## Quality Assurance Project Plan for Coho Sampling for Contaminant and Diet Analysis

**Biota Work Group** 

Mark E. Holey and Robert F. Elliott U.S. Fish & Wildlife Service Fishery Resources Office 1015 Challenger Court Green Bay, WI 54311

April 1994

## Quality Assurance Project Plan for Coho Sampling for Contaminant and Diet Analysis

## **1.0 Introduction and Project Description**

#### 1.1 Overview

The Great Lakes National Program Office (GLNPO) of the US EPA has initiated a Mass Balance Study for selected toxic contaminants in Lake Michigan. The mass balance effort will be part of a "Lake Michigan Enhanced Monitoring Program" which includes tributary and atmospheric load monitoring, source inventories, and fate and effects evaluations. In general, the primary goal of this enhanced monitoring program is to develop a sound, scientific base of information to guide future toxic load reduction efforts at the Federal, State and local levels.

A modeling team will construct a mass budget/mass balance model for a limited group of contaminants which are present in Lake Michigan at concentrations which pose a risk to aquatic and terrestrial organisms (including humans) within the ecosystem. Components to the mass balance model will be designed to predict contaminant concentrations in the water column and target fish species over a two year period, relative to loadings. Predictions of contaminant levels in three species of fish will be calculated as final output of the model. The target fish species include:

Lake trout (*Salvelinus namaycush*) Coho salmon (*Oncorhynchus kisutch*) Bloater chub (*Coregonus hoyi*)

The calibration of the food web model(s) for these target species requires data on contaminant concentrations and fluxes (diet) not only in these species, but also in the supporting trophic levels. The contaminant burden of each prey species varies based on feeding patterns at lower trophic levels. The concentration of contaminants in coho salmon will depend on what prey items they choose to consume. The diet information for coho salmon sampled by this project will enable the modelers to quantify the movement of contaminants from their source, through the food web, and ultimately the body burden in coho salmon.

The basic design and data requirements for the fish samples have been outlined in Tables 5 and 6 and in Appendix 4 of the Lake Michigan Mass Budget/Mass Balance (LMMB) work plan of October 14, 1993. This project addresses a subset of the work objectives for coho salmon, one of the target species described in the LMMB work plan.

The specific objectives are to:

- 1) Describe the diet of coho salmon in Lake Michigan from April-October 1994.
- 2) Collect representative samples of coho salmon from spring, summer, and fall in 1994 for the purpose of conducting contaminant analysis.
- 3) Review past published and unpublished information on the diet of coho salmon in Lake Michigan and report on the comparability of the data collected in 1994 to past data.

#### 1.2 Experimental Design

Spatial and temporal variations in coho salmon feeding habits and movement will require fish to be collected in spring, summer, and fall and from both the east and west shore of Lake Michigan. Based on coho migration patterns, spring samples will be collected primarily from the southern region of the lake, summer samples from the central region, and fall samples from the north central region of the lake near the egg collection facilities (Table 1.0). The 1993 year class (age 1.1) of coho will be sampled during the entire sampling period (Table 1.0). The 1994 year class will be sampled while in the hatchery (age 1.0) and once in the fall (Table 1.0). The hatchery sample will quantify the amount of contaminants the coho acquired, if any, from the hatchery before they enter the lake and began feeding on natural foods.

## Table 1.0. Sample Size Objectives for the Collection of Coho Salmon in Lake Michigan by Season and Location

Season	Location	Age	<b>Contaminants</b>	Diet	<u>Total</u>
Spring (April to r	nid-June)				
	Hatcheries	1.0	25	0	25
	East Shore (Indiana to Benton Harbor, MI)	1.1	25	75	100
	West Shore (Illinois waters)	1.1	25	75	100
Summer (mid-June	to mid-August)				
	East Shore (Benton Harbor to Ludington, MI)	1.1	25	75	100
	West Shore (Kenosha to Sheboygan, WI)	1.1	25	75	100
Fall (mid-Aug	ust to October)				
	East Shore (Ludington to Frankfort, MI)	1.0 1.1	25 25	75 75	100 100
	West Shore (Sheboygan to Kewaunee, WI)	1.0 1.1	25 <u>25</u>	75 <u>75</u>	100 <u>100</u>
Total			225	600	825

The most difficult part of this project will be the collection of the necessary samples of coho salmon. Netting techniques to capture salmon in the open water of the Great Lakes is difficult, expensive, and not widely practiced. For salmon, angling is the most appropriate method for addressing the specific needs of this project. Coho salmon collected for contaminant analysis will be obtained by contracting sport charter anglers from the areas sampled (Table 1.0). As necessary and available, samples from assessment netting or creel surveys by state or other research agencies will be used. Standard biological and site specific information (length, weight, age, sex, location, and season) will be recorded for all coho collected.

#### 1.3 Contaminant Sampling

The total number of coho required for contaminant analysis outlined in the LMMB work plan was been modified from 450 to 225 (Table 1.0). Samples will be packaged as required for contaminant analysis, frozen, and delivered to the NBS Great Lakes Research Center. To make these collections as representative as possible, samples will be taken throughout each season to the extent possible. Salmon for contaminant analysis will be collected primarily by contracted charter fishermen.

#### 1.4 Diet Sampling

The LMMB work plan did not have a sample size objective for describing the diet. Based on recent diet work describing variation typically observed in the diets of salmon from Lake Michigan (Elliott 1993), we estimate the sample size goal should be at least 100 fish per season per region (Table 1.0). To account for as much of the spatial and temporal variation as possible, sampling effort will be distributed throughout each season in the regions of the lake where the fish are commonly found. To achieve the 100 fish per season per region goal, 75 fish (per season per region) in addition to the salmon collected for contaminant analysis will have to be collected. Diet samples will be collected from contracting charter fishermen and from sampling sport angler catches at boat ramps (see section 4.0 for description of methods).

Historical data describing coho diet will be analyzed and summarized to complement the information collected from those coho sampled in 1994 and 1995. This will serve to put the 1994-95 diet information in perspective and minimize the dangers of having to assume that the diet of a relatively small number of fish collected in 1994-95 is representative of typical years.

Parameter	Sampling Instrument	Sampling Method	Analytical Instrument	Analytical Method	Reporting Units	LOD
Location (critical)	GPS, Loran, Port Location	SOP-1	NA	NA	Lake Regions	Basin- East, West- North, Central, Southern
Sample Date (critical)	None	NA	NA	NA	mo/day/yr xx/xx/xx	day
Coho length (critical)	measuring board ruler	NA	NA	NA	mm	1 mm
Coho weight (critical)	spring or electronic balance	SOP-1	NA	NA	Kg	0.1 Kg
Coho age (critical)	Knife and envelope	SOP-1 and Bowen 1983	scale projector	SOP-2	years	1 year
Diet Species (critical)	NA	SOP-1	NA	SOP-2	total number	Species-fish & Common invertebrates Order for less common invertebrates
Diet item Length (critical)	NA	NA	ruler	SOP-2	mm	1 mm
Diet item Weight (critical)	NA	NA	ruler or electronic balance	SOP-2	grams	0.1 gram
Sample Depth (non-critical)	echo sounder	operating instructions	NA	NA	meters	0.1 meters
Time of Sample (non-critical)	clock	NA	NA	NA	HH:MM	minutes
Water Temperature when sampled (non-critical)	thermometer	NA	NA	NA	degrees C	1°C

# Table 1.1 Summary of Critical and Non-Critical Parameter Measurements for the Evaluation of Coho Salmon Diet.

## 2.0 Project Organization and Responsibilities

644444444444444444444444444444444444444	4444444444	4444444444	44444444447	
+)))))))J))))))), +))))), +))))	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	))))))))).	+))))))J)))))))))))	
* John Gannon * *		1 Bertram		, ou Blume *
* NBS /))))0)))1		ject Offic		A Manager*
				5
		Co-Chair	* .)))))))H	1))))))-
		)))))))))-	5	
5 .))))))	))))))))))))))))	,	5	i i
944444444	4444447	*	1	5
	+))))))J	))2)))))))))))))))	)), 5	
	* Ma	irk Holey	*	5
	*	USFWS	G4444444444444	48
	* Dro-	ect Manag	*	10
	FIO			
	• • • • • • • • • • • • • • • • • • • •	)0)H)))))))))))))) * 5		
		5		
		* 5		
	+))))))))	)2)J)))))),	,	
	* Robe	ert Elliot	t *	
	*	USFWS	*	
	* Fiel	d Manager	*	
		)0)H))))))		
		2) 0) ) ) ) ) ) ) ) ) )		
		44=444444444		
+)))))))))))))))))))))))))))))))))))))			)) J) 2))))))))))))),	·
* Stewart Cogsw			o Temporary Posit	lons*
* USFWS	*	*	USFWS	*
* Field Samplin	ng *	*	Field Sampling	3 *
* Analysis	*	*	Analysis	*
.))))))))))))))))))))))))))))))))))))))	) -	.)))))))))))))))	))))))))))))))))))))))))))))	
	-			

S) ) Project communicationW44 QA communication

2.1 GLNPO Project Officer and Biota Co-Chair

The GLNPO Project Officer is the Agency official who initiates the grant, evaluates the proposal, is the technical representative for EPA, and is also co-chair of the Biota workgroup for the Lake Michigan Mass Balance Program. The Project Officer is responsible for:

Budgeting Program planning, scheduling, and prioritization Developing project objectives and data quality objectives Ensuring that project meet GLNPO missions Technical guidance Program and data reviews including audits Data quality Final deliverables

#### 2.2 GLNPO QA Manager

The GLNPO QA Manager (QAM) is responsible for ensuring that each project funded by EPA satisfies the Agency's requirements for QA programs. The QAM is responsible for:

Offering guidance on QA techniques Evaluating QA Project Plans (QAPjPs) and approving QAPjPs for the Agency Assisting in the coordination of audits

#### 2.3 NBS Biota Co-Chair

The Biota Co-Chair from NBS works in partnership with the GLNPO QA Project Leader to implement the Biota portion of the Lake Michigan Mass Balance Project. Duties are:

Program planning, scheduling, and prioritization Developing project objectives and data quality objectives Ensuring that project meets GLNPO missions

#### 2.4 USFWS Project Manager

The Project Manager is the USFWS official who initiated the proposal to perform the coho sampling portion of the LMMB project and is responsible for:

Developing the sampling plan for coho collection Administration of the coho segment of the Biota objectives Overall supervision of field work Ensures QA objectives are met Technical supervision Final deliverables Data Quality Assessment

#### 2.5 USFWS Field Manager

The Field Manager is the USFWS position that will provide daily supervision of the field collection activities and achievement of the QA objectives. This position is responsible for:

Collecting field data Directly supervise the field crew activities Reviews progress toward QA objectives Develops and implements sampling and analytical procedures Prepares reports and deliverables Trains field crews on sampling and analytical procedures Technical systems audits for field and laboratory activities Data quality assessments for lab and field segments 2.6 Field Sampling and Analysis Personnel

These positions are responsible for the majority of the field sampling and lab ID. They will receiving training and guidance from the Project and Field Managers, who will also audit their work to ensure QA objectives are met. These positions will be temporary positions hired at a GS-5 fishery biologist level. Minimum requirements for a GS-5 are six college credits of fishery related courses and 12 credits of related natural resources or animal science related courses or appropriate experience.

## 3.0 Quality Assurance Objectives

As outlined in the Lake Michigan Mass Budget/Mass Balance Work Plan, the proposed model output should be within a factor of two of the observed concentrations in the water column and target fish. It is also estimated that the required level of model accuracy can be achieved if loadings and contaminant mass in significant environmental compartments are determined to within +/- 20 to 30 percent of the actual value.

#### **Objectives:**

- 1) Within each season/region strata, collect as representative a sample of coho salmon as possible so as to minimize the spatial and temporal population uncertainty  $(Sp^2)$  to the extent possible (given the sample size that can be collected with the financial, logistic, and biological constraints of this project).
- 2) To collect, package, and transport each sample, and to record, summarize, and report each physical measurement with a level of precision, accuracy, detectability, and completeness that will ensure that Measurement Uncertainty (Sm<sup>2</sup>) will be nominal compared to Sp<sup>2</sup> and therefor not affect the interpretation of the results.

The level of population uncertainty can not be determined priori. That the contaminant levels in the coho collected will be within +/- 20 to 30 percent of the actual population values is a function of sample size and the collection procedures. The sample size for contaminants has been established by the LMMB Work Plan and subsequent modifications. The designed collection procedures described here attempt to make the most of the sample size target.

Variability in the diet of Lake Michigan salmon can be great, especially when examined from a lakewide perspective encompassing large scale spatial and temporal gradients. The desired sample size for determining diet is to a large degree constrained by the difficulty of collection of these fish. Presently coho abundance in Lake Michigan and therefor catch is very low.

#### 3.1 Measurement Quality Objectives

Measurement quality objectives (MQOs) are designed to control various phases of the measurement process and to ensure that total measurement uncertainty is within ranges prescribed by the DQOs. The MQOs can be defined in terms of data quality attributes; precision, accuracy, completeness, detectability, representativeness, and comparability. The first four can be defined in a quantitative terms, while the later two are qualitative. MQOs are listed in table 3.0.

*Precision*: A measure of mutual agreement among multiple measurements of the same property, usually under prescribed similar conditions. Precision will be evaluated through auditing of data collection activities to determine whether activities are performed in a consistent manner, and by established protocol.

*Accuracy*: The degree of agreement between a measurement (or an average of measurements of the same thing), and the amount actually present.

*Completeness:* For this QAPjP, completeness is the measure of the number of valid samples obtained compared to the amount that is needed to meet the DQOs. The completeness goal is 90%.

*Detectability*: The determination of the low-range critical value of a characteristic that a methodspecific procedure can reliably discern or is necessary to meet program objectives.

*Representativeness*: Express the degree to which data accurately and precisely represent characteristics of a population, parameter variations at a sampling point, a process condition, or an environmental condition.

Comparability: Express the confidence with which one data set can be compared to another.

#### 3.2 Field MQOs

The following information describes the procedures used to control and assess measurement uncertainty occurring during the field sampling. Field parameters in this section will include location, coho length, coho weight, and coho age. Since these measurements are straightforward, the measurement quality evaluations will be simple remeasurements.

The majority of the uncertainties occurring in the field can be alleviated by the development detailed standard operating procedures (SOPs), an adequate training program at appropriate frequency, and a field audit program. SOPs have been developed (appendices A and B) and training has occurred. Field audits will be implemented during the course of the program implementation.

#### 3.3 Precision

Another term for precision is repeatability. Repeatability in the field is very important to precision, as well as data comparability. Repeatability is controlled by the development of detailed SOPs and adequate training in those SOPs. Field precision will be checked by remeasuring 5% of the samples. Remeasurements must be within the acceptance criteria as stated in Table 3.1. Field precision can also be evaluated through the implementation of field technical systems audits. These audits will be used to evaluate the adherence to the SOPs. Audits are discussed in section 8.

#### 3.4 Accuracy

As stated earlier, accuracy is based on the difference between an estimate, derived from data, and the true value of the parameter being estimated. For the field measurements, with the exception of location, the true value is dependent on the calibration of the instrument (ruler or scale).

Following calibration procedures and precision requirements will provide an indication of accuracy. Following SOPs as written should reduce contamination as much as possible. Accuracy is also based on training. Therefore, during audits the trainer will remeasure 5% of the samples to determine accuracy. If accuracy requirements are not met, the trainer will review the methods with the sampler until agreement is reached.

#### 3.5 Detectability

Detectability in this study is a function of how accurate and repeatable the measuring instruments can be maintained. Rulers or tape measurements, unless broken, will be considered accurate. Therefore, detectability of coho length is a function of following the SOPs. Similarly, scales, if calibrated properly, should reflect an accurate weight unless various conditions (wind or rain) create a situation where an accurate weight (within detectable limits) cannot be met. The SOPs will discuss ways to measure samples within the detectability requirements.

#### 3.6 Completeness

Completeness for the field is defined as the successful collection of all viable samples in the appropriate time frame. A viable sample would be defined as any single sample whose integrity has not been effected during the collection process and would therefor not be flagged with a field qualifier. In some cases, the sampler has no control on the integrity (e.g., samples remaining in the sun too long) while in other cases the sampler might effect the integrity (e.g., contaminating a sample through improper handling).

In any case, the DQOs are based on the evaluation of a statistically relevant number of samples which are effected by all errors occurring in the field and laboratory. Therefore, the overall goal is a completeness of 90%. The completeness objective for the measurement phase is 100%. As with the other data quality attributes, completeness can be controlled through the adherence to the SOPs in order to minimize contamination and sampling errors.

#### 3.7 Representativeness

Representativeness, with respect to the overall program objectives is a function of the statistical sampling design and how well this design estimates the measurement parameters to this project. Variation in coho diet is expected seasonally but also from year-to-year, depending on the abundance of prey and environmental factors that might affect feeding behavior. Since the sampling period for this project is only one year, the review of past coho diet data will assist in determining how representative the 1994 diet of coho salmon is to the yearly variation that can be expected.

#### 3.8 Comparability

Comparability will be maintained by the adherence of the SOPs. Adherence of these SOPs by all samplers will allow for comparability of data among sites and throughout the project. Evaluation of comparability occurs through the implementation of the training program and the field technical systems audit.

Parameters	Sample Type	Frequency	Acceptance; Other Corrective Action
Location			The accuracy required is to regions of the lake.
Coho Length Precision	Remeasurement	5%	1 cm of original measurement - recalibrate instrument and remeasure sample to compare to closest.
Accuracy	Independent remeasurement	5%	1 cm of original measurement - review protocols and remeasure another sample
Completeness		NA	90%
Coho Weight Precision	Remeasurement	5%	0.1 Kg of the original measurement - recalibrate instrument and remeasure sample to compare to closest.
Accuracy	Independent remeasurement	5%	0.1 Kg of original measurement - review protocols and remeasure another sample
Completeness		NA	100% for salmon collected for contaminant analysis 0% for salmon collected only for diet analysis
Coho Age Precision	Length Frequency	100%	Confirmation with scale aging
	Re-age, inspection	5%	Direct match with original
Accuracy	Independent Re-age, inspection	5%	Direct match with original
Completeness		NA	
Diet Species Precision	Re-identify, inspection	5%	95% identification, precision will be maintained through training and periodic audits to verify accuracy of identification of prey items
Accuracy	Re-identify, inspection	5%	95% identification, to determine accuracy, samples will be re-identified and compared to reference samples.
Completeness		NA	

# Table 3.0. Measurement Quality Objectives for Parameters for the Evaluation of Coho Salmon Diet

Parameters	Sample Type	Frequency	Acceptance; Other Corrective Action
Diet Item Length Precision	Remeasurement	5%	+/- 2 mm of original measurement - recalibrate instrument, remeasure sample and compare to closest
Accuracy	Independent remeasurement	5%	+/- 2 mm of original measurement - review protocols and remeasure another sample
Completeness		NA	90%
Diet Item Weight Precision	Remeasurement		0.1 g of the original measurement - recalibrate instrument and remeasure sample to compare to closest
Accuracy	Independent Remeasurement		0.1 g of the original measurement - review protocols and remeasure another sample
Completeness		NA	90%

Table 3.0. Measurement Quality Objectives for Parameters for the Evaluation of Coho
Salmon Diet

## 4.0 Site Selection and Sampling Procedures

A site-specific sampling plan for coho salmon is not available prior to the sample period since it depends on the migration patterns of the salmon and how that pattern is affected by environmental factors. In each of the three seasonal periods (spring, summer, and fall), we will sample coho where ever they happen to be in their migration pattern. The exact location of our sampling will also be determined by the location the anglers who caught the fish chose to fish on any given day. Table 1.0 outlines the anticipated sampling regions by season.

#### 4.1 Sampling Procedures and Sample Custody

Detailed sampling procedures can be found in Appendix A. Method summaries are presented in this section.

4.2 Contaminant Sampling

We plan on collecting all the coho salmon used in contaminant analysis from contracted sport charter anglers or on board USFWS vessels. The field sample preparation procedures will follow the SOP guidelines. A Service biologist will be onboard during all the fishing to insure proper handling of the samples. After capture, the stomach of a coho salmon will be removed in such a way that all body fluids will be captured in the aluminum foil that the fish will be frozen in for analysis. After the fish has been put in the storage bag and labeled, it will be kept on ice until it can be frozen within 24 hours after capture. The samples will be transported frozen in a cooler to the Green Bay Fishery Resources Office where they will logged and placed in a chest freezer until delivery to the Great Lakes Center in Ann Arbor, MI. All samples will be delivered by Service

vehicle. Each transfer to a new location will be recorded on the sample collection sheets (Appendix C) and each sample will be labeled individually and recorded on a summary data sheet.

#### 4.3 Diet Analysis

Diet samples may be collected from contracted sport charter anglers, sport anglers, or from assessment activities of the USFWS. Each fish sampled only for diet will have the stomach removed as soon after it was caught as possible. The stomach will be placed in individually numbered whirl-pac bags, preserved with 10-15% formalin, recorded on a summary data sheet, and stored in a sealable five gallon plastic bucket. Diet samples will be transported to the GBFRO for analysis. Chain-of custody procedures for transported samples will be the same as those mentioned above.

The GBFRO is a small developing office and all staff will be involved in the sampling in some way. Those individuals include, Mark Holey, Robert Elliott, Stewart Cogswell, Pat Bouchard, and Bruce Peffers. These biologists will collect all field samples and prepare the field labeling of the samples. Each sample will be clearly identified with date, location, species, length, weight, and sampling gear (see attached table example).

## 5.0 Analytical Procedures and Calibration

Analytical procedures will follow those outlined in Bowen 1083, Elliott 1994, and Miller and Holey 1992. Standard Operating Procedures for the laboratory activities are included in the *SOP for Lab Analysis of Coho Salmon Stomachs and Data Entry*.

## 6.0 Data Reduction, Validation, and Reporting

The responsibility for data reduction, validation, and reporting will be shared between Mark Holey and Robert Elliott. This section is intended to describe the step by step procedure used to reduce the raw diet data into summary statistics, verify those statistics, and report them as products that describe the diet of coho salmon in the manner required for this project.

#### 6.1 Overview and Summary of Method

The raw data as entered and described in SOP-2 will be reduced so that the average diet of all coho within a given strata (age-region-season) can be reported. Diet will be reported for both coho that were sampled for contaminants, and for all coho sampled during this project. The primary descriptive statistic calculated and reported will be the percent that each prey type contributes to the average wet weight of all prey found in the stomach. The range and frequency distribution individual weight values and percent weight values from which the average values are calculated will indicate the variance associated with these data. The range and distribution of site specific and biological variables will characterize the coho sample within each major strata. Length distributions of prey fish in the diet will describe the characteristics of each species found in the stomachs of coho.

Data collected and results reported during other diet studies of Lake Michigan coho will be summarized to provide a framework with which to ascertain how valid and representative the diet information collected during this project is. It is assumed that the sampling design will provide a sample of coho having characteristics (including diet) that are representative of all coho available for capture by anglers, and that collected samples will be representative of the entire strata. Therefore, although variables such as date, general location, depth, time, temperature, sex, exact location, and gear etc. will vary within a strata, determining their effect on diet will not be necessary for this project.

#### 6.2 Reduction Procedures

Methods of data analysis will generally follow those outlined in the Lake Michigan Technical Committee's document entitled "Conducting Diet Studies Of Lake Michigan Piscivores, A Protocol" (Elliott et. al 1996).

In brief, using the database developed in SOP-2, calculate the percent that each prey type contributes to the average wet weight of all prey found in the stomachs of coho salmon as follows.

Within each strata (age, region, season), group coho and their associated data by general location (port) and date specific groups. This will generally result in groups of data that will describe the diet on a weekly basis in each region of the lake.

For each of the location-date specific groups, calculate the average weight (0.1g) per stomach, and percent (0.1%) of the total weight, for each prey category. Also calculate the percent (1%) of the stomachs found empty or void of prey. Omit data flagged as outliers from these and subsequent calculations.

Compute a grand average of all location-date specific average weight values. Then calculate the percent that these average prey weights are of the total grand average weight of all prey combined.

For each strata, calculate the range and the frequency distribution of individual weight values and percent weight values for each prey species. If necessary, adjust the weight value intervals to reflect fresh weights using conversion formula determined in SOP 2.4.3.

For each strata, calculate the range and the frequency distribution of prey lengths for each prey fish species. If necessary, adjust the lengths to reflect fresh lengths using conversion formula determined in SOP 2.4.3.

For each strata, calculate the range and frequency distribution of site specific and biological variables (coho length, weight, sex; time, water depth, capture depth, temperature, where captured etc).

Maintain updated/backed up independent copies of the reduced data (hard drive, disk, and hard copy printout) in the same manner as is done for the raw database (SOP 2.4.4) for the duration of the project.

#### 6.3 Validation Procedures

Verification of the raw database is described in SOP 2.4.4. Validation of reductions/calculations is divided into two procedures: validation of correctness, and validation of representativeness.

#### 6.4 Validation of Correctness

Reductions/Calculations result from manipulations of the database by a personal computer using a set sequence of commands and formula (a program). This ensures that all reductions/calculations are consistent and not subject to random error. Verify that the values resulting from the reduction/calculation procedures are correct by reproducing by hand the process carried out by the computer for a randomly selected portion of the database.

6.5 Validation of Representativeness

To determine if the results of the reductions/calculations of this data set are representative of the diet of coho in Lake Michigan for this year and for other years in recent history, data collected and results reported during other diet studies of Lake Michigan coho will be summarized and compared to the results produced from this database.

6.6 Reporting Procedures

For each strata, report graphically and/or in table form the following:

- The percent that each prey type contributes to the average wet weight of all prey found in the stomach.
- The range and frequency distribution individual weight values and percent weight values from which the average values are calculated.
- The range and distribution of site specific and biological variables.
- Length distributions of prey fish in the diet will describe the characteristics of each species found in the stomachs of coho.

Summarize the results of data collected and results reported during other diet studies of Lake Michigan coho and contrast and compared to the results produced from this database.

Raw data in paper and electronic medium, and copies of the reports generated from the data will be stored at the GBFRO for a minimum period of five years.

## 7.0 Internal Quality Control Checks

Quality assurance for this project will be achieved primarily through specific training both prior to sampling and during the sampling season. Several persons on the GBFRO staff are experienced in diet sampling (Miller and Holey 1993, Elliott 1994), and will provide training sessions on procedures in the SOPs and parameter measurement requirements in Table 1.1 before the sampling begins and while in progress. Field staff will work in pairs with experienced staff until such a time that the quality of their work justify them working independently. The quality of field staff work will be checked periodically throughout the project duration, roughly once or twice per month. The field staff hired will be required to have completed six credits of fishery related college course work and 12 credits of related natural resources or animal science courses, or have appropriate equivalent work experience.

Measurements of length and weight required for this project are straight forward, and their variation will be a function of the ruler or weight scale used than the person taking the measurement. The rulers or measuring boards will be examined prior to the field season to ensure the error between them is less than +/- 2 mm. The weight scales used for this project will be standardized against standard weights at the beginning of the project and compared to each other throughout the sampling period. The readability of the scales used is 0.1 g for small fish and prey types measured in g, and 50 grams for large fish measured in Kg.

## 8.0 Performance and Systems Audits

Specific Audits will not be conducted as part of this sampling project. Procedures required for this project are straight forward and not complicated. The duration of the project is also short enough that the periodic checks on performance of the field and lab staff will serve as audit checks for this project. The amount of staff involved in this project will be few, therefor, the ability to control the quality of the project will not require elaborate auditing procedures. Quality control audits at each stage of the field sampling and analysis will be conducted by the Project Manager, the Field Manager, or the EPA QA Manager. Audit reports will be kept on file at the GBFRO and available for review at any time.

Inadequacies in sampling procedures or the quality of the data collected will be addressed immediately by the Project Manager or Field Manager when discovered. All previous and current data collected by the person when the inadequacies will be review for accuracy. Additional training and supervision will be provided until the quality of work is appropriate.

## 9.0 Calculation of Data Quality Indicators

This QA Plan has defined the DQOs and MQOs (Section 3). This section describes the statistical assessment procedures that are applied to the data and the general assessment of the data quality accomplishments.

#### 9.1 Precision

The precision will be evaluated by performing duplicate analyses. Various types of duplicate samples are described in Section 3. Precision will be assessed by relative percent difference (RPD)

9.2 Relative Percent Difference (RPD)

$$RPD = \frac{(X_1 - X_2) * 100}{(X_1 + X_2)/2}$$

Relative standard deviation (RSD) may be used when aggregating data.

9.3 Relative Standard Deviation (RSD)

 $RSD = (s/\overline{y}) \times 100$ 

*Where:* s = standard deviationy = mean of replicate analyses

Standard deviation is defined as follows:

$$s = \sqrt{\sum_{n=1}^{n} \frac{(y_i - \overline{y})^2}{(n-1)}}$$

Where:  $y_i = measured value of the I the replicate$ y = mean of replicate analysesn = number of replicates

9.4 Accuracy

Accuracy will be based upon expert remeasurements of a percentage of samples.

Accuracy will be evaluated by determining whether the measurements are within the acceptance limits. Deviations beyond the acceptance criteria could be justification for retraining technicians.

Bias can be estimated from the theoretical "true" value of the expert measurement. "System" bias for the study may be calculated from individual samples and is defined:

$$Bias = \frac{\sum (Y_{ik} - R_i)}{n}$$

Where:  $Y_{ik}$  = the average observed value for the ith audit sample and k observations.  $R_i$  = is the theoretical reference value n = the number of reference samples used in the assessment

9.5 Completeness

Completeness for most measurements should be 90%. Completeness is defined:

$$Completeness = \frac{v}{n} \times 100$$

Where: V = number of samples judged valid n = total number of measurements necessary to achieve project objectives

The 90% goal means that the objectives of the survey can be met, even if 10% of the samples are deemed to be invalid. An invalid sample is defined by a number or combination of flags associated

with the sample. This value will be reported on a annual basis.

9.6 Representativeness

Based upon the objectives, the three seasonal collections (spring, summer, fall) represent different coho diet conditions. In order to determine whether a change is statistically significant, the samples must be representative of the population, and the samples must be collected and analyzed in a consistent manner.

Representativeness will be evaluated through variance estimates of routine sample in comparison to previous years estimates. These estimates can be performed at within-site and between-site levels. Analysis of variance (ANOVA) will be used to determine whether variances are significantly different.

#### 9.7 Comparability

Comparability is very similar to representativeness in that comparability is ensured through the use of similar sampling and analytical techniques. Comparability will be assessed through the evaluation of precision and accuracy measurements and technical systems audits.

## **10.0 Corrective Action**

Corrective actions are discussed in Table 1.1, the internal quality control section (7.0), SOPs, and in the performance and systems audit section (8.0). The Project Manager and the Field Manager will initiate corrective actions. Corrective actions will be documented in audit reports, through data flags, and revisions to the QA plan if methods are changed.

Table 10.0 List of Data flags								
LAC	laboratory accident	There was an accident in the laboratory that either destroyed the sample or rendered it not suitable for analysis.						
FAC	field accident	There was an accident in the field that either destroyed the sample or rendered it not suitable for analysis.						
ISP	improper sample preservation	Due to improper preservation of the sample, it was rendered not suitable for analysis.						
AVG	average value	Average value-used to report a range of values.						
UNK	unknown sex	In the case of species, indicates undetermined sex.						
EER	entry error	The recorded value is known to be incorrect but the correct value cannot be determined to enter a correction.						
OTL	data point outlier	When a series of data are plotted and analyzed, this point is obviously not within the normal distribution of the data, and eliminated from further analysis.						

Table 10.0 List of Data flags

## **11.0 Quality Control Reports to Management**

A progress report outlining the achievement of the Quality Assurance Objectives will be provided to the Program Manager at the end of the project. The Project Manager will be notified immediately, however, if substantive changes are made to the QAPjP. The Quality control report will include a summary of the results of audits that were conducted, data quality assessment, and the corrective actions that were taken. Quality control reports will be provided to the Project Officer and QA Manager at EPA-GLNPO and the Biota Work Group.

## 12.0 References

- 12.1 Auer, N. A. 1982. Identification of larval fishes of the Great Lakes basin with emphasis on the Lake Michigan drainage. 744 pp. Spec. Publ. 82-3, Great Lakes Fishery Commission, Ann Arbor, MI.
- 12.2 Bowen, S. H. 1983. Quantitative description of the diet, p. 325-336. In *Nielson, L. A. and Johnson, D. L. (eds.) Fisheries Techniques*. American Fisheries Society, Bethesda, MD. 468 pp.
- 12.3 Becker, G. C. 1983. Fishes of Wisconsin. 1052 pp. University of Wisconsin Press, Madison, WI.
- 12.4 Elliott, R. F. 1993. Feeding habits of chinook salmon in eastern Lake Michigan. M.S. Thesis, Michigan State University, Lansing, MI, 108 pp.
- 12.5 Elliott, R. F. and eight other authors. 1986. Conducting diet studies of Lake Michigan piscivores, a protocol. U.S. Fish and Wildlife Service, Green Bay fisheries Resources Office, Report No. 96-2.
- 12.6 Miller, M. A. and M. E. Holey. 1992. Diets of lake trout inhabiting nearshore and offshore Lake Michigan environments. J. Great Lakes Res. 18(1):51-60.
- 12.7 Nielson, L. A. and Johnson D. L. eds. 1983. Fisheries Techniques. American Fisheries Society, Bethesda, MD. 468 pp.
- Scott, W. B. and E. J. Crossman. 1973. Freshwater fishes of Canada. Bulletin 184. Fish. Res. Board Can. 966 p.

## Appendix A. Standard Operating Procedure for Sampling Coho Salmon

This SOP is intended to provide a step by step procedure for collecting measuring, preserving and transporting Coho salmon and stomach contents from coho salmon for the Enhanced Monitoring Program Lake Michigan Mass Balance.

## 1.0 Overview

Coho salmon samples will be collected at various region within Lake Michigan in order to measure contaminant concentrations in the fish tissue of PCBs, Mercury, and trans-nonachlor and to examine the diet of the salmon by evaluating the stomach contents. Specific details of the study are documented in the Lake Michigan Mass Balance work plan and in the QA project plan. Critical and non-critical associated information, as follows, will be recorded:

Critical	Non-critical
Location	Fin clip
Date of sample	Sex
Sample length	Stomach fullness
Sample weight	Sample depth
Age	Water temperature
Physical characteristics	
Capture Time	
Sample Time	
Preservation Time	

Two techniques will be used to collect samples: contaminant sampling and diet sampling. Of primary importance is the collection of fish samples for contaminant analysis which must be collected, prepared, and preserved as soon as possible for transport to the laboratory for analysis. These samples will be collected by USFWS personnel while on a chartered fishing vessel. Therefore, there is a good chance that both critical and non-critical measurements will be taken. Locational accuracy will also be much improved. Diet sampling will involve the collection of samples after they arrive from various fishing vessels and sport fisherman. Due to various types of locational equipment (some fisherman may not have sophisticated equipment), locational accuracy may be low and non-critical measurements may not be collected. However, critical measurements will occur when fish are collected and the same techniques will be used as those aboard the fishing vessel.

#### 1.1 Summary of Method

Samplers will visit the ports (weekly/daily) in the regions mentioned in the Sampling QAPjP to check for catches. Boats will be chartered as frequently as necessary in order to collect the minimum number of samples (25) for contaminant analysis is each region within the specified time frame. The following sampling activities will take place and are discussed in detail in the order listed.

- 1) Collection of sample
- 2) Size measurement
- 3) Scale collection
- 4) Stomach removal/preservation
- 5) Data reporting
- 6) Sample labeling
- 7) Sample preservation and storage
- 8) Waste disposal and clean-up
- 9) Sample shipment

#### 1.2 Safety

In any field operation, emphasis must be place on safety. Samplers must be aware of the potential safety hazards to which they are subjected. Follow all safety protocols and equipment guidelines, and be prepared for emergency situations. The sampler is responsible for his/her safety from potential hazards.

1.3 Equipment check and calibration

Check to make sure all equipment and supplies are available in required amounts. The following is a list of all needed equipment and consumables.

- 1.3.1 Serviceable Equipment
  - Fishing vessel equipped with navigational instruments and appropriate sampling gear to catch coho salmon.
  - Ice chests, including appropriate amount of ice or freeze packs
  - 5-gallon plastic bucket (diet sampling only)
  - Measuring board (mm markings required)
  - Spring or electronic scale (1-10 Kg, 0.1 Kg markings required)
  - Calibrating weight
  - Dissecting pan
  - Dissecting knives
  - Thermometer

#### 1.3.2 Consumable Equipment

- Dissecting gloves for preserving and handling fish
- Aluminum foil
- Fish storage bag
- Whirl-pac bags
- Formalin (10-15% and full strength for mixing)
- Sample labels
- Reporting sheet
- Marking equipment
- Scale envelopes
- Cleaning sponge and brush
- 1.3.3 Calibration and Standardization

Equipment necessary for calibration and the required frequency can be found in table 1.

Instrument	Calibration Technique	Frequency	Acceptance Criteria
Thermometer	Ice bath and boiling water	1/year	+/- 2 degrees
Locational Devi	eCalibration to a standard of known Long	Lat a <b>pe</b> r trip	+/25 Km
Measuring Boar	Check against second device	1/year	+/- 2 mm
Scale	Check against a standard S class we 1, 5, 10, 25 kgs.	eightsdaily	+/1 kg

 Table 1. Equipment Calibration and Required Frequency

## 2.0 Procedures

2.1 Collection Of Contaminant Samples

Contaminant samples will be collected on-board a chartered or USFWS owned vessel using angling equipment.

- 2.1.1 Throughout each season, contract charter operators to fish for coho salmon in areas where coho are currently or are most likely to be caught. Verify that chartered vessels will have on-board adequate instrumentation and gear to catch fish and establish the location, time, and depth of capture. Samples of age 1.0 coho before they are stocked into the lake will be sampled at the state fish hatcheries where they are reared.
- 2.1.2 For each coho salmon captured, record all site and sample identification data specified on the Field Data Sheet, on two I.D. Labels, and on a whirl-pac bag (see attached examples).

**Note:** Data recorded will include: Objective (contaminant, diet, audit) Gear, Lake, Region, Nearest Port, Lat/Long or Statistical Grid, Species, Date, I.D. number, Lake Depth/Capture Depth, Water Temperature, Time Of Capture/Time Of Sampling, Field Qualifier Flag, Collectors Name.

Immediately after capture:

2.1.3 Determine and record:

Maximum Total Length (mouth closed and caudal fin dorso-ventrally compressed to nearest mm) using the measuring board.

Total Weight (0.1 kg) using the spring or electronic balance. For the hatchery sample, weigh fish with an electronic balance to the nearest 0.1 g.

- 2.1.4 Remove at least five scales (from just above the lateral line and below the posterior insertion of the dorsal fin) with a clean knife and place in the scale envelope. Record on the label the fish length, weight if taken, date, location sampled, and sample number.
- 2.1.5 Line the examination tray or measuring board with foil and place the coho on the board or in the tray. Make a 3-5 inch incision with a clean knife in the belly of the fish. Determine and record the sex and physical characteristics. Pull out and remove the stomach (anterior esophagus to pyloric sphincter) and all its contents. The spleen and any other organs that may be attached to the stomach should be removed and left inside the fish. Make a small slit in the stomach to allow preservative to enter, and place in the whirl-pac bag. If the stomach appears empty, open the stomach completely to verify that it is completely void. Indicate so on the field data sheet. Void stomachs do not need to be kept. Pack the whirl-pac bag with stomach contents on ice until you return to port where they can be safely preserved (see 2.1.9).
- 2.1.6 Maintaining all body fluids within the foil, wrap the coho completely with the foil lining the measuring board and attach one I.D. label to the foil. Place wrapped fish in a 4 mil polyethylene bag, seal the bag and attach the other I.D. label.
- 2.1.7 Place the bagged fish in a cooler and pack with ice until it can be transferred to a freezer and frozen. Verify that the samples were frozen within 24 hours by recording the date and time when the fish was captured, sampled, and placed in the freezer.
- 2.1.8 Clean/rinse all equipment thoroughly that comes in contact with sampled fish between sampling each fish.
- 2.1.9 After returning to port, preserve the stomach contents in the whirl-pac bag with at least 2X their volume of 10% formalin. Seal the bag and place in the sealable 5 gallon bucket. When handling formalin, wear rubber gloves, keep away from fish, food, and other people, stay in a well ventilated area, and thoroughly rinse with water any object or surface that comes in contact with the formalin.

- 2.1.10 Keep all samples in your possession and in their preserved state (on ice, frozen, in formalin etc.) until they have been delivered to the laboratory where the subsequent analysis will occur. For foil-wrapped coho, this is the NBS-Great Lakes Center in Ann Arbor. For preserved stomachs and all Field Data Sheets, this is the FWS Green Bay FRO. Transport only in FWS approved vehicles. With each transfer between locations, record the date and sample ID number to verify sample integrity.
- 2.1.11 Contaminant samples will be composited by the GBFRO. Samples for contaminant analysis will be taken throughout each season sampled. The five fish composites will be prepared after each season has been sampled. Each season is roughly eight weeks long (56 days). Composites will be combine as similar as fish as possible based on size, location of capture, and when possible, sex in consultation with the LMMB modelers.

#### 2.2 Collection of Diet Samples

In addition to diet samples (stomachs) collected from coho sampled for contaminant analysis, diet samples will be collected at port from various fishing vessels.

- 2.2.1 As soon as anglers/operators return to shore, obtain permission to examine and sample their catch. Permanent cleaning stations located near boat launches and marinas provide ideal locations for this sampling. To ensure that as representative a sample as possible is collected, sample from as many boats as possible over all hours of the day, and sample all coho creeled by anglers aboard an individual boat.
- 2.2.2 For all fish sampled, record all site and sample identification data specified on the Field Data Sheet, and on a whirl-pac bag (see attached examples).

**Note:** Data recorded will include: Objective (contaminant, diet, audit) Gear, Lake, Region, Nearest Port, Lat/Long or Statistical Grid, Species, Date, I.D. number, Lake Depth/Capture Depth, Water Temperature, Time Of Capture/Time Of Sampling, Field Qualifier Flag, Collectors Name.

As soon as possible after capture:

2.2.3 Determine and record:

Maximum Total Length (mouth closed and caudal fin dorso-ventrally compressed to nearest mm) using the measuring board. Flex fish several times if rigor mortis has set in so that fish lays flat on the board.

Total Weight (0.1 kg) using the spring or electronic balance (when time permits).

- 2.2.4 Remove at least five scales (from just above the lateral line and below the posterior insertion of the dorsal fin) with a clean knife and place in the scale envelope.
- 2.2.5 Make a 3-5 inch incision in the belly of the fish. Determine and record the sex and the clinical condition of the fish. Pull out and remove the stomach (anterior esophagus to pyloric sphincter) and all its contents. Return the fish to the angler/operator. Make a small slit in the stomach to allow preservative to enter, and place in the whirl-pac bag. If

the stomach appears empty, open the stomach completely to verify that it is completely void. Indicate so on the field data sheet. Void stomachs do not need to be kept. Temporarily place the whirl-pac bag with stomach contents on ice until they can be safely preserved (see 2.2.7). Stomachs from hatchery sampled fish will not be taken.

**Note:** Step 2.2.5 may be done after the fish has been filiated if the angler/operator prefers to clean the fish before the stomach is removed.

- 2.2.6 Preserve the contents in the whirl-pac bag with at least 2X their volume of 10% formalin. Seal the bag and place in the sealable 5 gallon bucket. When handling formalin, wear rubber gloves, keep away from fish, food, and other people, stay in a well ventilated area, and thoroughly rinse water any object or surface that comes in contact with the formalin. If extra personnel are available, preservation can be done as soon as the stomach contents are removed. If not, wait until all fish have been worked up, packed, and stored.
- 2.2.7 Keep all samples and data sheets in your possession until they have been delivered to the FWS Green Bay FRO. Transport only in FWS approved vehicles. Upon return to the GBFRO, make photocopies of the original Field Data Sheets to be kept on file at a location other than where the original data sheets are filed. With each transfer between locations, record the date and sample ID number to verify sample integrity.

## Appendix B. Standard Operating Procedure for Lab Analysis of Coho Salmon Stomachs and Data Entry

This SOP is intended to provide a step by step procedure for examining and quantifying the contents of the stomachs sampled, and then entering all data on the computer as part of determining the diet of coho salmon for the Enhanced Monitoring Program Lake Michigan Mass Balance Study.

## 1.0 Overview

Contents of stomachs collected from Lake Michigan coho salmon will be identified, enumerated, and weighed. Data will be recorded on data sheets and entered into a computer data base.

#### Summary of Method

Stomachs will be rinsed to free excess formalin and allow for safe handling of the sample. Fish found in the stomachs will be identified to species, assigned a percent digested state, measured and weighed. Invertebrates will be identified into the appropriate taxon and weighed as a group. The age of the fish will be determined by a length frequency analysis and a subsample will be verified through scale aging. Reconstruction of the prey length will also be used to determine reconstructed weight. The data will be entered into database (FoxPro) and spreadsheet (Lotus) software, verified, and summary reports created.

### 2.0 Safety

In any lab operation, emphasis must be place on safety. Samplers must be aware of the potential safety hazards to which they are subjected. Follow all safety protocols and equipment guidelines, and be prepared for emergency situations. The sampler is responsible for his/her safety from potential hazards.

## 3.0 Equipment Check and Calibration

Check to make sure all equipment and supplies are available in required amounts. The following is a list of all needed equipment and consumables.

#### 3.1 Equipment

Serviceable Equipment

- Fume Hood
- Rinse Water Supply and rinsing bath
- Rinse Tray
- Dissecting Tray and Tools (scalpel, forceps, scissors)
- Dissecting Microscope
- Electronic Balance and calibration weights

- Plastic Ruler (mm divisions)
- Glass Specimen Jars
- Scale Press
- Scale Projector/Reader
- Computer & Printer (with hard drive, disk drive, and necessary software)

Consumable Equipment/Supplies

- Weighing trays
- Formalin (5%)
- Rubber Gloves
- Impression Acetate
- Paper Toweling
- Plastic Bags (2-5 gal)
- Reporting Sheets and Marking devices
- 3.2 Calibration and Standardization

Equipment necessary for calibration and the required frequency can be found in Table 1.

Instrument	Calibration Technique	Frequency	Accepted Criteria
Plastic Ruler	Check against second device	Start-end/season	±1 mm
Electronic Balance	Use calibration weight methods as prescribed by scale manufacturer	Daily	±0.1 g
Computer	Virus scan	Every boot-up	No viruses

#### Table 1. Equipment Necessary for Calibration and Required Frequency.

#### 4.0 **Procedures**

The following procedures will be discussed:

- Sample preparation
- Identification and quantification of prey items
- Numeration and estimation (for invertebrates)
- Length measurement and
- Weight measurement and estimation
- Archiving representative samples
- Mounting and ageing scales
- Data Recording
- Data Entry
- Verifying Data
- Determining conversion data and developing formula

#### 4.1 Analysis of Stomach Contents

Proceed with the following steps in a well ventilated (fume hood operating if necessary) area intended for work of this nature. Wear rubber gloves when handling preserved prey items, have equipment set up, calibrated and ready for use, and start with and maintain a clean work area.

- 4.1.1 Open whirl-pac bag, pour contents into rinsing container with 365 micron mesh screen, flush with rinse water until contents are free of excess formalin, remove from rinse container and allow to drip free of excess water.
- 4.1.2 For each prey fish, identify to species, assign an estimated percent digested state, measure (nearest mm) and weigh (nearest 0.1 g for large items and 0.02 g for small prey items). For identification of fish, Becker (1983), Scott and Crossman (1973), Auer (1982), and Elliott et al. (1996) will be used as reference material. In addition, during the training period we will develop our own reference specimens for identification purposes. Record data as indicated on the lab data sheet (see attached). Measure length to level of precision allowed depending on how much of the fish is remaining. Order of priority is: 1) maximum total length, 2) standard length, 3) vertebral column length, and 4) length of as many vertebrae as possible. For those fish or parts of fish that can not be positively identified, record as unidentified.
- 4.1.3 For invertebrates, group into appropriate taxon and weigh (nearest 0.02 g). Either count directly or estimate indirectly the total number based on weight (at least 0.5 g or 25 individuals) of a known number representative of the group. Determine an average length and digested state for each taxon group. Record data as indicated on the lab data sheet.
- 4.1.4 If the identification of a prey item is uncertain, the item will be examined by a second identifier and compared to the reference collection of diet items prepared for training. If an agreement on the identification can not be reached, the prey item shall be recorded as unidentifiable.
- 4.1.5 Throughout the stomach analysis, set aside and preserve in glass jars with 5% formalin, examples of each species of prey fish and taxonomic group of invertebrate. Examples should represent the range of both digested conditions and sizes of prey observed and be able to document the methods of identification and quantification used in this analysis. Label saved samples as to their source (sample I.D. number), their identification.
- 4.1.6 Package contents back into whirl-pac bag and preserve. To facilitate easy retrieval of samples for quality control verification, package samples from similar locations and dates together (groups of 10-25) into clear plastic bags. Maintain the reference collection for identification until the final project report is accepted by EPA.
- 4.1.7 Make photocopies of each completed Lab Data Sheet and file at designated separate locations.

4.2 Aging Coho Scales

The method aging fish by length frequencies or scales, and verifying age is adequately described in fisheries Techniques (Nielson and Johnson 1983). The following highlights the procedure to use.

- 4.2.1 Prepare a length frequency histogram by 10 mm increments of all the coho samples for each season sampled. Only two year classes of coho will be in the lake at any one time, therefore separation of age by length should be obvious. Based on the length of each sample, assign an age based on the age/length frequency histogram developed. To verify the ages determined from the length frequency analysis, especially if ages overlap in length, scales will be aged.
- 4.2.2 Remove scales from the envelope and clean them in a solution of 5% Clorox in water with brush or wooden stick.
- 4.2.3 Place cleaned scales on the glass plate of a microfiche reader, add a few drops of water, and cover with a glass slide. Examine all scales to determine which scale exhibits the most representative growth pattern of the available scales. Age that scale by counting annuli observed. Record the age using the European method (stream years . lake years) on the scale envelope along with the readers initials.
- 4.2.4 To verify, re-age those fish that would have different ages assigned using the two methods. Also, re-age enough additional fish that have sizes nearest the size division indicated by the length frequency analysis so that at least 5% of all fish are re-aged. Re-aging is to be done by both the individual who originally aged the fish and a second individual who has not yet aged that fish, both using the same methods as in Section 4.2.2. Assign and record final age on the envelope based on consensus reached by both individuals or by the majority if a third independent reader is necessary.
- 4.3 Standard Measurements for Developing Conversion Equations

To allow reconstruction of total prey length and weight from partial length measures, and to allow the conversion of total length and weight of preserved prey to length and weight of fresh prey (or visa-versa), the following procedures will be followed.

- 4.3.1 For up to 50 intact individuals representing all sizes of each prey fish species (5 per 1/10 of size range encountered from preserved stomachs), measure total length and weight, dissect the fish and measure (nearest mm) the standard length, the vertebral column length, the length of as many vertebrae as possible, and count the total number of vertebrae. Record these measures on a lab data sheet identified as Standard Measures.
- 4.3.2 When in the field, the Project Field Manager will conduct independent measurements of enough stomach contents (Section 4.1) so that representing all sizes and digested states will be identified and measured prior to preservation for later lab analysis. Data will be recorded on a lab data sheet identified as Standard Measures.
- 4.3.3 Enter all data from Standard Measurements Data Sheets into database in prescribed fields.

- 4.3.4 Develop the following conversion equations with associated errors for each prey species:
  - Vertebrae length to vertebral column length and total length
  - Vertebral column length to standard length and total length
  - Standard length to total length
  - Total length to wet weight
  - Preserved total length to fresh total length
  - Preserved wet weight to fresh wet weight
- 4.3.5 Compare to similar equations developed from other studies to determine validity.
- 4.4 Data Entry and Verification
  - 4.4.1 Maintain three independent copies of the data (on hard drive, on disk, and hard copy printout) in different locations and update/backup each on a daily basis when altered.
  - 4.4.2 Enter all data from Field and Lab Data Sheets into database in prescribed fields.
  - 4.4.3 Using equations determined in 4.3, calculate missing total length measures from partial length measures and add to the database.
  - 4.4.4 Identify and correct inaccuracies in data recording and entry, and identify outliers as follows:
    - Plot data variables, identify peripheral values, and cross-reference with original data records. Example plots include:

Predator length vs. weight	Predator length vs. date (by age)
Prey length vs. date	Prey length vs. weight (by length type)

- Query all data fields for values above and below expected values and cross-reference with original data records.
- Visually compare and verify each computer record with field and lab records on original data sheets.
- Resolve with the data collector any possible errors in recording.
- Identify data points as an outlier, that after completing the above, still appears to be outside the range of expected values.

# Quality Assurance Plan for Coho Sampling for Contaminant and Diet Analysis

CHIGAN MASS BUDGET non Contaminant Sample	SEASON REGION AGE SP SU FA WE - S C N 0 1 2 LENGTH (mm)	DATE: DATE: (YEAR - MONTH - DAY) FISH # COLLECTOR I.D.	Ear comple collection information contract: TICENVC Graan Bay Eicherr Decompose Office	-	LAKE MICHIGAN MASS BUDGET/MASS BALANCE PROJECT Coho Salmon Contaminant Sample	SEASON REGION AGE	SP SU FA WE - S C N 0 1 2 LENGTH (mm)	DATE: (YEAR - MONTH - DAY) FISH # COLLECTOR I.D. SEX (M,F)	For sample collection information contact: USFWS Green Bay Fishery Resources Office Mark Holey - Project Leader ph: 414-433-3803	LAKE MICHIGAN MASS BUDGET/MASS BALANCE PROJECT Coho Salmon Contaminant Sample	SEASON REGION AGE	SP SU FA WE - S C N 0 1 2 LENGTH (mm)	DATE: ( YEAR - MONTH - DAY ) FISH # COLLECTOR I.D.	For sample collection information contact: USFWS Green Bay Fishery Resources Office Mark Holey - Project Leader ph: 414-433-3803
CHIGAN MASS BUDGET non Contaminant Sample	SEASON REGION AGE SP SU FA WE - S C N 0 1 2 LENGTH (mm)	DATE: DATE: (YEAR - MONTH - DAY) FISH # COLLECTOR I.D.	Exercision information contract:     INSERVIC Green Back (M.,F')		LAKE MICHIGAN MASS BUDGET/MASS BALANCE PROJECT Coho Salmon Contaminant Sample	SEASON REGION AGE	SP SU FA WE - S C N 0 1 2 LENGTH (mm)	DATE: (YEAR - MONTH - DAY) FISH # COLLECTOR I.D. SEX (M,F)	For sample collection information contact: USFWS Green Bay Fishery Resources Office Mark Holey - Project Leader ph: 414-433-3803	LAKE MICHIGAN MASS BUDGET/MASS BALANCE PROJECT Coho Salmon Contaminant Sample	SEASON REGION AGE	SP SU FA WE - S C N 0 1 2 LENGTH (mm)	DATE: (YEAR - MONTH - DAY) FISH # COLLECTOR I.D.	For sample collection information contact: USFWS Green Bay Fishery Resources Office Mark Holey - Project Leader ph: 414-433-3803

			FLAG																
	e	lay FRO ta Sheet	FREEZE	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
GEAR	Date Page	USFWS Green Bay FRO Field Data Sheet	SAMPLE	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
			TIMES: PTURE	_	_	_	_	_	-	_	_	_	_	_	_	_	_	_	itered:
			TEMP CA	_	_	-	-	_	-	-	-	-	-	-	-	-	-	_	Data Entered:
		CONDITIONS	TIMES: CLINICAL-CONDITION-SCARS LOCATION: (Grid, LaULong) CAPTURE DEPTH_DOWN, TEMP CAPTURE :			_				_								_	Sheet Copied:
			LOCATION: (Grid, Lat/Lon																
VE		I	NDITION-SCARS 1	-	-		-	-	-	-	-	-	-	-	-	-	-	-	
OBJECTIVE		LOCATION		_	_	_	_	_	_	_	_	_	_	_	_	_	_	_	NBS LAB:
		<u>.</u>	STOMACH	_	_	_	_	_	_	_	_	_	_	_	_	_	_	_	
REGION	Z	COLLECTOR I.D.	Scales Head SEX	_	_	_	_	_	_	_	_	_	_	_	_	_	_	_	
	- S C N			-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
	W E -	FISH #'s		-	-	-	-	-	-	-	-	-	-	-	-	-	-	–	) Freezer:
SEASON	SU FA	DAY FIS	(_) HENGTH (	-	_	_	-	_	_	_	-	-	-	-	-	-	-	_	istory: GBFRO
SEA	SP	HLNOW	ES WEIGHT	_	_	_	_	_	_	_	_	_	_	_	_	_	_	_	Transfer DATE and TIME History: GBFRO Freezer:
LAKE	Ш	YEAR	FISH # SPECIES	_	_	_	_	_	_	_	_	_	_	_	_	_	_	_	Transfer DAT

#### Volume 1, Chapter 5

H#'s OLLECTORLB. LOCATION PRESERVED STATE vertebral 10. avertu. A. f. werd. A. f. werd. A. f. f. w	USFWS, Green Bay FRO, Lab Data Sheett	Data Sheett		<b>OBJECTIVE:</b>	VE:						<u>Shee</u> t Copied:	Data E	Data Entered: _	
**u/L       Anti * vert.       Fort * vert.       T::* * vert.       Anti * vert.         *vert.       Fort * vert.       Fort * vert.       Fort * vert.       Anti * vert.         *vert.       Fort * vert.       Fort * vert.       Fort * vert.       Anti * vert.         *vert.       Fort * vert.       Fort * vert.       Fort * vert.       Anti * vert.         *vert.       Fort * vert.       Fort * vert.       Fort * vert.       Fort * vert.         *vert.       Fort * vert.       Fort * vert.       Fort * vert.       Fort * vert.         *vert.       Fort * vert.       Fort * vert.       Fort * vert.       Fort * vert.         *vert.       Fort * vert.       Fort * vert.       Fort * vert.       Fort * vert.         *vert.       Fort * vert.       Fort * vert.       Fort * vert.       Fort * vert.         *vert.       Fort * vert.       Fort * vert.       Fort * vert.       Fort * vert.         *vert.       Fort * vert.       Fort * vert.       Fort * vert.       Fort * vert.         *vert.       Fort * vert.       Fort * vert.       Fort * vert.       Fort * vert.         *vert.       Fort * vert.       Fort * vert.       Fort * vert.       Fort * vert.         *vert.       Fort	ONTH DAY	MONTH DAY	MONTH DAY		FISH	∿# E		<b>DELECTOR I.D.</b>	LOCATION	PRESERV	ED STATE	Date Page	oť	
Wetherd (M)         Factor         Andi vect.         Real i vect.         Pact vec	E - S C N M E - S C N	I I I VILE Z												
	SPECIES D. STATE WEIGHT (0.1 gr) LENGTHS (mm): Total (TL) Standard (SL)	WEIGHT (0.1 gr) LENGTHS (mm): Total (TL)	L	L	ndard (SL)		tebral (VL)	# vert/VL		Post: # vert/L	??:# vert/L	- 1		FLAG
								_	-	_	_			
								_	_	_	_			
								_	_	_	_			
								_	_	_	_			
								_	_	-	_			
								_	_	_	_			
								_	_	_	_			
								_	_	_	_			
								_	_	_	_			
								_	_	_	_			
								_	_	_	_			
								_		_	_			
								_	_	_	_			
								_	-	_	_			
								_	_	_	_			
								_	-	_	_			
								_	_	_	_			
								_	_	_	_			
								_		_	_			
								_	_	_	_			
								_	_	_	_			
								_	_	_	_			
								_	_	_	_			
-								_	_	_	_			
								_	_	_	_			

Volume 1, Chapter 5

State of Wisconsin Department of Natural Resources	urces		Form 4100-145 CHAIN OF CUSTODY RECORD 7-87
Sample Collector(s)		Title/Work Station	Telephone No. (include area code)
Property Owner		Property Address	Telephone No. (include area code)
Split Samples: Offered? Q Yes C	0 No (Check One) 0 Yes (Check One)	(Check One) Accepted By:	
		Signature	
-			
Field Date Time ID No.	Sampie Type Comp Grab	Station Location Sample Description	Lab ID No. of Comments Number Containers
	L		
I hereby certify that I received, noted below:	d, properly handled,	and disposed of these samples as	
Relinquished By (Signature)	Date/Time	Received by: (Signature)	Disposition of Unused Portion of Sample
Relinquished By (Signature)	Date/Time	Received by: (Signature)	Dispose Retain for days
Relinquished By (Signature)	Date/Time	Recived for Laboratory By: (Signature)	Return Other

Audit Finding		
Audit Title:	Audit #:	Finding #:
-		
— Finding:		
Discussion:		

	t Finding nse Form		
Audit Title:		Audit #:	Finding #:
 Finding:			
Cause of the problem:			
Actions taken or planned for correction:			
Responsibilities and timetable for the above actio	ns:		
Prepared by:			