEQ 2006)

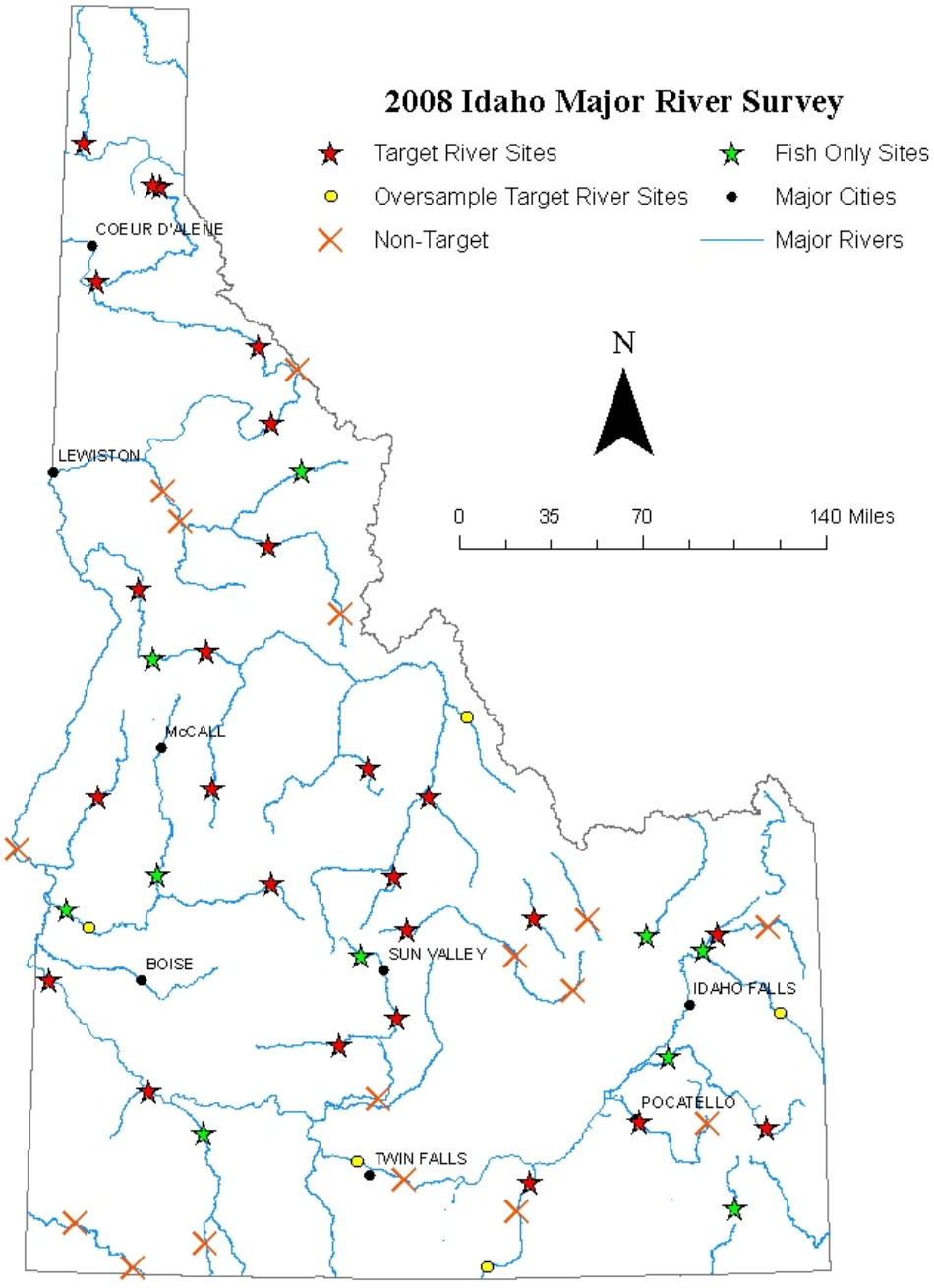
The name of the water body and exact location (latitude and longitude) are provided with the site coordinates, also known as the "x-site".. Because capture of fish will involve moving around the water body, GPS coordinates will be obtained at the beginning and end of the reach fished. Water samples will be obtained at the end of the reach to minimize time between collection and shipment.

B1.2 Sampling Times

The time of day sampling of water and biological communities occurs is not critical but will be recorded. Likewise, although fishing success may vary throughout the day, the exact time of collection is not critical to this study.

Overall sampling is planned for July through October of 2008. Because of Idaho's snowmelt dominated hydrographs and semiarid climate water levels in rivers can vary greatly from spring through summer. We will not begin sampling until flows have subsided enough that conditions are safe for floating and waters have cleared of typical spring turbidity. Biological monitoring protocols dictate that base flow is the best time to sample macroinvertebrate populations. Availability of seasonal help also constrains us to summer sampling. EPA's Guidance for Assessing Chemical Contaminant Data for Use in Fish Advisories, Volume 1 (Section 6.1.1.5, EPA 2000) recommends that the most desirable sampling time is from late summer to early fall. To minimize the limitations to sampling water level changes may present, sampling in this study will like start early in summer in drier southern portions of Idaho and progress north and into higher elevations.

Figure 1: Idaho Major River Survey sites for the 2008 field season, including oversample and non-target sites.



B1.3 Target Fish Species and Size Class

EPA (2000) recommends that when choosing the target species, the primary selection criteria should be that the fish is commonly consumed locally and bioaccumulates high concentrations of mercury. Additionally, the species should be abundant and easy to capture and identify.

The target species for this study in order of preference are: **rainbow trout, brown trout, smallmouth bass, mountain whitefish, catfish, and largescale sucker.** We would like two species from each site and need two species from at least 18 sites to meet our target of 53 fish samples. If preferred species are absent other species will be caught at the crew's discretion. Game fish are preferred.

Size of fish collected for analysis will vary based on species but all fish collected should be of legally kept size. Idaho Fish Consumption Advisory Program (IFCAP protocol, IFCAP 2004) specifies that individual fish must be a minimum of 10 inches in length, since larger fish generally bioaccumulate the most methylmercury. It is known the larger (older) individuals within a population are generally the most contaminated (EPA 1995). So to avoid the variance in mercury levels due to fish size, the largest fish of a species at a site should be no more than 150 percent of the length of the smallest individual for the species at that site. So if the smallest fish is 10 inches, the largest should be no more than 15 inches long. The length and weight of each fish caught will be measured and recorded.

B1.4 Target Analytes

For this study the primary analyte of concern in fish and water is mercury. Although the fish tissue water quality criterion is expressed in terms of mg of methylmercury per Kg of fresh weight tissue, analysis will be of total mercury. This is justified because 1) it is has been established that the vast majority of the total mercury in fish tissue is in the form of methylmercury (90% or more, EPA 2001a; Larosa and Allen-Gil 1995); 2) analysis of total mercury is easier and less costly than analysis of methylmercury; and 3) assuming the methylmercury concentration is the same as total mercury concentration thus provides a conservative bias for comparison to the criterion.

When composite samples are analyzed, most of the cost in fish tissue monitoring is in the obtaining of the fish tissue, rather than analytical costs. Adding additional analytes greatly enhances the information gained from this effort for relatively minor added cost, and with no further sacrifice of fish. Secondary analytes are total selenium, total arsenic, and inorganic arsenic.

Selenium is a known issue in southeastern Idaho's phosphate mining patch, but selenium release is also known to be associated with coal combustion and, like mercury, some kinds of metal smelting (Lemly 2002). Selenium is also used as a nutritional supplement for livestock and has been associated with feedlot runoff. Therefore investigation on a statewide basis is prudent. Arsenic is of interest because Idaho has an outdated human health criterion and efforts to update it in 2005 failed in part because of lack of information about arsenic bioaccumulation specific to species inhabiting Idaho waters. Part of the issue is the form of arsenic (inorganic or organic)

that bioaccumulates. Data on arsenic levels in fish tissue and water from this study should be useful to informing bioaccumulation rates pertinent to Idaho and application of or revision of current arsenic criteria in addition to providing a statewide picture of the extent of arsenic contamination in fish from Idaho's rivers.

B1.5 Sample Type

Water samples will be surface grabs from well mixed flow. Because of the multiple analytes and different sample container materials and preservatives each water sample will be split into three bottles.

For fish this study will define 'fresh weight fish' as the skinless, boneless fillet, which is the portion most likely to be consumed by anglers.

Most consumers in the general angling population do not eat the skin of the fish, justifying its removal for analysis. In addition methylmercury is concentrated in muscle tissue, therefore analysis of skinless fillets provide a more protective result than analysis of whole fish or fillets with skin attached. To maintain consistency, simplify sampling, and because the focus is human health and possible fish consumption advisories, selenium and arsenic analysis will use the same samples as total mercury.

Boneless skin on fillets will be prepared from each fish in the field. One fillet from each fish will be sent to the laboratory for analysis. The other fillet will be sent to the DEQ state office for archiving (Attn: **Don Essig, DEQ, 1410 N. Hilton, Boise ID, 83706 ph: 208-373-0119**). The laboratory will remove the flesh or portion of flesh from the skin for compositing. Leaving the skin on until preparation for analysis minimizes handling and thus contamination in the field.

In the field care will be taken to avoid exposure of fish to exhaust fumes and dust and contact with metal surfaces once filleting begins. Polyethylene cutting boards or other portable surface will be provided to each crew. In addition the same type of knife will be supplied to each crew for use in filleting only.

B1.6 Number of Fish per Sample

IFCAP protocol and DEQ Implementation Guidance for the Idaho Mercury Water Quality Criteria (Idaho DEQ 2005a) recommends a minimum of 10 fish from each species at each site. This number provides an adequate sample to provide statistical significance and strikes a balance between a high level of precision, good representation, and analytical costs. However, if ten individuals of the same species can not be obtained with reasonable fishing effort (1-2 hours), composites based on a smaller number of individual fish will be used.

Individual fish in a sample must all be of the same species and from the same waterbody, should be of similar size, and should all be collected within a 24-hour period.

B1.7 Fish Sample Compositing

For this project subsampling of fillets for compositing will occur at the contract laboratory.

Subsamples (nominally 10 grams) from one fillet from each individual fish (up to 10) for a species at a site will be ground together to form one composite sample for that species / site. Composite samples are a cost-effective method for estimating average tissue concentrations of analytes in target species populations to assess chronic human health risks (EPA 2000). To have a legitimate composite sample the fillets subsampled must be from different fish and this is why each fish (two fillets) is individually numbered.

This procedure is different from that which the USGS uses for the monitoring they conduct under the Statewide Trend Monitoring Coop under a joint funding agreement with DEQ. The USGS subsamples the fish in the field, cutting out an approximately 1 inch chunk of muscle from the side of the fish, removing the skin from this chunk while still attached to the fish, and placing the chunk in a plastic baggie with similar chunks of skinless flesh from other fish that make up a composite sample. This is all done with gloved hands, and a new scalpel for each fish. The USGS method lessens handling and thus opportunity for contamination. The degree of subsampling is the same under the USGS procedure as the procedures described herein, so we feel they are comparable.

A limitation of using composite samples is that information on extreme levels of chemical contamination in individual fish is lost. Individual fish data also allows calculation of statistical confidence limits to be placed around mean values. In order to preserve the opportunity for individual fish analysis at a later date should funds permit, the spare fillets not used in composite sample preparation will be saved and kept in frozen archive.

Sample composites will be prepared as follows:

1. Fillets should come from the field double bagged. Each individual fillet in its own Ziploc bag indentified by Sample ID (see section B2.3), with a set of fillets all from one species together in a second outer bag. Nominally there should be ten fillets, but some samples may consist of less than ten fillets.
2. The fillets will be inspected for integrity and allowed to thaw before processing. Compromised samples (e.g. broken Ziploc bags, unlabelled samples) will be discarded. Experience has shown that partially thawed fillets, with a few remaining ice crystals are easiest to work with. Fillets may be allowed to thaw for up 16 hours before processing, so long as spoilage is avoided.
3. A new disposable scalpel will be used for preparing each composite sample (set of ten fillets from one species / site). Used scalpels will be disposed of in a medical sharps container.
4. Each fillet will be rinsed with de-ionized water before proceeding with subsampling (next step).
5. Then a ~10 gram plug (subsample) is taken from the meatiest (thickest) section of the fillet using a clean scalpel. The plug is weighed on a tarred piece of aluminum foil. The weight should be recorded in the processing log to the nearest gram if not ~10 grams.
6. This plug will be placed in a stainless steel and glass grinder along with the other fish flesh plugs for that species and site. Repeat steps 4-6 until all ten fillets have been subsampled

7. The ten plugs will then be ground until blended into a consistent paste. Typically this will take at least 120 seconds of grinding.
8. Approximately 100g (½ cup) of blended flesh will result. A sterile scoop will be used to transfer the blended flesh to a mercury-free sample container.
9. Composite samples will be indentified by Site # + Species Code, and date processed. Field duplicates (Fish #'s 11-20 for a sample) will be identified by appending FD to the composite sample ID, and processing duplicates by appending a P suffix. A laboratory ID number may also be assigned.
10. Composite samples should be refrozen if not to be digested the same day.

Duplicate processing composites will be prepared identically, from a second set of ten gram plugs from the same set of fillets as the original sample. The remainder of the unused fillets will be discarded.

Between each sample, the blender will be cleaned with hot water and detergent, sterilized in 0.1% hydrochloric acid, and triple rinsed with de-ionized water. A new disposable scalpel and piece of aluminum foil for weighing will be used for each sample (set of up to ten fillets from one species and site). The scoop used for transferring the homogenate to its storage container may be reused with cleaning between composites.

A sample processing log will be maintained to record the time and date each set of fillets are taken from the freezer, subsample weights, and the time and date the composite is completed and returned to the freezer. On this log will also be recorded any discrepancies in field samples (samples not double bagged, or more than one species or site per cooler, apparently missing specimens, e.g. gap in numbering). The project manager will be notified of these discrepancies.

Composite tissue sample not used by the laboratory for analysis will be shipped back to DEQ within 30 days, or once no longer needed by the laboratory. These samples will be retained by DEQ for at least one year from time of sample collection.

B1.8 Sampling Quality Control

Field blanks will be generated for water samples. There are no field blanks for fish. We will test the possibility of contamination that the fish tissue compositing procedure may introduce through the use of processing blanks generated at the laboratory when the compositing takes place.

Field duplicates will be used for both water and fish. See section B5 Quality Control for details.

B2. Fish Sampling Methods

This section briefly discusses the three main methods that will be used to collect fish. A general discussion on sampling procedures then follows, and is applicable to all collection methods.

B2.1 Collecting Fish

A raft-mounted electrofisher will be generally used to collect fish. A backpack electrofisher may be used in smaller streams or near the shoreline. This will be operated by trained DEQ personnel. Electrofishing is the preferred method of capture, as it involves minimal handling of fish. However it is not effective in deep water, or for larger fish. Hook and line sampling may be used to augment electrofishing, or in the event electrofishing is not possible or effective.

Upon capture fish will be identified for eligibility to be kept as part of the sample. For this study, 'eligible' means fish of a target species and appropriate length. The length is defined as the distance from the anterior-most part of the fish (lips) to the tip of the longest caudal fin ray.

Additional eligibility guidelines:

- Dead specimens other than those killed in the process of collection will be discarded.
- Specimens with lacerations will be discarded.
- Specimens with sores or lesions will be discarded.

It is desired to avoid hatchery planted fish. This can usually be discerned in the field by fin abrasion that results from early life in a concrete runway. If fish are abundant obvious hatchery fish should be discarded. If fish are not abundant, hatchery fish should be kept but noted on the field form (Appendix B). The only species for which hatchery fish may be found are rainbow trout. It is highly recommended that the local fish and game office be contacted as to recent fish stocking and species likely to be encountered at each site.

Retained fish will be kept in a live well until fishing is done at a site. Filleting of fish will take place on-shore at the end of fish collection for the site. Each fish will be weighed (grams) and length measured (cm). This information will be recorded on the field form (Appendix B). Copies of these forms should be made and originals sent to DEQ (see section B2.4). Length of time spent fishing and general weather and water conditions should also be recorded. Weighing and measuring of each fish may be done either as fish are caught (desirable from standpoint of limiting size range) or on-shore before filleting. All sample containers will be protected in an ice chest that will be kept closed.

B2.2 Handling Fish and Labeling Samples

Clean Hands/ Dirty Hands techniques (EPA method 1669) are required in this study for collection of water samples for mercury analysis. These procedures are not necessary for collection of fish. Mercury levels in fish tissue are thousands of times higher than in water and thus the samples are much less subject to contamination, therefore allowing a less stringent sampling protocol. It is desirable that one person is dedicated to filleting fish. Other elements of EPA method 1669 to be used are:

- Fish will be rinsed with ambient water immediately prior to filleting to remove any mud. It is recommended to then wipe each fish with a rag to remove slime and ease handling. The cutting board should also be rinsed and wiped clean.
- In all cases, the person handling fish will avoid touching the sample flesh with bare hands. The crew member will be dedicated to filleting and will wear nitrile gloves while filleting the fish. There will be no contact of bare hands with the fillet. This

might take two people; one to pick up and clean the outside of the fish, and another to only touch the fish while it is filleted.
- Gloves will be discarded if they contact any environmental surface, especially metal surfaces, such as the raft frame.
- Each fillet will be placed in its own plastic zip-lock bag. It is also desired to combine bagged fillets of the same species from each site into one larger bag (e.g. kitchen garbage bag) or cooler.
- Between species and at the end of each day the fillet knife and cutting board will be cleaned. The cutting board should be scrubbed with a brush and washed down with a dilute soap solution, then rinsed—preferably with de-ionized water, but clear fresh stream water is acceptable. The fillet knife should be similarly cleaned, and also after any time that it is sharpened. Equipment should be stored dry.

Fish should be filleted as quickly as possible after removal from the live well. Each fillet should be carefully placed into a Ziploc bag. The full sample ID and date MUST be written in permanent marker on the outside of each bag with a waterproof marker. Pre-labeling of bags is recommended to expedite this process and usually results in more legible information. It is strongly recommended that one person hold the bag open, taking care not to touch the inside with ungloved hands, while the filleter with their gloved hands places the fillet in the bag. Bagged fillets will be promptly put in a cooler on ice. Samples should be frozen or placed on dry ice within 24 hours. Frozen samples may be held for up to a week for shipping. A daily record should be kept documenting that fish samples remain frozen.

Each site will have two designated fish coolers—one for fish to be sent to the lab, one for fish to be retained for archive purposes. It is desirable that fish from different sites not be packaged in the same cooler, but this is acceptable if all the fillets from each sample (ten fish per species at a site) are kept together in separate larger bags. A third cooler will be needed for water samples. Water samples must be kept cold but not frozen, i.e. on wet ice. The fillets must be kept on ice or frozen until processing for analysis. If fillets will be held more than twenty-four hours before shipping they should be frozen. Dry ice is needed for holding and shipping fish fillets.

All sample coolers will be brought back to the DEQ state office for handling and shipping, see section B2.4.

B2.3 Sample Identification Numbers

Each bagged fillet will be identified with a Sample ID number that consists of a Site # + Species Code + Fish #. Site #'s take the form of a 3-digit number (001, 024, 078, etc.) that identifies the waterbody from the site list in Appendix A. Species codes are 3-digit codes as follows:

Species code	Common name	Scientific name
008	kokanee	*Oncorhynchus nerka*
009	chinook salmon	*Oncorhynchus tshawytscha*
010	**rainbow trout**	***Oncorhynchus mykiss***
011	cutthroat trout	*Oncorhynchus clarki*

016	**mountain whitefish**	***Prosopium williamsoni***
019	**brown trout**	***Salmo trutta***
021	brook trout	*Salvelinus fontinalis*
022	bull trout	*Salvelinus confluentus*
024	Arctic grayling	*Thymallus arcticus*
027	chiselmouth	*Acrocheilus alutaceus*
030	common carp	*Cyprinus carpio*
042	Utah sucker	*Catostomus ardens*
043	longnose sucker	*Catostomus catostomus*
044	bridgelip sucker	*Catostomus columbianus*
045	bluehead sucker	*Catostomus discobolus*
046	**largescale sucker**	***Catostomus macrocheilus***
047	mountain sucker	*Catostomus platyrhynchus*
048	black bullhead	*Ameiurus melas*
049	brown bullhead	*Ameiurus nebulosus*
050	**channel catfish**	***Ictalurus punctatus***
052	flathead catfish	*Pylodictis olivaris*
061	**smallmouth bass**	***Micropterus dolomieui***
062	largemouth bass	*Micropterus salmoides*
065	yellow perch	*Perca flavescens*
077	whitefish	*Coregonus sp.*
078	Pacific salmon/trout (Oncorhynchus sp.)	*Oncorhynchus sp.*
079	whitefish	*Prosopium sp.*
080	Atlantic salmon/trout (Salmo sp.)	*Salmo sp.*
084	chub (Couesius sp.)	*Couesius sp.*
085	chub (Gila sp.)	*Gila sp.*
086	squawfish	*Ptychocheilus sp.*
089	sucker	*Catostomus sp.*
090	catfish	*Ictalurus sp.*
091	trout-perch	*Percopsis sp.*
093	bass	*Micropterus sp.*
095	perch	*Perca sp.*
116	yellow bullhead	*Ameiurus natalis*

Target species are indicated with bold text. The more common of these species codes are included on the field form in Appendix B. The project manger will be contacted before additional species codes are used to ensure all codes are unique and consistent through the project.

Fish #'s take the form of a 2-digit sequential number (01, 02, 03 etc.) for each individual fish of a species from a site. For example: 008-010-03 would be the sample code for the third rainbow

trout collected from the eighth river site on the sample list. This number is the same for both fillets from this fish.

If a fillet is too large to fit in a single quart-sized bag it is permissible to cut out and keep for further processing only a central (thickest) portion of the fillet. This portion should be as large as will fit in a quart sized bag. If such field sub-sampling occurs it will be noted on the field form.

Note: the Specimen ID is dropped from the Sample ID once a sample is composited. If necessary, Sample IDs will be reconciled with a laboratory-assigned sample number at a later stage.

Further field precautions:

- Filleting of fish will occur away from dust
- Sterile coolers will be used (wiped or rinsed with bleach solution, then three rinses with tap water).
- Regular ice is preferred to 'Blue' ice packs. Loose ice is to be avoided. Milk jugs filled with water and frozen have been found to work well. If this is not possible loose ice will be contained in large zipped bags, such that meltwater does not escape and contact the sample containers or fish.
- Sampling equipment obviously dirty will not be used.
- Measuring devices will be washed before each sampling day, and rinsed with ambient water between each species/sampling event.

Water and other samples will be identified by a site ID only. All samples will be identified with date of collection and names or initials of samplers as well.

B2.4 Field Materials

Site map
Electro-fishing boat
Nets
Satellite telephone
GPS unit
Digital camera
Case for equipment
Safety equipment
Bucket or container for rinse water (non-metallic)
Bleach (dilute 1:10 for rinsing)
Disposable towels
Nitrile gloves (100 pairs)
Fillet knives (2)
Knife sharpener
Cutting Board
Scrub brush (plastic)
13 gallon plastic garbage bags (120)
1 gallon zipped bags (120)
1 quart zipped plastic bags (1200)
Sample bottles
Milk jugs to contain ice for shipping
Packing tape
Dry Ice
Blank water
Permanent markers
Pencils
Field book(s)
Field forms (on waterproof paper)
Chain of custody/ analysis forms
Chain of custody seals
Cooler labels (on waterproof paper)
Coolers (20)
Butcher paper

B2.5 Handling and Shipping Samples

All samples will be brought back to the DEQ state office for handling and shipping. Frozen fish samples will be stored at DEQ until sufficient samples can be batch for shipping to Brooks Rand or taken to Boise Cold Storage for archiving. Water samples will be kept in a refrigerator and also batched for shipping and analysis. The shortest holding time, from time of filed collection, is 28 days for total mercury. This will be the limiting factor in holding water samples and therefore water samples should be sent so that they arrive at the lab at least one week prior to expiration of this holding time for the oldest sample in the batch.

Samples for analysis of As, Hg and Se in fish and water will be shipped to:
Attn: Amanda Fawley
Brooks Rand LLC
3958 6th Ave NW
Seattle, WA 98107
Ph: 206-632-6206

Water samples for nutrients and common ions will be hand delivered to:
Attn: Wally Baker
Idaho Bureau of Laboratories
2220 Old Penitentiary Road
Boise, ID 83712
Ph: 208-334-2235

Dry ice is a must for shipping fish. Water samples should be shipped on wet ice. Experience has shown even frozen fish on dry ice will not stay frozen more than a day during Idaho's hot summers. Thus all shipped samples will be sent via overnight shipping. Nine lbs of dry ice is usually enough to keep a cooler of fish frozen and is usually the maximum accepted by shippers. It is recommended that the ice be placed on top of the samples and excess space filled with packing material (air pillows, crumpled news paper, etc.). Analytical results from fish samples received unfrozen or waters samples received above 4°C will be flagged as a departure from protocol.

Each cooler will have a waterproof label that specifies the site and species ID, collection date and time, and shipping date and time, as well as the contact details of the project manager (see Appendix C). The project manager will notify the laboratory of each shipment, and retain a copy of the chain of custody form.

B3. Sample Handling and Custody

A chain-of-custody form / laboratory analysis-request form detailing the samples identities and specifying analyses to be performed must be completed and included with each cooler shipped or delivered to the laboratory. The laboratory will be responsible for maintaining integrity of samples until analysis is complete and results are accepted by DEQ.

Upon arrival at the laboratory the coolers of samples will be placed in the restricted-access clean room, and their arrival date and time noted in a log. Fish samples to be processed within twenty-four hours will have ample ice to keep the samples cool until processing. If composite preparation cannot take place within twenty-four hours they must be placed in a freezer and kept frozen until not more than 16 hours prior to processing. Fillets kept frozen may be held for up to 30 days before processing in order to facilitate processing in batches.

Water and blank samples will be kept refrigerated at the laboratory until analysis is complete and passes lab QA.

B4. Analytical Methods & Data Reporting

Copies of all field forms should accompany samples brought to DEQ. It is suggested these be placed in a Ziploc bag with the frozen fillets or water samples rather than delivered separately. The project manager or his designee will accumulate field data sheets for entry into a database.

Fish tissue concentrations will be reported on a wet (fresh) weight basis, in units of ng/g (ppb) for Hg and µg/g or mg/Kg (ppm) for As and Se. Processing blank results will be analyzed and reported as if fish tissue, i.e. in units of mass/mass. Concentrations in water will be reported in units of ng/L for Hg and µg/L for As and Se. The laboratory will apply blank corrections per laboratory SOP and note if this in done in their reports.

EPA Method 1631 Appendix A (USEPA 2001) will be used to prepare fish tissue samples.

EPA method 1631, Cold Vapor Atomic Flouresence (USEPA 2002b) will be used to analyze the fish tissue digests and water samples, including blanks, for total mercury. The typical working range for this method is 0.5 - 100 ng/L and the instrumental detection limit is 0.15 ng/L total mercury. The required method detection limit (MDL) for this project is 0.04 ng/g in fish tissue and 0.15 ng/L in water (blanks).

EPA Method 1632, As species, will be used for analysis of inorganic arsenic in fish tissue digests and water samples. The required MDL for inorganic As is 0.003 µg/g in fish tissue and 0.01 ug/L in water (blanks).

EPA Method 1638, Inductively Coupled Plasma – Mass Spectrophotometer will be used for analysis of total arsenic and total selenium in fish tissue digests and water samples. The required MDL for these analytes in fish tissue is 0.05 µg/g for total As and 0.1 µg/g for total Se. In water (blanks) the required MDLs are 0.1 µg/L for total As and 0.2 µg/L for total Se.

In addition to the above chemical analyses, the percent moisture content of each composite fish tissue sample will be determined by the laboratory so that reported wet weight concentrations may be converted to a dry weight basis.

Standard Method 10200H will be used for chlorophyll-a analysis. Samples will be filtered in the field, wrapped in foil, and frozen on dry ice until shipped to the contract laboratory.

EPA Method 353.2, Nitrate-Nitrite Nitrogen by Colorimetry, and EPA Method 365.2, Phosphorus by Colorimetry, will be used for nutrient analysis. Samples will be preserved with sulfuric acid and will remain on wet ice until shipment or delivery to the contract laboratory.

EPA Method 160.1, Filterable Residue by Drying Oven, EPA Method 120.1, Conductance by Conductivity Meter, and EPA Method 180.1, Turbidity by Turbidimeter, will be used to determine physical properties of the water sample. All samples will be placed immediately on wet ice until shipment or delivery to the contract laboratory.

B5. Quality Control Samples

FISH DUPLICATE SAMPLES: There will be three levels of duplicates employed in this project for fish – field, processing and laboratory. Each will be done at the rate of ten percent, based on the number of samples (sites x species) collected. Since the target number of samples is 53 this is nominally six duplicates of each type, and 18 total.

Field duplicates will consist of an additional set of ten fish collected and filleted as if an original sample from a site. We want ten percent duplication of fish samples collected, not sites, a sample being a set of ten fish of a species from a site. Since 53 such samples are planned this means 6 duplicate samples over the course of field sampling need to be obtained. Because availability of fish is unpredictable, field duplicates will be driven by plentitude of fish rather than pre-selection of duplication prior to field work. There will be no more than one duplicate for any one sample.

Processing duplicates will consist of a second set of 10g subsamples taken from the same fillets as the original set. Samples for processing duplicates will be randomly selected and may therefore by chance occur with a field duplicate.

Laboratory duplicates will be done according to the laboratory's standard operating procedures.

FISH BLANKS: A true blank for fish tissue is not possible. We will check for possible tissue sample contamination by use of a processing blank. These blanks will be generated at the rate of one for every ten fish samples (a set of ten fish), but not less than one for each day of fish tissue processing.

Processing blanks will be generated from a volume of de-ionized water equal to the final digest volume for fish tissue samples. This blank will be shaken once then opened and placed in the clean room during processing. At the end of processing one sample (ten fillets) a sterile scalpel will be stirred in the water. The blank will then be poured into a blender that has been cleaned and is ready for processing a fish tissue composite sample. The water will be blended for one minute, and then poured back into the bottle. This will then be prepared for analysis as a fish tissue sample. This blank serves as a check on the cleanliness of the equipment and room used in tissue composite processing.

Acceptable levels of blank quality are specified in Table 2 section A7.2. Any value above this level will trigger a review of sample processing procedures and appropriate flagging of results for samples processed that day as possibly biased high (See D1).

FISH SAMPLE SPLITS: Some samples may be split and sent to the State of Idaho Laboratory for analysis. Samples will be split after compositing.

WATER DUPLICATE SAMPLES: There will be two levels of duplicates employed in this project for water – field and laboratory. Each will be done at the rate of at least ten percent, based on the number of samples, but not less than one field duplicate per field trip. Since the target number of sites is 35 this is nominally 4 duplicates minimum of each type. Because sampling will take place over a three-four month summer field season, there likely will be several more field duplicates, as many as 12.

Field duplicates will consist of an additional sample of water taken immediately after the primary sample from the exact same location. Water duplicates will be labeled with the Site # + Dup (in place of species code) and the date.

Laboratory duplicates will be done according to the laboratory's standard operating procedures.

WATER BLANKS: There will be two levels of duplicates employed in this project for water – field and laboratory. Each will be done at the rate of at least ten percent, based on the number of samples, but not less than one field blank per field trip. Since the target number of sites is 35 this

is nominally 4 blanks minimum of each type. Because sampling will take place over three month summer field season, there likely will be several more field blanks, as many as 12.

Field blanks will be generated in the field from a bottle of de-ionized "blank" water taken to the field and used to fill a set of sample containers in the field. Sufficient volume of blank water is needed to fill the three samples containers that one water sample. One liter per sample should be enough for each blank. For the mercury sample container the same clean hands / dirty hands procedures will be used as for the ambient river sample. Water blanks will be labeled with the Site # + Blank (in place of species code) and the date. These blank samples will otherwise be treated in the same manner as ambient river samples.

Laboratory blanks will be done according to the laboratory's standard operating procedures.

Acceptable levels of blank quality are specified in Table 2 section A7.2. Any value above this level will trigger a review of sample processing procedures and appropriate flagging of results for samples processed that day as possibly biased high (See D1).

The analyzing laboratory conducts calibration of their equipment and also runs quality control samples to verify analytical methods are performing within specifications. They will provide a summary of their internal QA/QC with reporting on analytical results. All quality control results will be listed in the final report.

Quality control measures will be undertaken throughout the sampling effort, and are listed in their respective sections (especially section B2.2, 'Fish Handling').

B6. Instrument/Equipment Testing, Inspection, and Maintenance Requirements

The sampling equipment used in this study will be maintained by DEQ personnel. This includes all field equipment, fishing and filleting equipment, materials for measuring fish length and weight, and packaging samples for shipment.

B7. Instrument/Equipment Calibration and Frequency

DEQ's field crew will calibrate their electrofisher. The only criterion affecting quality is that sufficient fish are caught. This will be the responsibility of DEQ.

B8. Inspection/Acceptance of Supplies and Consumables

Water sample containers and de-ionized water for field blanks shall be provided by the contract laboratory.

Zipped plastic bags for fish samples shall be obtained from a local grocery store. It has been shown that these bags contain negligible levels of mercury (Frontier Geosciences, DEQ training presentation 2005).

B9. Data Management

Field data and paperwork will be kept in a dedicated folder, to be retained at DEQ's State Office for at least five years. Laboratory analysis data will be transmitted electronically via email to the project manager. Electronic data will be backed up onto a CD, which will be stored in the dedicated folder. Working copies of the data will be kept on the computer of the project manager.

Each receipt of data from the laboratory will receive a visual inspection. At this time analytical data will be rectified with field locations/IDs as necessary. Any questions that arise as to reported values or sample identity will result in the project manager consulting with laboratory staff and /or field crew until the question is resolved. Location information (latitude, longitude, and depth) will be added to the data, along with the number of fish in the composite.

Data will be available to the general public upon request. Copies of all data may be obtained by contacting the project manager. Fish tissue and associated water chemistry data will be reported by March 2009. A separate report on habitat and biological monitoring results will follow, after macroinvertebrate identification is complete.

No specialized software will be used in the handling and transmittal of the data. It is expected that Microsoft Excel will be the preferred format of near-term data transmittal. All data will be entered into a database by project's end.

GROUP C: ASSESSMENT AND OVERSIGHT

The elements in this group address the activities for assessing the effectiveness of project implementation and associated QA and QC activities. The purpose of assessment is to ensure that this plan is implemented as prescribed.

C1. Assessment and Response Actions

The project quality assurance officer (QAO) will have the lead role in assessing the QA and QC measures employed in this study, e.g. review of procedures and training and will have the lead role in data quality review. The QAO will work with the project manager to assure overall project objectives are met.

The QAO shall have access to and is responsible for inspecting field supplies and equipment so as to make sure they are adequate to deliver the quality of results specified in this QAPP.

As quality control data becomes available from the lab the QAO will review these results for compliance with the data quality objectives specified in section A.7. Any departure from quality objectives will be brought to the attention of the project manager and options for corrective action discussed. The QAO will document any such conversation via e-mail or a memo to file to become part of the project record. The QAO will compile all his/her observations into a review of the quality assurance measures used, to be included in the final report.

All project personnel are instructed to bring any serious quality control problems to the immediate attention of the project manager. Details of the incident will be included in the final report, along with any corrective action that was taken.

C2. Reports to Management

A final report will be prepared by the project manager and available in March 2009 to include:

- A summary of the field work conducted
- The results of the laboratory analyses, including QC results
- A QA and QC summary prepared by the QAO

This report will be provided to all contacts on the distribution list. No specific action will be required by any recipient of the report.

GROUP D: DATA VALIDATION AND USABILITY

The elements in this group address the QA activities that occur after the data collection phase of the project is completed. Implementation of these elements determines whether or not the data conform to the specified criteria, thus satisfying the project objectives.

D1. Data Verification

Data verification will consist of checking that the planned number of sites and locations, quality control samples, field data sheets and sample logs are completed according to this QAPP.

Upon receipt from the laboratory, sample analysis results will immediately be checked by the QAO for completeness, in order to assure that all the requested analyses were performed along with the correct methodologies and detection limit. If errors or omissions are noted during this step, then the laboratory will be notified immediately and the data will not be considered usable or reportable until those errors have been corrected and new reports issued from the laboratory.

Data will be subject to visual inspection and any questions as to values or sample identity will be resolved via line-by-line confirmation with the analyzing laboratory.

Data will also be checked to assure that the specified frequency of quality control samples specified in section B5 is obtained and that all data can be unequivocally associated with a site and species.

D2. Data Review, Validation, and Use

Data will be validated by comparison to the quality assurance criteria in section A.

The data will be rejected as unusable when serious deficiencies in meeting quality control criteria occur. Two possible deficiencies are:

1) When RPD exceeds 50% for processing duplicates in which analyte levels are greater than the practical quantification limit (PQL). In this case all results in the associated batch will be rejected.
2) When quantified blank results (> PQL) are more than 20% of sample results. Then those sample results less than 5 times the blank result will be rejected.

Unless otherwise defined by the laboratory, the PQL will be taken to be five times the method detection limit (MDL). Data rejection is at the discretion of the QAO. Rejected data will not be entered into the database, count toward meeting the data completeness objective, or otherwise be used.

All other data will be useable but may be flagged as described below.

Data Quality Flags

As a result of the data evaluation procedure, data qualifier flags may be applied to individual analytical results if qualification for project data usability is appropriate. Definitions of the flags are as follows:

Flag	Definition
B	Analyte confirmed present but the reported value is an estimated quantity. Used when the result is above the MDL, but less than the PQL.
H	Holding time exceeded or samples storage conditions not met.
J	Analyte confirmed, but the reported value is an estimated quantity. The associated numerical value is the approximate concentration of the analyte in the sample. Used when duplicate RPD is greater than specified QC limits.
J+	The reported value is an estimated quantity, and may be biased high. Used when associated blank value is above QC limit but less than 10% of sample result, or spike recovery is high, above upper QC limit.
J-	The reported value is an estimated quantity, and the result may be biased low. Used when matrix spike recovery is below the lower QC limit.
U	Analyte not confirmed present at or above the MDL.

Flagged data will be accepted and count toward meeting the data completeness objective. Flags may affect interpretation of results.

Unflagged data means the result meets all sample specific data quality objectives, i.e. accuracy and precision are within control limits, and there is no significant contamination in blanks. Additional data qualifiers may be developed at the discretion of the quality assurance officer.

D3. Reconciliation with User Requirements

Upon validation the data will be entered in an Excel spreadsheet. Data quality flags specified in section D2 will be associated with each analytical result as appropriate. If data are manually entered they should be double entered and the two versions electronically compared for any discrepancies. Once all discrepancies are resolved duplicate entries will be discarded. Data will remain on file at the DEQ State Office indefinitely, but for a minimum of five years.

Data will be geo-located, and an ArcGIS compatible shapefile will be provided with them.

Fish contaminant will be reduced such that each site is characterized by a single result for each analyte. Duplicate results for the same species will be combined as a simple average. Where more than one species is obtained from a site, the results from multiple species will be averaged. For comparison to methylmercury fish tissue criterion, the mercury average will be trophic level weighted as specified in Idaho's water quality rules (IDAPA 58.0102.210 footnote p).

References

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USEPA 2000 – Guidance for Assessing Chemical Contaminant Data for Use in Fish Advisories, Volume 1 Fish Sampling and Analysis Third Edition. EPA 823-B-00-007. November 2000. Office of Water. Washington, DC

USEPA 2001a - Water Quality Criterion for the Protection of Human Health: Methylmercury. EPA-823-R-01-001. January 2001. Office of Water. Washington, DC

USEPA 2001b – Appendix to Method 1631: Total Mercury in Tissue, Sludge, Sediment, and Soil by Acid Digestion and BrCl Oxidation, EPA 821-R-02-019.

USEPA 2002a – Guidance for Quality Assurance Project Plans, EPA QA/G-5

USEPA 2002b – Mercury in Water by Oxidation, Purge and Trap, and Cold Vapor Atomic Fluorescence Spectrometry, EPA Method 1631, Revision E.

Appendix A

Site Selection

Idaho Major River
Survey Design 2006-2008

Contact:

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Description of Sample Design

Target population: Major rivers in Idaho, as identified by Idaho.

Sample Frame: To identify the target population streams, Mary Anne Nelson provided the GIS stream coverage. It is based on NHD with only major rivers included. Note that it appears that run-of-the-river reservoirs were included in the GIS coverage. They were included in the design.

Survey Design: A Generalized Random Tessellation Stratified (GRTS) survey design for a linear resource was used. The GRTS design includes reverse hierarchical ordering of the selected sites.

Multi-density categories: None

Stratification: None.

Panels: Two panels to be visited in two different years: Panel_2006 and Panel_2008.

Expected sample size: Expected sample size 25 sites per panel.

Over sample: 200% (100 sites).

Site Use: Within State, the base design has 50 sites. Sites are listed in SiteID order and must be used in that order. All sites that occur prior to the last site used must have been evaluated for use and then either sampled or reason documented why that site was not used. As an example, if 50 sites are to be sampled and it required that 80 sites be evaluated in order to locate 50

sampleable stream sites, then the first 80 sites in SiteID order would be used.

If the design is implemented over two years, then use the sites in siteID order within year and then continue with the next siteID in the next year. If want to identify revisit sites, use the first 5 sites in siteID order that were actually sampled in the field each year.

Sample Frame Summary

Total stream length (in km) in the sample frame is 7384.939 km.

Site Selection Summary

Number of sites in sample

mdcaty	OverSamp	Panel_2006	Panel_2008	Sum
Equal	50	25	25	100
Sum	50	25	25	100

Description of Sample Design Output:

The dbf file for the shapefile ("ID Major Rivers 2006-08 Sites") has the following variable definitions:

Variable Name	Description
SiteID	Unique site identification (character)
x	x-coordinate from map projection (see below)
y	y-coordinate from map projection (see below)
mdcaty	Multi-density categories used for unequal probability selection
weight	Weight (in km), inverse of inclusion probability, to be used in statistical analyses
stratum	Strata used in the survey design
panel	Identifies base sample by panel name and Oversample by OverSamp
EvalStatus	Site evaluation decision for site: TS: target and sampled, LD: landowner denied access, etc (see below)
EvalReason	Site evaluation text comment
auxiliary variables	Remaining columns are from the sample frame provided

Projection Information

PROJCS["IDTM83",
GEOGCS["GCS_North_American_1983",
DATUM["D_North_American_1983",
SPHEROID["GRS_1980",6378137.0,298.257222101]],
PRIMEM["Greenwich",0.0],UNIT["Degree",0.0174532925199433]],
PROJECTION["Transverse_Mercator"],
PARAMETER["False_Easting",2500000.0],
PARAMETER["False_Northing",1200000.0],

PARAMETER["Central_Meridian",-114.0],
PARAMETER["Scale_Factor",0.9996],
PARAMETER["Latitude_Of_Origin",42.0],
UNIT["Meter",1.0]]

Evaluation Process

The survey design weights that are given in the design file assume that the survey design is implemented as designed. Typically, users prefer to replace sites that can not be sampled with other sites to achieve the sample size planned. The site replacement process is described above. When sites are replaced, the survey design weights are no longer correct and must be adjusted. The weight adjustment requires knowing what happened to each site in the base design and the over sample sites. EvalStatus is initially set to "NotEval" to indicate that the site has yet to be evaluated for sampling. When a site is evaluated for sampling, then the EvalStatus for the site must be changed. Recommended codes are:

EvalStatus Code	Name	Meaning
TS	Target Sampled	site is a member of the target population and was sampled
LD	Landowner Denial	landowner denied access to the site
PB	Physical Barrier	physical barrier prevented access to the site
NT	Non-Target	site is not a member of the target population
NN	Not Needed	site is a member of the over sample and was not evaluated for sampling
Other codes		Many times useful to have other codes. For example, rather than use NT, may use specific codes indicating why the site was non-target.

Statistical Analysis

Any statistical analysis of data must incorporate information about the monitoring survey design. In particular, when estimates of characteristics for the entire target population are computed, the statistical analysis must account for any stratification or unequal probability selection in the design. Procedures for doing this are available from the Aquatic Resource Monitoring web page given in the bibliography. A statistical analysis library of functions is available from the web page to do common population estimates in the statistical software environment R.

For further information, contact

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Bibliography:

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Web Page: http://www.epa.gov/nheerl/arm

Table 2: Randomly Selected River sites for the 2008 field season. "Panel 2006" in the panel column refers to sites that were sampled in 2006 and were electrofished again in 2008. "Panel_2008" in the panel column refers to primary sites and "OverSample" refers to Over-sample Sites. A total of 46 sites were initially evaluated prior to field season to determine target status.

	ID	Site_ID	FEAT_NAME	Latitude	Longitude	Panel	EvalStatus	EVALCOMMENTS
*	005	IDR06615-005	Blackfoot River	43 12 29.64	-112 12 14.48	Panel_2006	MONITOR	INACCESSIBLE
*	011	IDR06615-011	Big Wood River	43 46 50.84	-114 32 32.02	Panel_2006	MONITOR	
*	012	IDR06615-012	Salmon River	45 24 24.35	-116 11 30.71	Panel_2006	MONITOR	DONE
*	017	IDR06615-017	Bear River	42 21 37.33	-111 44 11.78	Panel_2006	MONITOR	DONE
	026	IDR06615-026	North Fork Clearwater River	46 43 12.73	-115 17 30.34	Panel_2008	MONITOR	
	027	IDR06615-027	North Fork Big Lost River	43 55 37.76	-114 11 16.07	Panel_2008	MONITOR	
	028	IDR06615-028	Salmon River	45 47 22.75	-116 19 12.21	Panel_2008	MONITOR	
	029	IDR06615-029	Teton River	43 52 54.25	-111 48 40.36	Panel_2008	MONITOR	
	030	IDR06615-030	Coeur d'Alene River	47 28 42.3	-116 44 8.72	Panel_2008	MONITOR	
	031	IDR06615-031	Weiser River	44 37 45.08	-116 35 9.18	Panel_2008	MONITOR	
	037	IDR06615-037	Blackfoot River	42 48 4.03	-111 29 6.54	Panel_2008	MONITOR	
	038	IDR06615-038	Coeur d'Alene River	48 0 47.07	-116 14 5.85	Panel_2008	MONITOR	
	040	IDR06615-040	Salmon River	45 27 18.08	-115 46 20.41	Panel_2008	MONITOR	
	043	IDR06615-043	East Fork Salmon River	44 13 20.75	-114 17 3.92	Panel_2008	MONITOR	
	044	IDR06615-044	Pahsimeroi River	44 39 31.9	-114 1 25.82	Panel_2008	MONITOR	
	046	IDR06615-046	Camas Creek	43 17 17.85	-114 42 13.81	Panel_2008	MONITOR	
	047	IDR06615-047	Snake River	43 36 23.55	-116 54 39.16	Panel_2008	MONITOR	
	050	IDR06615-050	Priest River	48 14 31.27	-116 53 1.92	Panel_2008	MONITOR	
*	051	IDR06615-051	Bruneau River	42 47 22.47	-115 43 3.6	OverSamp	MONITOR	
	054	IDR06615-054	Coeur d'Alene River	48 1 20.89	-116 17 35.23	OverSamp	MONITOR	
*	055	IDR06615-055	NF Payette River	44 12 49.08	-116 6 23.63	OverSamp	MONITOR	DONE
	057	IDR06615-057	Little Lost River	43 59 9.08	-113 12 40.51	OverSamp	MONITOR	
*	061	IDR06615-061	Camas Creek	43 52 54.36	-112 21 5.86	OverSamp	MONITOR	DONE
*	063	IDR06615-063	Payette River	44 0 12.85	-116 48 12.48	OverSamp	MONITOR	DONE
	068	IDR06615-068	Camas Creek	44 49 3.3	-114 29 33.64	OverSamp	MONITOR	
*	074	IDR06615-074	Lochsa River	46 27 31.49	-115 2 25.34	OverSamp	MONITOR	DONE
*	077	IDR06615-077	Henry's Fork	43 47 49.49	-111 55 37.7	OverSamp	MONITOR	DONE
	083	IDR06615-083	Snake River	43 0 52.52	-116 7 54.48	OverSamp	MONITOR	
	084	IDR06615-084	South Fork Salmon River	44 41 42.04	-115 42 5.63	OverSamp	MONITOR	
	085	IDR06615-085	Portneuf River	42 51 2.5	-112 26 30.37	OverSamp	MONITOR	RAINEY PARK, JUST ABOVE POCATELLO
	086	IDR06615-086	Saint Joe River	47 8 23.09	-115 24 29.02	OverSamp	MONITOR	

087	IDR06615-087	South Fork Payette River	44 10 17.03	-115 14 4.57	OverSamp	MONITOR
088	IDR06615-088	Selway River	46 2 44.38	-115 17 47.04	OverSamp	MONITOR
089	IDR06615-089	Raft River	42 31 39.55	-113 15 40.79	OverSamp	MONITOR
091	IDR06615-091	Big Wood River	43 26 3.52	-114 15 44.92	OverSamp	MONITOR
093	IDR06615-093	Raft River	42 3 28.79	-113 35 19.24	OverSamp	MONITOR
094	IDR06615-094	Lemhi River	45 6 1.9	-113 43 36.48	OverSamp	MONITOR
095	IDR06615-095	Snake River	42 38 7.66	-114 33 28.82	OverSamp	MONITOR
097	IDR06615-097	Snake River	43 26 8.74	-111 21 27.49	OverSamp	MONITOR
099	IDR06615-099	Payette River	43 54 2.98	-116 37 59.82	OverSamp	MONITOR

*INDICATES RE-FISH SITES FROM 2006

Appendix B

Fish Field Form

Idaho Fish Tissue Mercury Sampling Field Form

Site Information

Latitude: _____° _____' _____" Longitude: _____° _____' _____"
Datum: _____ Site #: _____ Site Name: _____
Site Description: _____
Reach Length (est in m) : _____

Collection Information

Date: ____ / ____ / 2008 Water Sample ☐ Duplicate ☐ Blank ☐
Weather Conditions (circle): Equipment (circle): *Electrofisher / Hook & Line / Other*
Windy / Sunny / Raining Fishing Start Time : _____ End Time : _____
Equipment Notes / Location Fishing Ended: _____
Field Crew: _____, _____, _____

Sample Information

Fish #	Species Code[1]	Length (cm)	Weight (g)	Comments (e.g. abraded fins, field duplicate)
1				
2				
3				
4				
5				
6				
7				
8				
9				
10				
1				
2				
3				
4				
5				
6				
7				
8				
9				
10				

[1] Species code: **019 = brown trout**, 021 = brook trout, 049 = brown bullhead, **050 = channel catfish**, 052 = flathead catfish, 062 = largemouth bass, **046 = largescale sucker, 016 = Mountain whitefish**, **010 = rainbow trout**, **061 = smallmouth bass**, 011 = cutthroat trout, 065 = yellow perch. The field manger should be contacted if additional codes are needed. Standard DEQ Taxa codes must be used

Site notes/comments:

Appendix C
Cooler Label

Site ID: _____ Species Codes: _____

Cooler _____ of _____ (for this site)

Collection date and time: _____

Shipping date and time: _____

This cooler contains frozen fish samples. These samples are to be analyzed for arsenic, mercury, and selenium contamination, and are time sensitive. Please do not disturb the contents.

For more information, please contact:
Don Essig of DEQ at 208-373-0119.

Appendix D

Processing Log

Fish Processing Log

Site ID: _____ **Species** _____

Time and Date fillets removed from freezer:
Date _____ **Start Time**: _____

Processed By: _____

Time and date composite tissue homogenate returned to freezer
Date _____ **End Time**: _____

Fish #	**Sample Integrity** Note if compromised, ✓ if good	**Sub-Sample weight (g)***

* ✓= nominal 10 grams, otherwise record weight to nearest gram

Composite Sample ID:

_____

Duplicate Processing Sample ID (if prepared)**:**

_____

Notes: