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Agency

Science and Ecosystem
Support Division
Region 4 and
Office of Research &
Development

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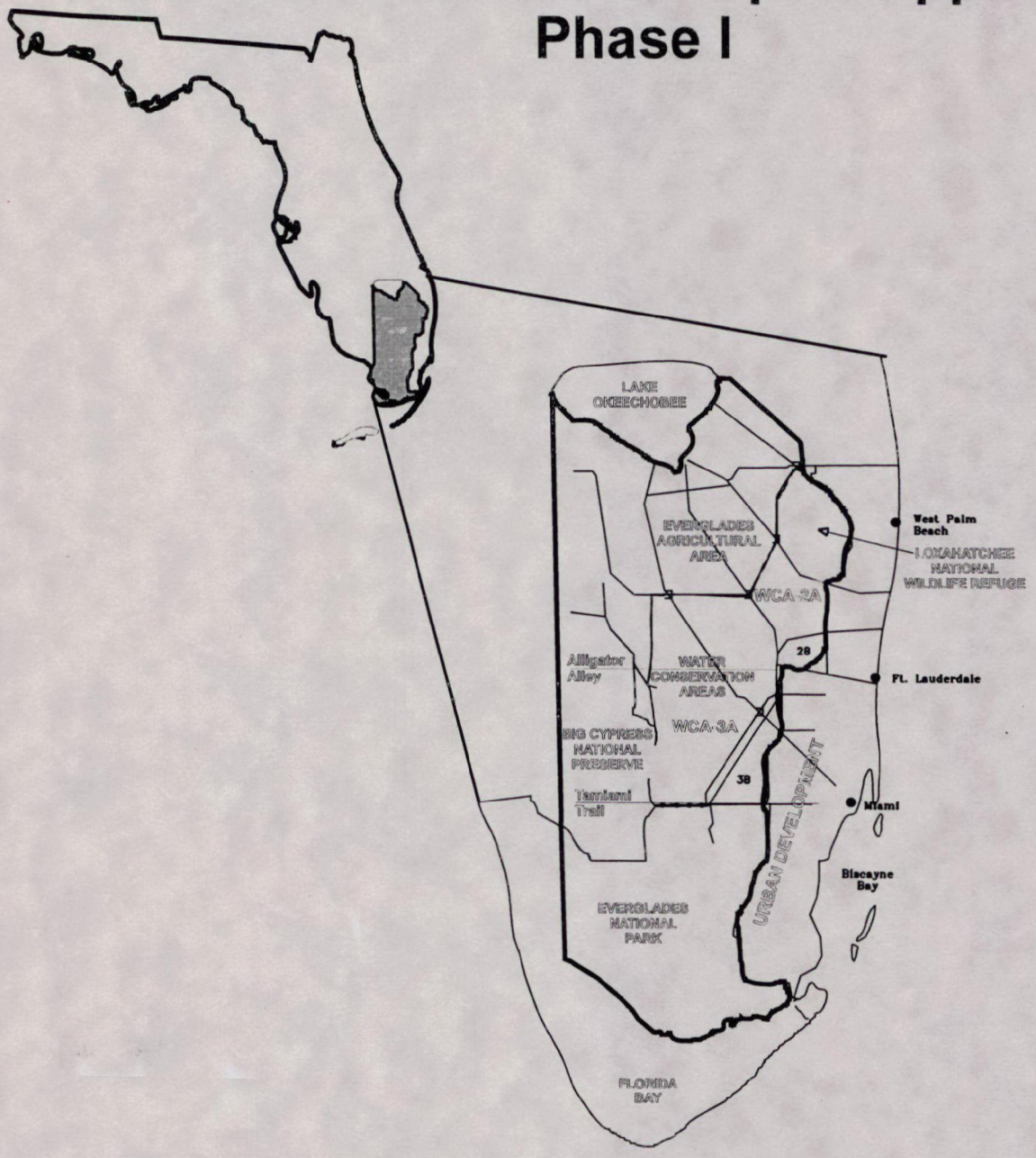
October, 1998

South Florida Ecosystem Assessment
Vol II. Final Technical Report Appendices
Phase I



Monitoring for Adaptive Management: Implications for Ecosystem Restoration

APPENDIX A

Sampling Apparatus

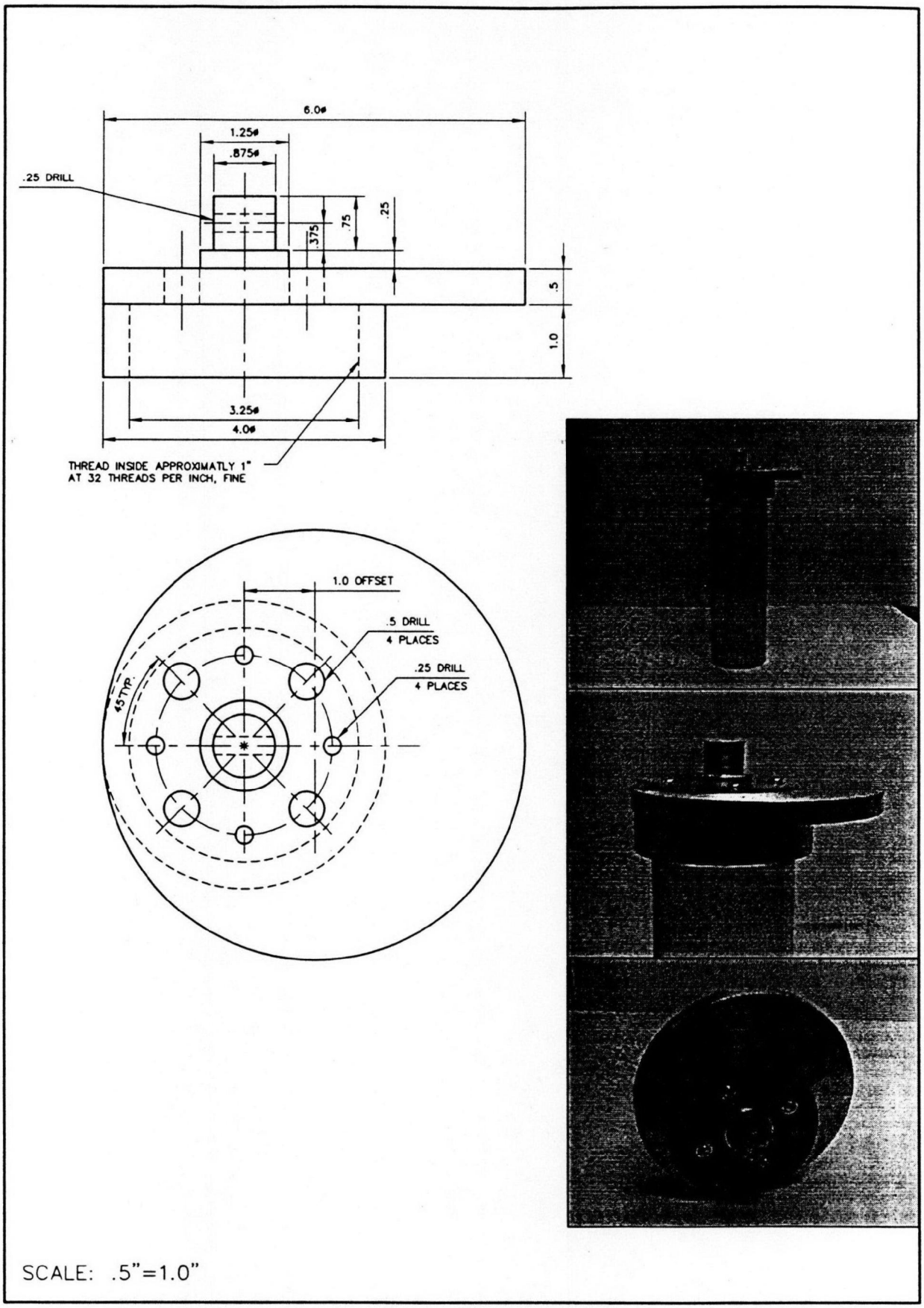


Figure A.1. Surface flange of soil cores.

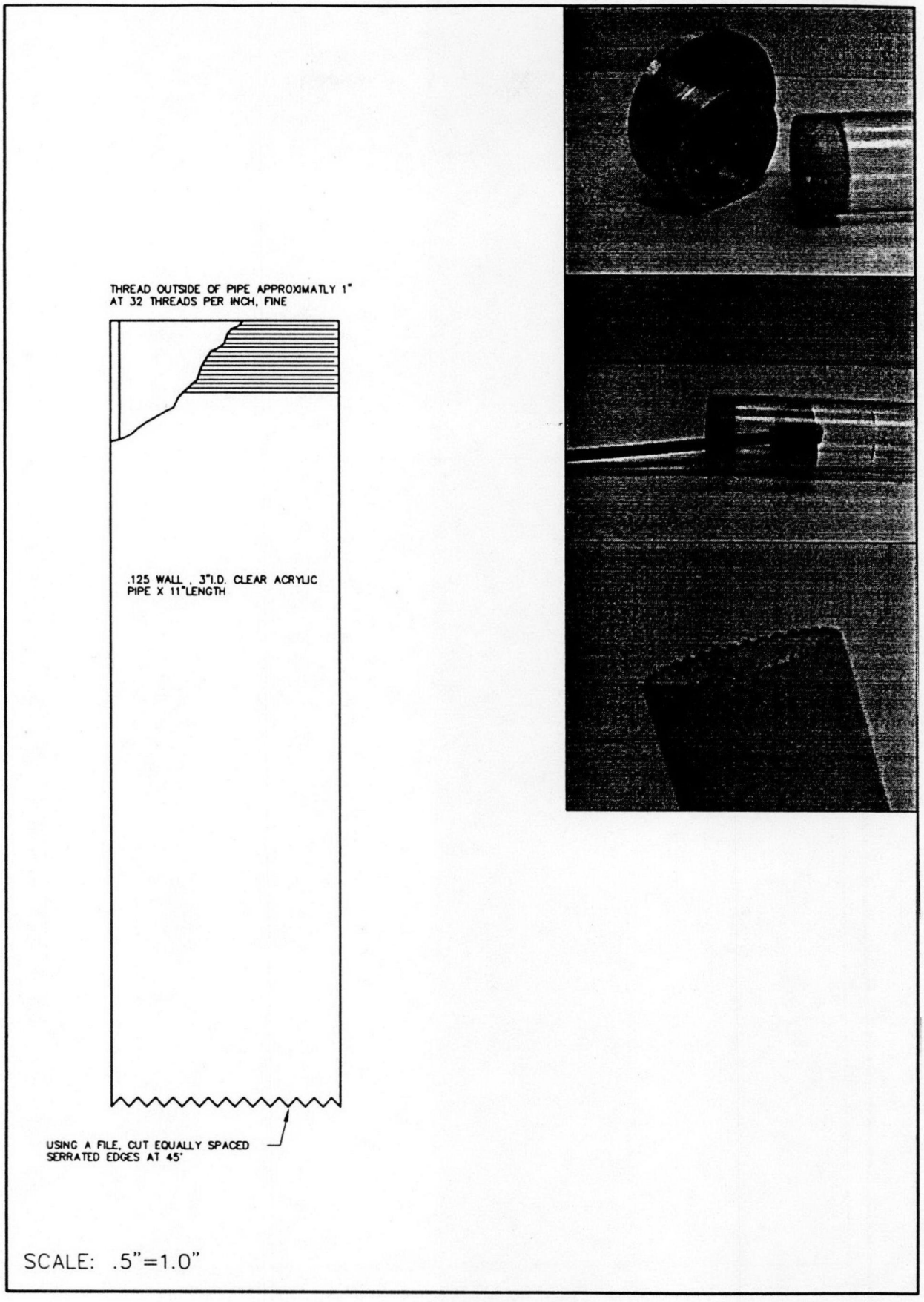


Figure A.2. Acrylic soil sampling tube.

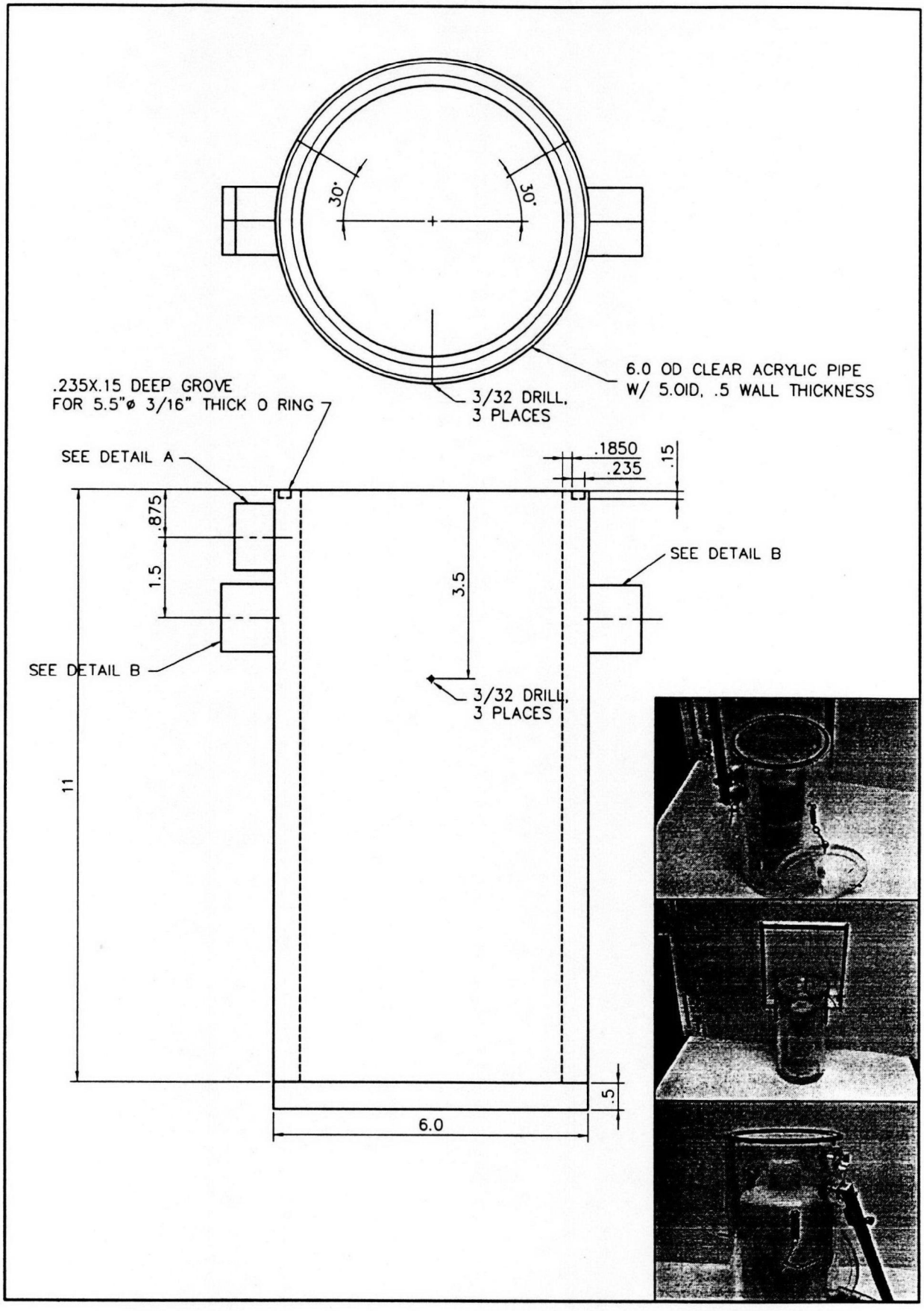


Figure A.3. Main body of vacuum chamber for vacuum water sampler.

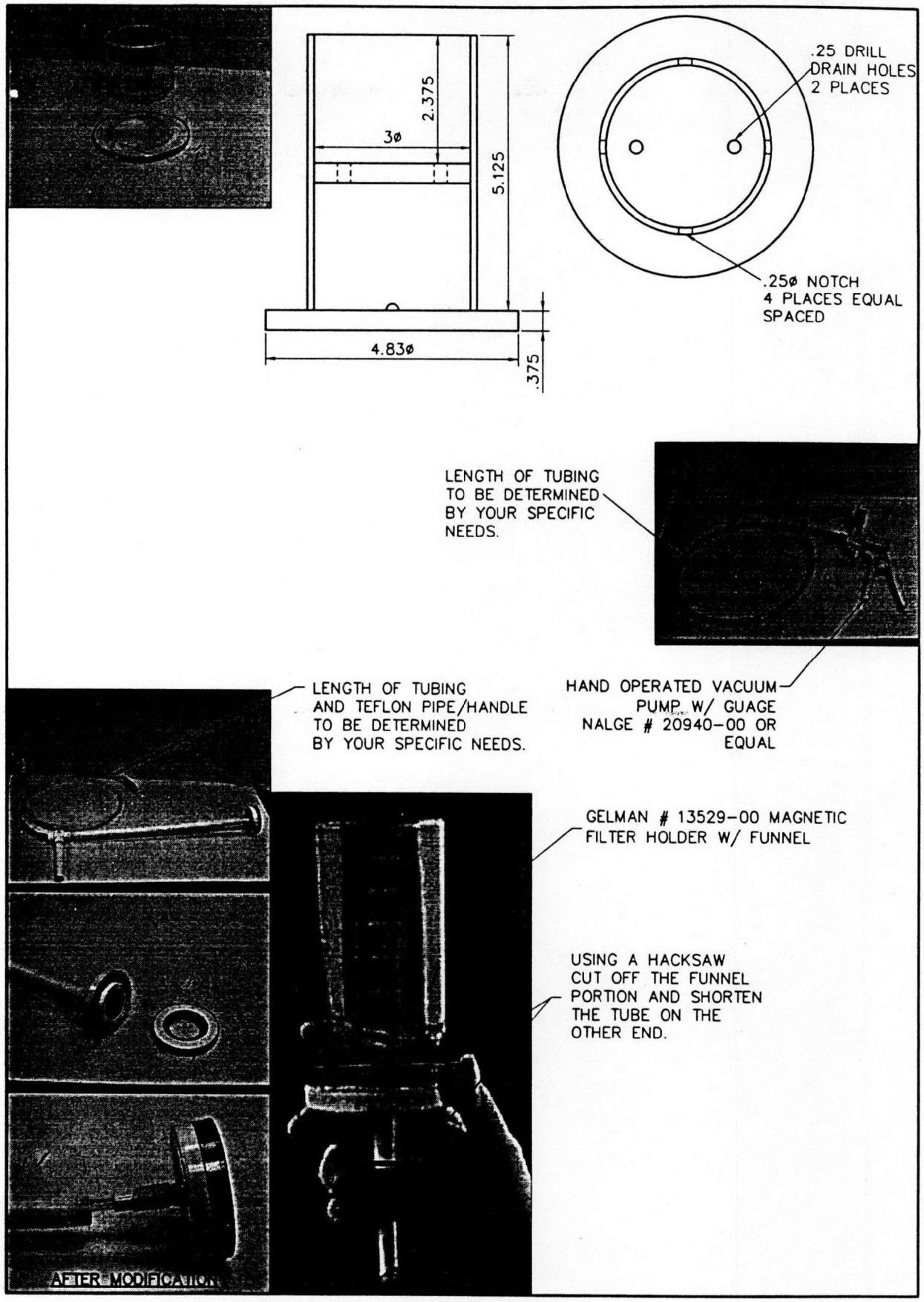


Figure A.4. Details of parts for vacuum water sampler.

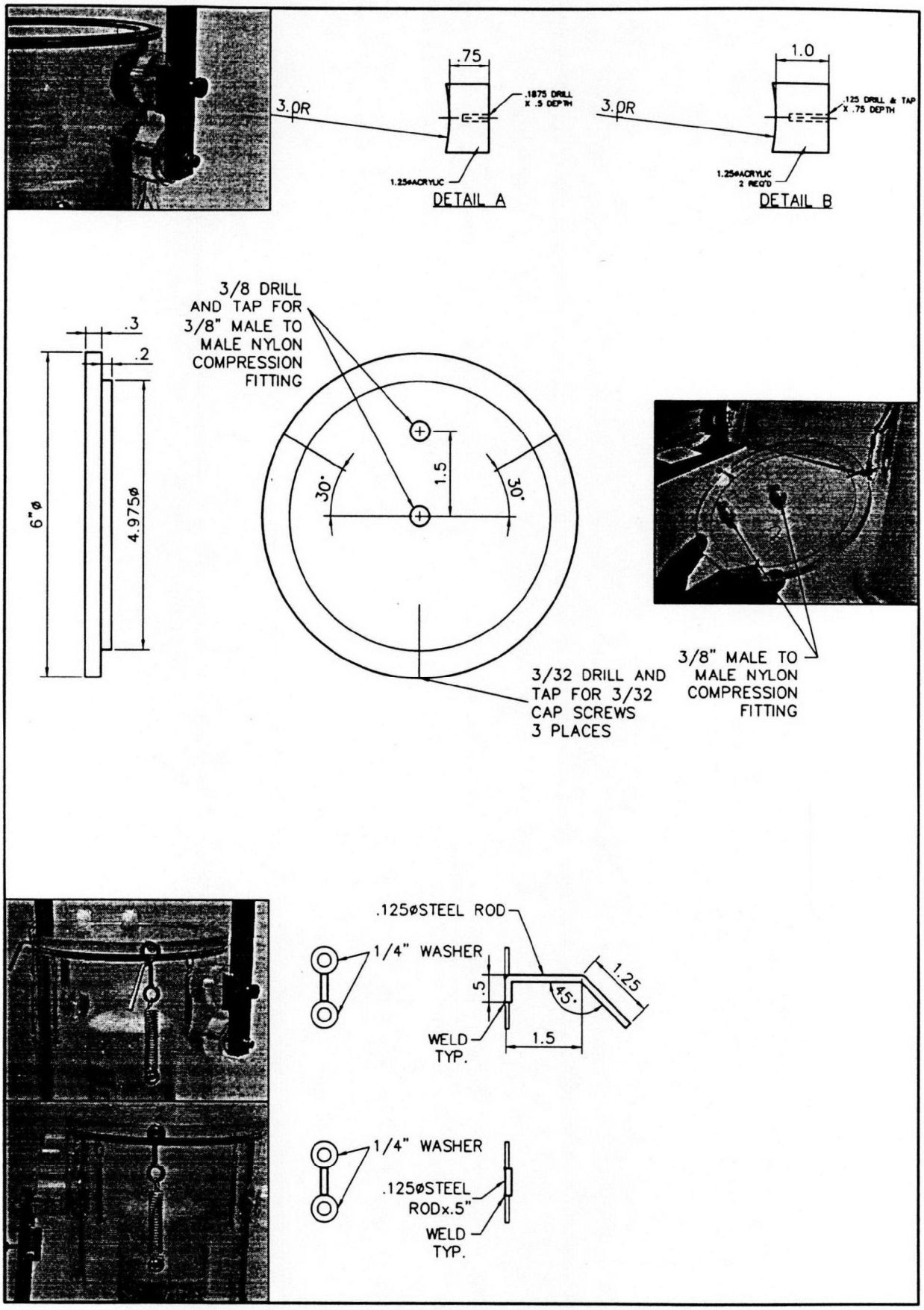


Figure A.5. Details of parts for vacuum water sampler.

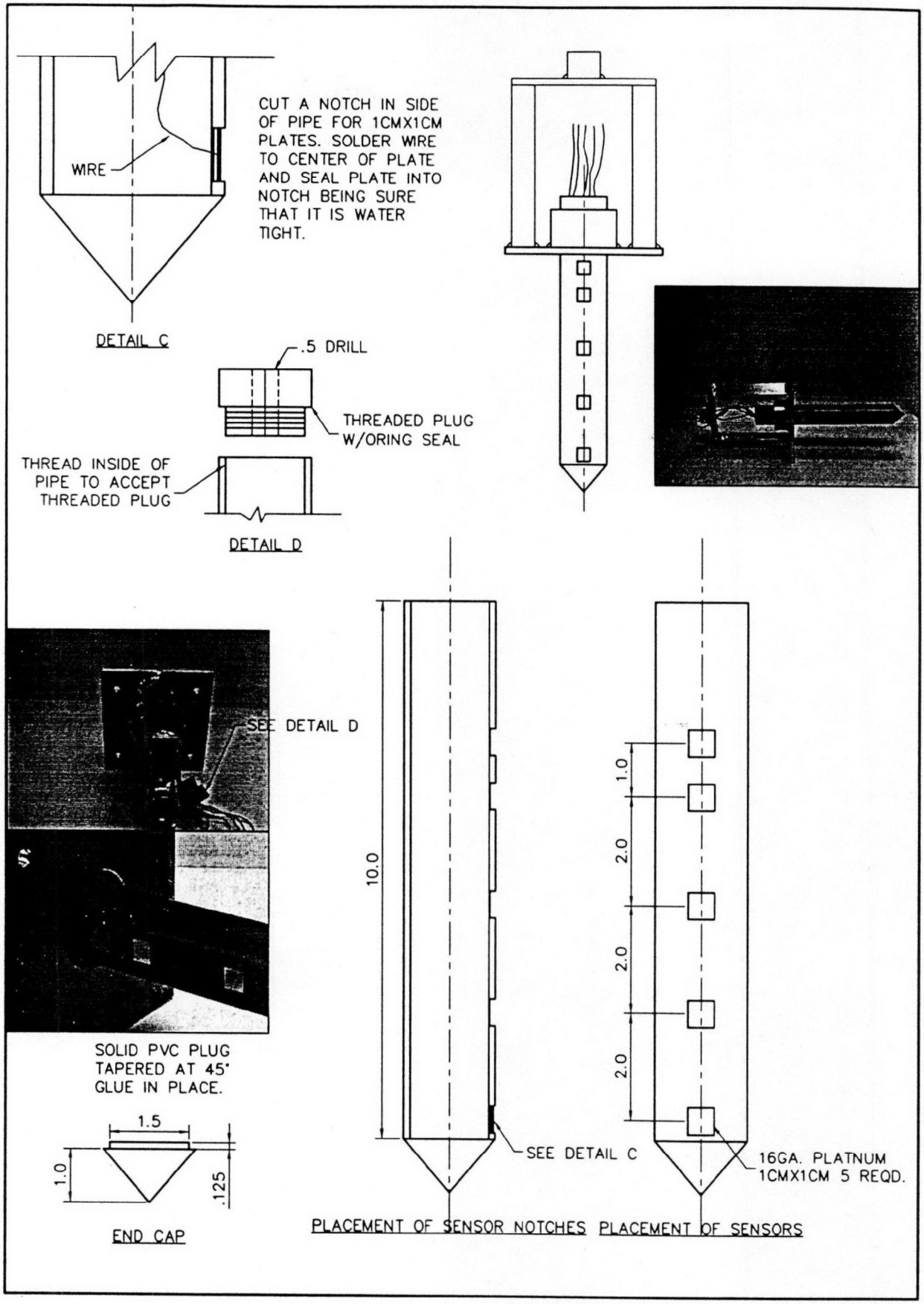


Figure A.6. Details of soil redox probe with sensors.

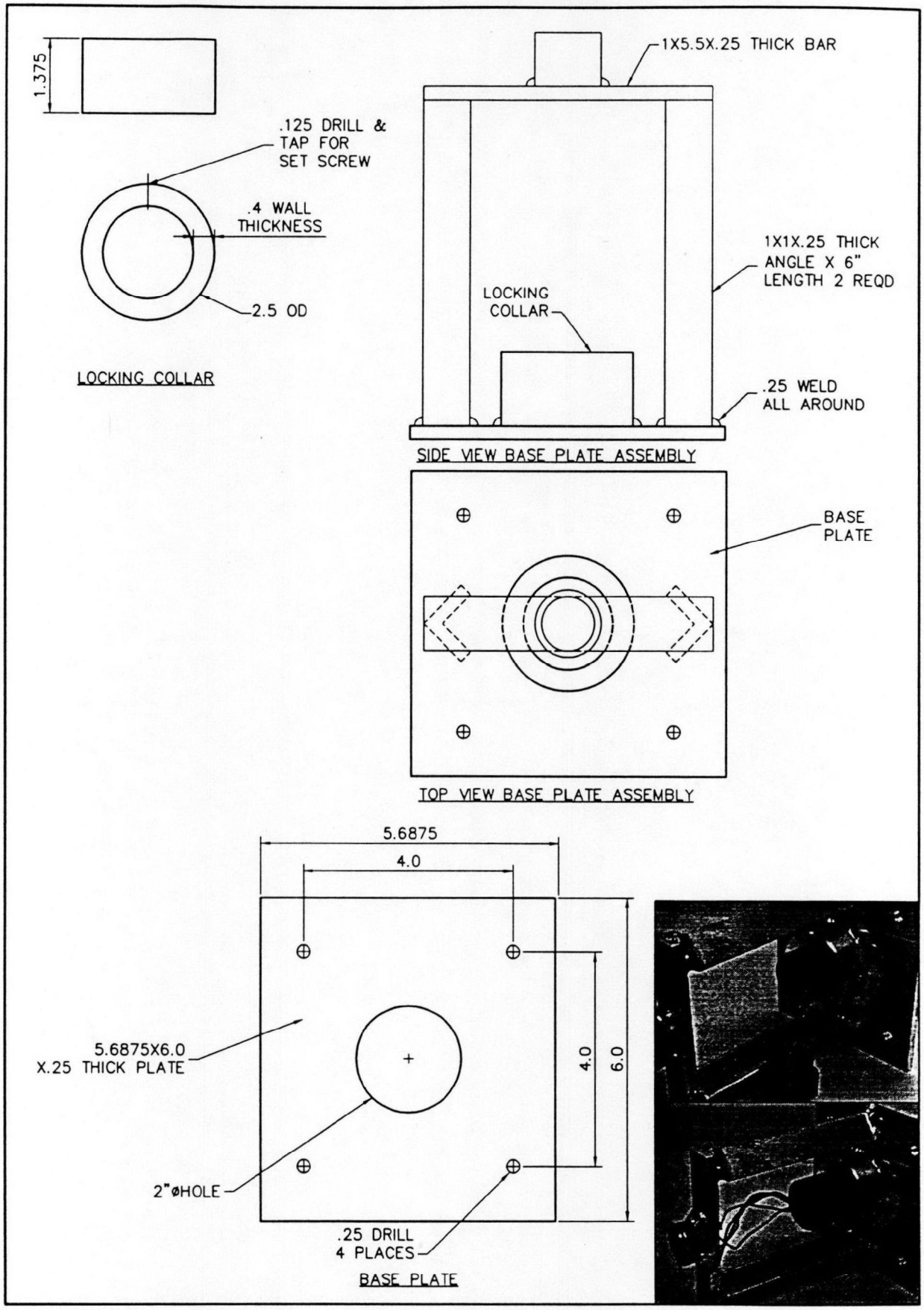


Figure A.7. Base plate assembly for redox soil probe.

APPENDIX B

Data Quality Objectives

SOUTH FLORIDA ECOSYSTEM ASSESSMENT PROJECT
DECISION-BASED DATA QUALITY OBJECTIVES

March 1997

South Florida Ecosystem Assessment Project

Decision-Based Data Quality Objectives

March 1997

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Data Quality Objectives

The Data Quality Objectives (DQOs) were prepared generally following the Guidance for the Data Quality Objectives Process EPA QA/G-4 (EPA 1994). This US Environmental Protection Agency (EPA) Guidance document, however, is not entirely appropriate for research projects. The EPA Quality Assurance Management Staff are in the process of preparing DQO guidance for research projects, but this guidance will not be available until late fall (John Warren, QAMS, personal communication). The South Florida Ecosystem Assessment Project is a research project that, in part, is developing risk-based criteria for decisions because the existing criteria are not appropriate or no criteria exist. Therefore, two separate, but complementary, approaches were used to develop DQOs. The first approach was to use the EPA QA/G-4 documentation as guidance in developing decision-based DQOs, which are discussed in this document. This document uses the EPA QA/6-4 report format. The second approach revised the DQOs originally proposed in the REMAP Research Plan (Stober et al. 1993). These revised DQOs are listed in Appendix A.

Background

In 1989, a Florida panther, an endangered species, died because of mercury toxicosis. Since then, over 2 million acres in South Florida have been placed under fish consumption advisories because of mercury contamination. The EPA Region 4 Science and Ecosystem Support Division (SESD), therefore, was charged by the EPA Regional Administrator to develop an action plan to evaluate the mercury issue and provide a scientific basis for evaluating options and strategies to eliminate mercury contamination in the South Florida Everglades Ecosystem. Subsequently, the Region 4 SESD prepared a research plan, had this plan peer-reviewed, and initiated the study as a Regional Environmental Monitoring and Assessment Program (REMAP) Project. As the Project planning and pilot Project proceeded, it became obvious that the environmental issues in South Florida (eutrophication, mercury contamination habitat alteration, hydroperiod modification) are highly interactive and need to be addressed through an integrated monitoring and assessment program. Therefore, the REMAP Project was expanded to become the South Florida Ecosystem Assessment Project addressing these multiple environmental issues. The variables being measured in this Project will permit answers to questions on these multiple environmental issues. A central goal of the Project, however, remains to answer assessment questions related to the magnitude, extent, trends, and transformation processes in mercury contamination of the South Florida Everglades Ecosystem.

State the Problem - a description of the problem(s) and specification of available resources and relevant deadlines for the study.

(1) *Identify the members of the team* - The team consists of the Region 4 Project Manager (i.e., fisheries biologist), SESD; Assistant Project Manager (natural resources manager), Water Division; Quality Assurance Officer (chemist); Southeast Environmental Research Program manager (microbial ecologist and chemist), Florida International University; spatial statistician, University of Georgia; and systems ecologist and data analysts, FTN Associates, Ltd.

(2) *Identify the primary decision maker(s)* - The primary decision maker is the South Florida Ecosystem Assessment Project Manager. Other decision makers include the Assistant Project Manager, Division Directors for the Water Division and Science and Ecosystem Support Division.

(3) *Develop a concise description of the problem* - Mercury contamination, nutrient loading, hydropattern modification, and habitat alteration are impacting fish and wildlife in the South Florida Everglades Ecosystem. The sources, causes, and interactions among many of these environmental stressors are unknown. Environmentally-sound, cost-effective restoration of the South Florida Everglades Ecosystem, however, depends on identifying these sources, causes and interactions. Almost one billion dollars are estimated to be spent on this restoration effort.

(4) *Specify the available resources and relevant deadlines for the study* - Approximately $1 million dollars/year are needed and available to determine the magnitude, extent, trends and possible causes of the mercury contamination, eutrophication, hydropattern modification and habitat alteration problems. This represents less than 0.1% of the proposed restoration expenditures. The relevant regulatory deadlines are listed in Table 1. These regulatory deadlines extend through 2004, with a major milestone in 1999 when the EPA mercury report is due to the South Florida Ecosystem Restoration Task Force.

Identify the Decision - a statement of the decision that will use environmental data and the actions that could result from this decision.

(1) *Identify the principal study questions* - The principal study questions were identified as part of the original proposal and specification of the DQOs. These seven policy-relevant questions are listed in Table 2.

(2) *Identify alternative actions that could result from resolution of the principal study questions* - The logical alternative actions and pathways that could result in answering these seven questions were identified during the initial phases of the Project. These pathways were incorporated into a Visual Basic computer program to show the logical development of these alternative actions. The expanded logic pathways from this computer program are shown in Figure 1. These logic pathways and alternative action formulations are a major part of the Problem Formulation phase of the Ecological Risk

Table 1. Mercury Related Legislative and Regulatory Deadlines.

Date	Federal	Florida
1995	NPDES Permit for the ENR project (CWA)	
1996	EIS for the Everglades Construction Project (NEPA)	
1996	404 Permit for the Everglades Construction Project (CWA)	
Oct 1997	404 Permit for STA-6 (CWA)	STA-6 NPDES Permit and 402 Certification
Sep 1998	USACOE Central & Southern Florida Project Restudy Plan Draft Report & Draft EIS (WRDA, NEPA)	
Dec 1998		Evaluation of water quality standards for the Everglades Protection Area & EAA canals (EFA)
Jan 1999	STA-1W, 2, & 5 404 Permits (CWA)	STA-1W, 2, & 5 NPDES Permits, 402 Certification (CWA)
Jul 1999	Final Restudy Report and EIS due to Congress (WRDA, NEPA)	
Dec 1999		Report to Governor and Legislature on status of EPA mercury study (EFA)
Dec 2001		Phosphorus criterion rulemaking for Everglades Protection Area and EAA canals (EFA)
Oct 2003	STA-3 & 4 404 Permits (CWA)	STA-3 & 4 NPDES Permits and 404 Certification (CWA)
Dec 2003		Revised water quality standards for the Everglades Protection Area & EAA canals (EFA)
2004	Approval of water quality standards for the Everglades Protection Area & EAA canals (CWA)	

WRDA: Federal Water Resources Development Act
EFA: Florida Everglades Forever Act
CWA: Federal Clean Water Act
STA: Stormwater Treatment Area
EAA: Everglades Agricultural Area
NEPA: Federal National Environmental Policy Act

Table 2. Policy-Relevant Questions Guiding the Project.

Status and Trends
1) What is the magnitude of the mercury problem? What are the current levels of mercury contamination in various species? What ecological resources of interest are being adversely impacted by mercury? 2) What is the extent of the mercury problem? (i.e., what is the geographic distribution of the problem? Is it habitat specific?) 3) Is the problem getting worse, better, or staying the same over time?
Diagnosis and Management
4) What factors are associated with, or contributing to, methylmercury accumulation in sensitive resources? 5) What are the relative contributions and importance of mercury from different sources (e.g., fossil fuel plants, waste incinerators, agricultural management practices, geologic pools, natural peat deposits, global atmospheric background, etc.)? 6) What are the relative risks to different ecological systems and species from mercury contamination? 7) What management alternatives are available to ameliorate or eliminate the mercury contamination problem?

Assessment Framework that forms the foundation of this study. Dichotomous trees were formulated for each of the logic pathways developed during the initial Project phases. These trees were developed prior to the initiation of the field sampling and were used to assist in the formulation of the preliminary project DQOs.

(3) *Combine the principal study questions and the alternative actions into a decision statement* - "Decide how the relative ecological risk from mercury contamination compares with the risks from nutrient additions, hydropattern modification, habitat alteration. Determine if controlling these other stressors will eliminate mercury contamination; if not, determine procedures that can be used to eliminate mercury contamination."

(4) *Organize multiple decisions* - Multi-decision pathways will be based on the outcomes from the logic pathway analyses shown in Figure 1. These logic and decision pathways will be refined as the Project proceeds and new information is collected and analyzed.

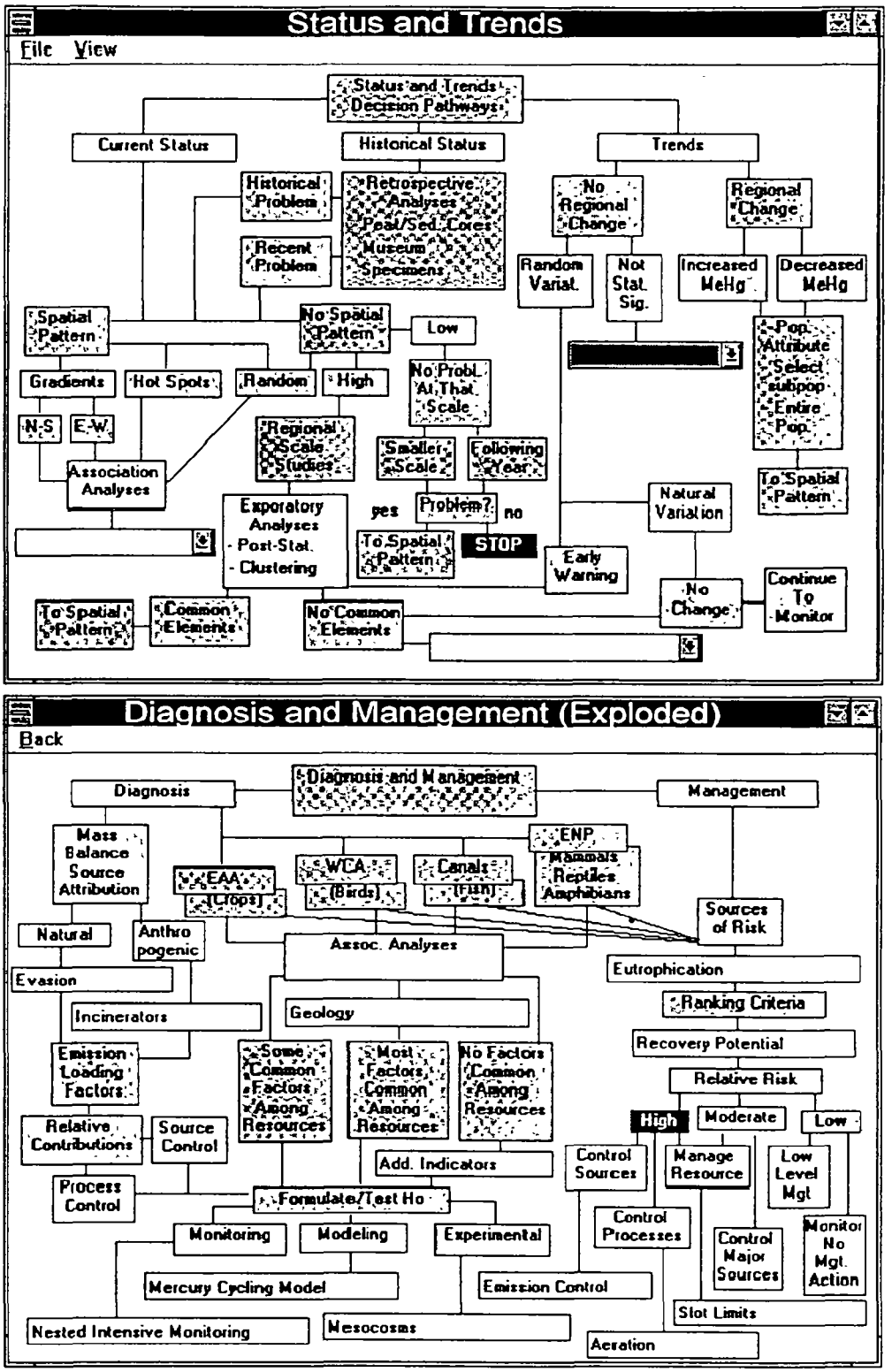


Figure 1. Logic pathways for decisions on status and Trends and Diagnosis and Management Questions. Pathways diagram information and analyses needed to answer the seven policy-relevant questions

Identify the Inputs to the Decision - a list of the environmental variables or characteristics that will be measured and other information needed to resolve the decision statement.

(1) *Identify the information that will be required to resolve the decision statement* - The information needed to resolve the decision statements is listed in Table 3.

(2) *Determine the sources for each item of information identified* - The South Florida Ecosystem Assessment Project (SFEA) is the primary source of the information needed to address the decision statements. The decision statements can not be resolved without this Project. Additional sources of information also are identified in Table 3.

(3) *Identify the information that is needed to establish the action level* - The criteria that will be used to establish the action level will be:

(a) Variability - ecological effects significantly different from natural variability
(b) Endpoints - reproduction, feeding efficiency, behavioral changes, and other ecologically relevant processes, in addition to toxicity
(c) Temporal scale - chronic versus acute effects
(d) Spatial scale - small versus large scale effects

For most constituents, regulatory criteria or standards do not exist. The decision will be made using risk-based action levels.

(4) *Confirm that appropriate measurement methods exist to provide the necessary data* - For conventional pollutants, EPA approved methods are being used to measure environmental variables with an approved QAPP. For some constituents, such as total phosphorus, existing EPA methods do not have the resolution needed to detect low-level background concentrations. For other constituents, such as methylmercury in water, soil, and sediment, there are no approved measurement methods. Therefore, experimental measurement methods are being developed for these constituents, with confirmatory analyses being conducted by independent laboratories.

Define the Boundaries of the Study - a detailed description of the spatial and temporal boundaries of the problem, characteristics that define the population of interest, and any practical considerations for the study.

(1) *Specify the characteristics that define the population of interest* - The target population or population of interest are all ecological resources in the South Florida study area. This includes the freshwater wetlands, open water and canals found in the Everglades National Park (ENP), Water Conservation Areas (WCAs), Big Cypress National Preserve (BiCY), and Everglades Agricultural Areas (EAA). The media to be sampled include soil, sediment, water, and biota. The emphasis is on mercury concentrations in biota, especially fish tissue. However, one of the desired outcomes of the Project is better estimates of the type and proportion of ecological resources and the impacts of other stressors on these resources in South Florida.

Table 3. Information Needs, Source and Method.

Measurement Variable	Source	Method
Physical Measurements		
Site location	SFEA	Global Positioning System
Weather	SFEA, NOAA	Visual observation, meteorological stations
Discharge, structure	SFWMD	Gage readings, pump capacity
Water depth	SFEA, SFWMD	Calibrated line, depth recorders
Temperature	SFEA	Thermistor
Peat depth	SFEA	Calibrated probe
Turbidity	SFEA	Turbidimeter
Bulk density	SFEA	Balance, weighing
% Mineral content	SFEA	Combustion furnace
Ash free dry weight	SFEA	Combustion Furnace
Chemical Measurements		
Dissolved oxygen	SFEA	DO probe
Specific conductance	SFEA	Conductivity meter
pH	SFEA	pH meter
Total organic carbon	SFEA	Total carbon analyzer
Total phosphorus	SFEA	New method development
Sulfate	SFEA	New method development
Total mercury	SFEA	New method development
Methymercury	SFEA	New method development
Alkaline phosphatase	SFEA	New method development
Redox potential	SFEA	Volt meter
Biological Measurements		
Resource class (canal, sawgrass marsh, cattails, etc.)	SFEA	Visual inspection
Periphyton presence/absence	SFEA	Visual observation
Chlorophyll a	SFEA	New method development
Soil/Sediment total mercury	SFEA	New method development
Soil/Sediment methylmercury	SFEA	New method development
Fish total mercury	SFEA	New method development

Table 3. Other Information Needs and Sources.

Information Needs	Sources
Water management operation records	SFWMD, COE
Atmospheric mercury deposition/evasion	FL DEP, EPA, FAMS, SFWMD, UFL, FSU
Nutrient loading estimates	SFWMD
Habitat changes	FWS NWI, NPS
Simulated natural hydropatterns	SFWMD
Vegetation patterns and production	NPS, FWS, SFWMD
ENR Project results	SFWMD, FL DEP
Periphyton production - nutrient relationships	SFWMD, FL DEP, FIU, UWI
Organic carbon speciation	USGS
Sulfate reduction/loading	SFWMD, USGS, FIU, UWI
Mercury methylation/demethylation	USGS, SFWMD, UMD, FIU, UFL, UWI
Fish and invertebrate impacts	FWS, NPS, FIU, UFL
Wading bird impact	FWS, NPS, UFL
Large mammal and reptile impacts	FWS, NPS, FIU, UFL, FSU, UGA

(2) *Define the spatial boundary of the decision statement*

(a) *Define the geographic area to which the decision statement applies.* The geographic area being studied, and for which decisions apply, is approximately 160 km long and 60 km wide, resulting in an area of about 9600 km^2. The exact boundaries are listed in Table 4 (next page) and shown in Figure 2.

Table 4. Geographic Area Boundaries.

Boundary	Description
Northern	West from Canal L8 to its junction with Lake Okeechobee and across to the Caloosachatchee River.
Western	Vertical line from the intersection of the Caloosahatchee River and Highway 833 south to the coast (the mangrove region is excluded from the target population).
Southern	Edge of the western mangrove east to the intersection with Highway US 1.
Eastern	Highway US 1 north to its intersection with Highway 27, then along the eastern boundaries of Water Conservation Areas to the Intersection with Canal L8.

(b) *When appropriate, divide the population into strata that have relatively homogeneous characteristics.* Strata of interest were based on the decision statement, rather than on homogeneity of variance. For example, there was less interest in defining the characteristics of the Big Cypress National Preserve (BiCY) than in other designated geographic areas. Therefore, BiCY was sampled with a lower inclusion probability (approximately 1/3 the density of other areas within the study boundaries). In addition, subsequent analyses have indicated the areas north of Alligator Alley, between Alligator Alley and Tamiami Trail, and south of Tamiami Trail have attributes that can influence management and policy decisions.

(3) *Define the temporal boundary*

(a) *Determine the timeframe to which the decision statement applies.* The decision statement applies from the time of the first data collection in April 1994 until at least 2004. The mercury-related legislative and regulatory deadlines are defined in Table 1. However, Project results are applicable to a longer timeframe because the South Florida Ecosystem Restoration Task Force has legislative mandates for hydropattern modification, habitat alteration and eutrophication deadlines beyond 2004 that can be addressed with results from this Project.

(b) *Determine when to collect the data.* Because time and space scales are inexorably coupled, the synoptic sampling approach spatially dictates that the temporal sampling frequency be seasonal. There are two distinct hydrologic seasons in Florida. The dry season extends from November to April and the wet season extends from June until September. May and October are transitional months. Sampling during only one season could result in biased and flawed decisions on

Everglades Ecosystem:

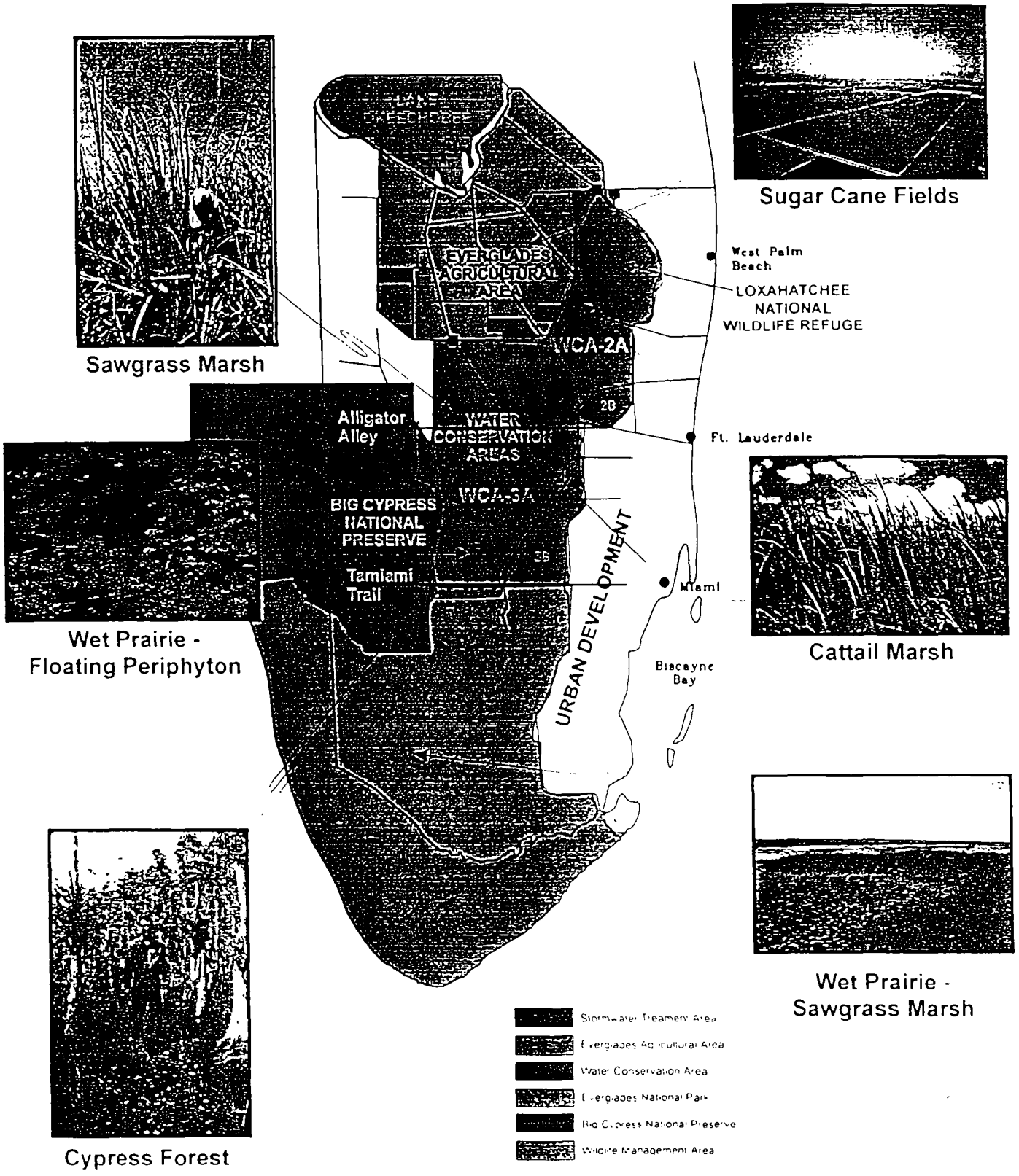


Figure 2 Study area boundaries, major management areas, and Everglades ecosystem community types

management or regulatory issues, because of seasonal variability. Sampling, therefore, needs to be done during both the dry and wet season. Decisions will be made over the next decade, based, in part, on spatial and temporal trends in information. These trends can not be defensibly determined with only one set (wet and dry season) of data at the beginning and end of the decision time frame. Two reference periods define change, not trends. Power analyses will be conducted to determine the number of sampling intervals needed to detect statistically defensible trends and contribute to the decision process.

(4) *Define the scale of decision making* - Decisions on mercury management and restoration issues must be made for the entire South Florida ecosystem. The environmental issues arose because of small-scale, piecemeal approaches to managing the system.

(5) *Identify practical constraints on data collection* - The large geographic area for sampling, and the need to collect synoptic samples requires that sampling be conducted by multiple teams using helicopters and airboats. The sampling period should be no longer than 10 days to minimize large scale changes in meteorology affecting water depth and quality measurements. The number of samples and sample volume need to be minimized to reduce weight and time for collection, but with sufficient volume to permit precision and accuracy requirements to be achieved. Clean sampling procedures are required for the mercury analyses, both in the field and in the laboratory. Low concentration nutrient analyses also are required because of the ultraoligotrophic condition of the Everglades wetlands.

Develop a Decision Rule - to define the parameter of interest, specify the action level and integrate previous DQO outputs into a single statement that describes a logical basis for choosing among alternative actions.

[NOTE: This DQO guidance statement is not compatible with the South Florida Ecosystem Restoration goals and objectives. The issues in South Florida are not independent; they are highly interactive. Multi-media decisions are required for multiple issues. There is no single statement can be formulated that will permit decisions among alternative actions. The greatest threat to the Everglades ecosystem is to assume these issues are independent and derive one single statement to address all issues. The Project, in part, will determine what the criteria should be for multiple issues such as phosphorus loading, water depth, distribution and timing, methylmercury concentrations in multi-media, and habitat types.]

(1) *Specify the statistical parameter that characterizes the population of interest* - REMAP is an exploratory research program so no single statistical parameter has been selected to characterize the population of interest. In addition, the emphasis is not on one single constituent, such as a hazardous material that might exceed a regulatory standard. Rather,

several statistical parameters are needed to characterize different population attributes, including:

(a) mean concentrations of selected constituents (see Table 3 for constituents)
(b) cumulative distributions of constituents, by season, by area
(c) distributional differences among constituents
(d) spatial patterns of constituents, and
(e) spatial/temporal associations among constituents.

(2) *Specify the action level(s) for the study* - Three action levels currently exist:

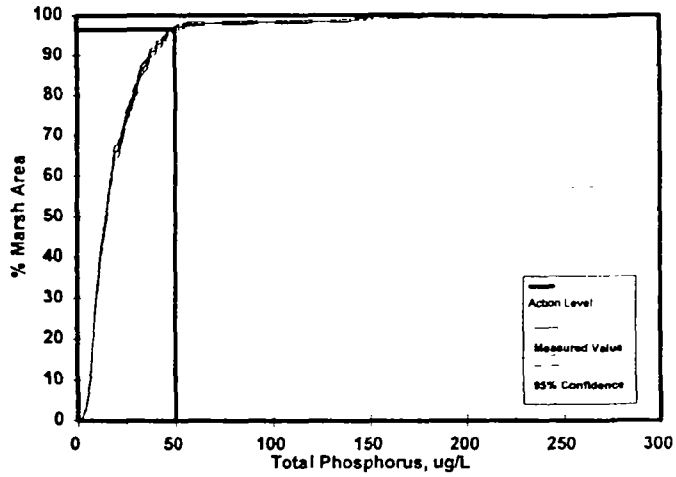
(a) Phase I control target for total phosphorus of 50 μg/L (ppb);
(b) Water total mercury criterion for protection of aquatic life of 12 ng/L (ppt); and
(c) Proposed predator protection level for mercury of 100 μg/kg (ppb) for prey species.

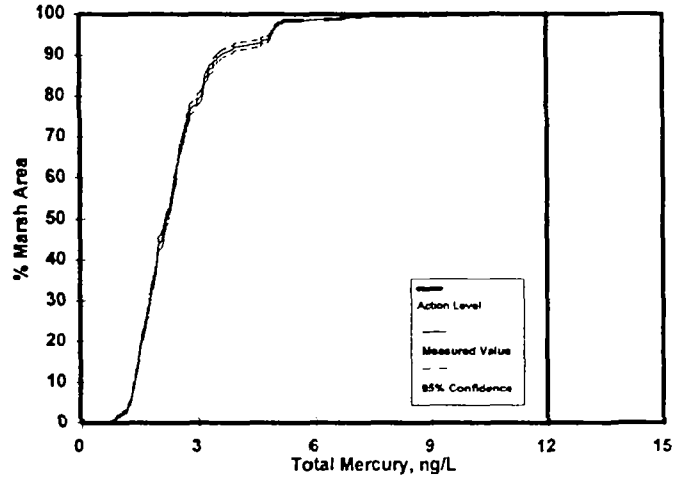
All three of these levels are underprotective. New risk-based action levels need to be determined. Currently, 95% of the marsh has total phosphorus concentrations less than 50 ppb; 100% of the marsh has total mercury concentrations less than 12 ppt, and 68% of the marsh has prey fish species with mercury concentrations greater than 100 ppb (Figure 3). Developing appropriate risk-based action levels for total phosphorus and mercury is one of the objectives of this Project. The detection and minimum quantitation limits for all three of these constituents are less than the respective criterion. Because risk-based action levels are needed, methods with increased sensitivity have been developed and are being tested.

(3) *Develop a decision rule (an "if...then" statement)* - Decision rules express what the decision maker ideally would like to resolve. The decision has been made that revised criterion are needed, based on the information developed to date from the Project. Preliminary decision rules, given this need, are listed in Table 5. Subsequent revisions of the DQO document will expand and refine these decision rules as additional information becomes available. Logic flow paths have been formulated (Figure 1) to increase the probability future information will improve the efficacy of the decision rules.

Specify Tolerable Limits on Decision Errors - the decision maker's tolerable decision error rates based on a consideration of the consequences of making a decision error.

(1) *Determine the possible range of the parameter(s) of interest* - The possible range of the parameters of interest are listed in Table 6. These ranges are based on this Project and other studies conducted in the South Florida Everglades ecosystem.





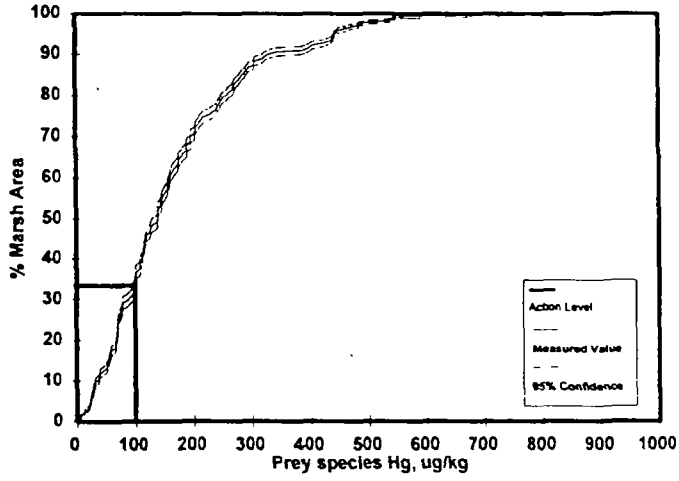


Figure 3. Distribution of total phosphors and total mercury in water and total mercury in a fish prey species, with 95% confidence intervals. **Note**: The existing action levels are under protective because ecological effects are observed from eutrophication and mercury contamination

Table 5. Preliminary Decision Rules for South Florida.

	Decision Rules
1a	If the South Florida Everglades Ecosystem can not be managed to achieve all desired ecological uses, then a comparative ecological risk assessment shall be conducted to determine which stressors, and their interactions, are placing the system at greatest risk.
1b	If the South Florida Everglades Ecosystem can be managed to achieve all desired ecological uses, then the management, regulatory and control practices shall be maintained.
	Based on the results of this comparative risk assessment, the following decision rules might be used:
2a	If phosphorus concentrations exceed a risk-based action level, then nutrient loads will be reduced until phosphorus concentrations are less than the action level.
2b	If phosphorus concentrations are less than a risk-based action level, then BMPs and other nutrient control programs will be maintained.
3a	If hydropattern modification varies by more than 10% from the desired natural hydropattern rule curve, then the hydropattern will be modified to match the desired natural hydropattern rule curve while maintaining flood control and water supply.
3b	If the hydropattern modification is within 10% of the desired natural hydropattern rule curve, and flood control and water supply purposes are satisfied, then the hydropattern management and operational programs will be maintained.
4a	If hydropattern modification varies by more than 10% from the desired natural rule curve and either flood control and/or water supply requirements can not be satisfied, then alternative flood control and water supply options will be investigated to return the hydropattern to within 10% of the desired natural rule curve.
4b	If hydropattern modification can not be returned to within 10% desired natural rule curve and achieve water supply and/or flood control demands, then a risk-based benefit/cost analysis will be performed to determine which alternatives have the lowest benefit/cost ratio and that use eliminated.
5a	If habitat alteration exceeds risk-based landscape action level metrics, then habitat alternation, a benefit/cost analysis will be done to determine if this habitat alteration including urban development or agricultural production, should be banned and habitat restoration under taken.
5b	If habitat alteration is less than risk-based landscape action level metrics, then habitat alteration will be permitted until these values are within 5% of the lower limit of the action level.

Table 5. Preliminary Decision Rules for South Florida (Continued).

	Decision Rules
6a	If mercury concentrations exceed a risk-based action level, then mercury sources will be controlled until mercury concentrations are less than this risk-based level.
6b	If mercury concentrations are less than the risk-based action level, mercury sources will be controlled to ensure the action level is not exceeded.
7a	If hydropattern modification greater than 10% from the risk-based desired natural rule curve aggravates mercury contamination of fish and wildlife, then the hydroperiod shall be modified to achieve the risk-based action level.
7b	If the hydropattern modification less than 10% of the risk-based desired natural rule curve aggravates mercury contamination of fish and wildlife, then a comparative risk assessment and risk-based benefit/cost analysis shall be conducted to determine which stressor places that system at greatest risk and has the lowest benefit/cost ratio; that stressor then will be reduced.
8a	If nutrient loading exceeds the nutrient risk-based action level and aggravates mercury contamination of fish and wildlife, then nutrient loading shall be reduced to achieve the risk-based action level.
8b	If nutrient loading is less than the nutrient risk-based action level and aggravates mercury contamination of fish and wildlife, then a comparative risk assessment and risk-based benefit/cost analysis shall be conducted to determine which stressor places that system at greatest risk and has the lowest benefit/cost ratio; that stressor then will be reduced.
9a	If habitat alteration exceeds risk-based landscape action level metrics and aggravates mercury contamination of fish and wildlife, then additional habitat alteration shall be banned and habitat restoration under taken.
9b	If habitat alteration is within the risk-based landscape action level metrics and aggravates mercury contamination of fish and wildlife, then a comparative risk assessment and risk-based benefit/cost analysis shall be conducted to determine which stressor places that system at greatest risk and has the lowest benefit/cost ratio; that stressor then will be reduced.

Table 6. Water Constituents Ranges in South Florida.

Measurement Variable	Range			
	Minimum		Maximum	
Physical Measurements				
Site location (deg.)	Latitude 25.30	Longitude 80.22	Latitude 26.93	Longitude 81.13
Weather				
Discharge, structure (m^3/s)	0			
Water depth (ft)	Marsh 0	Canal 0.5	Marsh 8	Canal 25
Temperature (°C)	18		36	
Turbidity (NTU)	0.1	80		
Chemical Measurements				
Dissolved oxygen (mg/L)	0		15	
Specific conductance (μS)	10		2150	
pH (s.u.)	5.5		8.8	
Total organic carbon (mg/L)	5		80	
Total phosphorus (mg/L)	0.001		0.500	
Sulfate (mg/L)	1.0		850	
Total mercury (ng/L)	0.02		12	
Methymercury (ng/L)	0.03		1.5	
Alkaline phosphatase	0.01		8.0	
Biological Measurements				
Resource class *(canal, sawgrass marsh, cattails, etc.)* (Numeric rank)	1		7	
Periphyton presence/absence (1,0)	0		1	
Chlorophyll a (μg/L)	0		100	
Periphyton total mercury (μg/kg)	4		600	
Periphyton methylmercury (μg/kg)	0.08		25	
Fish total mercury (μg/kg)	5.0		1000	

Table 6B. Soil/Sediment Constituents Ranges in South Florida.

Measurement Variable	Range	
	Minimum	Maximum
Physical Measurements		
Peat depth (m)	0	>4.25
Bulk density (g/cc)	0.05	1.4
% Mineral content (%)	3%	99%
Ash free dry weight (%)	1.0	96.0
Redox potential (mV)	-250	+600
Chemical Measurements		
Soil/Sediment total mercury (μg/kg)	3.0	500
Soil/Sediment methylmercury (μg/kg)	0.01	50
Soil/Sediment total phosphorous (μg/kg)	10	9000
Soil/Sediment sulfate (μg/kg)	20	850

(2) *Identify the decision errors and choose the null hypotheses*

(a) *Define both types of decision errors and establish the true state of nature for each decision error.* By convention, a Type I (false positive) error is rejecting the null hypothesis when it is true. A Type II (false negative) error is not rejecting the null hypothesis when it is false. The two types of decision errors for the Project are (I) deciding the risk-based action level is exceeded when it truly is not, and (II) deciding the risk-based action level is not exceeded when it truly is.

The true state of nature for decision error (I) is that the null hypothesis is true.

The true state of nature for decision error (II) is that the null hypothesis is false.

(b) *Specify and evaluate the potential consequences of each decision error.* The consequences of deciding the risk-based action levels are exceeded when they truly are not (decision error I) means there will be increased control costs associated with nutrient and mercury source reduction, restricted urban and agricultural development, habitat restoration, and restricted hydropattern modification around the natural hydropattern rule curve, which could result in flood damage or water supply shortages.

The consequences of deciding the risk-based action levels are not exceeded when they truly are (decision error II) means that ecological restoration of the South Florida Everglades ecosystem will not be successful.

(c) *Establish which decision error has more severe consequences near the action level.* Based on current laws and regulations related to the South Florida Everglades ecosystem (e.g., Everglades Forever Act), the decision II error has the more severe consequences near the action level because of the risk to both ecological and human health and ecological restoration. However, this consequence must be based on a comparative risk assessment and a risk-based benefit/cost analysis of the risks and impacts. The economic consequences are in the billion dollar category for both types of decision errors.

(d) *Define the null hypothesis (baseline condition) and the alternative hypothesis and assign the terms "false positive" and "false negative" to the appropriate decision error.* Null hypotheses for DOQs are not equivalent to experimental null hypotheses for statistical testing. Null hypotheses for DQOs reflect the decision error that has the most adverse potential consequences. The DQO null hypothesis is equal to the true state of nature that exists when the more severe decision error occurs. The null hypotheses for this Project, therefore, would be:

H_0 = The comparative ecological risk assessment indicates the interactions among stressors puts the South Florida Everglades ecosystem at risk.
H_0 = The risk-based action levels for nutrient concentrations are exceeded.
H_0 = The risk-based action levels for mercury concentrations are exceeded.
H_0 = The risk-based landscape action level metrics are exceeded.
H_0 = The risk-based action levels for hydropattern modification exceed by X% the natural hydropattern rule curve.

A "false positive" has the greatest consequences for each of these hypotheses.

(3) *Specify a range of possible values of the parameter of interest where the consequences of decision errors are relatively minor (gray region)* - The purpose of this research project is to determine the action level values. Until these action levels are defined, it is not possible to specify actual numeric values to an area of minor importance. It is, however, possible to indicate these areas of minor importance will be at the extremes of the distribution. In this portion of the action level curve, there will be a low probability of making either type of decision error.

(4) *Assign probability values to points above and below the action level that reflect the tolerable probability for the occurrence of decision errors.* - The QA G-4 Guidance manual indicates the gray region where greater tolerable errors are permitted are around the action level, with lower tolerable errors around the extreme values. The planning team disagrees with this concept. The greater tolerable errors are permitted at the extemes of the distribution because it is unlikely that large errors in the metric would alter the conclusion that the action level was either exceeded or not exceeded. However, near the action level, particularly as values approach the lower limit of the action level, decision errors can have significant consequences on subsequent actions (Figure 4). Tolerable error around the action level in this region should be no more than 10%.

Optimize the Design - The REMAP monitoring design for South Florida was revised to provide more resource-effective information at reduced cost without compromising the DQOs for the marsh samples. The canal samples will be selected as subjective samples because the first four sampling cycles indicated the marsh processes are more significantly affected by multiple environmental stressors than canal processes in South Florida. Sufficient samples will be collected in the canals so that temporal and spatial trends can be detected over time in loadings to the marsh, but not with the same precision as marsh trends. This improved design will permit answering the original seven policy-relevant questions guiding the Project and establishing the action levels needed for decisions.

Appendix A contains statements for data representativeness, completeness, comparability, precision and accuracy for each of the constituents measured in the EPA Region 4 South Florida Ecosystem Assessment Program. These quantitative DQO criteria will be revised as additional data become available to the program.

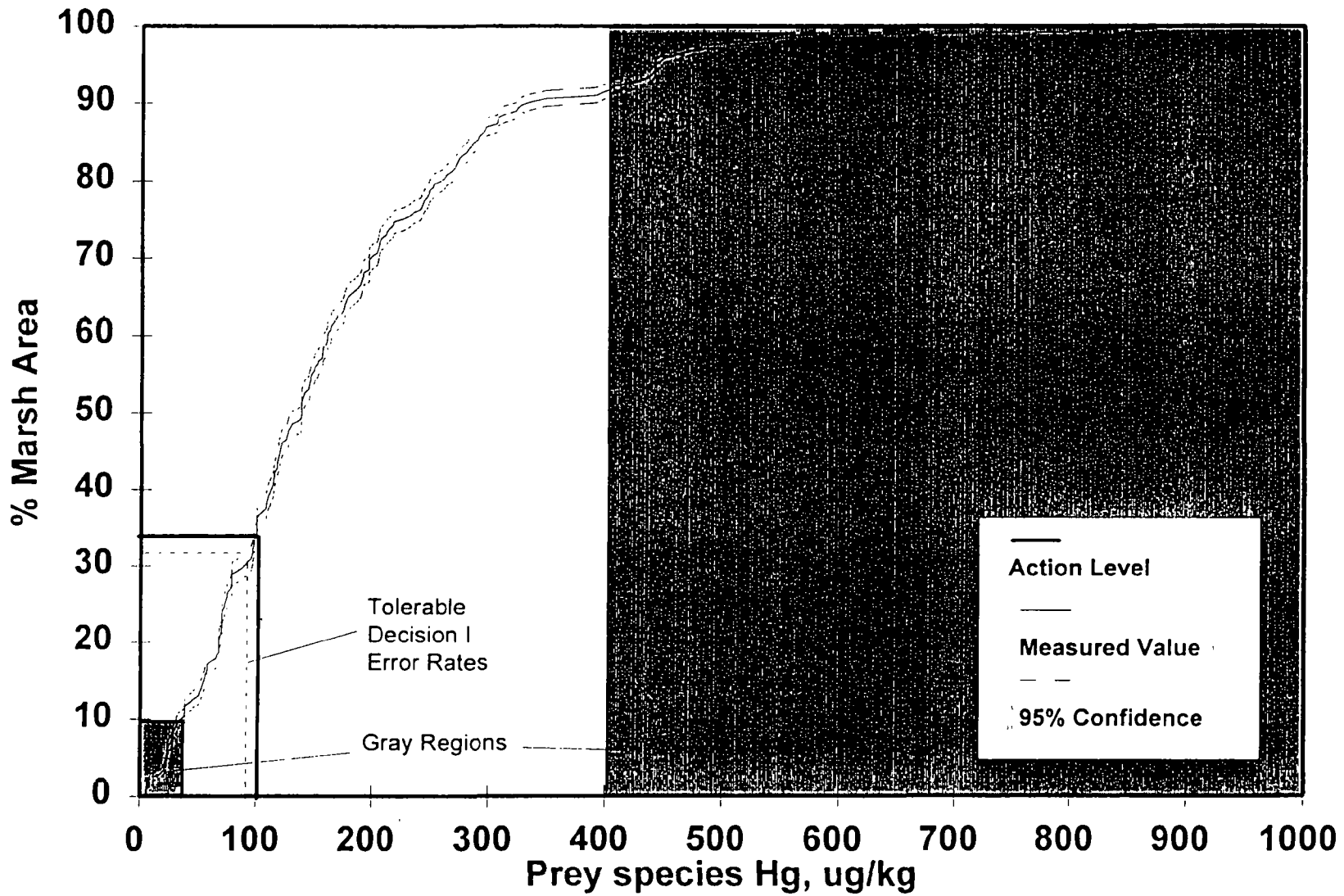


Figure 4. Proposed mercury Action Level for predator prey species identifying The Gray Regions and area for 10% Tolerable Decision 1 Error Rates.

Appendix A

Data Quality Objective Criteria

Table A1A Water Data Quality Objective Criteria.

Measurement Variable	Representativeness	Completeness	Comparability (Split Samples SOPs, Std. Units)	Field Precision 3σ		Duplicate Precision % RSD @ 95%		Accuracy (% Spike Recovery)
				Canals	Marsh	Canals	Marsh	
Physical Measurements								
Site location (deg.)	Design-based statistically representative	90%	SOPs			NA	NA	
Weather	"	"	SOPs	NA	NA	NA	NA	NA
Discharge, structure (m^3/s)	"	"	NA		NA	NA	NA	NA
Water depth (m)	"	"	SOPs	±15.0	±3.3	NA	NA	
Temperature (°C)	"	"	SOPs	±5.06	±9.05	NA	NA	± 0.15
Turbidity (NTU)	"	"	SOPs	±21.9	±34.6	±182 9	±136.4	NA
Chemical Measurements								
Dissolved oxygen (mg/L)	"	"	SOPs	±7.45	±8 52	NA	NA	± 0 2*
Specific conductance (μS)	"	"	SOPs	±1119	±760	NA	NA	± 1
pH (s u.)	"	"	SOPs	±0.96	±1 39	NA	NA	± 0.2*
Total organic carbon (mg/L)	"	"	SOPs, USGS	±37.14	±31.40	±28 87	±27.14	85–115
Total phosphorus (μg/L)	"	"	SOPs, SFWMD	±269 4	±106.2	±101 2	±68 8	81–115
Sulfate (mg/L)	"	"	SOPs, USGS	±112	±51	±12	±107	85–115
Total mercury (ng/L)	"	"	SOPs, Battelle hab.	±8.13	±8.11	±230 24	±139.16	70–130
Methymercury (ng/L)	"	"	SOPs, Battelle hab.	±1 230	±2 256	±83.69	±204.66	70–130

* Actual Units RSD Relative Standard Deviation SOP. Standard Operation Procedures

Table A1A. Water Data Quality Objective Criteria (Continued).

Measurement Variable	Representativeness	Completeness	Comparability (Split Samples SOPs, Std. Units)	Field Precision 3σ		Duplicate Precision % RSD @ 95%		Accuracy (% Spike Recovery)
				Canals	Marsh	Canals	Marsh	
Alkaline phosphatase	"	"	SOPs	±3 74	±3.48	±75.21	±61.18	70–130
Biological Measurements								
Resource class *(canal, sawgrass marsh, cattails, etc)* (Numeric rank)	"	"	SOPs, FWS, NWI	NA	NA	NA	NA	NA
Periphyton presence/absence (1,0)	"	"	SOPs	NA	NA	NA	NA	NA
Chlorophyll a (μg/L)	"	"	SOPs	±22.55	NA	±191.48	NA	NA
Periphyton total mercury (μg/kg)	"	"	SOPs	NA	±487.87	NA	±225.95	70–130
Periphyton methlmercury (μg/kg)	"	"	SOPs	NA	±9 51	NA	±115.59	70–130
Fish total mercury (μg/kg)	"	"	SOPs, Reg. 4	±257 42	±397.35	±159.36	±89.95	70–130

* Actual Units RSD. Relative Standard Deviation SOP· Standard Operation Procedures

Table A1B Soil/Sediment Data Quality Objective Criteria.

Measurement Variable	Representativeness	Completeness	Comparability (Split Samples SOPs, Std. Units)	Field Precision 3σ		Duplicate Precision % RSD @ 95%		Accuracy (% Spike Recovery)
				Canals	Marshes	Canals	Marshes	
Physical Measurements								
Peat depth (m)	"	"	SOPs	NA		NA	NA	
Bulk density (g/cc)	"	"	SOPs	NA	±0 77	NA	±35.56	NA
% Mineral content (%)	"	"	SOPs	NA	±93.82	NA	NA	NA
Ash free dry weight (%)	"	"	SOPs	±51 84	±93.82	±60.54	±52 79	NA
Redox Potential (mV)	"	"	SOPs	±386	±308	NA	NA	
Chemical Measurements								
Soil total mercury (μg/kg)	"	"	SOPs, USGS	±249 1	±196 9	±70 2	±89 5	70–130
Soil methylmercury (μg/kg)	"	"	SOPs, USGS	±32 6	±3 87	±383 3	±255 8	70–130
Soil sulfate (μg/kg)	"	"	SOPs, USGS	NA	±1412	NA	±107	
Total phosphorus in soil (μg/kg)	"	"	SOPs, USGS	±3904.00	±741.64	±108.54	±72.98	75–125

RSD Relative Standard Deviation
SOP Standard Operation Procedures
NOTE: Use largest standard deviation when two measurements of a parameter–top and bottom, FIU & BATTELL, floating and soil periphyton

APPENDIX C

Summary of Data Review Findings

EXECUTIVE SUMMARY

The EPA Region 4 South Florida Everglades Ecosystem Assessment Project is a pioneering research and monitoring effort to assess mercury contamination, eutrophication, habitat alteration, and hydroperiod modifications in the South Florida Everglades ecosystem using the ecological risk assessment process. Over a four year period, this Project made over 20,425 measurements on 20 constituents in 5 different media at about 200 canal stations, 550 marsh stations during the 9 field sampling events over the 10,000 km^2 area in South Florida. Included in these measurement were 7 structures sampled on a bi-weekly basis from February 94 thru February 97. The large scale patterns in total phosphorus and eutrophication, mercury contamination, and habitat types, observed for the first time because of this Project are described in the 1998 South Florida Ecosystem Assessment Technical Report, Volumes I and II (Stober et al., 1998). To accommodate this large sample load, and to analyze ultra-trace level concentrations of selected constituents, new methods also were developed as part of this Project. In addition to the quality assurance reviews that occurred as part of the data analysis, a rigorous quality assurance/quality control review was conducted on the data by scientists of the Region 4 Science and Ecosystem Support Division (SESD), Office of Quality Assurance (OQA).

OQA was requested to review all contractor data provided by laboratories supporting the SESD Everglades ecosystem project. Raw data were obtained from each laboratory involved and were reviewed against the four levels of criteria addressed in: 1. the Data Quality Objectives (DQOs), 2. requirements stated in Section 2.5; Data Quality Requirements in the EPA Project Research Plan, 3. the laboratories' respective Standard Operating Procedure (SOP) and 4. quality control (QC) practices, and procedures typically applied for regulatory and compliance reviews conducted by the OQA. The OQA goals were:

- to reconstruct the final reported data for the field, laboratory and QC activities,
- to determine if the contract laboratories' documentation was accurate and provided adequate defensibility of the monitoring data, and
- to verify if the data were of acceptable quality based on the Project design.

Review findings are documented in this *Summary of Data Review Findings* for each constituent by media and by cycle and applicable recommendations are included.

Quantitative and qualitative evaluations indicate the vast majority of the data were of acceptable quality, and met the overall DQOs of the Project design. All data sets will have some QA/QC deficiencies and certain QC procedures and documentation deficiencies also were noted in the Project data. Out of the over 20,425 measurements made over the four year period, only 15 percent of the data were qualified. There were 1,325 points flagged "NR"- not reviewed because raw data were not available from the laboratory, 1,783 assigned a "J" flag - analyte identification was acceptable for use but certain QC criteria were not satisfied and 256 values rejected and not reported in the database.

The data that have been qualified after this rigorous review are:

- "J" – Soil/sediment methyl mercury for the secondary laboratory
- "NR" – Water total mercury for September 1993 Canal samples and April '94 transect data for primary laboratory
- "J" – Water total mercury, excluding September 1993 Canal, for primary laboratory.
- "J" – Periphyton total mercury for primary laboratory
- "J" – Soil/sediment total mercury for May 1994, 1995 Canal cycles and May 1996 marsh samples for secondary laboratory
- "NR" – Soil total mercury for September 1995 marsh split samples for secondary laboratory.
- "J" – Mosquitofish total mercury for very low mercury concentration fish for primary laboratory
- "J" – Total nitrogen for the secondary laboratory for Sept. '96 marsh samples
- Reject – Water alkaline phosphatase activity for 123 samples for May 1996 marsh cycle from the primary laboratory
- Reject – Water sulfide for 123 samples for the September 1995 marsh cycle from the primary laboratory.

The Study was designed to have a minimum of 10 percent of the samples split for certain parameters with a second laboratory to provide between laboratory comparability and field duplicates to provide a combined field and laboratory precision. Samples for certain parameters were split at a higher frequency than 10 percent. The higher frequency of split sample data provided strong analytical support for data analysis and interpretation even though documentation or record retention may have been lacking in some cases.

The OQA finds that the data provided by all laboratories (EPA and supporting) permit assessment of trends and the characterization of the Everglades on a spatial and temporal basis and satisfy the Project objectives. Data qualified with a "J" code would not compromise the characterization and trend analysis or Project objectives. "J" coded data should not be used solely in the setting of environmental standards unless additional data are collected to substantiate these original values that has all supportive QC and documentation. These data can be used as part of the weight of evidence approach for establishing environmental standards.

This *Summary of Data Review Findings* report should be kept in perspective. The South Florida Everglades Ecosystem Assessment Project was a research and monitoring effort, not a regulatory compliance project. It developed and implemented new methods for rapidly analyzing a large sample load with ultra-trace level constituent concentrations. The QA/QC review, however, included rigorous QA/QC methods used in reviewing compliance monitoring data. Yet, out of the over 20,425 measurements made over the four year period, only 15 percent of data were qualified.. This data set should provide an important baseline for assessing the current conditions of the Everglades ecosystem and for monitoring future trends in this condition. The results from this QA/QC Review have already been, and will continue to be, used to refine the QA/QC procedures in future Project monitoring efforts.

INTRODUCTION

In 1993, EPA Region 4 Science and Ecosystem Support Division (SESD) initiated the South Florida Everglades Ecosystem Assessment Project. This pioneering 10,000 km^2 system wide research and monitoring project was conducted to assess mercury contamination, eutrophication, habitat alteration, and hydroperiod modification using an ecological risk assessment process. A statistical survey design was used to select 200 canal sites and 500 marsh sites to sample for 20 constituents measured in 5 different media during 4 different periods or cycles. In addition, transect sampling was conducted to test the marsh sampling procedures, constituents were measured on a bi-weekly basis at 7 structures in the South Florida system, and new methods were developed to accommodate the large sampling load and measure ultra-trace level constituent concentrations. Over a four year period, this project made over 20,425 measurements in the South Florida Everglades ecosystem.

Although QA/QC procedures were designed into the Project from its onset, a *Quality of Science Review* was requested from the EPA Office of Research and Development (ORD) National Exposure Research Laboratory (NERL) Environmental Sciences Division (ESD)–Las Vegas, in 1995. ORD was, and continues to be, in the process of trying to develop a Data Quality Objectives (DQO) process for research projects similar to the DQO process developed for regulatory and compliance programs (EPA 1994). This has proven to be a formidable task. The purposes of the NERL-ESD review were to (1) identify quality-related issues and provide recommendations for correction or improvement, and (2) to provide the project participants with the necessary tools to enable them to continue to monitor data quality. The *Quality of Science Review* report (Chaloud, Heitmar, and Birch 1996) was provided to SESD in April 1997. The *Quality of Science Review* report identified a number of strengths in the Everglades Mercury Study. Strengths of this project include:

- Project Design
- Personnel
- Field Operations
- Sampling Procedures
- Low Crew Variability
- Project Management
- Sample Representativeness
- Laboratory QA/QC Programs
- Low Error Rate in data Base
- Spatial Analyses

Recommendations suggesting positive corrective actions were commenced or implemented during the on-site audits in 1995 and continued during successive sampling events and data evaluation by the OQA and the laboratories involved. Three areas were specifically mentioned in the *Review* related to data quality. These were

* Data Package Issues
* Data Validation/Verification
* Total Mercury in Water Data

The *Review* recommended:

- that all raw data provided by contract laboratories through the Interagency Agreements with the U.S. Department of Energy and the U. S. National Park Service should be obtained and maintained on file at EPA,
- validate or verify the quality of data generated,
- use flag fields to indicate validation/verification results, and
- document validation and verification criteria in metadata files or in a user's guide.
- recover additional sulfate data

As a result of the recommendations made in the *Quality of Science Review,* the SESD Office of Quality Assurance (OQA) and Region 4's Environmental Service Assistance Team (ESAT) conducted a data review of all available contractor acquired data for the Everglades Assessment Project.

In addition to the above recommendations, the OQA also conducted selected reviews of other raw data collected/analyzed by EPA staff and EPA's ESAT contractor. The goal of the audit of data quality was to provide a quantitative and qualitative evaluation of the documentation and procedures associated with environmental measurements to verify that the resulting data were of acceptable quality.

Data Collection Activities

The gathering of data for review was conducted in two phases. First, the OQA and ESAT Work Team conducted a preliminary review of the data submitted by the Florida International University (FIU) Southeast Environment Research Program (SERP) laboratory for sampling events conducted in 1993, 1994, 1995, and 1996. Randomly selected raw data packages were obtained by the OQA and ESAT Team during a visit to the SERP laboratory on March 12–14, 1997. Total and organic mercury raw data packages were also requested and received in March 1997, from Battelle Marine Sciences Laboratory (Battelle), Sequin, WA. AmTest Analytical Service, Inc., Seattle, WA, a subcontract laboratory for Battelle provided quality control (QC) split sample data for total nitrogen and phosphorus for the Marsh Cycle 3 sampling event. Raw data from AmTest were obtained in March 1998.

Since the Quality Assurance Project chemist participated in the ORD *Quality of Science Review,* and was part of the data review Team, an additional on-site visit was not made to the Battelle laboratory. The AmTest laboratory was not visited since only 26 samples were analyzed during the last sampling event of 1996.

After an initial assessment of the raw data packages, the second phase of the OQA/ESAT audit included two subsequent visits to the SERP on May 14–15, and October 14–15, 1997 to obtain additional raw data for the nine sampling events. The visits were necessary to fulfill the recommendation of *Quality of Science Review* Team that all raw data should be resident at Region 4 SESD and to allow the SERP personnel time to gather the large volume of raw data. A

third visit was made during June 1998 to review data stored in electronic format for methyl mercury at the SERP laboratory. During each visit members of the data review Team discussed the methodologies used and observed the SERP staff conducting the on-site analyses.

Data Review Guideline

Data were reviewed using data review practices employed by the OQA and the ESAT Team for regulatory and compliance monitoring programs with standard methodology. These practices included, but were not limited to, such items as sample collection, receipt, storage, checks of holding time, preparation, analysis, reporting, and all associated QC. Preparation checks included the use of blanks (reagent, digestion, lab and field blanks) for background correction, use of calibration standards and application of linear regression analysis of calibration curves for linearity, utilization of standard reference materials (SRMs) or performance evaluation samples, use of matrix spikes for accuracy, analysis of both field and laboratory duplicates for precision statements and determination of detection limits. Data packages also were reviewed against criteria addressed in the Data Quality Objectives (DQOs) and quality assurance requirements stated in Section 2.5; Data Quality Requirements in the EPA Project Research Plan; against SERP's Standard Operating Procedures (SOP) for Total and Methyl Mercury analyses; SERP's Comprehensive Quality Assurance Plan for the Mercury Laboratory dated 7/27/93, and subsequent revisions dated 6/14/94 and 4/18/96; and SERP's Methods Validation for Micromolar Concentrations of Total Nitrogen in Natural Water dated 4/25/96. The latter included total phosphorus and total organic carbon (TOC) SOPs. Because method development was a Project objective, the SERP staff expended considerable time in developing and refining methods and modifying the SOP's. These modifications were necessary as procedures were continually improved to enhance the quality of the data as the project moved from a research-orientation to a research-production environment. The same data review procedures were employed for the other participating contractor laboratories.

All data entries in the EPA database were checked against the raw data sheets from all laboratories for completeness and correctness. Relatively few transcription errors were observed considering the vast amount of data provided. Corrections were provided to the Project Leader.

Data Review Qualifiers

Reported data that did not satisfy all the criteria specified in the **Data Quality Review Guidelines** were qualified with appropriate codes. Each parameter that has data qualifier codes assigned has remarks added to assist the reader in understanding the code and the reason(s) for the qualifier or data flag. A list of qualifier codes accompany each parameter or group of similar parameters. The codes were:

Code	Definition of analytical data qualifiers

NR A "NR" Qualifier Code indicates that these data were not reviewed, since raw data were not available from the laboratory. In these cases, EPA data reviewers were not able to reconstruct the QC and analytical activities.

"U" The analyte was analyzed for but not detected. The value preceding the "U" is the "minimum quantitation limit (MQL)".

***Minimum Quantitation Limit (MQL) – Every sample has a concentration level below which the variance of the results for a particular analyte (element or compound) exceeds the acceptable quality control criteria. This level is the MQL and is reported as the value preceding the "U". The MQL is determined from sample size, dilution required, and instrument sensitivity. The value often varies from analyte to analyte within a sample. Analytes are often detected at levels below the MQL and are reported as estimated values (J). Generally, analytes identified below the MQL will only be reported if concentration is greater than one tenth of the MQL.

"J" The identification of the analyte is acceptable for use, but certain QC criteria were not satisfied.

***Estimated Value--Every sample analysis has quality control criteria associated with the quantitative data which have been established based on similar analyses. When these criteria are exceeded, the value for that analyte or similar analytes is flagged. Examples are:

(1) calculated values were below or above an appropriate linear range
(2) calculated values were below the MDL or PQL of an analyte
(3) analytical holding times for analysis were exceeded
(4) surrogate recovery limits were exceeded
(5) some QC criteria were not documented or followed as specified in the laboratory's SOP or requirements necessary for confirmation were not met.

"A" The analyte was analyzed in replicate. The value preceding the "A" is an "average value" of the replicates.

***Average Value--Samples were often analyzed in replicate (usually in duplicate). Aliquots of the same sample were analyzed and the values are averaged. Sometimes replicate samples were analyzed and the values were reported as an average.

Specific data qualifier codes and explanations are provided with each constituent by media and by cycle and the associated data.

Summary of Data Review Findings

Summary results are presented for each constituent by media and by sampling cycle that data qualifier codes were assigned. Applicable recommendations are included. If there were no qualifying codes assigned, the data met the data quality review guidelines used to evaluate the data. Primary and secondary laboratories were used for sample analysis. The primary laboratory was the lead laboratory for the analysis with the secondary laboratory providing QA/QC on duplicate and split samples. The goal was to have QA/QC samples for about 10 percent of the total number of samples analyzed so the secondary laboratory had a significantly lower sample load.

1. Total and Organic Mercury in Soil/Sediment and Water by Battelle

Battelle served as the primary laboratory for methyl mercury in water and provided backup QC for total mercury in water splits/duplicates throughout the study and methyl mercury in soil/sediment after the Sept. 93 Canal sampling. The Battelle total and methyl mercury data for water and soils were reviewed for accuracy and compliance according to the same criteria used for SERP total and methyl mercury analyses. Linear regression was checked using the standards from the calibration curve. Quality control check samples were evaluated for recovery of the analyte from spiked samples and relative percent difference of duplicate samples. The final results were checked to ensure that the results were converted accurately from the instrument with the appropriate units. For example, the results for soil samples are converted from pg/ml to ng/g. Less than 5 percent of the methyl mercury samples for water were flagged "J" due to the quality control checks being outside of the acceptable limits: matrix spikes outside DQR limits (70 percent to 130 percent), duplicate samples results relative percent difference greater than 30 percent, or the calibration curve correlation coefficient less than 0.995.

After May 94, Battelle switched from an extraction to a steam distillation separation for soil/sediment samples. Matrix spikes were conducted and recoveries were acceptable. Duplicate splits with SERP were comparable with Battelle reporting values that tended to be higher than the SERP data. It has been reported that steam distillation could produce a methyl mercury artifact in samples containing elevated concentrations of total organic carbon (TOC) and total mercury (Hintelmann 1997). However, another study (Bloom 1997) using Everglades water and peat indicated this artifact would not be significant in Everglades samples. Methods for measuring ultra-trace level total and methyl mercury concentrations in water and soil/sediment are in the research phase. There currently is no standard EPA method for these constituents at these concentrations. Therefore, Battelle QA/QC soil/sediment data after May 94 should not be used solely in the establishing trends or environmental standards unless additional collaborative data are available to substantiate these original values

2. Total Mercury in Water by SERP

SERP served as the primary laboratory for the approximately 900 total mercury samples in water. These data were reviewed and validated. The September 1993 Canal and April '94 transect

data for total mercury were flagged as "NR," not reviewed. The reason was the raw data were unavailable for review from the laboratory.

Data also were reviewed against QC checks EPA considered as good laboratory practices for regulatory or compliance monitoring programs with standard EPA methods. An independent stock standard prepared from another source to verify the initial calibration was not included in the analysis, but it was not part of SERP's SOP. The lack of this independent calibration check would not change the values obtained but would have strengthened the quality of the data. However, no data were flagged because SERP followed its SOP and the independent stock standard was not part of the initial QA plan reviewed by EPA. The independent stock solution will be added to future sample analyses.

The SERP data prior to May 1996 did not include a matrix spike. The matrix spike strengthens the data in that it shows that the analyte is actually being recovered during the digestion of the samples. According to SERP CompQAP, the accuracy of the matrix spike should be between 90–110 percent recovery or the sample analyses must be repeated. In May 1996, the data includes a water spike at 2 ng/l. Six of sixteen data sets had recoveries ranging from 83–122 percent with an average for all sets of 103 percent. In September 1996, the data included a sample (matrix) spike at 2 ng/l with 9 of 17 data sets having recoveries of 49–138 percent. The average percent recovery of this matrix spike for all 17 data sets was 94 percent recovery. However, none of the samples analyzed with the spike recoveries outside of SERP's stated performance limits were re-analyzed.

Although reagent blanks were analyzed, the sample results did not include a consistent reagent blank correction prior to May 1996. Just prior to the May 1996 sampling event, the SERP staff instituted a correction method that accounted for the background contribution from reagents and glassware. Correction for any contribution by the reagents was made utilizing varying concentrations of the reagents added to mercury free water and calculating the response due to the reagents using linear regression. Data previous to May 1996, were corrected for any reagent contribution by using an average value (0.5 ppt) for the reagent blank correction. The reagent blank corrects for any contamination due to the reagents and could alter the result at the lower concentrations if the contribution was significant. Since raw data for the first two sampling cycles were not available for review, a statement could not be made about the quality of the data. However, the SERP staff did have an in house policy of checking for background contamination and if background contamination was determined to be significant, samples were to be re-analyzed. Recalculation of the data using a four-point curve and subtracting the reagent blank showed differences of as much as 1.0 ng/L in only 4 out of 552 cases prior to May 1996.

A detection limit for total mercury was established by SERP at 0.3 ng/L using the recommended procedure for establishing detection limits given in 40 CFR 136 Appendix B. A low-level 0.3 ppt standard was not analyzed to show this level could be achieved each day. To provide a day-by-day assessment of the method stability near the MDL, a practical quantitation limit (PQL) for total mercury was established by the OQA data review Team at 1 ppt., (three times the standard deviation of 21 readings from the 2 ppt spike and reagent blanks). Results below 1 ppt. should be used with caution, especially since the lowest calibration standard reported as

analyzed during the study was 2.5 ppt. The precision for field duplicates below 1 ppt ranged from 0–118 percent relative standard deviation (% RSD equals the standard deviation for duplicates divided by the mean time 100) with an average of 28 percent. Based on the review findings, the total mercury data in water by SERP were assigned a "J" flag except for the September 1993 canal data assigned "NR". The "J" flag indicates the data are acceptable for use, but did not satisfy certain QA/QC criteria. The SERP data below 1.0 ppt should not be used solely for setting regulatory limits based on field precision and blank correction.

QC data for total mercury in water from split sample QC data provided from the Battelle laboratory showed a positive bias compared with data submitted by the SERP. However, the bias between the laboratories diminished during the May and September 1996 sampling events. The quality of the total mercury data were strengthened in two ways: 1) the observed collaborating trends for split samples between the SERP and Battelle laboratories; and 2) consistent performance by SERP and Battelle was observed on performance evaluation samples supplied as unknowns by the Project Quality Assurance Chemist to each laboratory. (See Table 1). Continued performance evaluation corroboration is expected to improve as mercury methods and analytical procedures continue to be refined.

Table 1 Total Mercury in Water Performance Evaluation Data (ppt)

Date of Analysis	P.E. Value	SERP	Battelle	EPA
05/12/96 WP 30 C1	0.98	1.44	1.51	1U
05/13/96 WP 30 C2	2.10	1.73	2.30	2.20
05/14/96 WP 30 C1	6.23	5.71	6.14	6.20
09/15/96 WP 33 C1	5.81	5.02	5.73	5.60A
09/18/96 WP 33 C1	2.71	2.21	2.87	2.30
09/19/96 WP 33 C1	10.16	8.87	10.40	10.00
09/20/96 WP 33 C1	1.16	1.34	1.26	1.0UA

3. Total Mercury in Periphyton by SERP

SERP was the primary laboratory for the periphyton samples. QC check samples (SRM) were analyzed by SERP with each set of total mercury in periphyton analyses to verify the accuracy of the calibration standards and validate the digestion method from an independent source. SERP's acceptable range for these check samples was 90–110 percent recovery of the true value of the sample (60 ng/g). In 6 out of 16 data sets, this percent recovery was not achieved. The range of recovery was from 68–135 percent. The average recovery was 101 percent for the 16 sets.

A reagent blank was included in the analyses, but was not used to correct for reagent contamination in the final calculation of the sample results. The reagent blank correction versus no reagent blank correction showed a considerable difference (130 percent RPD) in sample results where peak heights were less than two times the peak height of the reagent blank. This impacted greater than 50 percent of the samples for April 1995 and May 1996, but only 10 percent of the samples in September 1996. These sample values should be recalculated to remove the contribution for the blank or the detection limits should be increased. A check standard of 500 ng/l prepared from the same source was used for verification. Therefore, all total mercury data for periphyton were given a "J" code. The data are acceptable for use, but did not satisfy certain QA/QC criteria. The FL DEP recommends percent recovery ranges of 75–125 percent for biological samples, rather than 90–110 percent. These changes would be implemented in future sample analyses.

4. Total Mercury in Soil/Sediment by SERP

EPA-SESD served as the primary laboratory and SERP as the secondary or QC laboratory. A soil SRM was analyzed by SERP with each set of total mercury in soil/sediment analyses to verify the accuracy of the calibration standards and validate the digestion method from an independent source. SERP's stated acceptable range for this check sample (60 ng/g) was 90–110 percent recovery of the true value. In the May of 1994 canal data, this check sample did not meet SERP's acceptance limits. A reagent blank was included in the analyses to correct for reagent contamination in the final calculation of the sample results in all the data except in the May 1995 canal data. The results for these data were "J" coded since the recoveries did not meet the SERP's stated limits. The check standard was the high standard, at 500 ng/l. The September 1995 marsh split samples were coded "NR" because raw data were not available for review. However, data compared favorably between EPA-SESD and SERP values on split samples. The average concentration reported for the splits for EPA was 117 ng/gm and for SERP 156 ng/gm with a average difference of 39.6 ng/gm. Both labs had comparable %RSD of 13.7 and 15.2, respectively.

5. Total Mercury in Fish by SERP

SERP was the primary laboratory for total mercury in fish analyses. Whole mosquitofish (Gambusia) were digested and analyzed for total mercury. When the recovery QC criteria for the SRM did not meet acceptance limits, the analysis could not be repeated because the entire fish was digested.. SRMs were included within the analyses to verify the accuracy of the data during the course of the analytical runs. For the September 1993 canal data, and one data set at the end of September 1994, SRMs were not analyzed. SERP set acceptance limits of 90–110 percent recovery of the true value for the SRM. The true value of the oyster tissue SRM was 64 ng/g and that of the dorm standard (Dog fish) was 4640 ng/g. The average percent recovery of the SRM (oyster std.) in September 1994 was 248 percent, and in April 1995, 287 percent. With the 64 ng/g oyster standard, there was not an adequate detector response following dilution, which contributed to the large variances for percent recovery. Four factors contributed to the wider range of recovery in the samples. First, the 64 ng/g SRM used did not provide an adequate detector response following dilution. Therefore, subsequent samples were run using the 4,640 ng/g SRM. Second, the inherent variability in biological samples results in larger percent recovery ranges. The FL DEP

recommends percent recovery ranges of 75–125 percent, rather than 90–110 percent. Third there might have been either an interference (effervescing due to CO_2 production) and/or fourth some of the SRM weights were documented incorrectly based on conversations with the analysts. Corrections for the elevated recoveries were not warranted due to the stated problems. SERP switched to the dorm standard because of sample handling problems. The average percent recovery for the Marsh sampling events were 92 percent for the April 1995 (range 77–99 percent), 98 percent for May 1996 (range 45–129 percent), and 80 percent in September 1996 marsh (range 45–100 percent).

Reagent blanks were included in the analyses, but were not used to correct for reagent contamination in the final calculation of the sample results. Reagent or glassware contribution in the blank impacted 110 samples analyzed from September 1994 to 1996. These sample values should be recalculated or the detection limits raised. Reagent blanks were not analyzed for 46 samples in September 1995 and 16 samples in September 1996. These data (172 of 704) were "J" flagged for no reagent blank correction. Matrix spikes were not analyzed because of the recognized difficulty to get a homogenous spike and the whole fish was used.

6. Organic Mercury in Soil/Sediment and Water by SERP

SERP served as the QA/QC laboratory for the Sept. '93 samples and as the primary laboratory for organic mercury in soil/sediment after Sept. '93. Battelle served as the primary laboratory for Sept. '93 but became the QC laboratory for solid/sediment duplicates/splits after this date. Samples and matrix spikes were analyzed in duplicate and results were corrected for recovery based on the matrix spike. SERP's policy in place during the time period of 1993–1996 was to store the electronic copies of the methyl/ethyl mercury chromatograms. No hard copies of the GC chromatograms were stored. Electronic chromatograms were only available for Sept. '96 for review. Selected analytical runs (electronic copies) were reviewed by the Team during the June 1998 visit to SERP. The Team was able to confirm qualitative and quantitative steps taken by the analysts for the selected data (soil/sediment) sets reviewed. Split water samples with the Battelle laboratory showed acceptable agreement between the field replicates for water samples even though the methods were different. The comparison for soil/sediment samples analyzed by both laboratories showed SERP data having lower average concentration (0.63 ng/gm) from samples collected at the same site than the Battelle average (0.96 ng/gm). Table 2 lists the comparison for the methyl mercury data between the two laboratories. There were some Canal samples that SERP had much higher concentrations than data reported by Battelle. This would indicate if there were a artifact for the distillation procedure it was not a consistent phenomenon. None of these data were qualified.

Table 2 Precision Data for Methyl Mercury			
Parameter	**Mean**	**%RSD**	**n**
Methyl Mercury – (Battelle) H_2O ppt	0.408[a] (0.39)[b]	14.68 (14.9)	68 (21)
Methyl Mercury – (SERP) H_2O ppt	0.21[b]	33.7	21
Methyl Mercury – (Battelle) Soil ng/gm	0.96[c]	-	62
Methyl Mercury – (SERP) Soil ng/gm	0.6[c]	-	62

a = mean of duplicate pairs
b = mean of duplicate pair from same distribution
c = average concentration for samples analyzed by the respective laboratory from the same location.

7. Total Phosphorus by SERP

The total phosphorus data in soil and water were reviewed for accuracy and compliance with the SERP SOP. SERP followed the procedures as outlined in their SOP with some documentation deviations. The laboratory did deviate from EPA's common laboratory practices in preparing the calibration curve. SERP ran a five-point calibration curve and if the correlation coefficient for this curve was greater than 0.995, indicating good linearity, the curve was recalculated as a two-point curve using the high standard and the origin. Values were recalculated by the review Team using all four points and there was no significant difference in the SERP reported values and the EPA calculated results. SERP determined a MDL of 0.6 μg/L using Appendix B to Part 136 of 40 CFR. A MDL of 1.4 μg/L was calculated by the OQA/ESAT Team from blanks and the lowest standard analyzed over a period of time, which corresponds well with the SERP MDL. SERP's practical quantitation limit (PQL) was stated to be 1.8–3.0 ug/L or 3–5 times the MDL. There were nine water values that had concentrations between 1.8–3.0 μg/L and one value below the stated PQl. All soil sample results were well above the calculated PQL 2.2 μg/gm, and no detection limits were changed.

Total phosphorus in water precision data from field replicates collected during the study from the canals and marsh show an average %RSD of 16.7 for the concentration range of 1.3–361 μg/L. Total phosphorus in soil/sediment precision data from field replicates from the canals and marsh show an average %RSD of 13.5 for the concentration range of 44–2452 μg/gm.

Split sample data between SERP and AmTest for the Sept. '96 sampling event showed acceptable agreement between the two laboratories. AmTest reported a MDL of 5.0 μg/L as compared to a lower MDL of 0.6 μg/L for SERP. No data were qualified.

8. Total Organic Carbon by SERP

Data obtained for TOC were reported from an instrument that operated in the concentration only mode. The output of the instrument was in concentration units that were entered directly into

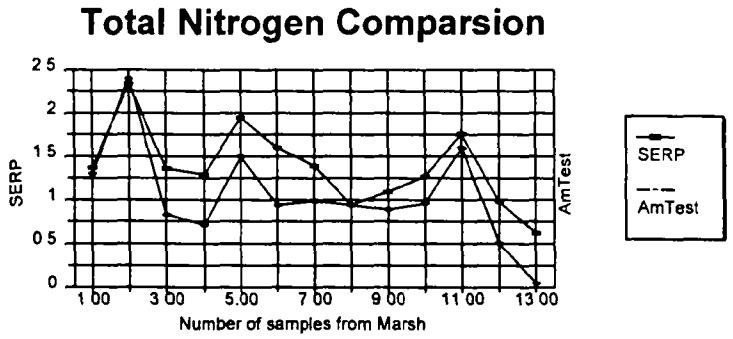
the spreadsheet. It was not possible to check the actual calculations used to generate the results for this parameter. Calibration curves and standards were checked and found to be in order. Therefore, the TOC data were not qualified.

9. Total Nitrogen Data for Water by SERP and AmTest

SERP total nitrogen analyses (TN) were performed using an experimental method developed with a ANTEK Model 7000N elemental analyzer by direct aqueous injection of the samples. Results were calculated using a 2.0 mg/L KNO_3 standard. No SRM nor additional standards such as glycine, urea or another stock source were analyzed during the analytical run. Since this was a direct injection a reagent blank was not necessary. Raw data for quality control split samples from AmTest were reviewed. Comparison of data between SERP and AmTest showed SERP values to average 0.33 mg/L higher. However, the SERP method measured total nitrogen and AmTest measured total Kjeldahl nitrogen (TKN). The results for AmTest would be lower since nitrogen as nitrate-nitrite would not be determined but would be included in the SERP data. The AmTest nitrogen data should be reported as TKN instead of TN. No SERP TN data were qualified. The AmTest data were qualified based on reporting as TN instead of TKN.

10. Alkaline Phosphatase Data by ESAT

The Data Review Team has reviewed all the water alkaline phosphatase data. Comparison of data for the May and September 1996, sampling events showed a difference in the fluorescence readings of the standard curves for the 2.5, 5, and 10 micromolar (uM) methylfluoroscein standards. The May standards appear to be approximately 1.5 times higher than the corresponding standards analyzed in September. Examination of the raw data showed the calibration curves for the analysis of both sampling events to be linear and recalculation of the curves by the data reviewer produced linear curves with correlation coefficients all greater than 0.990. The data reviewer was also able to reproduce the results for a selected random sampling of the samples for these sampling events. This would indicate that the calibration procedure and actual analysis of the data was performed correctly. It was not possible to compare samples analyzed in May and September to determine if the samples exhibited higher fluorescence in May, since there was no way to determine which sites, if any, were the same, or whether there should be a comparison. The method blanks, prepared daily from the methylfluorosceinphosphate substrate rather than the methyl fluorescein, were observed to see if the fluorescence readings for them were comparable between the May and September sampling events. Except for the method blank for the first analytical run in September, the fluorescence of the method blanks were reasonably consistent during all analytical runs in both May and September and compared quite



well. The fluorescence reading for the method blank for the first analytical run in September is approximately 3 to 4 times the fluorescence readings for the other analytical runs in both May and September. An interview with the analyst was conducted. The age of the standard used to calibrate the Shimadzu Spectrofluorophotometer was a possible reason for the elevated May results. The only quality control standard analyzed, however, was the high standard (10 uM) from the calibration curve, which always gave better than 90 percent recovery.

The difference in the fluorescence readings in May and those in September '96 could be explained by the use of an old standard or more likely explained by possible changes in the Shimadzu instrument. Raw data were obtained from SERP for alkaline phosphatase in other similar samples analyzed just before, during and after the May 1996 sampling event. These data showed similar fluorescence readings for the standards. There does not appear to be a practical way to recalculate the curves from the May sampling event so these samples match the curves from other sampling events, as stated above. It is not recommended that a factor be used to make the curve fit other data. Such a procedure would not be scientifically defensible. Therefore, the Team concluded that the May 1996 alkaline phosphatase data (123 samples) are highly questionable and recommends that the data be rejected and not reported. EAB should delete the May 1996 data from the database to avoid any misuse at a later date.

11. Sulfide in Water by ESAT

The EPA SESD/ESAT laboratory was the primary laboratory for total sulfide in water analyses. EPA records show that sulfide samples were collected for September 1995 sampling event. However, the method used for sulfide analysis during the sampling event was not followed as written. Samples were not mixed properly prior to adding the color developing reagent. This would cause the values to have a negative bias. The OQA Project QA chemist upon learning of the sampling handling deviation after debriefing the analyst, recommended in writing to the EAB Project Leader that the sulfide data be rejected. For this reason, 123 sulfide data points for the September 1995 marsh cycle were rejected and not reported.

12. Sulfate in Water by EPA-SESD

EPA-SESD served as the primary laboratory for sulfate analyses. Analysis of water samples were conducted using the turbidimetric method based on Method 375.4 during the period from Sept. '93 - Sept 95. Starting in May '96 an ion chromatography (IC) Method 300 was used to provide lower detection limits better recoveries for spiked water samples. EPA-SESD policy is to report all results less than the lowest nonzero calibration standard as below detection (flag data with a "U". The detection limit changed over the course of the sampling program. A 5.0 mg/L or "5U" reported detection limit was used for the period of Sept. '93 - Sept. '94. In May 1995, the detection limit was lowered to 2.0 mg/L or reported as "2U" to provide additional data except for a couple of samples where the detection limit was 1.0 mg/L. A review of the %RSD showed that the variability was generally less than 8 %RSD indicating excellent precision and did not show the typical degradation of precision as the detection limit was approached. This indicated the laboratory was producing valid results before the declared detection limits and data could be reported at a lower level without compromise. The ion chromatograph method was commenced in

May 1996, which allowed for reporting data down to 0.5 mg/L. Since many Marsh samples have concentrations below 0.5 mg/L a lower detection limit is needed for future studies to give added information for spatial display of the sulfate data.

13. Conclusions

The South Florida Everglades Ecosystem Assessment Project data set was reviewed against the Project DQOs, requirements defined in the Project Study Plan, each of the four laboratories SOPs and QC practices, and additional OQA procedures typically used for regulatory and compliance program reviews. Each individual datum was verified and validated. Over 20,425 measurements were made on samples analyzed by 4 laboratories for 20 constituents sampled in 5 media at about 200 canal sites and 550 marsh sites during the 9 field sampling events over the 10,000 km^2 area in South Florida. Included in these measurement were 7 structures sampled on a bi-weekly basis from February 94 thru February 97. The data that have been qualified after this rigorous review are:

- "J" – Soil/sediment methyl mercury (5%) for the secondary laboratory
- "NR" – Water total mercury for September 1993 Canal samples and April '94 transect data for primary laboratory
- "J" – Water total mercury, excluding September 1993 Canal, for primary laboratory.
- "J" – Periphyton total mercury for primary laboratory
- "J" – Soil/sediment total mercury for May 1994, 1995 Canal cycles and May 1996 marsh samples for secondary laboratory
- "NR" – Soil total mercury for September 1995 marsh split samples for secondary laboratory
- "J" – Mosquitofish total mercury for very low mercury concentration fish and SRM for primary laboratory
- "J" – Total nitrogen for the secondary laboratory for Sept. '96 marsh samples
- Reject – Water alkaline phosphatase activity for 123 samples for May 1996 marsh cycle from the primary laboratory
- Reject – Water sulfide for 123 samples for the September 1995 marsh cycle from the primary laboratory.

This *Summary of Data Review Findings* report should be kept in perspective. The South Florida Ecosystem Assessment Project was a research and monitoring effort, not a regulatory compliance project. It developed and implemented new methods for rapidly analyzing a large sample load with ultra-trace level constituent concentrations. This rigorous review occurred over about a 2.5 year period, being initiated in 1995 and completed in 1998. Out of the over 20,425 measurements made over the four year period, only 15 percent of data was qualified. There were 1,325 points flagged "NR"- not reviewed because raw data were not available from the laboratory, 1,783 assigned a "J" flag - analyte identification was acceptable for use but certain QC criteria were not satisfied and 256 values rejected and not reported in the database.

The OQA finds that the data set satisfies the Project objectives and permits the assessment of trends and a characterization of status and extent of ecological condition in the South Florida Everglades ecosystem. Data qualified with a "J" code would not compromise the characterization and trend analysis or the Project objectives. "J" coded should not be used solely in the setting of environmental standards unless additional data are collected to substantiate these original values that have all supportive QC and documentation. These data can be used as part of the weight of evidence approach for establishing environmental standards.

This data set should provide an important baseline for assessing the current conditions of the Everglades ecosystem and for monitoring future trends in this condition. The results from this QA/QC Review have already been, and will continue to be, used to refine the QA/QC procedures in future Project monitoring efforts.

APPENDIX D

Eastern Mosquitofish Studies

APPENDIX D

1.0 Eastern Mosquitofish

A predator protection criteria of 0.1 mg/kg THg for prey species has been proposed by the USFWS (Eisner et al. 1987). The eastern mosquitofish, *Gambusia holbrooki* whole body THg concentrations were presented in the proceeding section. About 15% of the canal miles and almost 70% of the marsh area have mosquitofish with mercury concentrations exceeding the predator protection criteria of 0.1 mg/kg. Because the mosquitofish is a prey species for piscivorous fish and birds and is an excellent indicator of Hg bioaccumulation, additional analyses were conducted on the mosquitofish populations in the canals and marsh. The purpose of these analyses were to determine if differences in population attributes or feeding habits among subareas or among latitudes might contribute to mercury bioaccumulation. The results are presented in the following section.

1.1 Descriptive Statistics

The descriptive statistics of the eastern mosquitofish were examined to determine the nature of the sample population and the possible relationships with Hg bioaccumulation, size, and condition factors. An additional test sample of mosquitofish was collected from near the marsh Hg hotspot and the Everglades ENR in July 1997.

1.1.1 Canal Fish

A length frequency histogram of the mosquitofish in the combined canal sample showed a normal distribution (Figure D.1). A total of 1,074 mosquitofish was analyzed with a median total length of 24.0 mm (0.95 inches), ranging in size from 10 mm (0.4 inches) to 38 mm (1.5 inches). Females made up 70.3% of the population sample and males made up 28.5% with the remainder made up by juveniles. The median length for males was 22.9 mm (0.9 inches), 1.2 mm (0.05 inches) smaller than the females at 24.2 mm (0.95 inches). A box and whisker analysis (Figure D.2) of the fish lengths by cycle indicated fish sizes in cycles 1 and 2 were significantly larger and smaller, respectively, than cycles 0 and 3. A box plot analysis of fish

length by geographic subarea showed significantly larger fish occurred in ENP, followed by EAA, WCA, and BCNP. Length/weight distributions and growth curves for each cycle show similar condition for each subpopulation (Figure D.3). A condition factor (CF) was calculated with the formula $CF = w/l^3$, where w = individual weight and l = individual length (Tesch 1968). The data combined by cycles showed significantly higher condition factors in fish from the EAA and ENP and lower condition factors for fish in the WCA canals and BCNP (Figure D.4).

1.1.2 Transect Fish

A length frequency histogram of the mosquitofish in the combined transect sample population is shown in Figure D.5. A total of 225 fish was analyzed, with 73.8% females, 25.3% males, and the remainder juveniles. The median total length was 25.6 mm (1.0 inch), and the sample population ranged from 14.2 mm (0.56 inch) to 34.4 mm. The median total length for males was 24.7 mm (0.74 inch), which was 1.9 mm (0.07 inch) less than females at 26.6 mm (1.05 inch). A box and whisker analysis (Figure D.6) of fish length found a significant difference between the LNWR and WCA3 in the fish sizes among the transects, however, all other distributions were not significantly different. Length/weight distributions showed similar condition among subareas (Figure D.7). The condition factors were significantly higher for mosquitofish from WCA2 and ENP transects than those on LNWR and WCA3 transects (Figure D.8).

1.1.3 Marsh Fish

A length frequency distribution of marsh fish included 2,158 individuals with a median size of 23.0 mm (0.04 inch), ranging from 9 mm (0.35 inch) to 39 mm (1.54 inches) (Figure D.9). The sample was normally distributed with 65.5% females, 25.1% males and 9.5% juveniles. Males had a median length of 22.4 mm (0.88 inch), which was 1.2 mm (0.05 inch) less than the females at 23.6 mm (0.93 inch). A box and whisker analysis (Figure D.10) of the data by cycle indicated the sample population had significantly larger fish in cycle 0 (April 1995) and significantly smaller fish in cycle 3 (September 1996) than the similar distributions in cycles 1 and 2. The September 1996 sample followed the driest dry period in spring 1996, which

apparently killed a large number of these small fish due to dry down and predation, followed by recruitment of young individuals during the following wet season. Length/weight distributions were plotted by cycle with associated growth curves(Figure D.11). There were no apparent differences in condition; however, in cycle 3 was somewhat less due to preponderance smaller individuals. The condition factor plotted by subarea for the combined data, indicated the highest condition factor occurred in fish from LNWR and BCNP and decreased in WCA2 and WCA3 with the lowest condition factor associated with fish in the ENP (Figure D.12).

1.1.4 Fish in 1997 Test Sample

A mosquitofish sample was collected from two marsh sites in 1997, the ENR and WCA3 near where the Hg concentrations in mosquitofish were previously found to be the highest in the marsh system. The ENR is an old agricultural field that has been converted into a prototype wetland stormwater treatment area designed to remove TP from stormwater. Agricultural fields were not sampled as a routine part of the REMAP study. A total of 153 fish was analyzed (Figure D.13). The median fish size at the ENR and WCA3 sites was 19.7 mm (0.78 inch) and 17.2 mm (0.68 inch), respectively. The fish ranged in size from 9.0 mm (0.35 inch) to 35.6 mm (1.40 inches) at the ENR site and from 7.6 mm (0.3 inch) to 34.0 mm (1.34 inches) at the WCA3 site. A box and whisker plot shows the ENR fish were significantly larger than the WCA3 fish (Figure D.14). The condition factor was higher for ENR fish (Figure D.15). The THg concentrations in fish from the ENR were found to be the lowest measured in fish during this study (Figure D.16). Samples analyzed by three different laboratories found that THg concentrations in fish from the ENR were less than 10 μg/kg while the THg in fish from the WCA3 site averaged about 150 μg/kg (Figure D.16).

It is apparent from these data that a consistent sample of mosquitofish was collected from both canal and marsh habitats throughout the course of this study. The sample collected is representative of the naturally occurring mosquitofish population in the system. The average size of the mosquitofish populations observed in the Everglades ecosystem are very small for the species found anywhere in their range (Trexler personal communication). The Everglades ecosystem is a food limited, oligotrophic system (Loftus personal communication) that reduces

the size of this species. The usefulness of this species as an important indicator for monitoring the bioaccumulation of Hg in the Everglades ecosystem is evident from the changes in THg concentrations in this species across the TP, TSO_4 and TOC gradients in this system. The consistent future sampling and analysis of mosquitofish can provide the information needed to assess changes and trends in Hg contamination in the South Florida Everglades ecosystem.

1.2. Mosquitofish Gut Contents

Bioaccumulation and biomagnification of MeHg through the food web is influenced by both the quantity and quality of the ingesta. An additional study was conducted to determine the gut contents of mosquitofish collected during the September 1996 sampling cycle.

Many species are known to switch diets both during development and based on food availability in their environment. Omnivorous species have the potential to yield important influences on their prey by switching their diet choice with regard to its changing abundance, targeting it in times of plenty, and ignoring it in times of scarcity. Furthermore, the ability to switch foods, even going from herbivore to carnivore, may buffer a species from fluctuations in food supply and permit it to sustain larger population densities. Ontogenetic changes in diet may also reduce competition among age classes (Werner and Gilliam 1984). One possible outcome in such a case is that individuals may begin to specialize in subsets of their potential prey (Magurran 1993). Also, local populations of omnivores may diverge substantially in their feeding biology, and role in local ecological communities, as a result of local environmental conditions and food availability.

In spite of these possibilities, few studies have characterized the diet of an omnivorous species over a large spatial scale, probably because of the inherent difficulties of collecting adequate specimens to do so. The eastern mosquitofish (*Gambusia holbrooki*), and its western congener, *Gambusia affinis,* are known to be aggressive omnivores (reviewed in Meffe and Snelson 1989) with the potential to yield greater effects on the ecosystems where they live than might be predicted based on their small size (Courtney and Meffe 1989). They have been widely introduced world-wide for mosquito control and have been responsible for the extinction of native fishes in some cases through consumption of larvae (Meffe et al. 1983). Experimental

ecological studies have documented their potential to regulate the dynamics of other community members through direct consumption and competition (Hurlburt et al. 1972, Hurlbert and Mulla 1981, Harris 1995, Belk and Lydeard 1994, Schaefer et al. 1994). Mosquitofish are also cannibalistic (Krumhotz 1948, Meffe and Crump 1987). Mosquitofish are known to feed on plant matter and detritus, in addition to these predatory predilections.

1.2.1 Methods

1.2.1.1 Data Gathering

Mosquitofish were collected by dipnet from 101 locations scattered across the Florida Everglades (Figure D.18) from September 18 to 23, 1996. The sites were selected by a stratified-random procedure described earlier. Immediately following capture the fish were placed in 10% formalin in the field to rapidly stop the digestion of the stomach contents. Twenty fish were collected at each site of which 12 to 14 specimens were analyzed for stomach contents. Duplicate samples were taken at 10 sites.

Individual mosquitofish were dissected and their gut contents removed and separated into six categories: (1) plant matter (pooling algae, vascular plant, and detritus), (2) cladocera, (3) aquatic mites, (4) chironomid larvae (midge larvae), (5) adult midges, and (6) other (primarily spiders, ants, aquatic beetles, and fish). Counts of the number of items in all animal categories were recorded for each mosquitofish, along with their sex and standard length. Males could be identified readily by the presence of a gonopodium, and females were identified by presence of mature ovaries or by standard length exceeding 18 mm (0.71 inch). Juveniles were all fish below 18 mm (0.71 inch) standard length lacking a gonopodium. The presence or absence of plant matter was recorded for each specimen, and if no food was present this was also noted. All food items for the fish from a single population sample were pooled and the mass of each food category was determined. The sum of these masses provided an estimate of the total mass of food consumed by that sample of fish.

1.2.1.2 Statistical Analyses

The pattern of food choices by individual fishes were examined, followed by analyses of the population samples. No mass estimates were available for the food items of individual specimens, particularly for plant food, so the presence or absence of food types was examined. Principle components analysis was conducted to investigate patterns in the food choices. The covariance matrix was factored because all data were scored as present or absent rendering all variables on a similar scale (Stevens 1986); varimax factor rotation was used. The effect of fish size and sex on food choices were examined by logistic regression of presence/absence of food types in individual fish gut contents (Trexler and Travis 1992). Fish with empty stomachs were excluded from these analyses. The presence/absence of food in the gut of individual fish was coded as a dependent variable and this was examined separately by logistic regression with fish sex and standard length as independent variables.

The percentage of each food category in the diet of fishes from each population sample was calculated from the mass data. These percentages were analyzed in analyses of covariance by grouping populations into geographic regions of the study area using two schemes. First, populations were grouped according to the water management region where they were found: LNWR, WCA2, WCA3, ENP, and BCNP. There are general north to south gradients in productivity across the Everglades following patterns of nutrient enrichment from agricultural runoff (Davis 1994, Stober et al. 1996). The effects of this pattern by grouping the populations into 6 regions by latitude from north to south were examined: (1) > 26.4°N; (2) 26.4°N to 26.2°N; (3) 26.2°N to 25.9°N; (4) 25.9°N to 25.7°N; (5) 25.7°N to 25.5°N; and (6) <25.5°N. The average standard length of fish from each collection was retained as a covariate in these analyses. In all cases, data were examined for consistency with the assumptions of standard statistical procedures such as normality, and transformations were applied as needed to fulfill the assumptions of analyses (Zar 1984).

1.2.2 Results

1.2.2.1 Overview

Over 1200 fish averaging 15.9 mm (0.63 inch) standard length, most of which were juveniles were examined (Table D.1). Males were typically smaller than females by over 2 mm (0.08 inch). A conversion between standard and total length for males, females, and juveniles with an r^2 over 0.9 is : Standard length = -1.337 + 0.886 (total length). Periphyton comprised 36% of the diet of mosquitofish based on biomass in gut contents, with insect, crustacean, arachnid, and piscine prey accounting for the remaining 64%. Adult midges, undoubtedly gleaned from the water surface, accounted for 34% of the biomass of the diet, and midge larvae, probably taken from floating, epiphytic and benthic periphyton mats, accounted for an additional 9.6%. Two fish (both *Heterandria formosa*) and an assortment of spiders, ants, and beetles account for 15% of the diet by biomass. About 50% of the individual fish had plant matter present in their guts, and about 45% had adult midges (Table D.2). Chironomid larvae and "other" prey were both found in about 10% of the fish, while mites were present in around 8.0% and cladocerans in only 3.0% of the fish examined. Very few of the fish had empty stomachs (53 out of 1,265 fish examined).

1.2.2.2 Individual Fish

Both the size and sex of fish influenced the likelihood that they had empty stomachs. Larger specimens were more likely to have empty stomachs than smaller ones, though even the largest juveniles had less than a 5% chance to have empty stomachs because of their relatively small size (always less than 18 mm (0.71 inch); Figure D.19). Females and juveniles did not differ in the probability of having food present in their gut once size differences were accounted for; however, males were more likely to have empty guts than females or juveniles at the same size (Figure D.20). Still, the likelihood of not having fed prior to collection was low in the sample; the model estimated that even the largest specimens had only a 20% likelihood of no gut contents.

Principal components analysis indicated that patterns of consumption of the six food categories did not overlap. No factor component loaded heavily on more than one food type,

although the first two components explained over 60% of the total variance. A component loading heavily on the incidence of plant matter explained 31.4% of the variance in gut contents, one loading on adult dipterans explained a similar amount (30.9%), and two other components loading on chironomid larvae and "other" each explained an additional 11.8%. The lack of structure in the data revealed by this analysis led to the consideration of each food class individually in subsequent analyses.

Size was a significant factor in the likelihood that mosquitofish had consumed plant matter immediately preceding collection. The likelihood of finding periphyton in the gut decreased with size (Table D.3) from approximately 60% for 10 mm (0.4 inch) juveniles to about 35% for a 25 mm (0.99 inch) adult female (Figure D.21). Males were less likely to have consumed periphyton than females (adult fish only, model: Pr [periphyton in gut] = constant + sex + size, $t_{212} = 7.47$, $P = 0.032$, size $t_{515} = 2.28$, $P = 0.015$, sex $t_{515} = 2.14$, $P = 0.012$; Figure D.22).

Some categories of animal prey decreased in frequency in the diets of mosquitofish as they got larger, others increased, and some were unaffected by size. The incidence of cladocerans decreased with size, while adult dipterans and "other" prey increased in frequency as fish got larger. Chironomids and mites were equally likely to appear in the diets of all size of fish (Table D.4). In general, the diets of males and females did not differ regarding the incidence of animal prey, with the exception of "other" prey. However, this difference was explained by the size difference between the sexes (adult fish only, model: Pr("other" in gut) = constant + sex + size +sex by size , $t_{213} = 12.01$, $P = 0.009$, size $t_{515} = 2.26$, $P = 0.015$, sex $t_{515} = 1.63$, $P = 0.11$, sex by size $t_{515} = 1.7$, $P = 0.09$).

1.2.2.3 Geographic Variation

The average size of fishes examined differed among the five water management units of the Everglades. Population samples of mosquitofish from LNWR and WCA3 contained significantly smaller fish than average, and those from WCA2 and BCNP tended to be larger (Table D.1). In light of the findings of diet changes with mosquitofish size, analyses comparing these water management areas must adjust for the size of specimens in samples. And, as

expected, the mass of food found in the guts of fishes is related to the average size of specimens in the population samples (Table D.3). However, differences among management areas persist after size variation is accounted for: WCA2 and WCA3 fishes tended to have more food in their guts than average, while those from LNWR and BCNP had less than average (Table D.4). When the data were sorted into six latitudinal categories, similar patterns were revealed.

Some food items appeared to vary across the water management units in their prevalence in the diet of mosquitofish. The relative amount of plant matter in the diets displayed a tendency to vary among regions (Table D.3; P=0.088, dropping to P=0.056 when length was excluded from the model) with BCNP fishes displaying notably less plant matter than the fish from the other four regions examined; this result was obscured when regions were created along a north to south gradient (Table D.4). Chironomid larvae displayed a more marked variation among regions in their inclusion in diets. They accounted for over 20% of the diets in LNWR and WCA2, but dropped to below 10% in all other regions (Table D.4). This is also seen as a north to south gradient when the data are grouped by latitude (Table D.4). "Other" prey items appeared most frequently in the diet of fishes from BCNP, and least in data gathered from fish from LNWR, though this result was obscured when samples were compared along the latitudinal gradient (Table D.4). Adult midges and mites displayed no regional patterns in their incidence in mosquitofish diets (Table D.3).

Table D.1 Descriptive statistics of size of the fish examined. Standard length (mm) and 95% confidence intervals are reported, with sample sizes below.

Data Grouping	All Fish	Males	Females	Juveniles
All Populations				
	15.93 ± 0.20 1270	17.12 ± 0.34 108	19.66 ± 0.28 407	13.73 ± 0.07 748
By Region				
WCA1	15.04 ± 1.16 11	—	—	—
WCA2	17.20 ± 1.70 8	—	—	—
WCA3	15.20 ± 0.26 40	—	—	—
ENP	16.18 ± 0.75 33	—	—	—
BCNP	17.83 ± 1.54 9	—	—	—

Table D.2 Relationship of diet to size in juvenile and female eastern mosquitofish. Results from logistic regression of incidence (presence/absence) of food item in gut contents on female size measured as standard length. In one case, adult dipterans, a significantly better model fit was obtained by use of ln-transformed length. Average Size, n-columns indicate the average size (mm) and sample size of fish with each item absent or present. The Pr (Present) is the probability estimated at the grand mean size. Slope is the probability that each food item will be present in gut contents with a 1-mm increase in standard length, except for adult dipterans which were best fit on a log scale.

Food Type	χ^2	P	Item Absent Average Size, n	Item Present Average Size , n	Pr (Present) Observed	Pr (Present) Predicted	Slope Pr(Present) vs Standard Length
Periphyton	11.6	0.001	16.2, 580	15.5, 581	0.500	0.500	-0.014
Cladoceran	8.1	0.006	15.9, 1117	14.3, 39	0.034	0.031	-0.004
Adult Dipteran	3.9	0.05	15.7, 645	16.0, 511	0.442	0.442	0.127*
Chironomid Larvae	0.5	>0.4	15.8, 1042	15.6. 114	0.099	0.098	
Mite	1.8	0.185	15.9, 1058	15.3, 98	0.085	0.084	
Other	17.9	<0.001	15.7,1040	17.2, 116	0.100	0.094	0.090

* indicates slope on a ln (standard length) scale

Table D.3 Geographical analysis of the percentage of total mass attributable to each food type from the gut contents of mosquitofish. Two geographical groupings were used, Water Management Units refers to comparisons of population means among management units, while Latitude refers to grouping populations by latitude. No interactions were significant.

		Water Management Units			Latitude		
Food Item	Effect	F	DF	P	F	DF	P
Plant Food	Length	0.004	1,95	0.843	0.060	1,94	0.807
	Region	2.088	4,95	0.088	1.410	5,94	0.228
Cladocera	Length	0.115	1,95	0.736	0.541	1,94	0.464
	Region	0.727	4,95	0.576	0.785	5,94	0.563
Adult Midges	Length	0.430	1,95	0.514	0.373	1,94	0.543
	Region	1.033	4,95	0.395	1.558	5,94	0.180
Midge Larvae	Length	8.379	1,95	0.015	9.034	1,94	0.003
	Region	4.130	4,95	0.014	5.042	5,94	0.001
Mites	Length	0.227	1,95	0.635	0.191	1,94	0.663
	Region	1.175	4,95	0.327	0.725	5,94	0.606
Other Prey	Length	1.974	1,95	0.163	4.046	1,94	0.047
	Region	1.900	4,95	0.117	1.224	5,94	0.304
All Food Mass	Length	7.373	1,95	0.008	6.85	1,95	0.010
	Region	2.878	4,95	0.027	2.580	5,95	0.031

Table D.4 Adjusted means from analyses of food items by geographical groupings. All means are adjusted to the grand mean fish size of 15.93 mm standard length.

Grouping	Plant Food (%)	Midge Larvae (%)	Other (%)	All Food (mg)
Water Management Units				
WCA1	27.3	25.1	6.1	0.973
WCA2	30.1	21.1	13.1	2.328
WCA3	42.7	7.9	16.5	2.352
ENP	36.5	6.0	13.1	1.348
BCNP	8.6	4.7	32.4	1.096
Latitude				
> 26.4	26.7	25.1	6.7	0.968
26.4 - 26.2	35.4	20.7	10.0	2.636
26.2 - 25.9	34.6	5.4	22.4	1.678
25.9 - 25.7	34.6	2.9	20.8	2.033
25.7 - 25.5	50.3	5.7	14.2	1.863
< 25.5	21.6	6.6	13.2	0.942

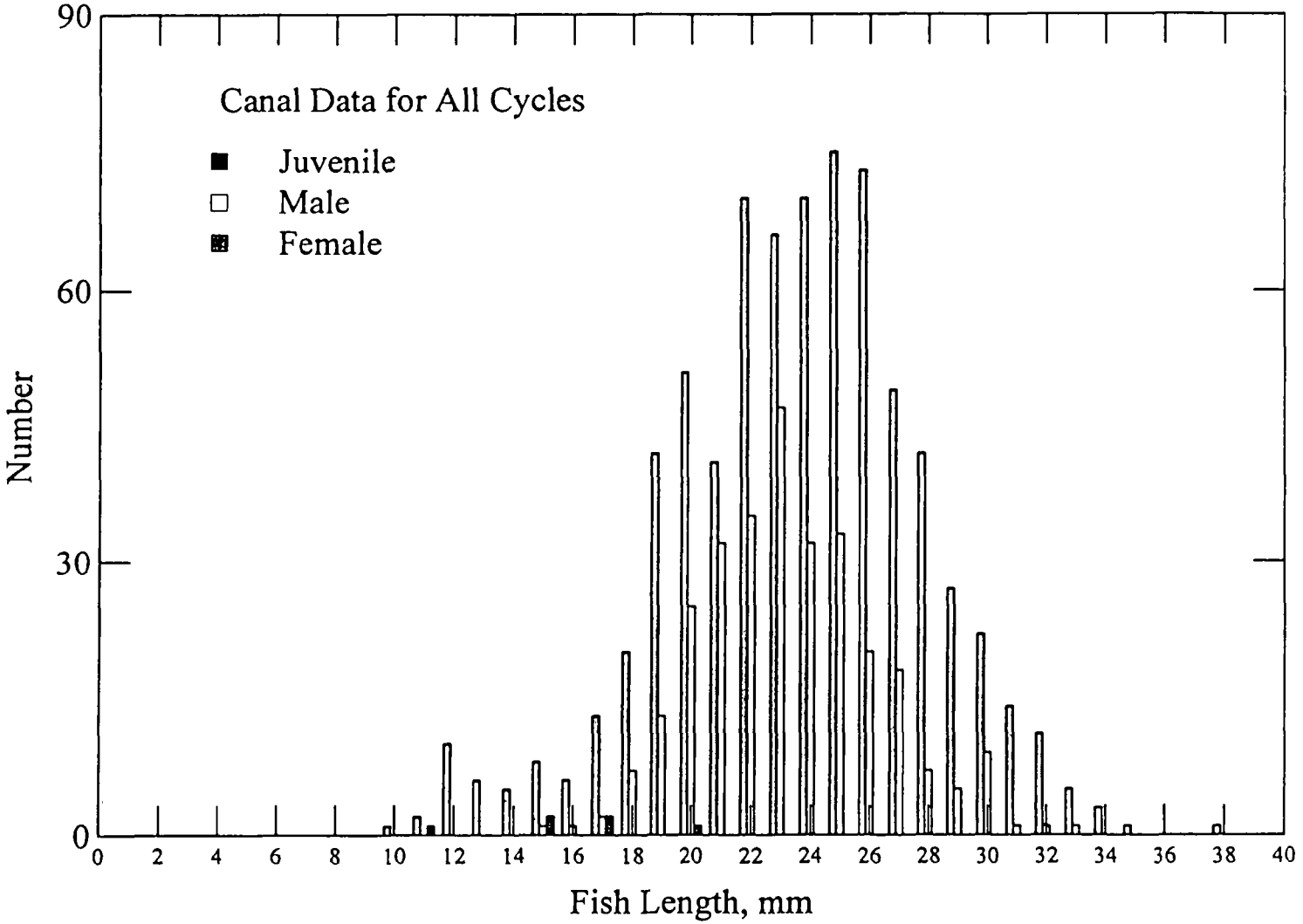


Figure D.1 Length frequency histogram of mosquitofish collected in the canals.

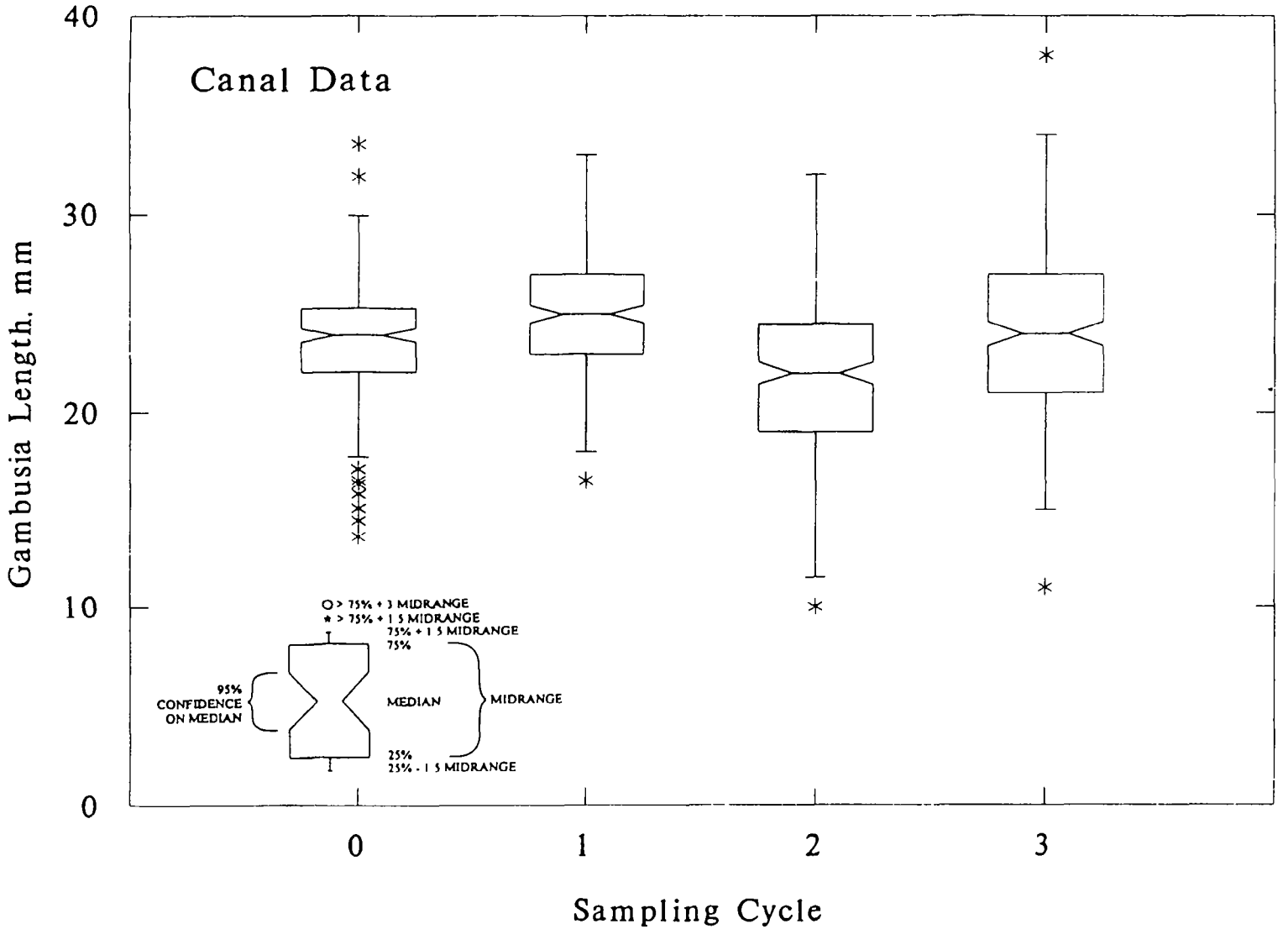


Figure D.2 Notched box and whisker plot comparing mosquitofish length for each sampling cycle.

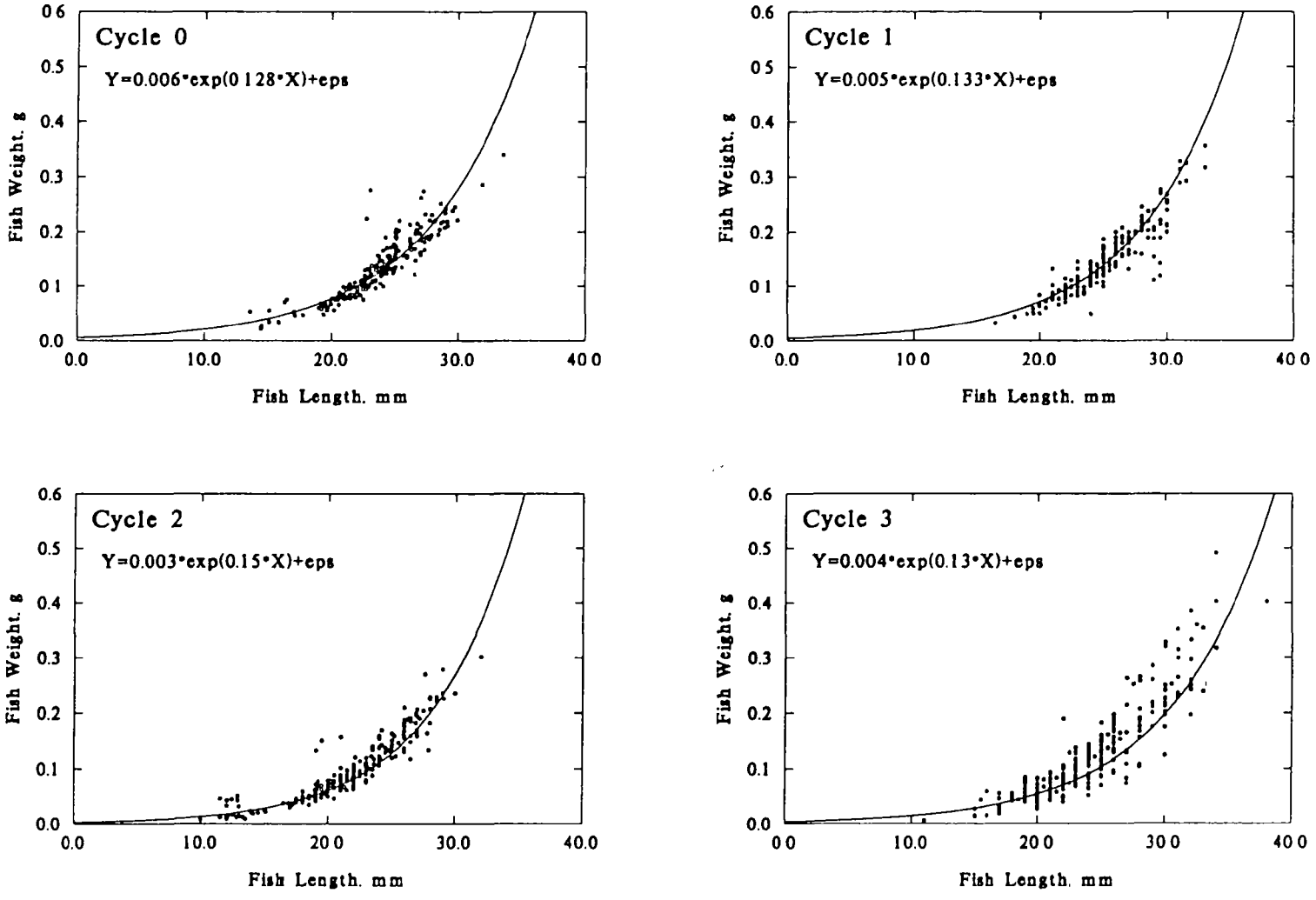


Figure D.3 Canal mosquitofish length versus weight with derived growth curves for each sampling cycle.

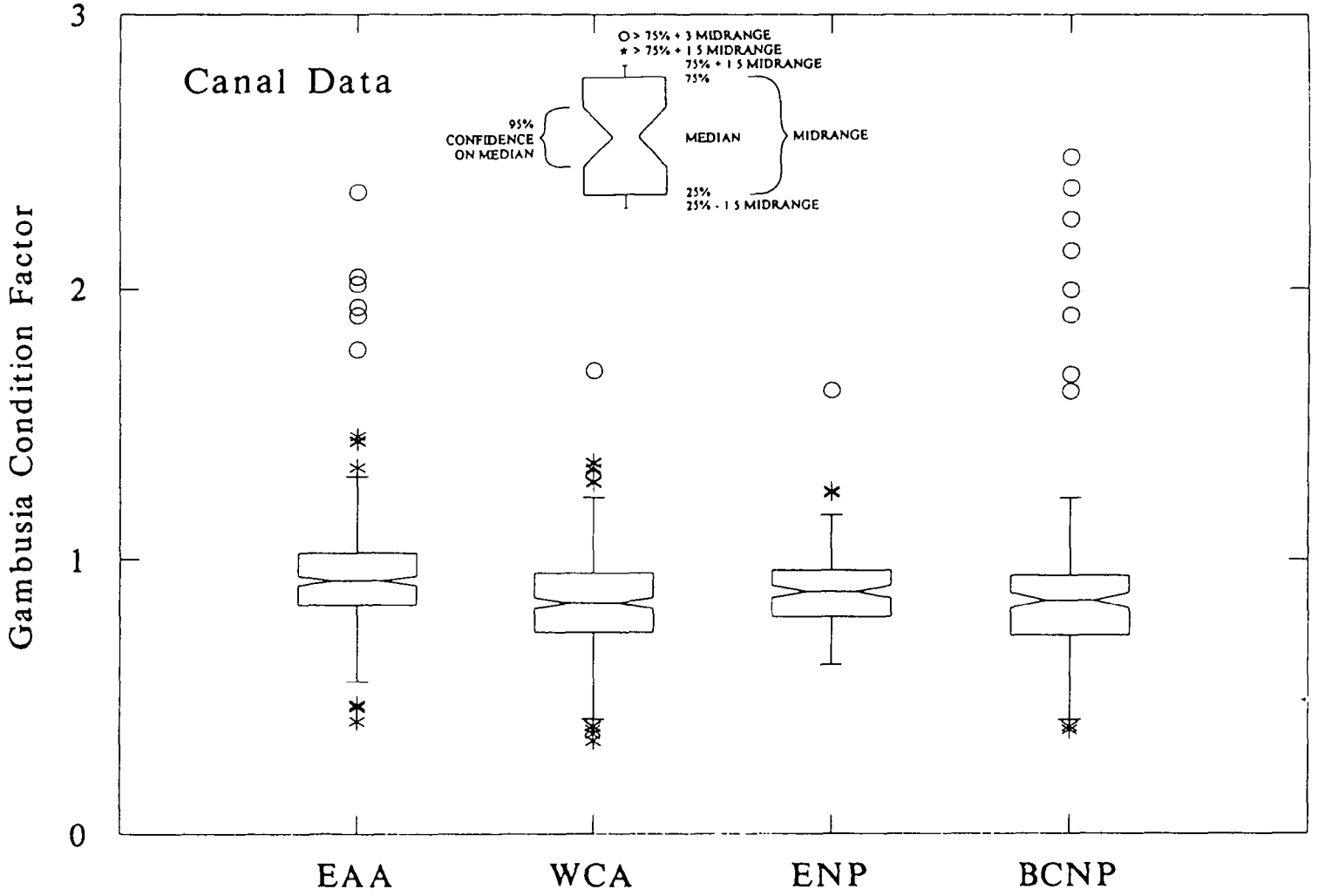


Figure D.4 Notched box and whisker plot comparing condition factors for canal mosquitofish in subareas.

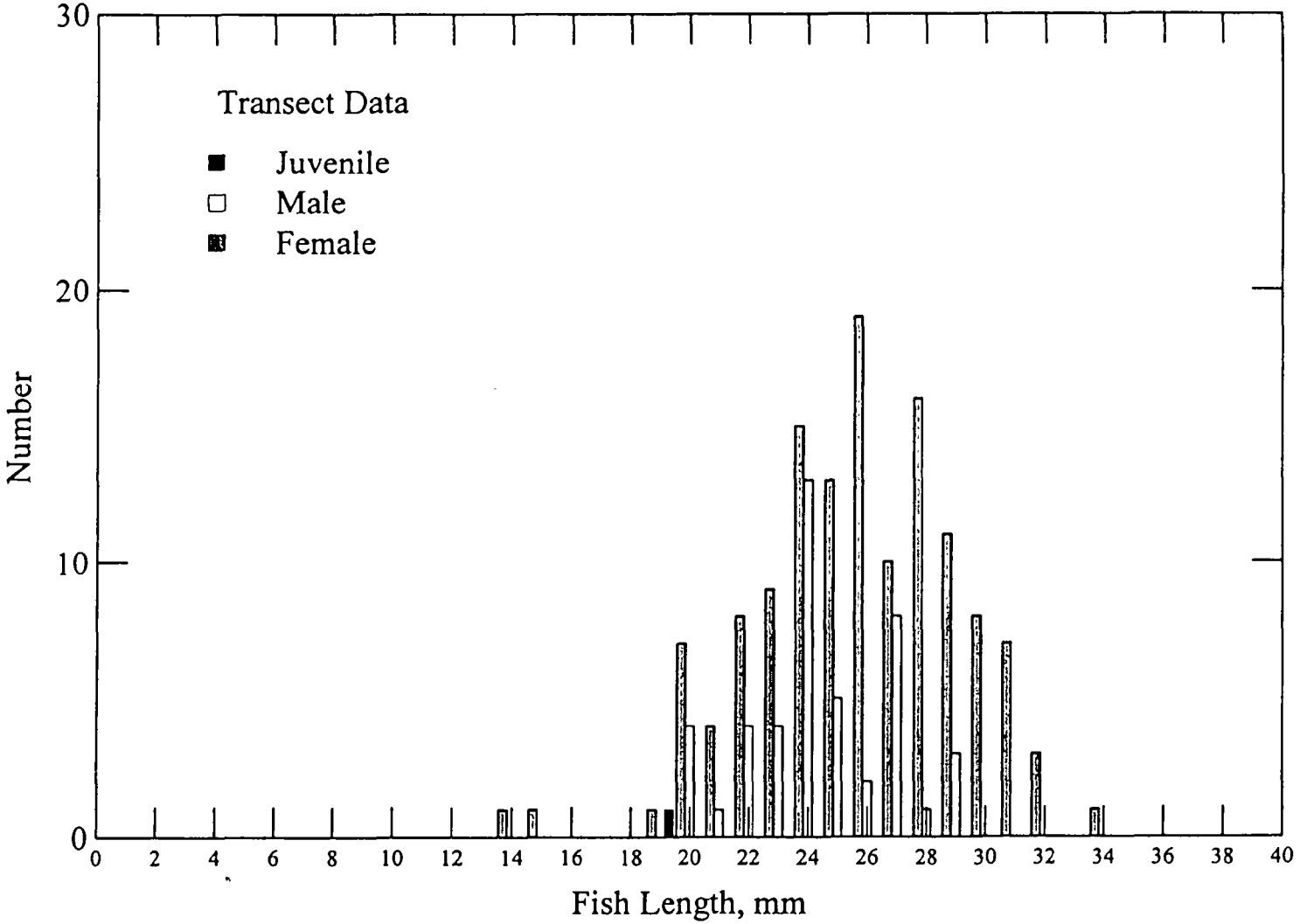


Figure D.5 Length frequency histogram of mosquitofish collected along the marsh transects.

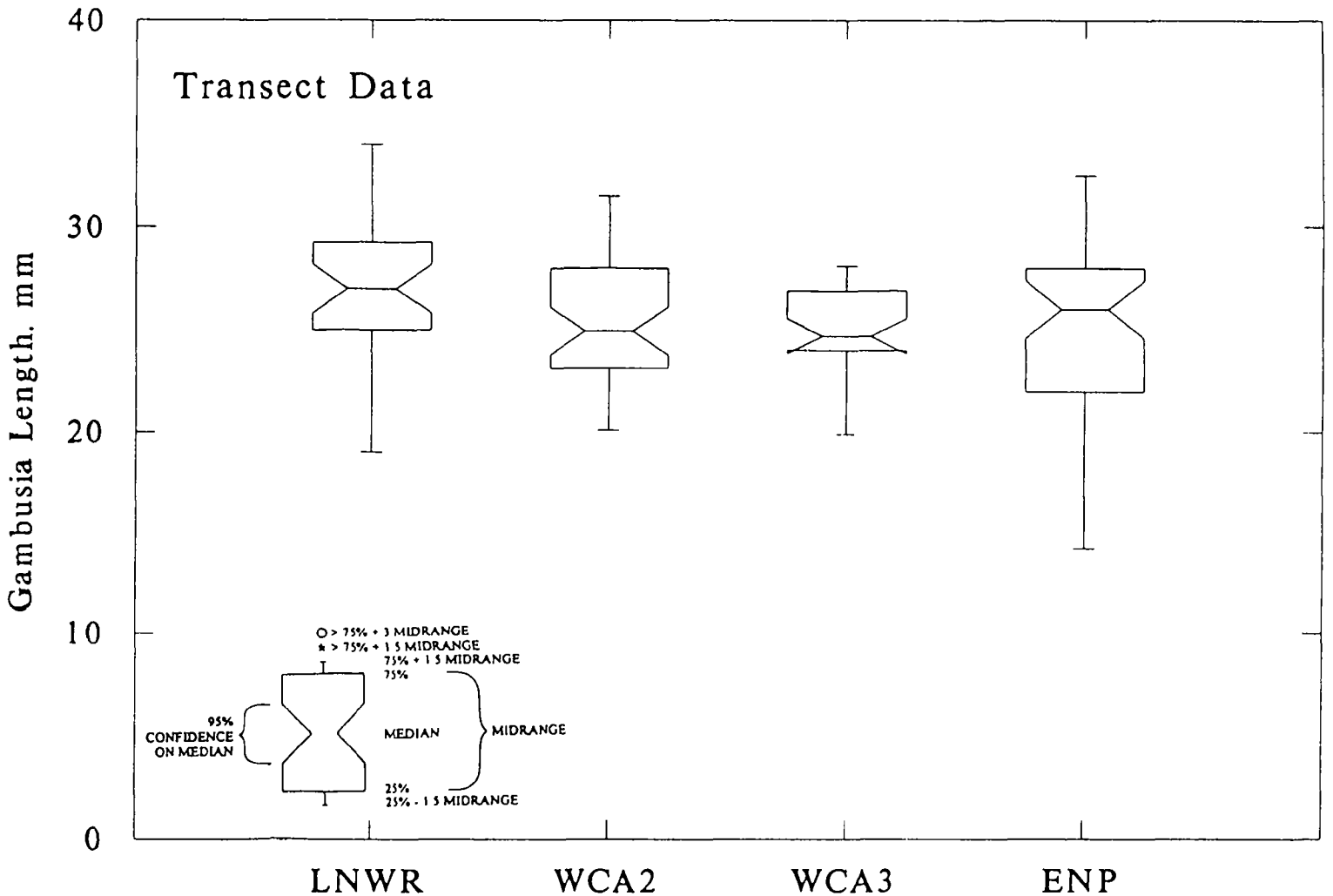


Figure D.6 Notched box and whisker plot comparing mosquitofish lengths in each of the marsh transects.

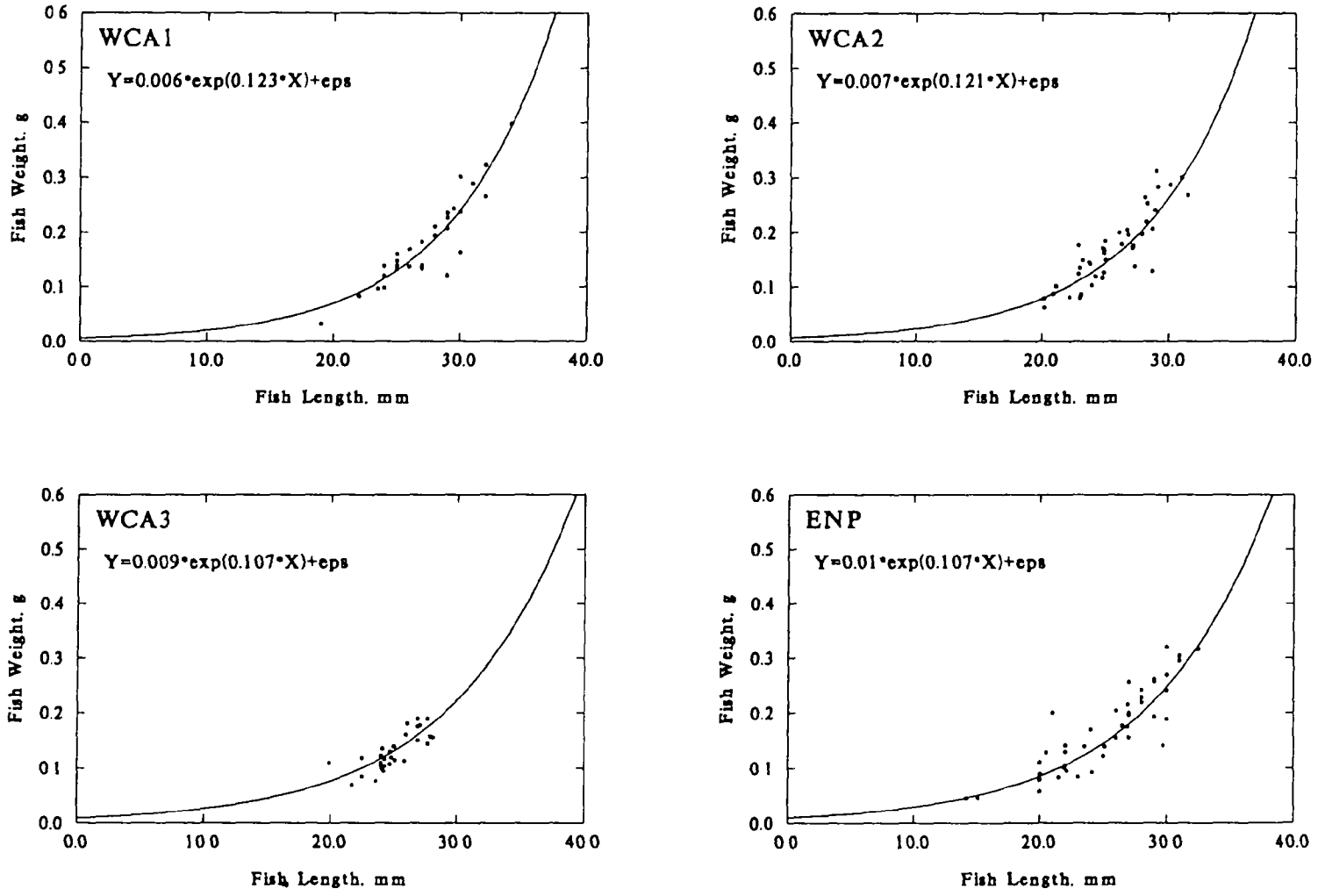


Figure D.7 Mosquito fish length versus weight with derived growth curves for each marsh transects.

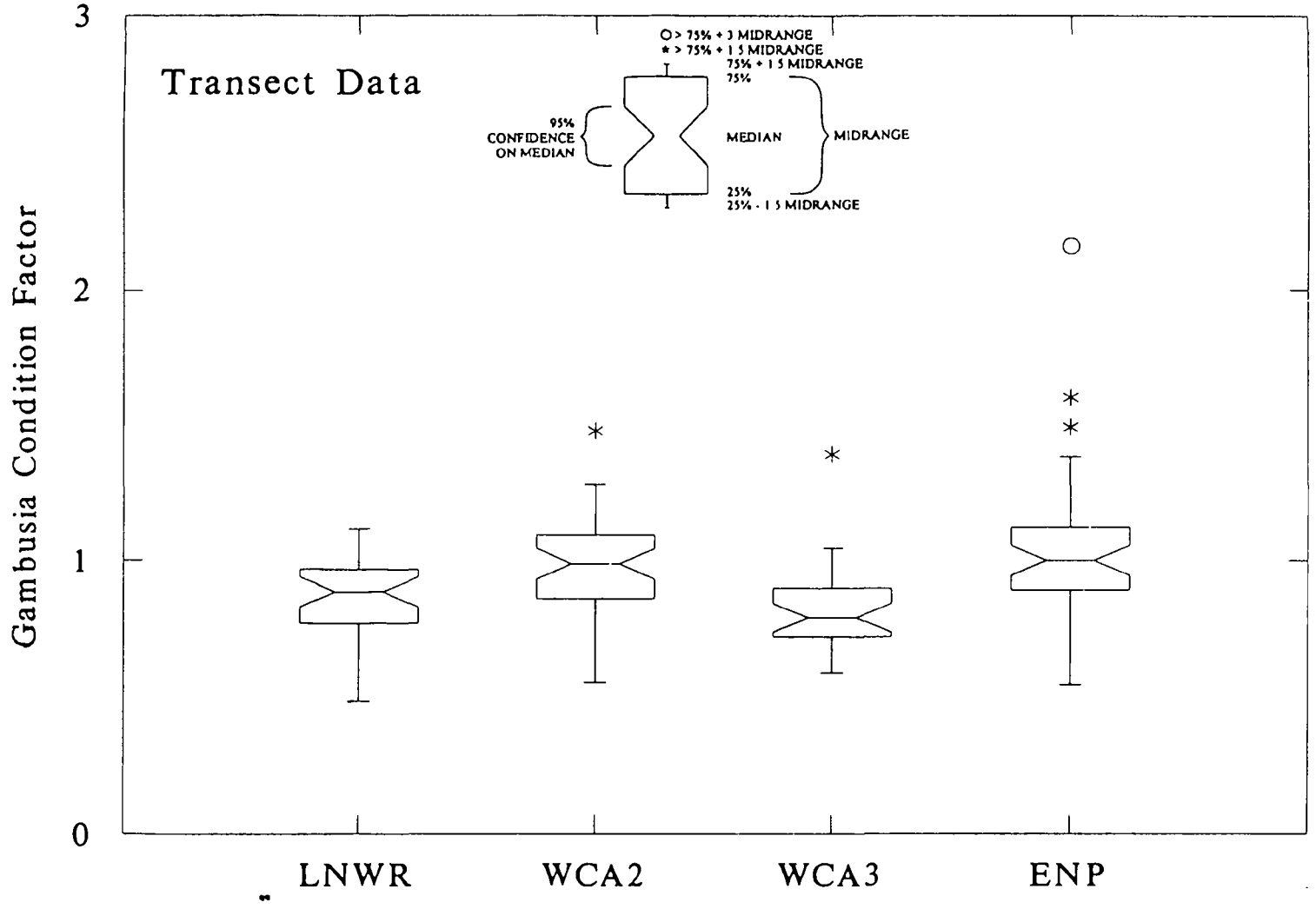


Figure D.8 Notched box and whisker plot comparing mosquitofish condition factors for transects.

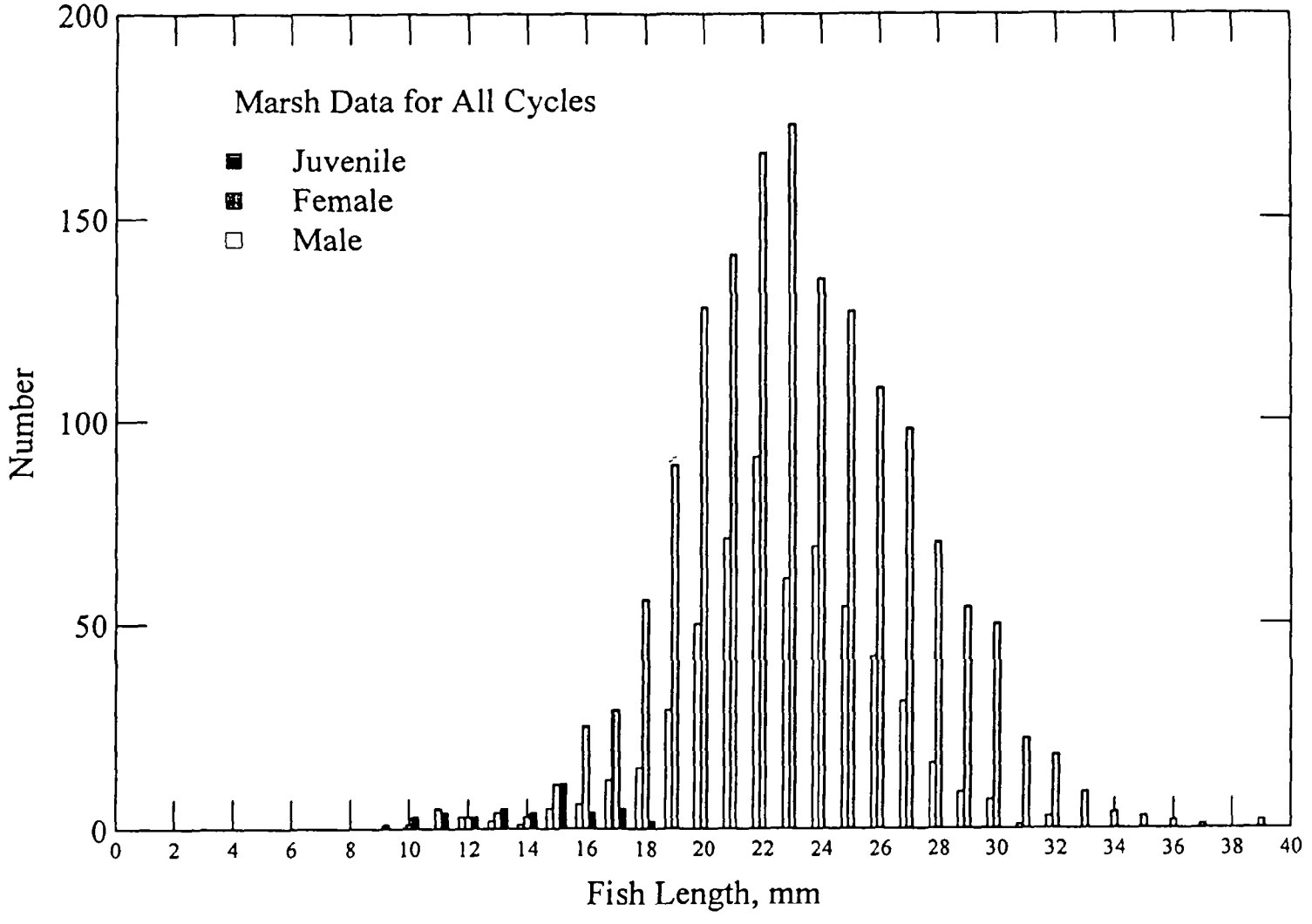


Figure D.9 Length frequency distribution for mosquitofish collected in the marsh.

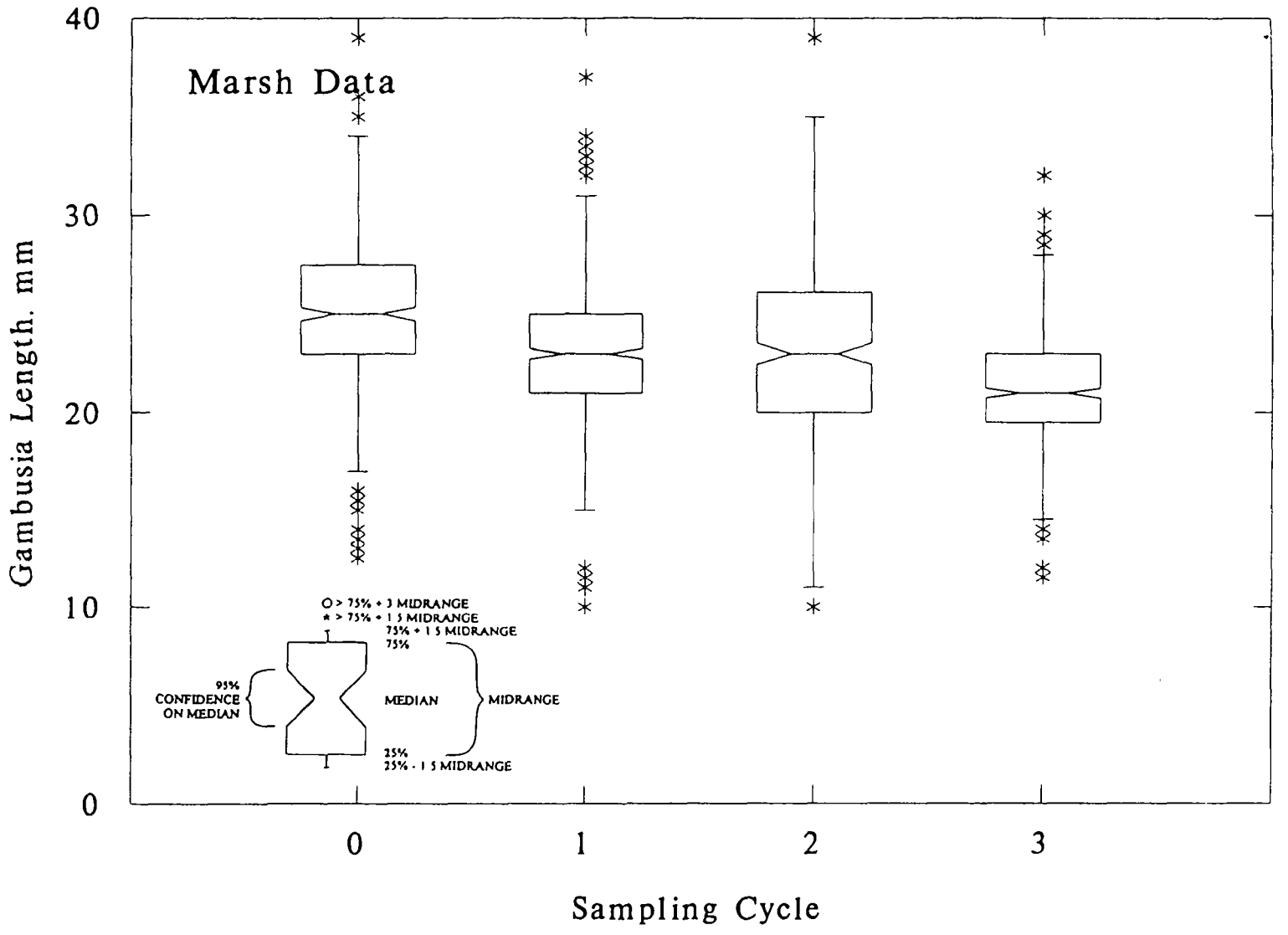


Figure D.10 Notched box and whisker plot comparing lengths of mosquitofish collected in the marsh for each sampling cycle.

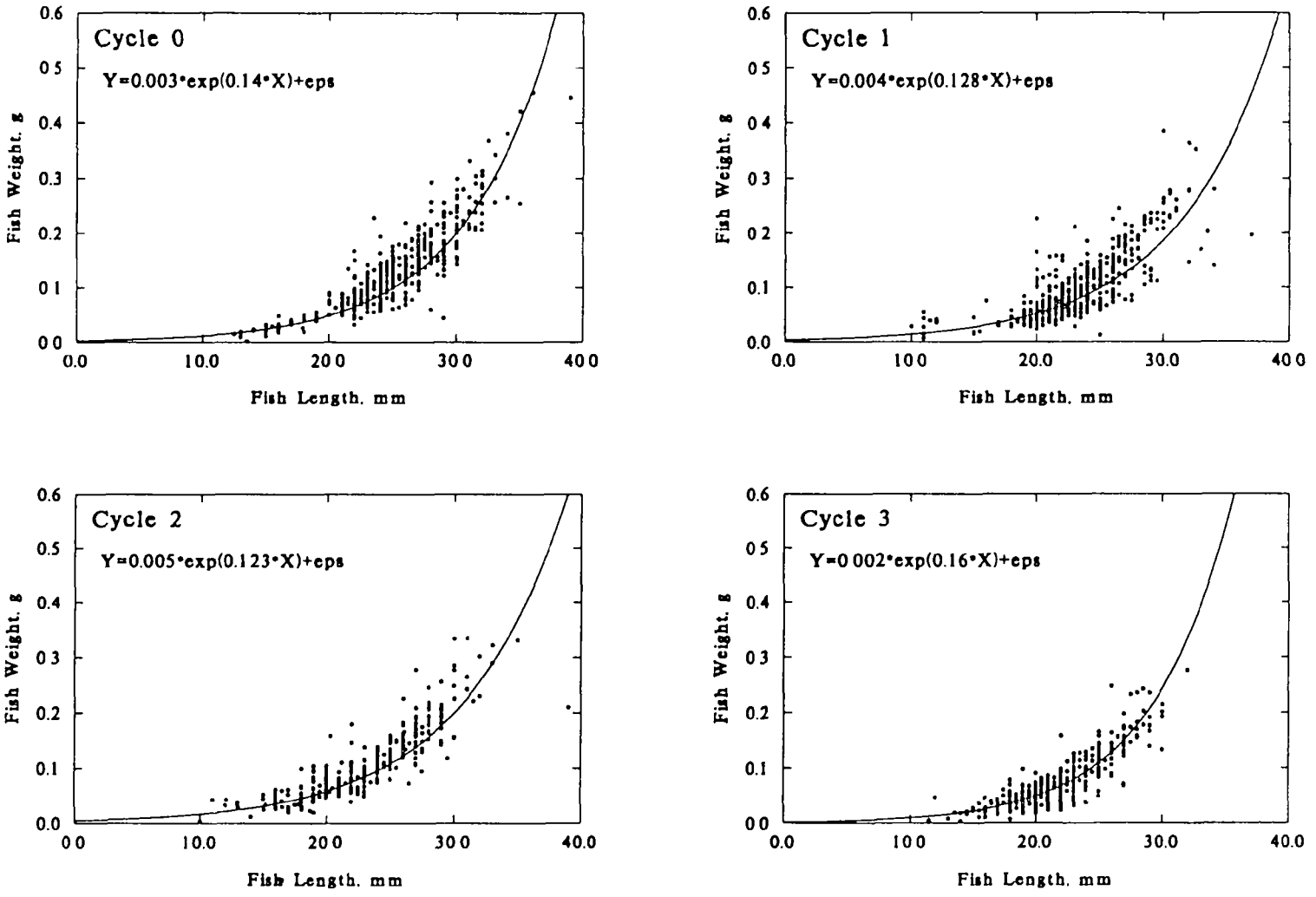


Figure D.11 Length versus weight of mosquitofish collected in the marsh during each sampling cycle with derived growth curves.

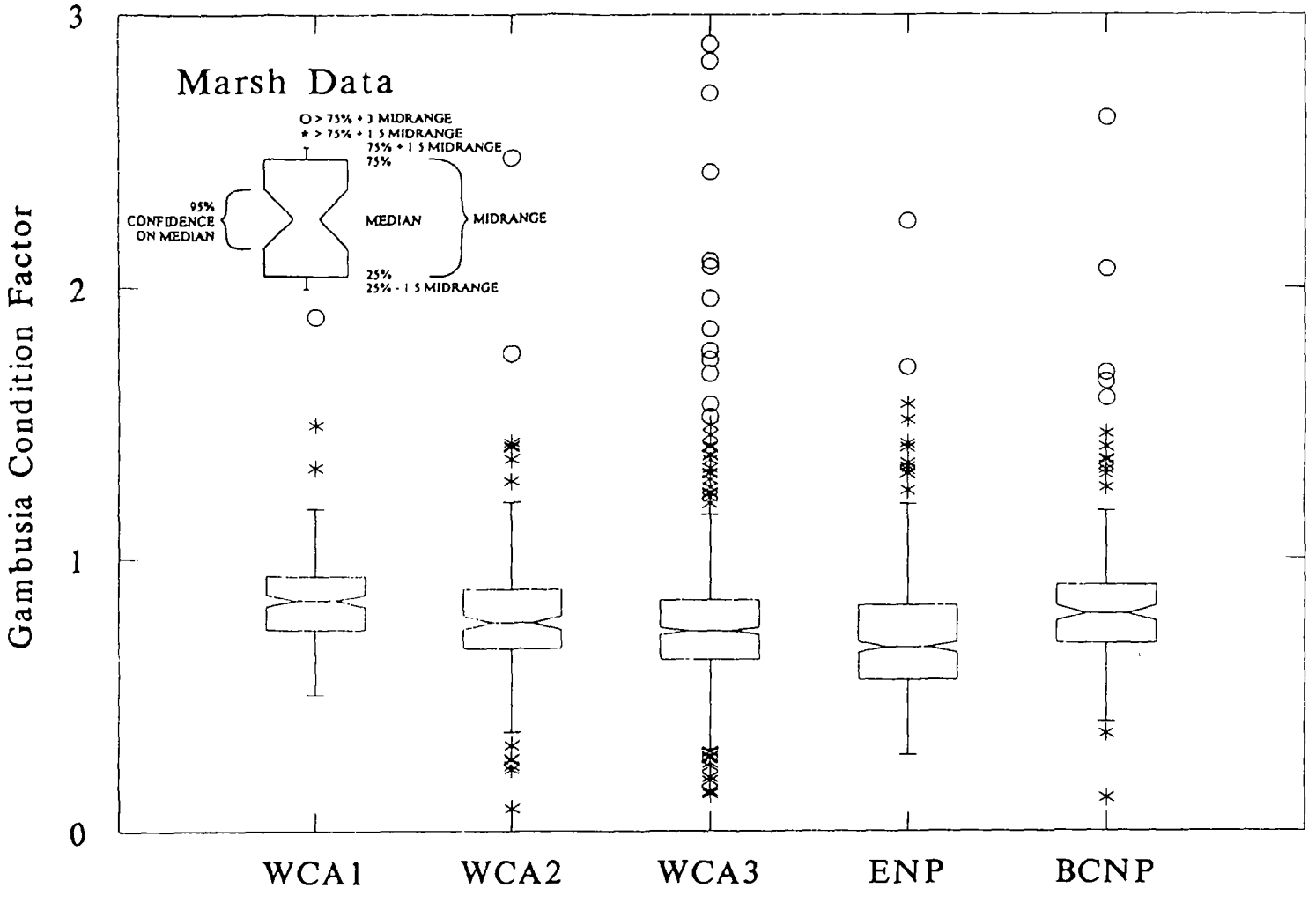


Figure D.12 Notched box and whisker plot comparing condition factors for marsh mosquitofish collected in subareas.

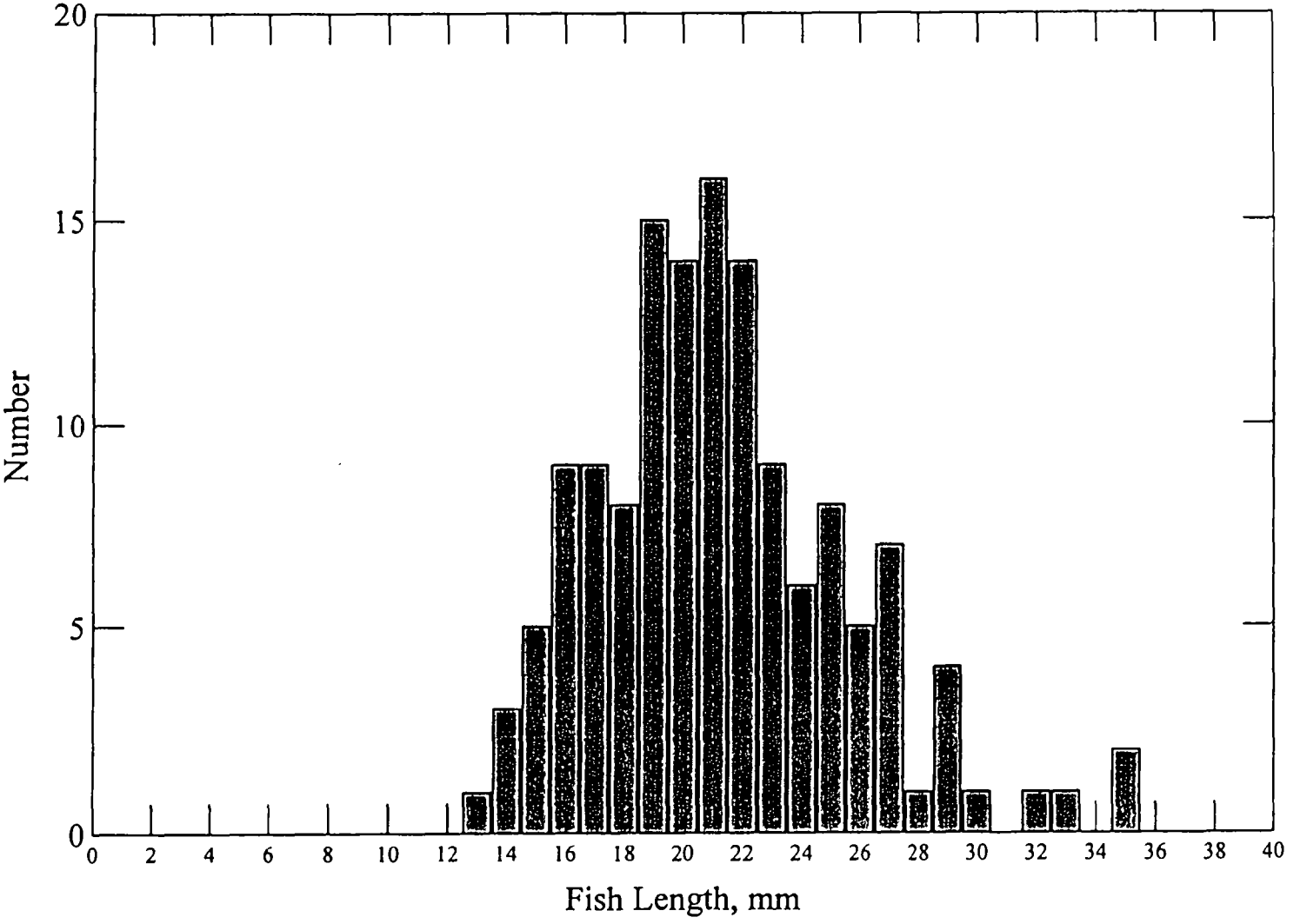


Figure D.13 Length frequency distribution of mosquitofish in the 1997 test sample.

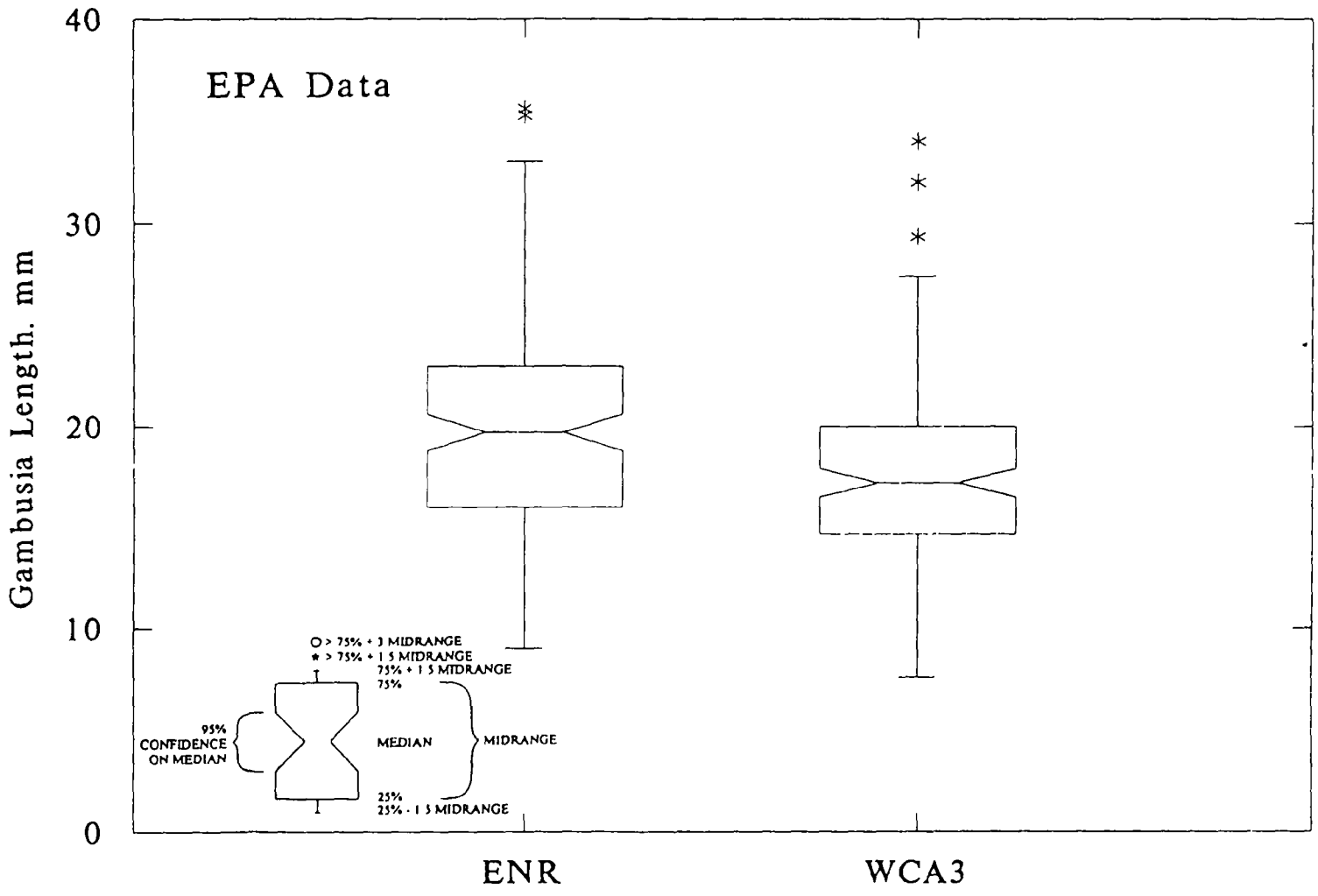
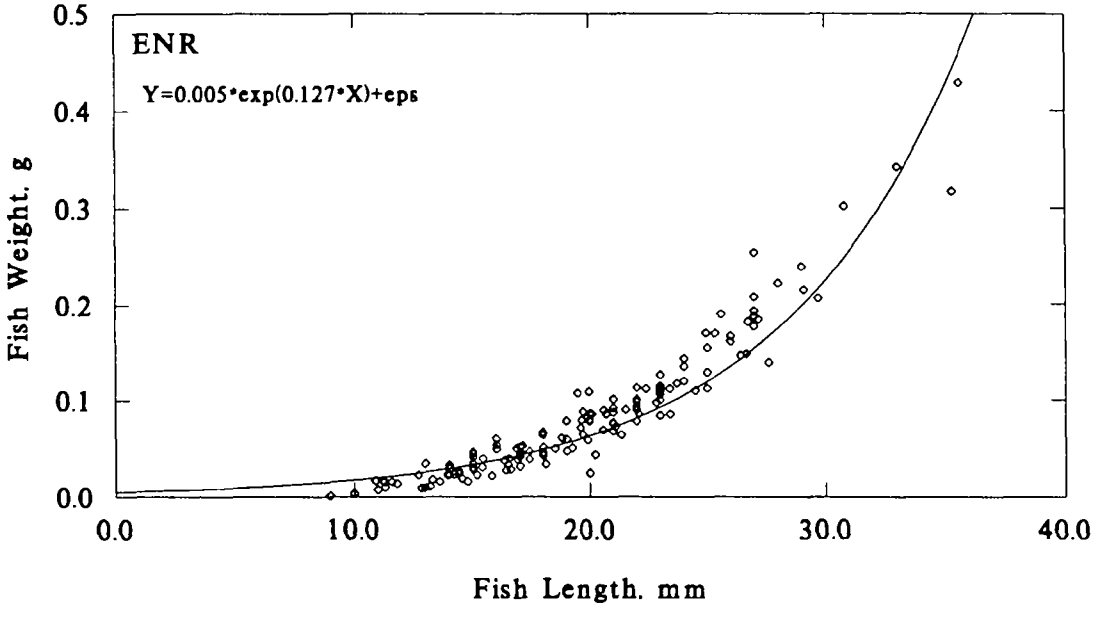
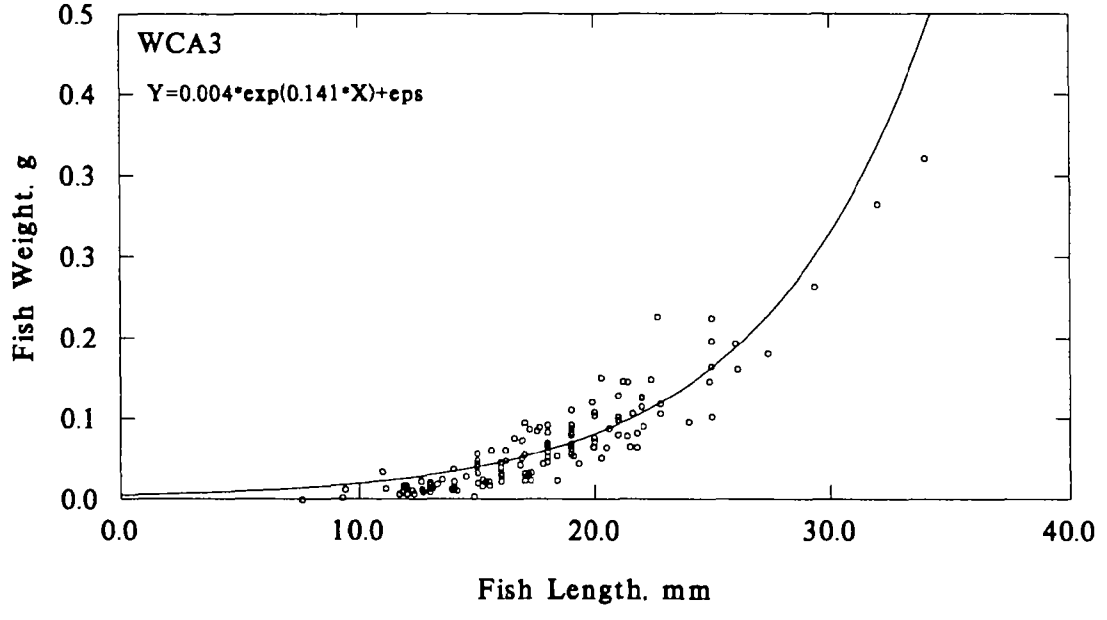
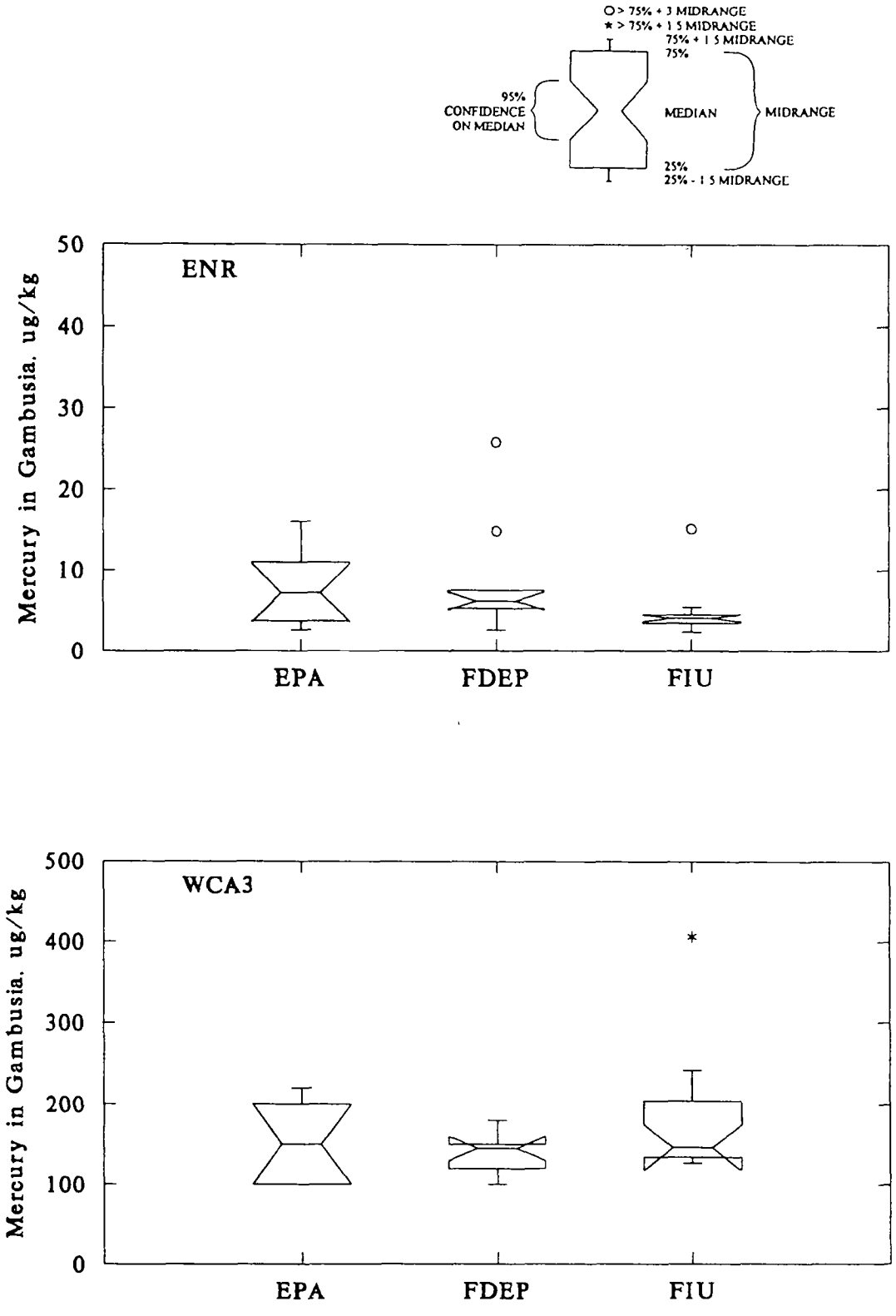


Figure D.14 Notched box and whisker plots comparing length of mosquitofish at the two marsh sites from the 1997 test sample.





D.15 Length versus weight of mosquitofish from the two sites in the 1997 test samples with derived growth curves.



D.16 Notched box and whisker plots comparing THg in mosquitofish analysis results from 3 labs.

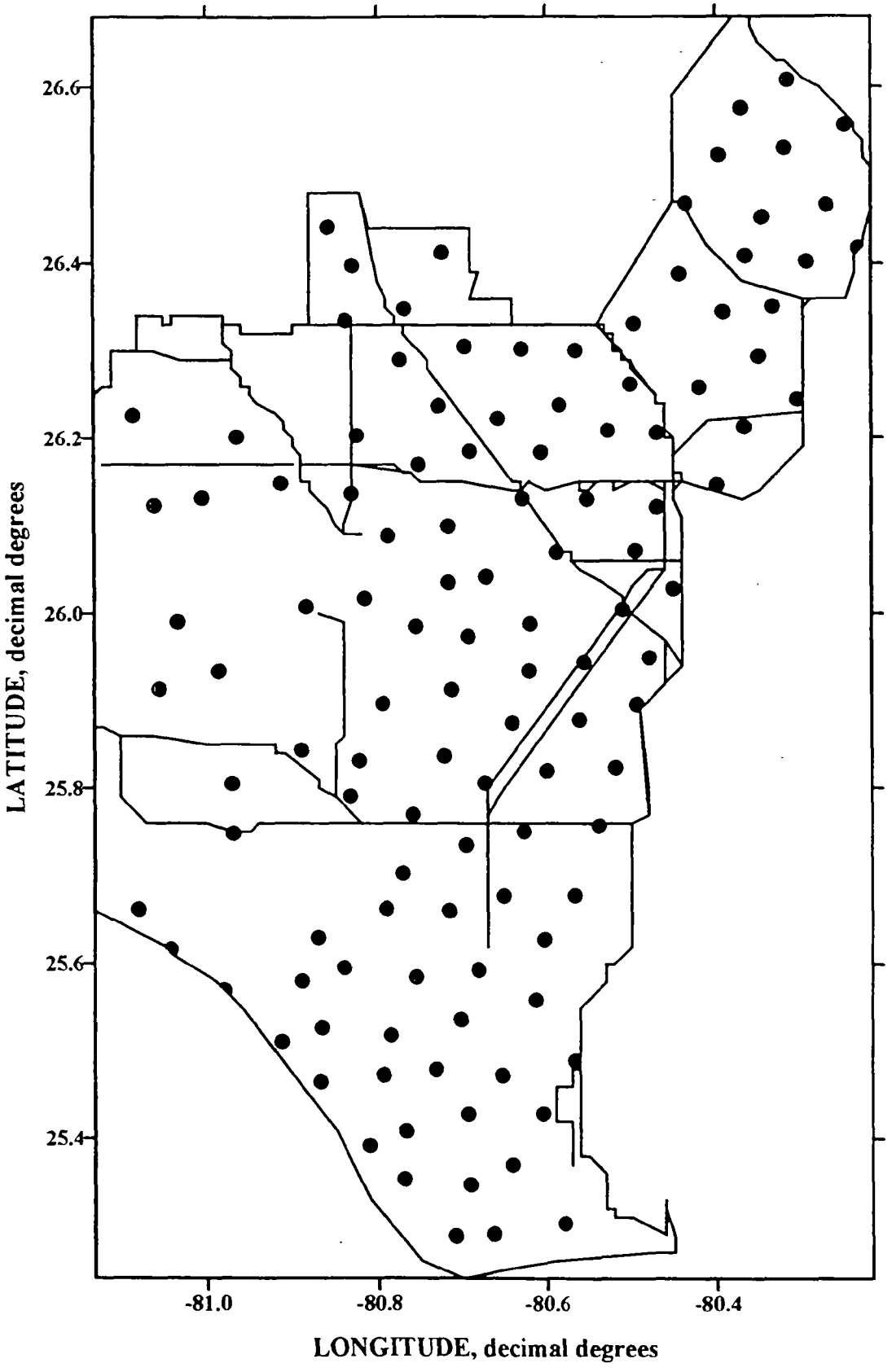


Figure D.17 Map of the study area indicating sites of sample collection.

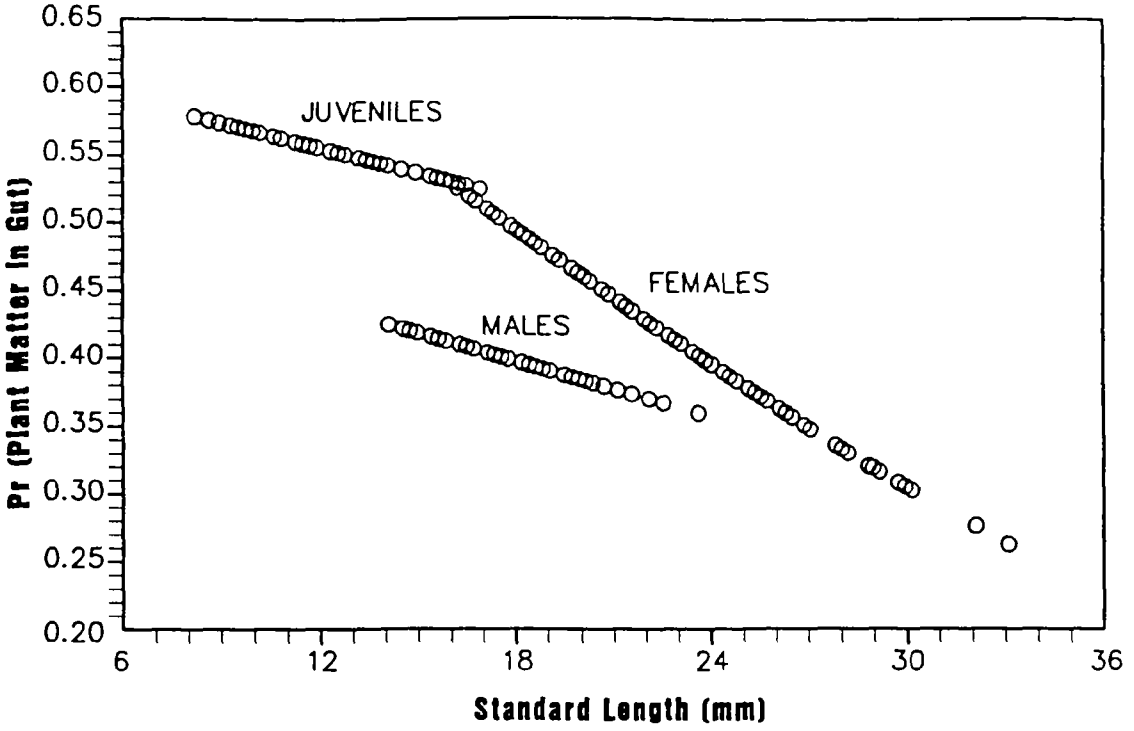


Figure D.18 Probability that an individual fish will have plant matter in its stomach relative to standard length. Based on logistic regression. Results for juveniles, males, and females are plotted separately.

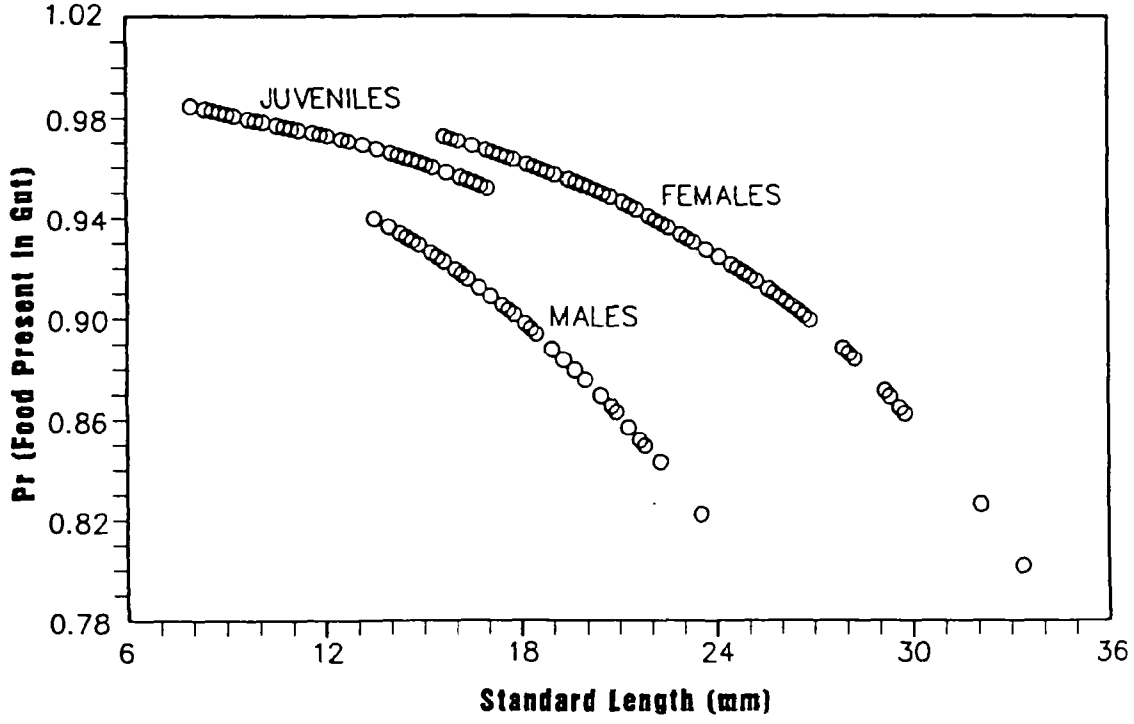


Figure D.19 Probability that an individual fish will have food in its stomach relative to standard length. Based on logistic regression. Results for juveniles, males, and females are plotted separately.

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APPENDIX E

Response to Peer Review Comments

Peer reviewers for the South Florida Ecosystem Assessment Final Technical Report.

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1) Tony Olson, EMAP, Corvallis**
2) John Stoddard, EMAP, Corvallis
3) Steve Paulsen, EMAP, Director
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5) Gil Veith, NEERL, RTP, Assoc. Director
6) Kevin Summers, Gulf Breeze Lab
7) Anthony Carlson, REMAP**
8) Rose Russo, NERL, Athens
9) Tudor Davies, OW,OST, Hdqs
10) Arnold Kuzmack, OW, Hg Task Force, Hdqs

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12) Ronnie Best, USGS, BRD
13) Dave Krabbenhoft, USGS, WRD
14) Bill Loftus, USGS, BRD
15) Aaron Higer, USGS, WRD
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20) Sue Jewel, LNWR**
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22) Tom Fontaine, SFWMD
23) Tom Atkeson, FDEP (Don Axelrad)**
24) Jerry Keeler, U of Michigan
25) Larry Fink, SFWMD(combined)**
26) Lorraine Heisler, FGFWFC
27) Ted Lange, FGFWFC
28) Col. Joe Miller, USACE
29) Terrance "Rock" Salt, Restoration Task Force
30) Terry Rice, South Florida Task Force
31) Billy Cypress, Miccosukee Tribe
32) James Billie, Seminole Tribe(Craig Tepper)**
33) Ron Tipton, Everglades Coalition
34) Bubba Wade, US Sugar
35) Ed Barber, Sugar Cane Coop
36) Pete Rosandahl, FloSun**
37) Don Porcella, EPRI
38) Ed Zillioux, FPL
39) Scott Osborne, FL Power Coord. Group
40) Thomas Corcoran, Audobon Society

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**Review Comments

41) John Rudd, FWI, Canada
42) Hans Hultberg, Goteborg, Sweden
43) Brian Rood, Mercer U., Macon
44) William Walker, for ENP**
45) Chris McVoy, SFWMD
46) Terry Haines, USGS, BRD
47) Ben McPherson, USGS
48) John Davis, ES&P**
49) Carol Kendall, USGS**
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55) Richard Harvey
56) Jerry Stober
57) Dan Scheidt
58) Ron Jones, FIU
59) Joel Trexler, FIU**
60) Steve Rathbun, UGA
61) Don Stevens, Dynamac
62) Mike Birch**
63) Linda Anderson-Carnahan and Stacy Howard**

*Acknowledgment
**Review Comments

Department of
Environmental Protection

June 26, 1998

Dr. Jerry Stober
Science and Ecosystem Support Division
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Thank you for the opportunity to review your draft report. This work reported on therein clearly has been a massive undertaking in the field and laboratory, and this work itself a massive data reduction effort. It is very informative and useful-and in fact-it has already contributed to hypothesis generation for research we hope to fund on MeHg bioconcentration by Everglades periphyton. Our only problem with your report, and one that I am sure you and your colleagues have (and are) struggling with, is how to comprehend so much data!

I am returning the first volume of the report with many marginal notes. Please see my handwritten comments and suggested edits in Chapters 8,9, and 10, as well as on previous pages marked by post-its. In some of my comments I have been assisted by Tom Atkeson, and Mike Turtora, a USGS water-quality statistician working with Tom Atkeson and me on mercury in lakes and streams outside the areas covered by your report. Anders Andren will also be submitting comments within a few days. Specific comments are as follows:

Chapter 8 Mercury

There seems to be little direct exploration of relations between variables in this report. In addition to implying relations by comparing trends in geographic medians among variables, direct regression or PCA of variables over the individual sampling units should be considered as it may prove to be more informative. Multiple regression of 'independent' variables on Hg concentrations or regression of the first few principle components on Hg concentrations would be valuable. In addition, maps could be prepared of principal component scores at each site. Finally, maps of the regression residuals may reveal interesting patterns.

Response: Exploratory analyses were used extensively during data analysis. The primary variables, DOC, Sulfate, TP, and methylmercury in water and total mercury in mosquitofish were identified through PCA and factor analysis, as indicated on p 4-7. The large array of statistical analyses performed on the data are presented in text and tables on pages 3-14 to 3-19. Unfortunately, displays of exploratory analysis results, in many instances, are not very illuminating if the reader is not familiar with the procedures. Therefore, these displays were not included in the report even though the analyses were conducted and the results used to identify multivariate relationships Information is presented using the most appropriate of the many statistical analyses performed.

While median values appear on most plots, parametric ANOVA on log-transformed data is implied. The plots would benefit by changing the vertical axis to a log scale and a nonparametric ANOVA would probably be more useful. A nonparametric ANOVA would eliminate the need for outlier analysis and a log plot scale would produce more interpretable graphics.

Response: Median values were chosen as a conservative analytical approach in which 50% of the data fell above and below placing less weight on outliers in the data set since no data were

removed. The data were log transformed for some analyses as indicated in Table 3.2. Some log scale graphics were incorporated in the report.

Chapter 10 Synthesis and Integration
10.2 (p. 10.1)

There is much data presentation here. Consider placing these data in Chapter 8, leaving this chapter reserved for "Synthesis and Integration."

Response: While there are data in Chapter 10, it represents a synthesis of information presented in Chapter 8. Additional emphasis was placed on synthesis and integration in this chapter to reduce introducing new information.

As you state, the LNWR as a rain driven marsh system is chemically distinct from other areas in the north to south flowway. Much of the thrust of this work is looking for patterns among water chemistry variables as they change along the gradient from northern WCA 2A down through ENP. As WCA 1 and Big Cypress are distinct from this gradient, perhaps a number of the plots of data might be more informative if these two areas were treated separately. It seems that one major thrust of your work is to describe the interrelationship between various water quality variables as they change along this gradient. Omitting distracting or confounding comparisons as caused by data from areas that don't fit along the gradient should improve the fidelity of the relationships you see.

Response: The Big Cypress information is not included in these plots. The WCA1 data is included because it does not overlap with the other WCAs and does present a contrast with WCA2 and comparison for ENP.

10.3 Conceptual Models (p. l0-6)

It is confusing in going from 5 to 3 to 6 and back to 3 geographic areas for marsh data presentation and discussion.

Response: The different latitudinal divisions were established because there are different patterns that emerge from these different divisions. However, it can be confusing for the reader. A more logical approach for building these different perspectives was investigated and presented.

10.3.1 North of Alligator Alley

One factor you believe to be contributing to low MeHg concentrations in *Gambusia* here is high TSO_4 because it (or it's product, sulfide) can complex Hg and reduce its availability for methylation. However, MeHg concentrations are in fact high in water, periphyton and soil in this area, suggesting that high TSO_4 is not reducing methylation rate sufficiently to explain the low MeHg in fish. Perhaps I am missing the nature of your argument here and it could be explained more clearly.

A second factor you believe to be contributing to low MeHg concentrations in *Gambusia* is high TOC because it can complex MeHg, making it 'unavailable for uptake. However, the fact that periphyton MeHg levels are high, indicates that the MeHg is available for uptake.

Response: The presence of high methyl mercury in water in WCA-2 in the presence of high sulfate indicates that MeHg should be available for biouptake, however, the fish remain low. We think this is largely due to the biodilution effect of high to low (north to south) phosphorus gradient and its stimulatory effects on primary level biomass production.

As regards the low MeHg in *Gambusia* in the north, it seems to me that rather than TSO_4 or TOC explaining this, either the food preferences of *Gambusia* in this eutrophic area, or low transfer efficiency of Hg from eutrophic periphyton species to *Gambusia*, are more likely responsible. (Perhaps *Gambusia* can't digest the periphyton occurring in eutrophic areas.) You have little discussion of the fish gut contents data from Appendix C here.

Response: The information on fish gut contents has only been briefly discussed because data are for a single wet season. A comparison is needed with a dry season sample to allow a more meaningful interpretation. The primary mechanisms controlling bioaccumulation operate at the cellular level which has been suggested in Chapter 10, but which is beyond the scope of this study.

Since most of the biomagnification of Hg in the food chain occurs by algae bioconcentrating Hg from water (BCF = 10^4-10^6), and BCFs are affected by Cl^- ionic strength, and pH, it would be of great value to plot periphyton BCF vs. pH and Cl concentration similar to what you have done for geographic areas. We are planning to support some experimental work following the lead of a paper by Mason et al. 1996 on this topic. It would be useful to see if any real-world patterns of interactions between pH and Cl^- follow Mason's findings.

Response: The bioaccumulation of mercury in mosquitofish is also thought to be low in the north due to changes in the food chain resulting from the stimulation of pollution tolerant species and a less complex food web and lower transfer efficiency as you suggest. The pH gradient over the marsh is quite small. The average pH is circumneutral throughout the marsh, except for WCA1. We did not measure Cl^- so this plot is not possible.

Regrowth dilution, do calculations indicate that the increased *Gambusia* growth rate can account for much of the lower MeHg levels in the *Gambusia*?

Response: No. We don't have growth rates for mosquitofish.

10.3.2 Alligator Alley to Tamiami Trail

Increased bioconcentration of MeHg by periphyton due to increased periphyton productivity that provides sites for methylation is hypothesized. However it's not clear to me that MeHg concentrations in periphyton are higher here, or that *Gambusia* are consuming more periphyton, or proportionally more periphyton relative to other dietary items which could explain the increased MeHg in *Gambusia* in this area.

Response: Methyl mercury in floating periphyton, soil periphyton, *Gambusia* and Great Egret chicks south of Alligator Alley shows remarkable consistency. The production of floating periphyton also appears to be higher here. The changes in the food chain seem to be apparent from north to south, however, although some of these changes in the food habits of *Gambusia* are indicated to some extent in the one food habits analysis done so far the results are not yet consistent enough to draw a conclusion. The food habits analysis needs to be repeated and is planned for Phase II REMAP.

Didn't Gilmour find maximum methylation rates here, but were these in sediment or periphyton?

Response: Gilmour found maximum methylation rates in sediment cores collected from WCA2 and WCA3A. However, the sediment periphyton were considered part of the sediment in her studies. Sediment periphyton mercury concentrations were analyzed separately from sediment mercury concentrations in the EPA study.

10.3.3 South of Tamiami Trail

Doesn't Gilmour believe that SO_4 concentration limits methylation rate here, which would affect MeHg in *Gambusia*?

Response: Gilmour hypothesizes that low SO_4 concentration does limit methylation rates south of the Trail. Data from the present study suggest that the combination of low SO_4, TP, and lower DOC influence Hg methylation rates, but that the increase in food chain complexity also results in increased BCF's in *Gambusia*.

10.4 Testable Hypotheses
Table 10.4 (p. 10-11)

For North of Alligator Alley, I suggest adding a testable hypothesis: Elevated TP concentrations alter periphyton community composition favoring species that have low transfer efficiency of MeHg to fish.

A testable hypothesis relevant to all three geographic areas and worth adding is:
Cl^- and pH affect MeHg uptake by algae and consequently affect periphyton BCF and fish BAF.

Response: Hypotheses added.

Don Axelrad, Ph.D.
Mercury Program

Jerry Stober, Ph.D.
U.S. Environmental Protection Agency Region 4
Science and Ecosystem Support Division
980 College Station Road
Athens, GA 30605-2720

RE: Peer Review of South Florida Ecosystem Assessment Vol 1. Technical Report Review Draft and Vol 2. Appendices

Dear Dr. Stober:

Enclosed are our peer review comments from the Mercury Studies Program Staff on the subject peer review draft documents. I apologize for the delay, but due to the scope of the documents and the number of comments generated, it was not possible to meet your original deadline. I hope that you will still find these comments timely and helpful. These comments originate with the Mercury Studies Program and therefore only represent our position [and do not represent the position of the South Florida Water Management District].

The REMAP study was a monumental undertaking. Its conceptual, methodological, logistical, and financial challenges cannot be overstated. We have also faced such challenges, albeit on a far smaller scale in the ENR Project, in an attempt to achieve the optimum balance between what is scientifically rigorous and what is reasonably achievable in practice with the resources available. The trade-offs between scientific rigor and practicability must be made explicit, however. Whether such trade-offs should extend into the realm of quality assurance (QA) has also been much discussed by the Mercury Studies Program staff in recent months. In our specific comments we have tried to convey that unresolved quality control and methodological issues in the South Florida REMAP Study could weaken some of its key conclusions. From our experience, the application of traditional QA criteria to ultra-trace mercury sampling and analysis may not always be appropriate. Nevertheless, such changes cannot be made arbitrarily or in a vacuum. We believe that all participants in the South Florida Mercury Science Program would benefit from a thorough reevaluation of QA performance criteria for such specialized applications.

Such a reevaluation may best be carried out in the context of an overall reassessment of data to be used to define the mercury baseline conditions in the Everglades. The reassessment is to be carried out over the next six months in conjunction with the activities of the Everglades Mercury Baseline Oversight Committee created by the Technical Oversight Committee Principals at its May 27, 1998, meeting in West Palm Beach.

Let me know if any of our comments require clarification. I can be reached at (561) 682-6749.

Sincerely,

Larry E. Fink, M.S.
Sr. Supervising Environmental Scientist
Mercury Studies Program Manager

Comments on South Florida Ecosystem Assessment Vol. I Technical Report Peer Review Draft
By Larry Fink, Pete Rawlik, and Darren Rumbold, Resource Assessment Division, Water Resources Evaluation Department, South Florida Water Management District

Preface

The REMAP study was a monumental undertaking. Its conceptual, methodological, logistical, and financial challenges cannot be overstated. We have also faced such challenges, albeit on a far smaller scale in the ENR Project, in an attempt to achieve the optimum balance between what is scientifically rigorous and what is reasonably achievable in practice with the resources available. From our experience, the application of traditional quality assurance criteria to ultra-trace mercury sampling and analysis may not be appropriate, and we believe that all participants in the South Florida Mercury Science Program would benefit from a thorough reevaluation of quality assurance performance criteria for such specialized applications. Such a reevaluation may best be carried out in the context of the reevaluation of the mercury baseline data that affects District permit compliance over the next six months in conjunction with the activities of the Mercury Baseline Oversight Committee created by the Technical Oversight Committee Principals at its May 27, 1998, meeting in West Palm Beach.

It is in this context and spirit that we offer our review comments on the on South Florida Ecosystem Assessment Vol. I. Technical Report Peer Review Draft.

Summary

- General
  - clarify audience and write consistently to that audience
  - add Background section
  - bring conceptual model/initial hypothesis discussion into Study Design chapter
  - summarize tesselated probabilistic sampling design in Study Design chapter and append the technical details
  - document could benefit from more diligent editing for spelling, more complete citations of key concepts and data not originating with the REMAP study, and more thorough checking of calculations (e.g., check units on sediment concentrations: ug/cc should probably be ng/cc)
  - add decision data quality objectives to performance data quality objectives

- Conceptual Model
  - reexamine conceptual model regarding the interrelationships of eutrophication processes, reducing conditions, and methylmercury production in light of the sulfur cycle and potential deficiencies in the redox measurements using the Eh probe
  - omission of sulfide from analytes list for surface and pore water severely compromises the ability of the study design to discriminate the validity of hypotheses in which phosphorus is the primary determinant of mercury speciation, fractionation, and transformation versus those in which sulfide is the primary determinant, as mediated by surface and pore water chemistry influenced by eutrophic conditions fostered by excess phosphorus

- Sampling Methods
  - justify and validate new methods with standard methods or accepted standards of practice by competent practitioners, where standard methods do not exist sampling from helicopter pontoon is not considered an accepted standard of practice in Everglades sampling by the Everglades Technical Advisory Committee (ETAC)

- please provide results of study showing equivalence between REMAP and ETAC approved helicopter sampling method

- Analytical Methods
  - no pore water chemistry
  - justify and validate new methods with standard methods or accepted standards of practice by competent practitioners, where standard methods do not exist
  - Practical Quantitation Limits for THg and MeHg using these new methods?
  - What is significance of EtHg if no data are included or analyzed EtHg?

- Quality Assurance
  - Appendix B omits ORD quality assurance report. Cannot perform thorough evaluation of data upon which report is based without a complete quality assurance data set and analysis viz-a-viz performance criteria.
  - the use of non-standard quality assurance performance criteria should be avoided (e.g., 3 standard deviations of mean of entire data set to evaluate duplicate sample performance viz-a-viz the use of ±25% RPD or RSD, which is based on paired duplicate sample results)
  - if non-standard quality assurance performance criteria must be used, include detailed rationale
  - no results of paired splits correlation analysis of inter-laboratory comparisons

General Comments

By my recollection, the primary purpose of the REMAP study was to determine the nature, magnitude, extent, and trends of mercury concentrations, exposures, and risks in South Florida that defined the mercury problem. A secondary purpose was to determine whether the cause or causes of the mercury problem could be discerned from an association between the meteorological, hydrological, chemical, and/or biological characteristics of the environment and the spatial or temporal patterns of mercury speciation, fractionation, and accumulation in that environment. A more complete presentation and analysis of the data collected under REMAP, including the quality assurance data, would make it possible to determine the degree to which the REMAP Study has achieved its primary and secondary purposes. The subject report contains an extensive analysis of mercury species concentration data, but we could find no preliminary or scoping-level ecological risk assessment. This should be added. Absent any other guidance, the Great Lakes Initiative Water Quality Criteria to protect wildlife could be used to quantify screening-level hazards.

Response: The ecorisk assessment planned from the outset by this study has been deferred due to the need to analyze and interpret the large amount of data collected. Additional sampling is planned in 1999 to complete baseline assessment of key plant indicators and to initiate trend monitoring. An initial ecological risk assessment will be based on the interpretation of the 1993-96 information followed by an update when the 1999 data becomes available in addition to input from studies from other agencies.

An ancillary benefit of the REMAP study was the generation of calibration data sets for the hydrodynamic, water quality, and landscape ecology models that are being used by agency scientists and engineers to organize, synthesize, filter, and integrate all of the data gathered to date under the various monitoring and research projects to support management-relevant predictions about the responses of the Everglades to various stressors under various water management scenarios. Had REMAP provided no other benefit, it would have justified its cost.

Please clarify the audience for this report: general reader? ... educated layman? ... legislative aide with interest in environmental issues? ... environmental manager? ... scientist with no specific knowledge of Everglades

environmental issues? The content and tone of the Introduction suggests a target audience of an educated layman, while the following chapter on Study Design requires advanced academic training in statistics to understand. The other chapters tend to fall between these two extremes. I think the target audience should be an environmental manager, whose decisions you intend to guide with the information contained in the report. An educated layman's version can be prepared for a subsequent 4-page, 3-color fact sheet. The detailed statistics lesson in Chapter 2 might better be appended to the document, reducing the discussion in Chapter 2 to a management-level summary of the approach and its benefits over other approaches, with a reference to the appended scholarly discussion.

Response: The target audience is the environmental manager as well as the scientist. The detailed statistical discussion up front is to provide the information on the statistical survey design which is frequently misunderstood. A summary of the main statistical design points will be added to the text.

Please summarize key background discussion in one paragraph for the Introduction chapter and add a new Background chapter that summarizes what was known about mercury sources, disposition, fate, bioaccumulation, and effects at the time of study design. This will allow the highlighting of the contributions of the REMAP study to our knowledge base in Chapter 10.

Response: Chapter 1 has been rewritten to better summarize key background issues. A background section on mercury has been added to Chapter 8. This avoids having to revise all subsequent table and figure numbers, while providing a reference of REMAP contributions in Chapter 10.

Citations should be provided to support characterizations of the system using the results of studies other than this one. There are a number of instances where such citations have been omitted (e.g, meteorological, hydrological, geological, and habitat data).

Response: Additional citations have been added.

Specific Comments

1.0 Introduction

1.4.1(1-7). Purpose of this Report. Move to beginning of this chapter to assist reader in appreciating why the report is being written sooner than later.

Response: Purpose moved.

1.3.1 *"Clearly, the greatest change in the Everglades ecosystem is the change in the natural hydropattern..."* Debatable. One could make a stronger case that greatest change is the loss of between 25 and 50% of the Everglades acreage.

Response: Text has been modified.

1.3.2 (1-3). Florida Bay advisory was issued in October 1995, not in 1994.

Response: Date changed.

1.3.2(1-3). The statement of the relative contributions of the various exposure pathways mercury exposures mixes mercury species. There is virtually no MeHg in air or drinking water. Nearly 100% of MeHg

exposure occurs via food, and more specifically, fish. For elemental mercury, most exposure occurs via air, not water or food. For Hg(II), most exposure occurs via air associated with particles and drinking water. If mercury is treated as one entity, the statement is probably accurate. However, it is not accurate to treat all mercury species as one entity, as their sources, fates, BAFs, exposure pathways, and toxicities are very different.

Response: Text was modified to reflect different exposure pathways for different mercury species.

1.3.2(1-4). What vast areas of Hg contamination? Hg in water and sediments is within normal ranges for aquatic ecosystems. Its MeHg in water and more importantly in biota that is higher than background over a vast area of South Florida. Please clarify

Response: Text was clarified to reflect the appropriate mercury species and media.

"While Everglades Hg contamination ... since 1989, the sources, etc., remain unknown." This statement seems out of date. Perhaps it was written in 1992 for the original proposal or work plan for the REMAP study and was overlooked in the early editing. It ignores the important contributions made by Delfino et al. (1993) and more recently Robbins and Keeler (1997) on the rate of deposition of mercury as a function of time, the Florida Atmospheric Mercury Study (FAMS) (see for example Pollman et al. 1995; Landing et al., 1996; and Guentzel 1997) to our understanding of the Hg(II) deposition via bulk rainfall, and the screening-level calculations of Landing (1996) on the potential role of reactive gaseous mercury deposition in making up the difference between the measured deposition rate of Delfino et al. and Robbins and Keeler (1997) and the bulk rainfall rate. Keeler and his students are now making important contributions to our ability to quantify local source-derived wet and dry deposition to the Everglades. It also ignores the contributions of the USGS ACME team to our understanding of the factors that are controlling net methylmercury production, bioavailability, and bioaccumulation, especially the role of excess sulfate derived from EAA runoff and perhaps acid rain deposition in determining the location and magnitude of the sulfide concentration gradient in the Everglades, the modeling of Hg(II) fractionation, and support to USEPA/ORD's efforts to refine an existing mercury cycling model for application to the Everglades.

Response. The text was clarified to indicate that atmospheric emissions, transport and deposition represents a major source of the mercury, and, perhaps, the source of mercury, but the cycling of mercury through the system and the fate of mercury in peat is still an unknown. There has been up to 2 m of peat lost over the past 50 years. It is also unknown what proportion of the atmospherically deposited Hg is methylated and accumulated through the food chain.

Although REMAP made limited contributions to our understanding of the fate, bioaccumulation, and effects of mercury in the Everglades, it did make important contributions to our understanding of: (1) the semi-annual spatial dispositions of THg and MeHg in water, sediment, periphyton, and mosquitofish in the Everglades that reflect its distinct ecophysiological regions, (2) the present-day insignificance of THg and MeHg in EAA runoff to the new annual loads to the Everglades, (3) the inverse relationship between the peat accretion rate and the THg concentration in peat, (4) the absence of an inverse relationship between phosphorus in water and mercury in mosquitofish in the southern Everglades, and (5) the strong correlation between mosquitofish hotspots and wading bird hotspots in WCA-3A.

Response: Contributions incorporated in the text in Chapter 10.

I would modify the above identified problematic statement to say that "While . . . , various degrees of uncertainty still remain in our quantitative understanding of sources, etc. that limit our ability to make fully informed management decisions at this time. These uncertainties are expected to be reduced substantially over the next two years as the monitoring, research, modeling, and assessment data collected over the last five years are further analyzed, synthesized, and integrated into a more complete, accurate, and precise predictive quantitative model. The REMAP studies are expected to make an important contribution to this effort."

Response: This statement was substituted for the problematic statement in Section 1.3.2.

1.3.3 (1-5): "A *combination of agricultural . . . is being implemented in an attempt . . .*" I would use "natural" rather than "historic" levels.

Response: Natural was substituted for historic.

Strategy for Study Design

In a fixed or static study design, samples of specified type and volume are collected at specified locations and times and characterized and analyzed for the constituents of interest. The study is carried on for a specified period of time. The data are analyzed, synthesized, and integrated into an understanding of the problem, and from that understanding a preferred solution emerges that is eventually adopted and implemented. In an adaptive or dynamic study design, the study evolves over time as the data collected in the early or reconnaissance and scoping phases is analyzed, synthesized, and integrated into a more complete, accurate, and reliably conceptual model of mercury sources, cycling, and exposures in the Everglades. This more robust conceptual model then forms the basis for a more refined set of management questions to be answered by the study and a more refined study design to answer those questions, and so forth, until the questions are answered to the satisfaction of the resource managers. The REMAP study appears to have followed the static study design strategy. This should be stated explicitly.

Response: The study design is clearly described in Chapter 3. There is no need to characterize the design as "static" versus "adaptive". In particular, the study was designed around management questions, not around specific hypothesis regarding Hg. The design was intentionally focused on achieving broad population coverage to provide some power for addressing a variety of hypotheses.

Conceptual Basis for Study Design

Hypotheses to be Tested: Rather than rely solely on management questions to introduce the reader to the study design, I think it would be more appropriate to bring the initial hypotheses to be tested from Table 8.1 (8-22) into Chapter 2.0.

Response: The study was not designed around the null hypothesis. It was designed around the policy/management questions.

Conceptual Model: Perhaps most notably absent from Chapter 2 is a conceptual model that includes the transport, disposition, transformation, accumulation, and effects of mercury species in the Everglades that warrant study). I think it would be beneficial to bring the initial conceptual model displayed in Figure 8.2 (8-23) into Chapter 2.

Response: The conceptual model is directed specifically at the mercury issue. The project and the report is directed at an assessment of the ecosystem. The conceptual model is most appropriate in chapter 8.

Data Quality Objectives: Not defined until much later in the document and not complete and definitive. What are the acceptable limits of quantitation and detection of each factor? How were these determined? With what resolving power, probability, and confidence level must the study design be able to discriminate true differences in a factor between any two cells? any two times? For adjacent cells or times, this ability to resolve true spatial or temporal differences will determine the minimum gradient or trend that can be quantified accurately, precisely, and reliably at the sampling grid scale and time step adopted.

Response: Data Quality Objectives are documented in a separate report, which is referenced in this report. Additional information can be obtained from that document. In addition, an analysis of variance was conducted for each variable collected as part of the REMAP program to assess the within site versus among site variance estimates. The EMAP goal was to have within site variance estimates about 10% of the among site variance estimates For almost all the variables except fish, this goal was achieved. For *Gambusia* mercury concentration, the within site variance was 14% of the among site variance. Future sampling efforts will collect and analyze 7 individual fish per site rather than the 5 fish collected previously. This will reduce the within site variance to 9% of the among site variance and satisfy the EMAP goal. Finally, EPA Office of Research and Development is in the process of preparing DQO guidance for research programs such as the REMAP project because the DQO guidance documents under which the REMAP DQOs were developed are not applicable for research programs.

Grid Scale: Reason for selection not defined. The unbiased statistical sampling model was adopted as the basis for the design of this study. It assumes no *a priori* knowledge of the spatial distributions or interrelationships of the factors to be studied. Therefore, one cannot determine *a priori* the appropriate grid scale to overlay upon the system of interest to ensure that nothing significant is missed in terms of differences between sites. As an alternative, one could have made the grid scale a random variable to test the hypothesis that the selected grid scale was inappropriate for each factor of interest. This would have required random subsampling at randomly selected finer and broader grid scales, until the desired resolution emerged for each factor of interest. This, of course, presupposes that well-defined data quality objectives had been adopted with which to discriminate appropriate from inappropriate grid scales. This was not the case. This also presupposes that there was enough time and money to conduct the required scaling scoping studies to achieve this objective. This was also not the case.

Response: The approach suggested by the reviewer is not feasible The time and resources required to mount the study suggested by the reviewer are enormous. Furthermore, the reviewer assumes that an "optimum" design exists, which is not the case. A general rule is that a study cannot be simultaneously optimized for more than a single objective. This was definitely a multiple-objective study, so the best we can do is a design that addresses all our objectives with an acceptable degree of uncertainty.

The design relies on several fundamental principles to achieve "acceptableness":

(1) Design-based inference and variance estimates must be possible This is achieved by the choice of the RTS design.

(2) We assume that the phenomena being studied all have spatial pattern, in the very general sense that two locations near one another are more likely to resemble one another that two locations far apart.

(3) Major results will be reported as a distribution quantile, e.g., "x% of the resource population is in classification y".

(4) More samples are always better than fewer, but 50 is enough to give acceptable confidence intervals for resource condition statements as in (3). With a sample size of 50, the worst case (estimating the 50th percentile of a population with no spatial pattern) confidence interval will be no more that + 12 percentage points.

(5) Given a spatial pattern as in (2), there is a result in spatial statistics that implies that regular point spacing is better than a purely random sample. Some of the relevant literature is cited in Section 2.1.1. The result is model-based and not strictly applicable to design-based estimation. Moreover, strict-systematic spacing does not provide a design-based variance estimate. (In the terminology of classical statistics, there is only one degree of freedom, so no variance estimator is possible.) The RTS does provide a variance estimator, and it can be shown, again using model-based methods, that it captures most of the efficiency of a strict systematic design. These results are in the references cited in Section 2.1.1.

As there was insufficient time and money to carry out such studies, the choice of grid scale was inherently arbitrary, based on what could reasonably be expected to be sampled and analyzed with the available transport, staff, and analytical resources over a period of time considered short relative to the response times of key transport, fate, and bioaccumulation processes. This is touched on the introductory discussion in Chapter 2. That's good. It could be made even more explicit, however.

Time Scale: Semi-annual sampling corresponded to the late spring and early fall, or the late dry season and the late wet season in subtropical South Florida. The timing allowed one to detect differences in periods of rapid plant growth in the late spring but not the rapid onset of senescence in the late fall or early winter. The maximum external loadings of new mercury to the system occur in the mid-summer and are minimal in winter. In this context, perhaps quarterly sampling at half as many sites may have provided greater insight into the temporal dynamics without losing significant information about spatial distribution.

Response: We were not attempting to establish or study temporal dynamics. The two sampling times were picked to examine the system at two extreme hydrologic regimes. The design does provide limited information about temporal dynamics, but that comes as a side-benefit, not as a factor that controlled the design.

Splitting the total effort into 4 quarterly cycles per year, as suggested by the reviewer, is not desirable. It changes the objective of the study, and substantially dilutes the spatial resolution of the sample. Estimates for a subpopulation that are feasible with 30-50 samples are not with only 15-25 samples.

What were key transport, fate, and bioaccumulation processes and their response times with which to determine the appropriate time scale for sampling? Perhaps a table with the ranges of response times of the various components of the system would help guide the reader in evaluating the logic of a 10-day period for synoptic sampling.

Response: The synoptic sampling period was a function of how rapidly sampling could physically be accomplished over the 9600 km^2, rather than based on time and length scale analyses of processes and transport times. The 10 day period was the maximum time to sample the marsh from the mangrove fringes to Lake Okeechobee (i.e., south to north). The last sampling cycles were conducted within 7 days. It might be useful at a later date to consider time-length scale analyses, but this information was not available for inclusion in the report. Process and transport studies relate to different study objectives than those of the REMAP project.

Factors of Interest: The conceptual model should have guided the choice of factors of interest. It is not possible to determine if this was the case from the organization and content of this report. Perhaps the choice of these key factors

derived from the literature review prepared by ORD-Athens. If so, I would recommend that it be distilled down and added to the Background chapter in the report or append it for ready reference.

Response: The conceptual model did guide the choice of factors of interest. The conceptual model was developed by Dr. Ron Jones prior to the REMAP proposal which preceded the ORD literature review by a couple of years.

In addition to pointing out what was included in the study, it is also important to point out what was excluded. For example, sediment pore water was not sampled and analyzed for THg and MeHg, not because such information was not thought important in defining the disposition of factors that determine the susceptibility of an ecosystem to methylmercury production, transport, and bioaccumulation but rather because the laboratory used by USEPA for this work had no such capability at the time of the study. Also excluded was an estimate of the flux of THg and MeHg from the sediment to the overlying water, because sediment strata were not analyzed separately. Only a sediment composite was analyzed. While this may have been sufficient for calculating the magnitude of storage compartments and saved significant cost, it lost significant information. Perhaps the random selection of a subset of cores for such more detailed analysis might have added to the conceptual power of the study without increasing its cost dramatically.

Response: Sediment pore water was not sampled for total and methylmercury because a reliable sampling method which would produce the required volume of uncontaminated pore water in less than one hour could not be designed. The sampling requirement remains a challenge with the above constraints.

The original study plan proposed sampling soil at the surface, 25 and 45 cm depths. This strategy was tested on the transects with frequent loss of one or both of the deep samples along with complications resulting from compression of the deep cores. Since only the top 10 cm of soil could reliably be collected a composite sample with the periphyton layer removed was used as a standard sample. In hindsight it might have lost some information on the strata, however, at the time there was no spatial information describing the mercury species in soil.

2.1.4 Variable Probability Estimation

There are a number of other probabilistic models for distributing sampling resources than the tesselated probabilistic sampling design. Why were these rejected?

Response: The comment does not pertain to Section 2.1.4, rather, it questions the choice of the RTS design as opposed to some other design. The rationale for selecting the RTS design is detailed in Section 2.1 Design Rationale and Section 2.3 Design Summary. The response to the question on conceptual basis for the design also addresses this comment.

The tesselated probabilistic sampling design is only one component of the statistical basis for study design. Without statistical DQOs for hypothesis testing, it is not possible to determine whether the study design was adequate to resolve statistically significant differences in a factor between sites or times. The ability to discriminate significant differences between sites is important in locating hotspots, identifying and quantifying gradients, and spatial correlation analysis relating changing magnitudes of factors to changing states and vice versa. The ability to discriminate significant differences between times is important in quantifying transport velocities and response times. in identifying and quantifying lag correlations between states, factors, or both, and in identifying and quantifying temporal trends.

Response: The study was not specifically designed to test hypotheses, but rather address management questions. As stated earlier, there was an EMAP guideline to have within site variances ≤ 10% of the among site variances. This was achieved for almost every constituent. The data permit hotspot identification, gradient quantification, and spatial correlation analyses. The systematic selection of probability sites is a design feature that contributes directly to these capabilities.

3.0 Materials and Methods

Figure 3-1(3-9) was there a need to develop new methods for THg or MeHg in any matrix, when such capability existed at Frontier Geosciences, Brooks Rand, and Battelle Northwest? The PQL and MDL for THg and MeHg in water for the Bloom method (Bloom and Fitzgerald, 1988) were 0.2 and 0.05 ng/L and 0.02 and 0.005 ng/L, respectively, while those adopted in this study were unspecified and 0.2 ng/L and unspecified and 0.01 ng/L (sulfhydryl cotton method), respectively.

Response: At the time of initiation of this project there was no approved USEPA standard method for ultra trace level analysis of either THg or MeHg analysis in any matrix. FIU-SERP was under contract by the USDI, NPS, Everglades National Park to develop an ultra trace level analytical capability. USEPA Region 4 was planning a large REMAP project which was going to generate a large volume of samples from intensive field surveys in multiple matrices and it was clear that in order to meet holding times additional laboratory capability would be needed preferably located in South Florida. In an effort to meet the high sample through-put required by the project, emphasis was placed on flow-through methodology. Dr. Ron Jones (FIU-SERP) was approached as a primary cooperator both because of his detailed knowledge of the Everglades ecosystem and his ability to develop new analytical methods on a wide variety of matrices and parameters. However, due to the lack of USEPA standard methods a three laboratory design was developed for the project for QA/QC purposes and Battelle Marine Sciences Laboratory was included. During the initial pilot studies it was found that some existing methods were not sensitive enough to provide the required unqualified concentration values for MeHg in soil/sediment and additional analytical methods development was carried out by FIU-SERP. The PQL and MDL for Thg and MeHg in water for the methods used in this study were 1 ng/L (USEPA OQA) and 0.3 ng/L and 0.008 ng/L, respectively.

3.2 (3-11) No discussion of why a new method was needed for the determination of MeHg in water samples. If it is only to allow for the identification and quantitation of EtHg, one must ask whether there is any significance to the presence of EtHg in the environment? Are there other organo-mercury compounds that are markers of the key processes that transform mercury species by microbially mediated processes? Questions like this might have been answered with the development of a conceptual model of mercury cycling in the Everglades. EtHg is nowhere subsequently discussed in the report.

Response: A new method was not needed for the determination of MeHg in water samples because it was the primary project responsibility of the Battelle Marine Science Laboratory to analyse MeHg in water using the Bloom method throughout this project. Only after the need for a more sensitive analytical method for the analysis of organic mercury in soils/sediments became evident was a new method developed which lead to the subsequent discovery of EtHg. The discovery of EtHg was an unexpected benefit from this project, however, it has been reported in several European mercury studies and the Bloom method cannot detect EtHg. EtHg has not been discussed in this report because we have left this aspect to the academicians who have published numerous articles (e.g., Alli et al., 1994, Jones et al.,

1996; Cai et al., 1996; Cai et al., 1997; Cai et al., 1998) on analytical methodology and the presence of EtHg in the Everglades ecosystem. Phase II REMAP will include analyses of additional plant materials in the Everglades ecosystem which may develop a more complete understanding of EtHg in this system. Once a new organic mercury method was developed it was applied to water and found to compare very well with the results for MeHg obtained by Battelle MSL.

The helicopter sampling method chosen is inconsistent with that adopted by the SFWMD and the Everglades Technical Advisory Committee (ETAC). While there is a time and cost rationale for this difference, it should be made explicit.

Response: The Everglades Technical Advisory Committee (ETAC) served as a technical forum for development of a standard method for field sampling shallow Everglades surface water. FDEP subsequently published the outcome as a recommended method, which SFWMD adopted for their field sampling efforts. The method involves walking away from the helicopter to dip a marsh surface water sample. These samples were to be analyzed primarily for phosphorus. This field protocol is not an appropriate clean sampling method for trace level determination of mercury. During development of the marsh sampling protocol the dip method was tried both in the immediate vicinity and away from the helicopter. It became apparent that the dip method had several intrinsic problems: 1) It was impossible to minimize the particulate matter in the sample 2) it was not possible to fill the bottle with zero head space if the water depth was less than 6 inches due to the diameter of the 2 L teflon bottle ; 3) neither of these problems facilitated a clean sampling protocol for mercury; and 4) samples collected under the helicopter when compared to those collected away from the helicopter showed no difference in mercury concentrations nor did the field blanks show any mercury contamination. However, a formal study to compare the two methods would have been confounded by the high particle loading in the dip samples. It was clear that a clean sampling device designed to correct these problems as well as allow sampling very shallow water (approx. 2.5 cm deep) was needed. The vacuum sampling device with a 105 um mesh screen was developed and used in this study. The time and cost rationale was of secondary importance With the above significant limitations the wade and dip method was not a useful method consistent with obtaining clean water samples from the marsh unimpacted by the presence of the sampling effort

3.1 *"Water samples collected at various distances from the helicopter as well as the field blanks indicated no contamination of the samples was evident."* Include these study results.

Response: The dip method of sampling was used during the first canal (pilot) cycle and the transect (marsh pilot) study which fully demonstrated the limitations of the method. The vacuum sampler was used for all subsequent sample cycles in the canals and the marsh. A comparison of the pilot study data with the subsequent vacuum samples showed no significant changes in the mean and median concentrations, however, removal of large particles may have reduced total mercury in some samples.

Concerning soil slurry measurements taken in the laboratory. Measurements of temperature, pH and Eh on samples transported in coolers, in helicopters, exposed to the atmosphere, mixed, cleaned of large debris and then mixed with deionized water are not representative.

Response: These measurements were part of the laboratory protocol in the preparation of soil samples. These measurements were not taken to describe the environment from which the samples were taken but to develop an internally consistent set of measurements in the laboratory which could detect potential differences during processing.

"Soil pH and Eh along the marsh transect were measured on site by inserting an Eh electrode into an intact soil core." This is not a standard method. Compression of the sample by the soil corer will destroy existing stratifications. Furthermore, even though the core is encased in the corer diffusion of air into the sediments is rapid and will result in oxic measurements that are dependent on exposure time.

Response: This method was only used in the transect pilot study.

"The in situ Eh probe was inserted into the marsh soil. . ." Contradictory to previous statements. Was the Eh probe inserted into the marsh soil or the marsh soil core?

Response: During the marsh transect (pilot study), an Eh probe was inserted into an intact soil core This procedure was eliminated in subsequent marsh sampling in favor of the in situ Eh probe. The in situ Eh probe was inserted into the marsh soil following the pilot study. A design drawing of the probe system is presented in the appendix.

"This probe was allowed to equilibrate for 15 minutes. . ." This is insufficient time for equilibration. See Koch-Rose et al. (1994).

Response: The probe was tested in the marsh over time and little change was found after 15 minutes had elapsed.

"Water samples were collected by filling bottles underwater at each site." Unclear. Is this is a deviation from canal operations? If not, it is inconsistent with previous statements. If this is a deviation for canal operations this means that the canal samples were not screened for large particulates.

Response: As described above the dip sampling method was used on the first canal and transect pilot sampling efforts. The inclusion of large particulate matter especially in the marsh water samples resulted in great difficulty in obtaining comparable sample splits. The vacuum sampler was developed with a 105 μm mesh intake screen to remove the large particles in the water while allowing the considerable fine particulate load to be included in the sample.

"Soil pH and Eh were measured by inserting the respective electrodes into an intact core collected at the site. Soil cores were sampled to a depth of 45 cm along the transects." Unclear. This description comes after the deviations statement and therefore could be describing the canal sediment samples but the ponar dredge would not bring up soil cores. The use of the word "transects" implies that these may be marsh samples rather than canal samples. The description of inserting the probe into the core is consistent with some previous statements but contradictory to others.

Response: The appropriate changes in Section 3.1.4 Sampling Routine were made to alleviate any confusion.

Pg. 3-4. Value of Eh readings of soil mixed in a pan with DI water under laboratory conditions. What does this measure?

Response: It was an effort to determine if any changes could be detected at this level of sample processing. None were and the in situ data were used exclusively in the analysis.

2.1 Laboratory Analyses

The section presents several problems. The MDLs are stated up front as 0.3 ng/L for THg and 0.02 ng/L for MeHg. Later on in the text the description of the THg method notes the limit as 10 ng/L without preconcentration. The next statements are presumably concerning the process of preconcentration, but no final MDL is stated. Later on in the text the MDL for MeHg is given as 0.01 ng/L for the sulfhydryl cotton method. Maybe a table with method name, performance ranges, and date at which it was brought on-line would be helpful here.

Response: Text in the Laboratory Analyses section was clarified to alleviate this confusion.

2.2 QA/QC

A summary of the lab and field QA protocols would be helpful here, especially as regards bottle blanks, trip blanks and spikes, field blanks, equipment blanks, and field duplicates.

Response: The text in the QA/QC section indicates that the QA/QC evaluation of the database is attached as an appendix to this report. The QA/QC samples which were analyzed are included in the database which will be uploaded to the Internet.

Appendix B does not define decision DQOs, only field and laboratory performance DQOs.

Response: Correct. Decision DQOs were not defined for the project.

The absence of the ORD QA Report from Appendix B presents severe problems for the reviewer. With what frequencies were accuracy, precision, and completeness criteria violated in each study cycle? If the frequencies were unacceptable, what corrective actions were taken? What were the new frequencies of violation in the remaining study cycles after the corrective actions were implemented? If samples were split with an outside lab, what was the correlation relationship between the study and reference lab? If the correlation was unacceptable, what corrective actions were taken?

Response: The ORD report recommended that the EPA Region 4 Office of Quality Assurance (OQA) conduct a review of all data obtained from contract laboratories. The Region 4 OQA report was in preparation at the time the draft Technical Report was distributed for review and is the reason it was not included at that time. The QA/QC report is now final and has been included in Appendix B The QA/QC report also will be uploaded to the Internet along with the data set as a metadata file.

4.0 General Characteristics of the Water Regime

There were no multivariate analyses performed.
There was no lag correlation analysis performed.

Response: The purpose of this section was not to conducted extensive analyses on the water regime, but rather to provide the reader with some perspective on the general flow patterns and wet and dry season characteristics during the sampling period. The SFWMD and US Army Corps of Engineers, as well as others, have, and are, conducting exhaustive analyses of the water regime. These studies should be consulted if detailed information is desired on hydropatterns within the water conservation areas or the Park.

Pg. 4-1.
Table 4.1 is out of order. Pg 4-7.

Response: Corrected.

Flow path analysis should be expanded to include information on topography, vegetation types and densities, as well as water chemistry of tracers like chloride, sulfate, and conductivity. Here it would be very helpful to add District maps of the types and distributions of vegetation and USGS maps of Everglades topography to enhance our understanding of the relationships between water depth, vegetation communities, and water flow path. Overlay of the most probable flow path of each associated variable should provide a probabilistic contour of most probable flow path.

Response: The purpose of the general characteristics of the flow regime section was to provide the reader with perspective, rather than an exhaustive review and analysis of the flow regime. While the suggested information would be useful, it is beyond the scope of this report to include.

4.2.4 This section compares surface DO measurements with bottom DO measurements. However the methods refer only to a single-depth measurement of DO (page 34).

Response: The text has been modified to indicate the mid-depth sampling was conducted in the marsh and DO profiles were taken in the canals. Surface and bottom DO concentrations were compared at canal sites.

2.3.1 Temperature
Based on the timing and duration of temperature measurements at a given site, the change in water temperature from a minimum just before dawn and a maximum in the afternoon is missed. It is not valid to compare temperature results from different sites collected at different times of day. Furthermore, the diurnal temperature change is mediated by vegetation type and density, so any site intercomparison must take into account these differences, as well.

Response: Diel temperatures, indeed, change significantly at a specific site. However, the temperature comparisons in the report focused on differences in median temperatures between the wet and dry season and not differences among individual sites. The focus was on seasonal differences among water conservation areas.

4.3.4 This section describes the dependence of DO on a diel cycle, but does not properly caveat its conclusions concerning low DO measurements in the marsh. Furthermore the statement ". . . it is improbable that many native Everglades species are tolerant of prolonged oxygen depletion . . ." is open for debate. This statement should be limited solely to fish, not reptiles or amphibians, and further caveated with the observation that several species of Everglades fish species are surface gulpers, which enhances their ability to respire under low DO conditions. Thus, fish like the gar may be at a competitive advantage over fish species less tolerant of low DO conditions.

Response: The referenced statement was taken from the McCormick et. al. (1997) citation, as noted in the text. A sentence was added to caveat this conclusion for fish only.

4.3.5 The conclusion that turbidity is inversely related to water depth could be more an artifact of the way the samples were collected (by helicopter within a 15-minute period) than a true characteristic of the system.

Response: The paragraph following the paragraph referenced above states that the elevated turbidity at the shallow water sites might have been disturbance due to the intake probe when sampling very shallow waters.

Pg. 4-33. Conductivity in WCA2-B is shown to be high, but consistent with surrounding areas. This is noteworthy given the fact that this area is isolated and its hydrology is rain and seepage driven.

Response: So noted.

5.0 Habitat

Here it would be very helpful to add District maps of the types and distributions of vegetation and USGS maps of Everglades topography to enhance our understanding of the relationships between water depth, vegetation communities, and mercury contamination, if any.

Response: This chapter describes the habitat characterization conducted as part of the REMAP project. The purpose was to describe population attributes rather than individual site characteristics.

5.1 ". . . . *cattail (Typha domingensis), a highly invasive species that outcompetes native species...*" Provide citation. Change to *(Typha spp.)* To account for latifolia and hybrids. Change "native" to "more slowly growing species adapted to the low-nutrient environment of the unimpacted Everglades"

Response: Changes made.

Pg. 5-4. The L67 canal is discussed in third paragraph (other canals are ref. in text elsewhere), but canals are not labeled in any of the maps. Please label canals for quick reference.

Response: The L67 canal and other major canals have been labeled.

6.0 Soils

6.1 *Soil redox (Eh) was measured along the transcets* [typo]. *Comparison of soil redox data among transects, however; is not appropriate because the sampling methods used differed from transect to transect.* "Why did sampling methods differ from transect to transect? If methods were not comparable, one might consider removing Eh from data reporting and analysis.

Response: Transect sampling methods did not differ from transect to transect (see page 3-4). The soil redox method for the transects did differ from the marsh grid soil redox sampling method.

Pg. 6-5. The report should define Everglades Protection Area for general audience.

Response: Agreed. The area is defined.

Pg. 6-5. With regard to percent organic matters and bulk density: is this data for top 10 cm?

Response: Yes.

Pg. 6-5. Be consistent with landmark references, i.e., switching from Alligator Alley to I-75.

Response: Noted.

Pg. 6-6. The redox condition in soils is of course critical to the biogeochemistry of Hg as well as other parameters. Reducing conditions are defined relative to the hydrogen electrode, such that negative values are reducing and positive values are oxidizing. The activity of the hydrogen electrode is a function of pH, so it is important that Eh is standardized for $pH-E_7$. In addition, general statements such as" the presence of Eh less than 100 mV indicates anoxic reducing conditions are occurring..." need to be supported with citations. We know of no such citations. Sulfate is generally reduced by *desulfovibrio* bacteria under reducing conditions in the range -30 to -100 mv (ref?).

Response: The point at which no available molecular oxygen occurs in the soil is around 100 mv When oxygen is depleted those microorganisms thriving under anaerobic conditions will thrive, further reducing the Eh.

7.0 Nutrient Conditions

7.1 *. . . TP has been hypothesized as being one or several variables that influences mercury methylation processes in the Everglades ecosystem.,'* What kind of influences are being referred to here? Direct agnosim or antgonism or indirect influences via DO, biomass turnover, and the production of DOM, colloids, and particles? Also, it would help to refer reader to the section of the report where this hypothesis is evaluated.

Response: The hypotheses are that TP might have an indirect influence by increasing DOM production, stimulating periphyton production (internal environment for methylation), stimulating microbial decomposition and creating an environment suitable for methylating bacteria, and other indirect influences. This hypothesis was not evaluated as part of this report. Intensive, site-specific studies, such as those being conducted by the USGS ACME team are needed to test these hypotheses. REMAP can assist in identifying where these studies might be conducted.

'An extensive characterization of nutrients... was initiated." The Methods section contains no mention of analytical methods for nutrients, chlorophyll or APA. Please add.

Response: Citations to publications and methods have been added to the methods section in Chapter 3.

8.0 Mercury

8.1 *"The sources, extent and magnitude, transport, transformation, and pathways of Hg through thew Everglades ecosystem are poorly known."* Is this intended to apply to the time prior to study design or still. If the latter, see papers by Landing, Gill, Krabbenhoft, Hurley, Cleckner, Gilmour.

Response: The statement was intended to apply to the time before the comprehensive mercury studies began.

"Among the possible Hg sources in south Florida are. . . and agricultural operations." How were these possible sources identified? For example, were there high mercury concentrations in EAA soils? ... high transport rates of EAA soil particles? What are the background concentrations in Lake Okeechobee water upstream of the EAA? Study design was deficient in this regard.

Response: This is a general statement intended to indicate possible sources of mercury to the system. There is mercury in Everglades soil. We don't think the study design was deficient by not including Lake Okeechobee water. The study monitored the four main structures biweekly for three years to determine the seasonal concentrations of total and methyl mercury in the

water leaving the EAA. There is also no evidence of a mercury problem in fish in Lake Okeechobee.

"Various hypotheses have been put forward to account for the apparent susceptibility of the Everglades to Hg impacts . . Reference SFMSP Science Plan.

Response: This reference has been included along with USEPA, 1993.

"For fish hg contamination to reach concentrations that have ecological and human health consequences. three factors must exist: (1) Hg source; (2) unique combination of environmental conditions; and (3) bioaccumulation and biomagnification through the food chain." Split thought and deal with human health and ecological health in separate sentences. For ecological health: Capitalize Hg. Change "and" to "or". Change "factors" to "conditions". (1) might be interpreted to imply anthropogenic *"Hg source"*. Natural mercury deposits together with favorable conditions of natural or anthropogenic origin can create an aquatic mercury problem. Change (I) to "The presence of mercury in locations, forms, and concentrations accessible to natural aquatic bacteria." (2) is not accurate. Might better delete "unique" and add after "conditions" the phrase" favorable to a high rate of net methylmercury production and bioavailability." Also, there should probably be a fourth and a fifth factor for consideration: (4) There must be a significant rate of exposure to the contaminant by consumption of contaminated food. (5) One or more species of sensitive wildlife consuming contaminated food at normal rates accumulate methylmercury to toxic levels.

Response: Text changed to reflect this suggestion.

8.2 Initial Conceptual Mercury Cycling Model

A summary of this discussion should appear in Chapter 2.0 Study Design.

Response: A summary has been included in Chapter 2.

Page 8-4. *"Organic soils and sediments, such as those found throughout Everglades wetlands, would be expected under appropriate conditions to sequester Hg. The natural processes, by which this occurs, however, have been altered by water management and other anthropogenic activities in South Florida..."* What is meant by sequester? ... bind irreversibly such that it is not available to methylating bacteria? This implies that the efficiency of sequestration has decreased over time and that this is why there is more Hg available for methylation. I don't think anyone knows what the sequestered fraction was under historical conditions vs today. However, if the increase in sulfur in the Everglades environment has increased the incorporation of sulfur in plant and microbial proteins, or sulfide concentrations in the soil pore waters, then the ability of present day soils to sequester Hg may be greater than historically. Eutrophication has increase the peat production rate, in which case the naturally deposited mercury is being biodiluted. However, associated with this higher production rate are higher concentrations of DOM, that can prevent the formation of or resolubilize insoluble mercury sulfides. This would decrease sequestration efficiency of soils. To support this simple statement you have to get into the heart of the thermochemistry and kinetics of the mercury cycle, which is well beyond~ the scope of this study and won't be sorted out for several more years. Haven't mercury loadings also increased during this period? This has somewhat offset the effect of biodilution. Due to the complexity of the underlying logic of this statement, I would delete as either unnecessary or unprovable. Or perhaps could rephrase as a hypothesis to be tested in process studies to be conducted by others.

Response: The use of sequester in this context means burial of mercury below the soil recycling zone. The remainder of the comment goes well beyond the general intent of the statement.

Pg. 8-4. The interaction of the sulfur cycle and sulfate-reducing bacteria with the chemistry of Hg present numerous questions for understanding the processes affecting Hg contamination in the Everglades. "Why stop here? Present some of the questions and discuss how the sulfur gradient runs parallel to the phosphorus gradient.

Response: The interactions between sulfur and mercury intimately involve the process aspects of mercury cycling which was beyond the scope of this study A closer look at the results in the report indicate that the sulfur and phosphorus gradients are not parallel suggesting the interactions are spatially limited to WCA-2 where most of the sulfate is reduced to sulfide.

Pg. 8-4. *"Under anoxic conditions, inorganic Hg is converted to MeHg by sulfur bacteria and bioaccumulated in the food chain";* How can you reconcile the magnitude and disposition of the methylmercury problem in the Everglades with the statement made on Pg. 6-6 that "most of the areas had oxic soil conditions."

Response: It is possible that mercury methylation occurs in the diverse marsh system at sites other than soil. REMAP data indicate that the most probable methylation sites are not found in the soil except in the highly reduced soils which are primarily found in areas with high phosphorus concentrations. The methylation of mercury can also apparently occur in thick periphyton (floating, epiphytic or soil mat) which may be anoxic during the dark phase. The statements can therefore be reconciled due to the shifting sites of methylation. Given the diversity of microhabitats habitats and conditions throughout the vast marsh system one would reasonably expect microhabitats where it is anoxic in one area, and oxic in another.

Pg. 8-6. *"Thus despite the presence of flooded soils, Hg methylation may be expected to be limited in the absence of eutrophic conditions.* "Is the implication here that the naturally oligotrophic Everglades soils are oxic and therefore there will be only inconsequential methylmercury production? Then what is producing the high methylmercury concentrations in the Park waters and fish? I haven't done the calculations, but decomposing plant litter has to exert a BOD in the water column that exceeds the physical reaeration rate of the Everglades in the absence of photosynthetic activity, so nighttime DO must drop to near zero throughout the water column, except on exceptionally windy evenings. Same for sediment. However, since REMAP was a daytime study, would have missed this effect. During the day, at and below the sediment/water interface, decomposing surface peat must exert an oxygen demand that exceeds the rate of oxygen production by epibenthic periphyton. So, with the exception of the vicinity of cattail roots, which passively and actively pump air into their roots during night and day, respectively, Everglades sediment cannot be oxic, unless sawgrass litter and peat are so refractory that the BOD and SBOD are low compared with the daytime physical reaeration and oxygen production rates and oxygen readily diffuses into the litter and peat to significant depth. I don't think so. Have the Eh measurements been verified with DO measurements in the sediment pore water?

This statement also implies that we can ignore sulfur and increases in the mercury deposition Hg flux as potential causes of increased rates of methylmercury production. I don't think so.

Response: Yes. Methylmercury in water is the lowest in the Park when compared to the rest of the system and lowest in the southern half of ENP which is the area farthest from the phosphorus source. The concentration of mercury in mosquitofish is high in northern ENP and equivalent to that found in the hotspot south of Alligator Alley, however, the mercury concentration in

mosquitofish is lowest in the southern ENP where the phosphorus is also the lowest. The bioaccumlation factor (BAF) in mosquitofish increases consistently from north to south and is highest in the southern ENP indicating the uptake is strongly affected by a food chain which is very likely recovering (least affected) from the impacts of excess phosphorus from the north. Eh was sampled at over 500 sites across the system many of which occurred in sweet brown fibrous peat soils. It should be expected that for peat soil to occur and accumulate, decomposition must be minimal and these conditions are found in ultra oligotrophic marsh systems. It should not be inferred that sulfur or mercury deposition can be ignored. The key processes are likely to change from one part of the system to another due to the strong gradients from agricultural runoff that affect the interactions of multiple processes throughout the diverse marsh ecosystem.

8.3 Results

8.3.1 Hg load determinations require structure flow measurements. How were calculations carried out? Not shown in Chapter 9. Please add and cross-reference.

Response: Chapter 9 is not a mass balance; it is mass estimates. The intent was NOT to compute a mass balance where inputs and outputs would have to be estimated, but rather to compute instantaneous mass estimates for the sampling cycle only, so that a relative comparison could be made of the mass of mercury in various media (water, soil, periphyton, fish). This does not include flux, turnover, etc., but does provide a relative indication of how the mass distribution changes by season and among years. This will be made clearer in Chapter 9.0.

8.3.2 Water Quality Patterns

Is there a way to map out the most probable water flow paths in the interior marshes by season using something in addition to conductivity (p. 4-7), such as a combination of water depth, conductivity, and/or a conservative tracer (e.g., chloride). When this is done, what are the ranges of travel times between stations?

Add graphs of box/whisker plots of ratios of THg in fish vs MeHg in water, sediment, benthic periphyton, and floating periphyton at a specified latitude by season vs longitude to quantify north-south bioaccumulation factor (BAFs) gradients?... THg and MeHg in floating periphyton vs THg and MeHg in water and sediment. ... THg and MeHg in epibenthic periphyton vs THg and MeHg in water and sediment with and without normalization to organic fraction.

Add multivariate analysis of water BAFs vs water constituents by season and sediment BAFs vs sediment constituents by season for each distinct ecosphysiographic region and the whole system. (Can one do the same thing using latitude and longitude as variables in such an analysis?)

Perform a pairwise station similarity analysis for THg, MeHg, and MeHg/THg in water, sediment. periphyton, and fish. Same for water, sediment, and periphyton BAFs for fish and for water and sediment BAFs for periphyton. Which stations are the most similar and most different?

Plot minimum and maximum rates of change of each of the water quality variables and reduced variables (e.g., BAFs) between adjacent stations as a function of space by season. Plot kriging contours.

Building on the work in Appendix C, what is the relationship between the mosquitofish size distribution vs. MeHg concentration in each distinct ecosphysiographic region by season? I'm especially interested in THg/unit length vs water depth per season and THg/unit length vs. water or sediment TP per season

Response: The primary purpose of the Technical Report review was to ensure the scientific analyses and conclusions were sound and supported the conclusions. Additional analyses will be performed on the data in the future, but the most important current objective is to publish the report and the data set so the information is available to the scientific community and other users of the data. Conducting the proposed analyses would delay publishing the report. The data set will be uploaded to the Internet so the reviewer will be able to perform these analyses, if desired.

Misnumbered sections.

Response: Corrected.

8.3.2 *"The methylation of inorganic Hg to its bioaccumulated form (i.e., MeHg) occurs under a unique set of environmental conditions."* Substitute "favorable" for "unique". As noted before the methods section provides no analytical methods for nutrients.

Response: Substitution made.

Pg. 8-9. Is THgF - THg filtered samples or THg in fish? Ambiguous notation.

Response: THgF is total mercury in fish. The notation will be made clearer.

Pg. 8-10. Arguments of inverse relationship between Hg and nutrients fall away when looking at gradients south of Alligator Alley - this should be underscored in text.

Response: This is part of the conceptual model, which also reinforces the threshold concept between TP in water and methylation of mercury. When TP concentrations decrease below a certain threshold, MeHg concentrations in the water and sediment and fish Hg concentrations significantly decrease. The latitudinal gradients indicate these inverse relationships nicely, until the TP threshold is reached. Below the threshold, the inverse relationship is no longer apparent.

8.3.3 Soil Parameters

One of the most frustrating things about this section is the choice of units of ug/cc as the only way to quantify mercury species in peat soil. This has value, but nobody works solely in these units. Please include traditional wt/dry wt units, as well, to make it possible to calculate fish and periphyton BAFs relative to sediment. Also, if one includes such units, include the bulk density data used to calculate these values, so that the ambitious reader can perform the conversion calculations for himself or herself.

Response: These data will be uploaded with the report. Bulk density and soil mercury in weight units are part of this data so the reviewer will be able to perform these analyses.

See recommendations in 8.3.2 for additional data analyses.

8.3.4 Marsh Characteristics

See recommendations in 8.3.2 for additional data analyses.

9.0 Mercury Mass Estimates

What about equations for calculating flowing water loads and rainfall deposition loads?

Omits THg and MeHg mass stored in submerged and emergent macrophytes and discussion of significance of same. (I know you didn't sample macrophytes, but you could use emergent vegetation coverages and densities and plant/water or plant/sediment ratios from ENR Project studies to estimate range of plant mercury concentrations and bound the values of this storage reservoir.)

Response: As indicated above, mercury mass balances or budgets are NOT being computed in this section. This section provides a relative perspective of mercury among media for snapshots in time, not mass balances or budgets over time. Loads and fluxes are not needed to estimate relative mass units at a single instant in time.

Is this only for mosquitofish? If so, then use of generic "fish" is misleading. How were "fish" densities obtained? No references. Were these densities correlated with season, water depth, TP, or other water quality constituents?

Response: Estimates are only for mosquitofish so the generic "fish" will be replaced by mosquitofish. Density estimates were obtained from J. Trexler and do not reflect seasonal differences.

Proper periphyton mass storage equation includes a coverage factor. See Bob Ambrose for his estimates of same. How were periphyton densities obtained? Were these densities correlated with season, water depth, TP, or other water quality constituents? How much of difference between seasonal mass estimates was due to difference in densities or coverages vs difference in THg or MeHg concentration?

Response: Periphyton densities were estimated from sample weights for each station. Seasonal differences due to densities, TP, etc. were not estimated.

What is the ratio of the new mercury load from EAA runoff and rainfall deposition to the stored mercury load by compartment for both total mercury and methylmercury. If floating and epibenthic periphyton MeHg is newly produced MeHg, what is the production rate per unit area required to produce the stored MeH load?

Response: These estimates were not computed.

10. Synthesis and Integration

I think the additional analyses recommended in Chapters 8 and 9 will help formulate and test existing and new hypotheses here.

10.3 (10-6) Conceptual Models

Authors propose three revised conceptual models based on the segmentation of the system: above Alligator Alley, between AA and Tamiami Trail (TT). Testable hypotheses are proposed for each segment. This is good. While such a segmentation has conceptual value, it misses other relationships that transcends such segmentation. For example, a cursory review of Table 10.3 (10-10) suggests that the bioaccumulation factors in mosquitofish increase monotonically from the northern to southern Everglades. What is the hypothesis to be tested that accommodates this observation? Is this evidence of biodilution, as total phosphorus concentrations fall (nearly) monotonically from north to south? ... rainfall dilution of the factors that limit MeHg production by diluting out sulfate, shifting sulfide/sulfate to favor the formation of the bioavailable neutral sulfide species? ... slower growth rates in fish from north to south

due to lower average stage-duration in south than north? ... slower growth rates in fish from north to south due to lower average mass and energy throughput due to nutrient limitation? combinations of the above?

Response: A hypothesis to be tested which most likely would explain the increasing BAF from north to south would be: what fundamental alterations in the structure of the food chain (e.g., pollution tolerant species to species native to the original ultra oligotrophic marsh) have been brought about by the phosphorus gradient? Biodilution is very likely a part of the explanation with the proliferation of pollution tolerant biomass in the north. Rainfall dilution and sulfur interactions, and growth rates in fish are probably less likely to be large factors in explaining the rapid change in the BAF.

10.3.1(10-7) North of Alligator Alley

Reference is made to increased growth and condition of mosquitofish in the ENR Project that results in biodilution, which explains the low MeHg concentrations in these mosquitofish. May be confusing growth dilution associated with rapidly growing fish with biodilution related to total biomass per unit area or volume. Fish biomass densities in ENR Project are actually lower than expected based on extrapolations from interior marsh studies of similar habitats in lower phosphorus concentration regime (Jordan, 1996). Growth rates of mosquitofish have not been measured. Peak concentrations correspond to or lag by one quarter the onset of the summer rainy season when the deposition rate of soluble, reactive mercury is at a maximum. MeHg concentrations in the ENR Project interior water are in the concentration range of 0.04-0.13 ng/L, with the mean 0.07 ng/L. This is about one-eighth the concentration in LNWR and AA-N. Mosquitofish concentrations are in the range of 1-30 ng/g, with a mean of around 7 ng/g, which yields a BAF of about 1×10^5. This is about double that reported for the LNWR, but a little less than half that in AA-N.

The more likely explanation for the lower mean MeHg concentration in ENR Project water is the much higher sulfate concentrations, which, in the presence of eutrophic and anoxic conditions, produces an abundance of sulfide that poisons the methylation process. This can be seen in ENR sediment MeHg concentrations, which are about an order of magnitude lower than those in WCA-3A.(Gilmour et al., 1998). The more likely explanation for the peak in MeHg concentrations in the interior water in the summer is that with the onset of summer rainfall, not only is more soluble Hg(II) delivered to the system, but both sulfate and phosphate are diluted, reducing the rate of production of sulfide, shifting conditions to favor the formation of the neutral sulfide complex that is readily taken up by methylating bacteria.

Response: Growth dilution does not appear to be a factor that occurs to any significant degree. Therefore there is no confusion with the concept of biodilution. If growth dilution were a factor there should be a relationship between Hg concentration and fish size. Hg uptake in small fish is apparently very rapid and the concentration does not decline with size. The biodilution process begins at the base of the food chain and is much more likely to be influenced by TP than other variables in this system.

11.0 Missing

Response: Section has been added.

12.0 Missing

Response: Section has been added.

13.0 References

Complete incomplete citations.

Response: Complete citations added.

Appendix A. Sampling Equipment

Appendix B. DQOs

Appendix B has an Appendix A. Perhaps better Appendix I.

Response: Change made.

Appendix B does not define decision DQOs, only field and laboratory performance DQOs.

Response: As explained above decision DQOs were not established due to the research nature of this project.

Three standard deviations does not define outliers per se.

Three standard deviations not applicable to evaluate performance relative to %RSD of %RPD.

Response: The guidance found in USEPA QA/G-4 (USEPA 1994) which is not entirely appropriate for research projects was followed.

Table 3A This table lists the methods used in the study. A large number of these methods are listed as new method development. Provide citations for peer reviewed method comparisons. If not available, provide data for method validation studies.

Response: The peer reviewed citations are included.

Table 6A The maximum value for water depth in the marsh is listed as 6 meters, this seems excessive. Probably 6 feet or 2 meters.

Response: Units corrected.

Table AlA The lack of text associated with this table makes it difficult to interpret. It is apparently the data quality objective targets for the project. If this is the case, then the precision targets for duplicate samples were 3s which is significantly higher than the more typical 2s. Were these criteria relaxed because of the research or scoping nature of the study? If so, make explicit. Furthermore, a secondary criterion of %RSD @95% is listed. This is difficult to interpret and appears to state that duplicate data will be accepted up to the 95th percentile confidence levels of the RSD. This is also very different from the standard of *+/-25%.* Same question regarding relaxation of QC criteria for research or scoping study?

Response: This table is the result of the application of the guidance provided in the document cited above for Appendix B.

Table B.1.A The lack of text associated with this table makes it difficult to interpret. It appears to be a summary of the field data quality results. If this is the case then the precision targets for duplicate samples were 3s which is significantly higher than the standard 2s. Furthermore, these targets appear to be used to identify outliers for removal from the data. If this study followed standard procedures than duplicate analyses were performed on about 10% of all samples and each duplicate pair is meant to represent a certain associated subset of data. Thus each pair represents a minimum of around ten data points. It is from these duplicate pairs that the calculation of precision is made. These values are not compared to the mean of the data but to each other. Similarly, it is inappropriate to qualify one value as failing the QA standard, rather it takes both values. So single points in the outliers list are not appropriate. Furthermore, if a duplicate pair fails QA the pair is not considered an outlier, rather the associated data set is flagged as suspect.

Response: This table follows guidance cited for Appendix B above in assessing the database as prescribed.

Appendix C. Eastern Mosquito Fish Studies Mosquitofish is one word.

Nice work as far as it goes. Would benefit from an analysis of the correlation of fish size distribution in each distinct ecophysiographic region vs mean water depth, season, presence or absence of periphyton, concentration of TP.

Response: This is the contribution of Dr. Joel Trexler and students who want at least another season of data (preferable dry season) before carrying the analyses further.

Author: Mike Birch at REGION4
Date: 6/23/1998 2:15 PM
Priority: Normal
TO: Jerry Stober
Subject: comments on report

Comments on South Florida Ecosystem Report

1. Page i Executive Summary "total sulfate" should be just sulfate. Since the method only gives ionic sulfate and would not measure sulfate bound in particulate matter.

   Response: Agreed.

2. Page iii list of Acronyms TSO_4 total sulfate should be SO_4 ionic sulfate. The acronym for ThgF is listed in the report but not explained here.

   Response: Total ionic sulfate has been added to list of acronyms ThgF definition has been added to the acronyms.

3. Page xxi People left off list. There is no mention of Doug Winters, Jim chandler, Sandra Allen, Charlie Appleby of the ESAT team, or EPA EMSL-LV ORD Deb Chaloud or Ed Heithmer.

   Response: These individuals were added to the list.

4. Page 2-14 Table 2.3 there were some Total phosphorus values for the media periphyton.

   Response: Total phosphorus was analyzed in periphyton samples from the April 1994 marsh transects. Only water, soil and sediment were analyzed for TP for the marsh grid samples.

5. Page 3-3 "lexan" does not have a trademark symbol.

   Response: Trademark symbol added.

6. Page 3-7 Last sentence of first paragraph, "this unit is equipped with a stirrer for accurate DO determinations." The stirrer maintains a constant flow of water across the membrane to prevent oxygen sag, but the stirrer does not increase the accuracy.

   Response: Sentence was revised.

7. Page 3-10 third sentence should be rewritten. For the analysis of THg in soil, sediment, and fish, the tissue samples were placed in glass ampules with concentrated nitric acid, sealed and autoclave to facilitate digestion.

   Response: So noted.

8. Page 3-11 last sentence, the reference should be "Bloom".

   Response: Reference added.

9. Page 3-13 The correction was not made in the standard curve but in making an appropriate correction for the reagents contained in the samples, blank and standards.

Response: So noted.

10. Page 3-17 Table 3.1 Under the column for Battelle for Total nitrogen and Total phosphorus there were splits and duplicates.

Response: Addition made.

11. Page 4-10 Section 4.3.6 Is it possible that the decrease in alkalinity is due to nitrification?

Response: The data do not permit this type of analysis.

12. Page 8-B what is THgF? Not defined in List of Acronyms

Response: ThgF is total mercury in *Gambusia*. It has been added to the acronym list.

Volume II comments

13. Page 16 Table 6A specific conductance. Conductivity according to Standard Methods is customarily reported in micromhos per centimeter (μmho/cm) . In the International system of Units (μS) is used. Is everything else reported in the International system?

Response: Yes.

14. Page 16 Table 6A Does alkaline phosphatase have units?

Response: The units are μMol/hr.

15. Page 8-17 where the data compromised by the detection levels reported or was the ability to define the significance compromised. The term "high" detection limit is a misnomer. The method is only suppose to go down to 1 mg/L. Both statement tends to say the data were "inaccurate" or lead to that impression.

Response: The sulfate analysis conducted by USEPA Region 4 SESD had MDLs of 5, 2 and 0.5 mg/L throughout the course of the study which resulted in a left censored data set These MDLs were certainly adequate to identify the major sulfate gradients across the system, however, it is unknown whether a detection level below 0.5 mg/L would have been able to further elucidate gradients in the southern part of the system.

16. Table AlA. Total Mercury and Methyl mercury under the column comparability (split samples SOPs, Std. Units) SOPs Battelle hab???

Response: This column in this table simply indicates with whom the comparability was made.

17. Table B.l.A. Canal data field precision. This table is not reproducibility between replicates, but how well a distribution agrees with it self. The title needs to be changed.

Response: Agree. Title has been changed. The table does indicate strong consistency within the sizable database.

18. Table B.1.A No statement about ethyl mercury but ethyl mercury is talked about in the report.

Response: Ethyl mercury data is not presented in the report, however, the method developed detects ethyl and methyl mercury. Ethyl mercury was not found consistently in all soil or periphyton analyses.

FLO-SUN INCORPORATED

JUNE 15, 1998

Jerry Stober, Ph. D.
Project Leader
Everglades Ecosystem Assessment
USEPA, Region 4
Science and Ecosystem Support Division
980 College Station Road
Athens, GA 30605-2720

Dear Jerry:

Thank you for providing an opportunity to comment on the South Florida Ecosystem Assessment Peer Review Draft report. Given the unavailability of the raw data, our comments are based solely on the content of the report. In addition, due to the short review time, do not construe that a failure to comment on a specific section implies that we concur with conclusions contained in that section.

My comments follow:

Section 1.4 & 7.2.4

Pgs. 1-7 & 7-9

Although this study collected a large amount of information, it was conducted over a short period under a narrow range of hydrologic conditions. The report states that the data "was collected over two very different years with respect to precipitation." This conclusion is unsubstantiated by the rainfall data presented. Based on Table 4.2, the report should have concluded that studies were conducted during a normal period. No data was collected during dry periods. In addition, since large volumes of regulatory releases were made from Lake Okeechobee during the study period, historical long-term comparisons of surface inflows need to be made. Dry and wet season comparisons also need to be provided.

Response: Table 4-2 does not reference either a dry or a wet period, but states the flows in 1994-1996. Since the flow through 12D is strongly influenced by water management decisions, the total Shark River Slough flow (S12ABCD+S333) was substituted for a frame of reference. With this frame of reference, all three years were wet with 1995 being exceptionally wet. The Shark River Slough flow was 4 times the average flow experienced in WY 1979-1993 and 1.6 times the maximum flow. A table with these computations was added to the report. We agree long-term comparisons need to be made. We also agree that wet and dry year comparisons should be made. Therefore, it is proposed that the REMAP program continue to monitor conditions in the marsh and canals so these comparisons will be possible in the future.

The scope of this study provides little insight into two of your policy questions - trend and cause. It would be speculative to base conclusions of long-term trends on this short-term data collected under a narrow range of hydrologic conditions. In addition, inferring causal relationships using synoptic data is speculative.

Response: There was no attempt to base any conclusions in the report on temporal trends, but spatial trends are especially applicable for this synoptic sampling approach. Because the data were collected using a statistical survey design, it is possible to provide population estimates of resource condition with known confidence. The design also permits spatial analyses and

associations that provide insight into functional relationships. Without such a design (e.g., judgmental samples selected at a single site), it is speculative to make conclusions about conditions or relationships anywhere else, but at that site.

Section 3.3

The Florida Department of Environmental Protection recently released an audit of the FIU-SERP laboratory. The audit covered the lab analysis of both the phosphorus and mercury. DEP concluded "it remains unclear whether past data can be defined as legally defensible and of know quality". In addition, EPA has conducted a quality control audit of the FIU lab and it is generally understood that the audit has identified major shortcomings. The results of these audits should figure clearly and prominently in the QA/QC section of your report Conclusions based on the FIU-SERP data should be flagged for the reader. Furthermore, EPA should reach no specific conclusions based upon this data.

Response: The audit by USEPA, Region 4, SESD, Office of Quality Assurance of the South Florida Ecosystem Assessment Project database found that of the over 20,425 measurements made during the four year REMAP study period, only 15 percent of the data were qualified after being reviewed using rigorous QA/QC compliance monitoring methods. Most of the data within the 15 percent remained usable in the assessment and characterization of the system. The results of the EPA audit are included as an appendix to the final report. In addition, the data set and qualifiers will be available on a world wide web site.

Section 1.3.3

Pg. 1-4
The report states that Nutrient loading from the EAA and urban areas has significantly increased nutrient concentrations, particularly phosphorus, in the downstream WCAs and ENP". The references cited in support of this statement are outdated. Recent analysis of concentrations entering ENP clearly demonstrate that phosphorus concentrations have declined since 1986 and are at the same level as in 1978 (Walker 1997, MacVicar, Federico & Lamb Inc., 1996).

Response: The marsh phosphorus data collected during this study show lower concentrations in 1996. However, whether the observed concentrations are due to the initiation of phosphorus control efforts, water management practices, the varying hydrologic conditions that occurred during a particular sampling event, or some combination of these factors, is unclear.

Pg. 1-5
FDEP has not concluded that eutrophication of the Everglades results in violations of water quality standards. The report referenced was and still is a draft that has never been adopted by the Department. It is misleading, therefore, to state the FDEP has reached the conclusions contained in the report

Response: Disagree. The report cited is a draft FDEP publication. In addition, FDEP did clearly stipulate these violations in the 1991 federal settlement agreement concerning eutrophication of the Everglades.

Section 7.3

Pg. 7-11
It is unclear how the data collected during this short-term study provide any insight into the effectiveness STAs.

Response: The paragraph refers to the synoptic sampling providing a baseline for comparison of conditions in the future following the implementation of the STAs. We agree without continued monitoring it will be difficult to distinguish the effects of hydrometeorological variability from STA performance. Text has been modified.

Section 7.2.4

Pg. 7-8
This section relies on logistic regression models to describe the relationship between phosphorus and cattail/periphyton abundance. However, the referenced figures (Figures 7.22 & 7.23) neither display the raw data used in the regressions nor the regression statistics. Without. such information the conclusions reached are unsupported and should be deleted.

Response: The results from the logistic regression models were highly significant, but the proportion of variance explained was not judged to be satisfactory by the Project Team. Therefore, these results and conclusions were removed.

Section 10.2

Pg. 10.5
The application of the data collected under this study to the determination or inference of a phosphorus threshold concentration is an inappropriate extrapolation. The report associates low ranges of phosphorus concentration to greater periphyton biomass. However, the Ecosystem Assessment study did not collect periphyton biomass data (only presence/absence was noted). Therefore, the inferences were based on causal observations at best. The determination of a phosphorus threshold concentration is too critical an issue to be based on such casual relationships. Government agencies and the EAA Environmental Protection District are spending millions of dollars on the experimental determination of phosphorus threshold concentrations The report should delete all references to phosphorus threshold concentrations.

Response: The association of plant responses with total phosphorus concentrations along the "river of grass" is too obvious to ignore. The inference of a phosphorus threshold will more clearly be stated as a hypothesis.

We would appreciate receiving all the raw data in electronic form in order to conduct independent analysis. Please consider this a formal request for the data. If this is not sufficient, please let me know what is necessary in order to receive an electronic copy of the data upon which this report is based.

Sincerely,

Peter C. Rosendahl, Ph.D.
Vice President of Environmental Relations

William W. Walker, Jr., Ph.D.
Environmental Engineer
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MEMO

To: Jerry Stober, U.S. Environmental Protection Agency, Region 4

Copies: Mike Zimmerman, Everglades National Park
Su Jewell, Loxahatchee National Wildlife Refuge

Subject: Comments on "South Florida Ecosystem Assessment, Vol.1, Technical Report, Peer Review Draft"

Date: June 18,1998

I appreciate the opportunity to provide comments on the above draft report. Your letter of May 11 indicated that if you did not receive my review by June 15, you would assume that I "fully concur with the substance of the report." The following comments reflect the level of effort that I have able to commit to the review within this time frame. Important items may have been overlooked. Because of time limitations, I did not review Chapters 8-9 and some of Chapter 10. Please do not assume that I fully concur with these or other parts of the report that are not discussed.

To some extent, my review is handicapped by the fact that I have not been able to get a copy of the data set, though I understand the need for quality control. It is difficult to form opinions on spatial and temporal patterns and relationships discussed in the report without working with the data. One important aspect is consistency with other regional data sets (in particular, those derived from SFMWD's long-term monitoring programs). Overlaying the EPA data sets with others may provide important insights. Given the spatial intensity, the water-column and soils data could be extremely useful for modeling and other purposes. I hope that you will be able to provide a complete copy if the data set in the near future.

p.1-5 "The effectiveness of these controls in reducing nutrient concentrations to near historical levels, however, is not yet known". Considerable information is available on these controls. In ~3 years of full BMP implementation (1995-1997), EAA phosphorus loads were reduced by an average of 51%, relative to 1979-1988 levels and adjusted for variations in rainfall (SFWMD Everglades Regulation Department). The Everglades Nutrient Removal Project (small-scale STA) removed an average of more than 75% of its influent phosphorus load between August 1994 (startup) and November 1996 (SFMWD, Everglades Nutrient Removal Project, 1996 Monitoring Report, March 1997).

Response: Controls have reduced phosphorus concentrations below the 1979-1989 levels, but although the phosphorus concentrations in the ENR effluent have been consistently less than 30 μg/L, they still exceed the Everglades marsh background concentration of 10 μg/L. The text will be clarified so that historical refers to natural phosphorus concentrations of 10 μg/L or less. The effectiveness of Phase I controls in achieving these phosphorus concentrations over the long-term remains unknown.

Chapters 2/3, also p.5-2. It would be helpful if more detail could be provided on the criteria used to classify sites based upon vegetation. If a single cattail (or periphyton mat) was visible from the sampling location was this enough to indicate "presence"? Were any density criteria applied? Did the vegetation have to be within a certain distance of the sampling location in order to be considered "present"?

Response: The simple presence of a cattail or periphyton mat was enough to indicate existence when visible in two routine 35mm slide photographs taken at each sampling site: 1) a ground level shot showing the typical plant types and 2) an oblique shot from an altitude of about 100 feet of the sampling site. General habitat types were identified by field personnel when on station, however, after all photographs were in hand and catalogued the same two people viewed all photos and classified the presence of selected plant types which were used in this analysis. No density criteria were used. No distance criteria were used except that the plant type had to be identifiable in the photo.

Chapter 3. It would be appropriate to document in greater detail the method used for selecting the precise spot for collecting a water column and soil sample. Once the helicopter lands at a site, the sampling crew has to decide precisely where to collect the sample Was there a tendency to collect samples in open waters (vs densely vegetated areas)? Or was this aspect also randomized?

Response: The GPS used was estimated to be accurate to within about 10 m. The sampling site was located with the GPS and the helicopter was landed as close to the point as possible. An effort was made to collect all media at each site whether or not in wet prairies or dense vegetation.

P 3-3. Collection of Soil Cores. The text does not define the sampled "soil". Marsh bottoms are often coated with periphyton and loose floc/detritus/soupy stuff. Was this material included or excluded from the 10-cm "soil" cores? The interface between the soup and the soil can be difficult to determine. Were plant roots picked out of the cores before analysis? These aspects of the sampling protocol could be very important for interpreting the soil data and comparing them with other data.

Response: Some of these questions are answered on pages 3-7 and 3-8. The intact soil periphyton was removed from the soil core and analyzed separately as soil periphyton. The overlying water or soup was not sampled but allowed to run off the top of the core. The large plant roots were picked out of the soil and the remaining soil was the sample.

P 4-2, Table 4-2. The flow through S12D is listed as a reference for hydrologic conditions. Since this flow is strongly influenced by water management decisions, the total Shark Slough flow (S12ABCD+S333) would provide a better frame of reference. It should be noted that all 3 years were wet and 1995 was extremely wet. The Shark Slough flow was 4 times the average flow experienced in WY 1979-1993 and 1.6 times maximum flow. A row with long-term-average flows could be included in Table 4.2. have recomputed the flows as follows:

WYear	55A	S6	S7	S8	S12s+S333
1994	13.4	13.8	9.9	11.0	33.5
1995	16.2	23.9	18.0	25.0	97.5
1996	11.5	12.7	9.7	14.0	58.0
1979-1993	9.1	6.4	8.8	11.4	24.5

Flows in cubic meters per second, Water Years ending Sept 30. values differ somewhat from values cited in the draft table. Note that the S5A values differ somewhat from values cited in the draft table.

I was curious about stage values. Stage is generally (temporally) correlated with water column P concentrations in the Everglades, particularly at more enriched sites. The following figure shows monthly mean stages for 1980-1996 at station P33 in ENP. As a consequence of the high stages during marsh sampling events, I would expect that the measured distribution of phosphorus concentrations at marsh stations is below the long-term average. Since water levels were higher during marsh sampling than during canal sampling, some of the apparent differences in P concentration between marsh sites and canal sites may be related to hydrologic variations.

Response: Thank you for the analyses. We agree and will cite this in the text with reference to Dr. Walker.

pp.4-3,7. Tables 4.3 & 4.4 are useful summaries. A reference should be given for the formula to compute confidence intervals for the median. The meaning of "confidence interval'. is not clear (see comments on Tables 10.2 & 10.3 below).

Response: Reference added.

Figure 4.19, 4.28. Since turbidity values are highly skewed, the Box plots would be more informative if the y axes were logarithmic. Likewise for all total P Box plots & frequency distribution plots (Figures 7.2, 7.3, 7.9.7.10, 7.12)

Response: Comment noted.

Figure 4.23. The legend should provide more detail on the frequency curves. Are these derived from the long-term stage record or from the stage record during the period of monitoring? The figure for P33 suggests that the sampling regime is representative (points are spread over curve). This is misleading in light of the above figure. Stages and water depths were well above normal in 3 out of the 4 marsh sampling dates.

Response: The legend will be expanded and the analyses will be reviewed. The frequency analyses were for the period of record, not just the sampling period.

Tables 5.1 & 5.2. This information is extremely interesting & useful. Did the habitat types change from one sampling event to another? Is there any way of computing the uncertainty associated with the habitat distribution estimates?

Response. Each sampling event was a new set of randomized stations. The plant species analyzed were the same for each event and did not change. Due to the relatively short time span of the study the plant responses were not expected to change and therefore were pooled over the entire system combining about 500 observations. Uncertainty estimates were not calculated.

Table 6.1. p.6-3. Are the bulk density & %organic matter values for the entire soil column or for 0-10 cm only? Are the statistics based upon pooling data from 4 sampling rounds and all stations within each Subarea? i.e. do the standard deviations reflect spatial variations, temporal variations, or a mixture? Were soil properties measured in each sampling round?

Response: Bulk density and % organic matter were based on samples from the 0-10 cm soil profile. The statistics, including standard deviations, represent pooled data for all four cycles. Soil properties were measured in each sampling cycle.

Table 7.1 p.7-3. Standard errors of the geometric means seem appropriate here.

Response: Standard errors of the geometric mean do not transform and therefore are not provided with the means.

p.7-7, top. What is meaning of "marsh water" in the EAA? Holeyland/Rottenberger?

Response: The EAA was not sampled, however, the Holeyland/Rottenberger area was included in marsh sampling. The text should have read "were highest in WCA2, WCA3, and LNWR and lowest in ENP and BCNP regardless of season."

p.7-7 bottom. Where did total N data come from? This is not on the measured parameter list (Tables 2.1, 2.2). Was total N sampled at the same frequency and locations as total P? If not, average N/P ratios should be computed based upon paired values only.

Response: Total N was measured only in the last two cycles. It has been added to the parameter list. N/P ratios were not calculated, but would be done on a paired basis.

Figures 7.22 & 7.23. Some measure of goodness of fit should be provided (classification error, etc.). At least show the data points upon which these curves are based. If the curves integrate over all regions and data, they may be misleading because the relationships may vary with habitat type, region, and hydroperiod.

Development of similar curves within regions with similar hydroperiod would be more informative and may give very different results. These curves may be very misleading and could be misused. In any case, much more statistical detail is warranted if they are to be included in the report.

Response: The results from the logistic regression models were highly significant but the proportion of explained variance was not judged satisfactory by the Project Team. Therefore, these results were removed.

p.10-2. last paragraph. "This plot shows an inverse relationship between TP in water and THg in mosquitofish.." The plot does not show a relationship, only a spatial correlation. The causal "relationship" for THg could be with another spatially varying factor. The report should be careful about assigning or implying causality based upon spatial correlations.

Response: So noted.

p.10-7. Section 10.3.2. "In the marsh, TP concentrations are between 10 to 20 μg/L, which is a level currently within the assimilative capacity of the system and that stimulates periphyton production and increased periphyton biomass." Where in the report has the "assimilative capacity" been determined? This sweeping statement does not seem to be supported in the report. The last part of the sentence is in direct contradiction with results of ongoing threshold research by SFWMD which, I believe, demonstrates experimentally that native periphyton mats tend to decrease at P levels above 10 μg/L. Is this sentence intended as a hypothesis (not a conclusion)? If so, it should be clearly stated as such.

Response: The statement is intended as a hypothesis and has been so indicated in the text.

Tables 10.2,10.3. The meaning of the "95% confidence interval about the median" is unclear. Is this the confidence interval for the median in space and time during the total sampling period? If the interval is calculated by pooling data from all stations and sampling rounds within each Subarea, then it is probably inaccurate because the samples are not statistically independent. A one-way ANOVA (or nonparametric equivalent) would probably indicate significant variations in Subarea means across sampling dates. The effective number of samples for representing temporal variations is only four. The effective sample size for computing the spatial & temporal median would be much smaller than the total number of samples presumably (?) used to calculate the confidence interval.

Response: The meaning of the 95% confidence interval about the median is given in Section 2. The median estimator and its CI are design-based, so that the only lack of statistical independence that needs to be accounted for is that introduced by the spatial restriction of the sample design. As noted in Section 2, under the condition of spatial pattern, the CI is conservative (in Walker's terms, corresponding to under-estimating the degrees of freedom, not over-estimating as suggested by Walker).

In particular, the value estimated is the median of the population that existed at the four sampling times. It is not a model-based estimate of the "real" median of some hypothetical population subject to stochastic spatial and temporal perturbations. So-called "temporal correlation" has no bearing on the estimator.

Table A1A - Water Data Quality Objective Criteria. The meanings of the terms should be clearly defined. Some of the numbers do not seem to make much sense. For example, Duplicate Precision (%RSD) value for Turbidity in canals is 182.9. Does this mean that duplicates can be expected to differ by as much as 183%? Likewise, duplicates for total phosphorus and mercury, by 101% and 230%, respectively? These seem like situations to be avoided, not OBJECTIVES. Does "Field Precision" reflect variance across samples taken at the same location and time? What program generated this table? How many replicates, duplicates, spikes etc.?

Response: The terms are more clearly defined in the table and the reader is referred to EPA DQO and QA documents cited in the reference section. The information in the table was generated by EPA QAMS based on QA programs which they developed. The number of replicates, duplicates, spikes, etc. can be found in the field and laboratory methods manuals which are cited in the reference section.

Table B.1.A. What methods were used to detect "Outliers"? Were outliers included or excluded from the analyses conducted in the report? It seems like one of these columns should contain a value for relative standard deviation (coefficient of variation) across replicate samples. It is not at all clear what the numbers mean or how they were generated.

Response: Outliers were detected if values were greater than $\pm 3\sigma$ or were outside range checks for Everglades constituents. However, all data, including outliers, were used in all statistical analyses and are included in the results presented.

This concludes my comments. I hope that you find them useful. The report and data sets will contribute substantially to Everglades research and management. Please contact me if you have any questions.

Author: ckendall@usgs.gov at IN
Date: 6/14/1998 5:11 PM
Priority: Normal
BCC: jerry stober at PEGION4
TO: STOBER.JERRY at IN
CC: ahiger@usgs.gov at IN, dpkrabbe@usgs.gov at IN, ckendall@usgs.gov at IN
Subject: review of technical report

Hi Response:

Thanks for sending me a copy of your report after the recent meeting. Here are a variety of comments based on spending a day reading it (alas, the first blue-sky weekend day we have seemingly had in months!) . I only read Joel Trexler's part of the appendix, not the rest of it. It was a shear delight getting to see the rest of your data, after so long. In particular, I have been really wondering about your vegetation data and what that might tell me about the spatial patterns in dl3C of the periphyton and *Gambusia*. When I get a spare moment, I will do all sorts of comparisons to see if the changes in relation dominance of the plants might explain some of the isotope variability. For example, is there a difference in the *Gambusia* in places with and without periphyton mats?

I also look forward to working with you and Joel to make sense of the spatial variability in *Gambusia* and to rationalize Joel's gut contents data with the isotope data. If neither of you has time or interest in taking the lead on such a comparison in the near future, I might see if Cecily Chang, the biologist in our group who has a moderately good statistical background, might want to make a start on the comparisons. What do you think? I will send a note to Joel to see what he thinks, too. I am sure that there is room for more than one paper relating these massive datasets.

This is an impressive document, sure to give readers lots of things to think about and hypotheses to test with other datasets. I hope that you will now have time to take chunks of these data and publish them in the white literature. You have LOTS of neat stuff here.

You have made me a believer in box and whisker plots. I finally see why you divided the system into latitudinal bands and used these plots to show broad regional changes. I like the idea of viewing the system as a "wide shallow stream", and I guess it is OK that the divisions are not perpendicular to the flow lines. Did you try it the other way? I understand that there are some fancy 3-D visualization packages that let you rotate the data in space to see in what plane the patterns are strongest. cool! These box plots work pretty well for showing big patterns, and there are not many readily available tools for comparing spatial patterns, but I am still hoping to find some better tools to deal with the smaller patterns.

There is a large amount of relevant ACME project work that should be cited in your report, as well as recent work by other USGS FL groups such as Bill Orem's sediment group. One paper by Gilmour is the only one I saw from these groups. There have been lots of published abstracts about our work in the proceedings of the annual USGS meeting and other international meetings, several papers have been published, and I know that a lot of papers were submitted to a Hg conference in Hamburg, and these certainly could have been cited as in press before (but now I think they are published). It seems like some of your Hg-related speculations and hypotheses are partially based on some ACME work or could be partially validated by reference to some ACME work. The FL missions of the two agencies are nicely complementary. I realize that this is an EPA report and you naturally want to emphasize your data and their patterns, but when you start interpreting the data, these USGS studies are as much a part of the public literature as the many academic or SFWMD papers that you cite extensively.

Below are miscellaneous comments I scribbled as I read through your report:

Executive summary: you need to explain what biodilution and BAF mean. I don't think BAF was defined anywhere in the text, nor was anything said about its units (if any). The whole concept of biodilution seems weird to me– just because a fish grows large, its MeHg is considered diluted?

Response: The concept of biodilution is used in reference to eutrophic systems. In eutrophic systems there is high nutrient availability which usually results in high primary plant production (periphyton or phytoplankton). As a result there are more cells per unit volume each with a portion of the available mercury in the system thereby diluting the mercury broadly across the base of the food chain. The grazers cannot utilize all of the available plant biomass and the efficiency of the bioaccumulation is lower at all higher trophic levels.

Response: The BAF is the bioaccumulation factor. The BAF for methylmercury is defined as the concentration of methylmercury in whole fish divided by the concentration of dissolved MeHg in the water. In this case it has only been used for the mosquitofish prey species which is about a trophic level 3 species. A BAF can be calculated for each trophic level up to the top predator.

I had not realized that you collected both soil and floating periphyton. Are all my samples floating ones or are they a mixture? If the latter, please tell me which is which. If the former, do you or Jones still have any soil periphyton archived?

Response: All periphyton sent to you was floating. We may have a limited number of soil periphyton samples archived.

I note that sometimes the tables (especially the larger ones) are at the ends of chapters, with the figures, but sometimes they (especially the smaller ones) are in the text. I found this confusing.

Response: Report has been revised with all tables embedded in the text. Because the figures are so numerous, they have been provided at the end of each chapter.

Page 4-8 -- I think the high conductivity of the EAA discharge is probably due to the leaching of highly weatherable materials from the rapidly oxidizing soils, plus agricultural amendments.

Response: So noted.

Chapter 5: you should define what you mean by wet prairie and what a Muhlenbergia is. Do you have some table where you list the dominant and secondary communities at each site? I would like this.

Response: Wet prairies include a group of low stature, graminoid marshes. They are found over peat and marl, and each soil type supports distinct communities. Wet prairies over peat occur in the central, wetter areas of the Everglades. Those over marl are found in the southern Everglades on higher, drier sites (Craighead, 1971; Davis, 1943 and Gunderson, 1994). Muhlenbergia occurs in the latter. See Tables 5.1 and 5 2 for listings.

You need to better explain figure 7.2 -- I spent a lot of time wondering what you meant by "% canal length" -- I kept figuring you were trying to actually show how the concentrations changed from N to S.

Response: % canal length refers to the population estimates where each sample represents a specific portion of the population. This is the strength of the EMAP design-you can estimate population attributes from the samples. In this example, the population is defined as the total length of the canals sampled. Explanation provided in revised report.

I think Fig 7.4 (and others like them) were especially effective, even more so that the box plots.

I found your ANOVA tables a bit confusing (eg Tables 8.2, 8.3). When you star the averages that are statistically significantly different -- it is not so clear what they are different from -- both of the unstarred values one of them (which one?). Sure, I could mostly figure them out after some thought, but I don't think this was a very clear way to present the data.

Response: Table revised and differences made explicit.

If you look at the spatial plot of % calcite I sent you, you will note that the area where periphyton "% calcite is especially low corresponds pretty well with your WCA3-C area, and also matches up with the large E-W band of marsh where I showed that the dl3C of *Gambusia* was inconsistent with bulk periphyton (or something of comparable d13C) being a major food source (I think I sent you this plot too). Perhaps the non-calcite-forming periphyton is not yummy? Or is the lack of calcite a response to some environmental condition that favors the production of different food sources? Interesting questions!

Response: It looks like there might be something there which could relate to the type of periphyton. In order to interpret your analyses and understand how they relate to the community composition requires some level of taxonomic identification of the samples.

I think you should revise and replace Figures 10.3 to 10.5. These are your final summary figures and they should be dazzlers -- you want the patterns to leap from them! These figures are hard to decipher because they are rather busy and the lines are not sufficiently distinct.

Response: Agreed. Figures revised.

I found section 10.2 in the synthesis difficult to read, partly because of all the descriptions of patterns. This would read much better if you subdivided it into mini-sections, each with a different heading and purpose.

Response: Section was revised for greater readability.

Page 10-5: you state that the MeHg in the canals comes from the marshes -- I missed how you determined this important point.

Response: The MeHg in water is consistently higher in the marsh than in the canals, indicating methylation is higher in the marsh system than in the canal system. This does not necessarily mean that the marsh is the source of canal MeHg.

Appendix C: It would be useful if there was a bit more discussion of what Joel's "Pr models" were and how they work. He explained it to me once, and I understood for only a brief moment!

Response: These are logistic regressions indicating the probability that various sizes of juvenile, male and female mosquitofish are likely to have plant matter or food in its stomach based on the relative standard length.

Comments on your "five questions" on the cover letter:

1) Yes.

2) I wish you had done more to try to relate changes in water chemistry to habitat, soil characteristics, or hydrologic data – or the habitats to the other parameters. I think that with such a huge dataset there might have been more gleaned from the spatial patterns with fancy statistics. Your division of the marshes into 7 bands is a good start, but there is a lot high-powered GIS type might have more sophisticated tools available for comparing these strong spatial patterns. Do you have such a person on your team or could you contract with one? I am facing the same problem with my isotope data – how do I express, in some statistically meaningful fashion, the fine-scale spatial similarities in patterns?

3) Yes. But as mentioned in #2 above, I think you might have done more to compare spatial patterns among plant, environment, and chemistry – which could have lead to more testable hypotheses. I feel that correlations that are only apparent after lots of fancy statistics are not very satisfying (which is perhaps why you didn't do this), but they are useful for developing testable hypotheses.

4) My copy of the report was missing Chapter 11 – so I couldn't comment on the conclusions. Was this an oversight?

Response: No. This chapter and chapter 12 were not released for peer review since they were still under internal review.

5) My copy was missing was missing Chapter 12 – so I couldn't comment on the recommendations. Was this an oversight?

Response: No. See response above.

I hope theses comments are useful and I look forward to working on papers with you in the near future – and in continuing our isotope collaborations in REMAP-II. When this gets funded, we can discuss the suggestions I listed above to see what you can accommodate and how to make things as easy for the field team as possible.

--Carol

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345 Middlefield Road, MS 434		
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SEMINOLE TRIBE OF FLORIDA

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June 15, 1998

Jerry Stober, PhD
Project Leader, Everglades Ecosystem Assessment
Science and Ecosystem Support Division
U.S. Environmental Protection Agency, R4
980 College Station Road
Athens, Georgia 30605-2720

RE: South Florida Ecosystem Assessment:Peer Review Draft

Dear Dr Stober:

Thank you for the opportunity to review the above referenced documents Be advised that though sent to Chairman Billie's office in a timely fashion, these documents were not received by our Department until June 5. While we appreciate the opportunity to review the materials provided and to comment on the approach used in the study, time constraints and limited resources preclude us from providing formal comments on the submitted materials at this time

While we wish to reserve providing you with any substantive comments until a later date, some preliminary observations/questions are as follows:

1. The appendices lacked the referenced materials/raw data addressed by the document.

   Response: We hope the important references have been added and the raw data will be released on the Internet with this report serving as the metadata.

2. What Quality Assurance/Quality Control protocols were obeyed in the collection of the data?

   Response: The QA/QC protocols used in the field and laboratory have been included as an Appendix to the report.

3. What are the qualifications of the principal scientists peer reviewing the papers submitted in this report?

Response: A list of the peer reviewers is presented in the response to comments.

Should you have any questions at all regarding this matter please do not hesitate to call me.

Sincerely,

Craig D. Tepper
Director

Author: trexlerj@fiu.edu at IN
Date: 6/18/1998 5:00 PM
Pricrity: Normal
BCC: jerry stober at REGION
TO: stober.jerry at IN
Subject: mercury report

Hi Jerry,

Sorry this is a couple of days late, I have been going crazy with some summer syposium talks I have to give. I examined the vol II issue because it is the section related to my kind of work (I don't know much about mercury chemistry) . I have two suggestions.

1) page C-3. This isn't my section, but it is not correct to claim length weight curves indicate growth rate. The slope of those curves is simply condition factor. Without some data on age at a given size, you can not address growth rate. This is mentioned twice on this page Sorry, but I don't buy it.

Response: Text corrected accordingly.

2) There are two typos on my section, page C-S. Both appear in grey, and should be easy to find. They are both where the computer couldn't decipher my chi-square symbols. On line 12 from the top, it should read chi-square sub 2 = 7.47, and on line 21 it should be chi-squre sub 3 = 12.01. Maybe you could use a capital X instead of the greek chi symbol if your wordprocessor can't make greek letters.

Response: Done.

Hope this is of some help. The rest looks great. I will be in Vancouver until next Wednesday, in case you need to reach me.

Joel

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
REGION 4
ATLANTA FEDERAL CENTER
61 FORSYTH STREET
ATLANTA, GEORGIA 30303-8960

SUBJECT: South Florida Ecosystem Assessment Volume 1, Technical Report, Peer Review Draft

FROM: Linda Anderson-Carnahan, Chief
Air Planning Branch

TO: Jerry Stober, Project Leader
Everglades Ecosystem Assessment

Attached please find comments on the subject report. If you have any questions on the accompanying comments, please call me at (404) 562-9074 or Ms. Stacy Gent-Howard at (404) 562-9063. Thank you for the opportunity to review this research report.

Attachment (1)
1. Air Planning Branch Comments

Comments/Recommendations on the South Florida Ecosystem Assessment
Volume 1 Technical Report
Peer Review Draft

1. Under the "APTMD" section, on the list of participants in the Everglades Assessment Project, add the name Stacy Gent-Howard

Response: Done.

2. Move the listing of acronyms on page iii to a location after the Table of Contents and identify it within the Table of Contents. Also, all acronyms used within the report should be listed on the acronym page; i.e. some acronyms such as "ENP-N, AA-N, and WCA-3S" are not included.

Response: Done.

3. Add a figure to the Executive Summary depicting the changes in mercury (Hg) in mosquito fish with total phosphorus (TP) (ref: para 6, Executive Summary) and a map of the study area indicating where each sample point is located, if possible.

Response: The Executive Summary is intended to be one or two pages. While the mosquitofish mercury-total phosphorus relationship is one part of the story, there are other factors that also would have to be added as figures, increasing the length of the Summary. This figure, therefore, was not added.

4. Place all graphs and figures within the text of the report near where they are referenced. This provides continuity for the reader.

Response: Done.

5. Move the "Purpose" section, section 1.4, of Chapter 1 to the beginning of the Introduction, ahead of the Background section. This section sets the tone for the reader and will assist them in understanding the need for the research.

Response: Done.

6. Given the current national attention on mercury, add a table or other figure to Section 1.3, that provides the reader with some perspective on how high/low the Hg levels in fish are in the study area as compared to other regions where mercury is a concern.

Response: The text provides the reader with the concentrations for Hg in bowfin and states that these are the highest recorded nationwide. The EPA Mercury Report to Congress was added to refer the reader to Hg levels in other regions.

7. Provide a frame of reference in Section 1.3.2, at the end of paragraph 1 where the discussion on human health effects of mercury is located to provide the reader with a basis for determining if the mercury levels noted are a concern, and to what extent.

Response: Reference to EPA Mercury Report to Congress was added to provide the reader with a citation for further information relative to human health issues.

8. In Section 1.3.3, the second paragraph mentions the natural water phosphorus concentration of 10 ppb. The significance of this number should be discussed in the text; i.e. what events are triggered/retarded at levels above this concentration?

Response: A reference for this phosphorus concentration has been added.

9. Section 1.3.5 mentions that non-indigenous plant and animal species within the Everglades need to be controlled, but provides no justification for this statement.

Response: Some justification has been added. The intent was to introduce issues, not provide a detailed discussion.

10. Section 1.3.6 states that previously mentioned issues are interdependent, but no discussion of this interdependence and its ramifications are provided. Further discussion of this topic should be provided.

Response: Again, the intent was to introduce the issues, not provide detailed discussion. This particular topic is discussed frequently throughout the report. Some additional discussion has been added to Section 1.3.6.

11. Section 1.4, paragraph 1 mentions the South Florida Interagency Ecosystem Restoration Task Force for the first time. A background and history discussion for this group and its relevance to this study should be provided within the Background section, Section 1.1.

Response: This information is provided in Section 1.2.

12. The last paragraph of Section 1.4 states that this study focused on a subset of hypotheses," but does not identify these hypotheses, nor are they identified elsewhere in the report.

Response: Done.

13. The purpose of this report, as defined in Section 1.4.1 is to provide preliminary answers to the four policy relevant assessment questions of 1) magnitude, 2) extent, 3) trend, and 4) cause. The text of the report currently does not explain how the hypotheses the study focused on assisted in answering each policy relevant assessment question. Additionally, an explanation of how the chapters following the purpose statement relate to and answer these questions needs to be discussed.

Response: Chapter 11 on Management Implications discussed these questions and the answers obtained by the study as well as the their use in making appropriate management decisions now and in the future. However, this chapter was not available for peer review since it was still under internal review.

14. Chapter 8, the Mercury Chapter, does not fit into the report; i.e. no relationship between mercury, the preceding information, the hypotheses tested, and the policy relevant questions is established in the prior chapters. A relationship needs to be established prior to presenting the mercury related information.

Response: The interface between mercury and these other issues was initiated in Section 1.3.6, and in the initial conceptual model discussion. The report now references these earlier sections so the relationships are more evident.

15. The report provides a great deal of data and information, but does not establish any relationship between the data and the conclusions drawn, leaving the reader to ponder the relevance of what is presented.

Response: The Synthesis and Integration Chapter brings the interrelationships of the data together and formulates a number of hypotheses to be tested based on the associations indicated by the data. Management implications of the study are stated in bullet form in Chapter 11 which was omitted in the peer review draft.

16. Figure 8., $TS0_4$, is an excellent visual aid for providing an understanding of where reported concentrations of compounds are found. More figures like this should be used in the text to aid the reader in visualizing the study area and drawing conclusions from the data.

Response: Various figures have been modified to improve visual clarity.

17. Sections 8.3.2.1 and 8.3.2.2 discuss canal water quality and mention the compounds TOC and TSO_4, but are located in the mercury section. No relationship between these compounds and mercury has been established. The correlation (if any) between these two compounds and the behavior or presence of mercury should be established prior to this discussion.

Response: The statistical tests which identified the associations were mentioned in the first paragraph, however, additional discussion will be added to indicate why these parameters have important interactions with mercury

18. Place Section 8.3 at the beginning of Chapter 8 to provide some perspective and lead-in to the rest of the chapter.

Response: Section 8.3 is the Results Section. It is in the appropriate place.

19. Section 8.3 cites complex relationships among the water depth, TOC, sulfate, and total phosphorous concentrations, but does not justify this claim or elaborate on what these relationships might be. Similarly, this Section states that canals play a major role in the transport of total phosphorous, but does not explain what this role is or its relevance to the policy relevant assessment questions.

Response: See question 17 above.

20. In Section 10.1, the third sentence begins "Other sections discuss the conceptual model for Hg..." It is suggested that this sentence be reworded to refer to "Previous sections..." or 'Chapter 8..." and follow with "...new models are offered later in this Chapter" to more accurately identify the context of the conceptual models in this report.

Response: Sentence reworded as suggested.

21. Data presented on page 10-3, Section 10.2, paragraphs 3, 4, and 5 would be better presented in a table with surrounding text providing perspective on the data and discussing its relevance and meaning.

Response: The data are in tables, Tables 10.2 and 10.3. The text has been modified to make it more readable.

22. Page 10-5, Section 10.2, paragraph 7 indicates there are important management implications of a threshold TP concentration, but does not elaborate upon this claim, or what the implications might be.

Response. We are stating what is indicated by the data in the TP gradient and the apparent associated plant responses as one proceeds downstream from north to south. A threshold TP concentration must exist and it will be stated more clearly as an hypothesis.

23. Page 10-5, Section 10.2, paragraph 9 remarks upon the small maximum sizes of mosquito fish in the Everglades and refers to sizes of mosquito fish found in other parts of the mosquito fish's "natural range." Provide additional information on what the "natural range" is and to what magnitude Everglades mosquito fish are deficient.

Response: The two authors cited have a paper in preparation.

24. Section 10.4 refers to "testable hypotheses" and "conceptual models"; Some distinction should be made as to whether these are the hypotheses/models developed prior to this study or subsequent to this study and upon which future work should be based.

Response: Distinctions have been made.

25. Chapters 11 and 12 are missing from the document provided for review. The "Summary and Conclusions" and "Recommendations and Future Directions" Chapters are very important and should be circulated for peer review as well.

Response: These chapters will be circulated for review at a later date.

United States Department of the Interior
FISH AND WILDLIFE SERVICE

Arthur R. Marshall
Loxahatchee National Wildlife Refuge
10216 Lee Road
Boynton Beach, Florida 334374796

June 12, 1998

Dr. Jerry Stober
Project Leader
Everglades Ecosystem Assessment
U.S. Environmental Protection Agency
980 College Station Road
Athens, GA 30605-2720

Dear Dr. Stober:

I have received the "South Florida Ecosystem Assessment" peer review draft that you sent to me. Unfortunately, I do not have sufficient time to review it as you requested. I have "skimmed" it and have found one item that I found confusing. In Volume II (Appendices), Table 1 shows the deadlines for permits. However, I know at least one deadline has not been met (NPDES for the ENR). The table makes it appear as if the deadline has been met, since it doesn't say otherwise.

Response: Missed permit deadlines are a regulatory issue beyond the scope of peer-review of this scientific report.

I have another issue that was brought to my attention independently recently and that is of concern to me. I have heard that there are various methods of analyzing mercury samples, just as there are for phosphorus. We have found that the results between labs can vary for phosphorus for split samples, as demonstrated by the Everglades Technical Advisory Committee's Round Robin sampling events. I have heard that the mercury results can vary significantly. Could this be a factor in your results for this document? Are you investigating this? I would like to have your input, because, aside from the effect on results such as in this report, I must deal with mercury due to its relationship with phosphorus in the Everglades as part of my responsibilities here at the refuge.

Response. The interagency Mercury Program Management Committee has initiated laboratory intercomparisons and a round-robin exercise in order to determine the comparability of data across the various mercury laboratories and Florida mercury research efforts. In addition, numerous sample splits have occurred throughout this study across the three analytical laboratories. The potential mercury-phosphorus relationship issue is included within Chapter 8.

Response: Because of different analytical methods for various mercury species, we had both primary and secondary laboratories designated for all analyses with the secondary laboratory providing QA/QC analyses. The mercury methods used in the study have all been published in the peer-reviewed scientific literature. In addition, the project participated in several mercury round robin analyses.

Thank you for soliciting my review. I hope you can shed some light on the Hg analysis problem.

Sincerely,
Susan D. Jewell
Senior Biologist

Environmental Services & Permitting, Inc.

June 15, 1998
File: 295-94-01

Jerry Stober, Ph.D.
Project Leader
Everglades Ecosystem Assessment
U.S. Environmental Protection Agency, Region 4
Science and Ecosystem Support Division
980 College Station Road
Athens, GA 30605-2720

Re: EPA PEER REVIEW DRAFT REPORT "EPA-904-R-98-XXX" SOUTH FLORIDA ECOSYSTEM ASSESSMENT

Dear Dr. Stober,

We appreciate receiving the draft report for review. The report contains a lot of data. We would like to have the raw data in electronic format so that we could review it and conduct our own analyses. This would enable us to make more meaningful comments. We also need more time to review the data and the report. We only received the report May 26 and have not been able to drop all our other projects to review this one. Given the importance of this issue we feel more time is needed and the raw data need to be provided.

We are also concerned that EPA may have the proverbial cart before the horse. Much of the data in the report was apparently collected by FIU. The Florida Department of Environmental Protection (FDEP) Quality Assurance section has recently audited the FIU mercury laboratory and found several major deficiencies that caused them to express some serious concerns about the quality and reliability of the mercury data from the FIU laboratory. We understand EPA is aware of these concerns and has been looking into them. We feel these issues need to be addressed prior to using the data in a report. We do not want to spend a lot of time working with the data or reviewing the report prior to these issues being definitively resolved. It would be inappropriate to use questionable data in statistical analyses and then draw conclusions about the results. Once the EPA QA\QC reviews are completed and made public, the report needs to be revised to include the QA\QC assessment and any questionable data need to be removed from the data sets and the analyses rerun. If none of the data are found to be questionable, then we need additional time to review the raw data and conduct our own analyses.

Response: The data have undergone extensive review and the QA/QC report has been included as an appendix. In addition, the database with all qualifiers will be available on a world wide web site.

We are also concerned about the synoptic nature of the data. The data clearly need to be collected over a much broader range of hydrologic and seasonal conditions.

Response: We agree and propose the project be continued so a greater range of hydrologic conditions can be monitored.

The report needs to be updated to use the most recent nutrient data and not rely on outdated and draft reports. The impacts of the Everglades Construction Project on future nutrient loads and their impact, if any, on the mercury issues needs to be addressed.

Response: The nutrient data reported in this study were collected from 1993 through 1996. The purpose of this report is to summarize this data set only. The text has been modified to include other published data on TP concentrations and results of ENP and BMPs, however, a detailed discussion of the ECP is not warranted in this report.

Please keep us on the list to receive all drafts and data distributions relative to Everglades issues. If you would like to discuss any of our comments, please call.

Author: federico@mfl-inc.com at IN
Date: 6/15/1998 4:47 PM
Priority: Normal
BCC: jerry stober at REGION4
TO: stober.jerry at IN
Subject: Pete Rosendahls' comments on Ecosystem Assessment Report

Pete Rosendahl requested that I e-mail you his comments on the Ecosystem Assessment Report. He will be sending a hard-copy in the mail.

Author: tolsen@mail.cor.epa.gov at IN
Date: 6/15/1998 11:08 AM
Priority: Normal
BCC: jerry stober at REGION4
TO: stober.jerry at IN
Subject: South Florida Assessment report

Jerry,

I looked through the two volume South Florida Ecosystem Assessment report. It appears to be a comprehensive and well-written technical report. The sections describing the statistical design and analysis are well written and communicate the essential information. The statistical analyses use exploratory data summaries, design-based estimates for populations and model-based (Kriging) estimates. Each has role to play in understanding the data and the techniques appear to be used correctly. In several instances additional information could have been given concerning the uncertainty of some of the estimates. It may be sufficient to present such information for several typical situations and then state that similar levels of uncertainty accompany other similar summaries. For example, a 4-cycle and 1-cycle variance contour map from kriging estimates may be used to illustrate the uncertainty in other maps (assuming that such is the case!)

Overall the project provides extensive data on a critical problem

Tony

Anthony R. Olsen
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Western Ecology Division
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UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
NATIONAL HEALTH AND ENVIRONMENTAL EFFECTS
RESEARCH LABORATORY
MID-CONTINENT ECOLOGY DIVISION
6201 CONGOON BOULEVARD. DULUTH, MINNESOTA 55804-2595

OFFICE OF
RESEARCH AND DEVELOPMENT

June 3, 1998

MEMORANDUM

SUBJECT: South Florida Ecosystem
Assessment Draft Report

FROM: Anthony R. Carlson
Research Aquatic Biologist

TO: Jerry Stober
Project Leader
Everglades Ecosystem Assessment

This report clearly provides answers to policy relevant questions related to the magnitudes, extent, trends, and causes of current ecological conditions in the Everglades. The statistical survey design sampling data and its synthesis provides the baseline and conceptual models for use in assessing future trends in ecological condition related to mercury contamination, eutrophication, habitat alteration and hydropattern modification issues. I agree with the report conclusions and recommendations for future studies. I believe this technical report is an excellent example of the EMAP approach and recommend that it be published.

Dr. Jerry Stober
U.S. Environmental Protection Agency, Region 4
Science and Ecosystem Support Division
Athens, Georgia

Response:

In response to your written request of May 11, I regret that I cannot provide a review of the South Florida Ecosystem Assessment, Volumes 1 & 2.

Last fall, our Center Director (USGS Upper Mississippi Science Center) transferred to a position in our Reston National Office, and I spent months serving as Acting Center Director. As a result of the associated heavy administrative and managerial workload, I have fallen behind schedule in several areas and for the foreseeable future, am declining all requests for reviews. On the positive side, I am very pleased to report that our new center director, Dr. Leslie Holland-Bartels, will arrive here in June.

My sincere regrets, Jerry, to you and your coauthors, but it's evident that good intentions don't get reviews done in such busy times. Please let me know if you would like me to return the subject documents to you.

Regards.

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Author: jgentile@rsmas.miami.edu at IN
Date: 06/03/98 10:26 AM
Priority: Normal
ECO: jerry stober at REGION4
TO: stober jerry at IN
Subject: Review: South Florida Ecosystem Assessment

Jerry,

Thanks for the opportunity to review the SF Ecosystem Assessment. Unfortunately I will be unable to review it within the time frame you needed. So I can't say I concur with the conclusions and didn't want you to assume that I did if I didn't respond. I'm sure I will get to it but not at the moment.

Regards,

Jack

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6 May 1998

United States Environmental Protection Agency
Region 4
Science and Ecosystem Support Division
980 College Station Road
Athens, Georgia 30605-2720

Attention: Dr. Jerry Stober, PhD, Project Leader
Everglades Ecosystem Assessment
stoberjerry@epamail.epa. .gov

Dear Jerry,

Your report did not arrive until 22 May and it was delivered in rather poor condition. It must have got a rough ride from Athens to Ottawa. Nevertheless, it provided excellent reading during my trip to and from Athens. I am very impressed with the care and quality of the work. It was a monumental task and will be a classic study that will be scrutinized by scientists for years to come. This means that we need to make it as tight as we can.

A hard copy will also be sent by courier.

I think that the problem that is discussed is not restricted to the Everglades. Here in Ontario, bogs and wetlands were drained from 1850 to the present and farmers have been diverting water into them and adding nutrients as well. Consequently, I think that the mobilization of mercury is widespread and your study will have rather broad application.

Please send me a copy of the Stober et al. 1996 report so that I can see what was discussed earlier (in case I get asked questions about it).

Attached are my reviews and comments on the Technical Report.

Answers to your questions are given below:

1. Is the purpose of the report clear and does the report achieve this purpose? Yes.

2. Are the report sections appropriate? Yes but there is some redundancy.

Are there additional hypotheses that might be appropriate to address with this monitoring design? Yes, see below. I think that the color of the water has inhibited the photoreduction processes which would remove the mercury which comes in the precipitation.

3. Are the conceptual models reasonable? Yes but photoreduction must be quantified. Are there additional hypotheses that might be appropriate to address with this monitoring design? Yes. The role of DOC and other ligands in changing the BAFs in the upper reaches.

4. Are the report conclusions supported by the information collected both from this program and from other on-going or completed projects? Are there other sources of information that support or refute the conclusions? I think that the proportion of MeHg to Thg is very high. Other literature values should be given.

5. Are the recommendations for future studies reasonable? Yes

Are there additional studies that should be considered and why? Yes, but include the part about the interaction with DOC both as it influences BAFs, photoreduction of Hg^{2-} and photodecomposition of MeHg.

Sincerely,

David Lean
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Review of Technical Report by Stober et al. "Monitoring for Adaptive Management: Implications for Ecosystem Restoration"

One of the strong aspects of the work is the spatial coverage and the selection of the stations. Too often in the past people went to easily accessible locations that would bias the analyses (because they were already impacted). The strength of the present study is the extent of the survey. This should be emphasized more.

Response: The spatial extent and the ability to make population estimates has been emphasized more in the revised report.

Executive Summary: covers the topic but ends without any forceful conclusion. As discussed below there are some conclusions that are unequivocal. Phosphorus and DOC inputs to the Everglades have done what? The principal component analyses was not sufficiently developed. I know what was found but some additional help to the reader in the interpretation would add to the weight of the discovery.

Response: Conclusions have been added to the Executive Summary.

Chapter 2. Study Design This report provides a lot of information and to get to the heart of the subject effectively, I would move supporting information to appendices. For example, I would suggest taking the essential information from pages 2-5 to 2.10 and moving it to an appendix. We don't want the reader to get bogged down with too much technical stuff At the same time, this is vital information that should be available.

Response: While the information is technical, this provides the support and documentation for one of the report's strengths (see comment above) - the spatial extent and population estimates. The information will be retained in the chapter.

Table 2.1 is especially useful. The only part that I question is the statement that low pH promotes methylation. Perhaps consider rewording this to say methyl mercury is often found in low pH waters. There must be a sufficiently low pH were methylating bacteria are no longer active.

Response: The sentence will be modified. However, there can be abiotic methylation occurring at low pH (e.g., pH < 5.0).

Chapter 3. Materials and Methods I thought this was especially well done. There was enough information for the reader to appreciate what was done but a reference was provided for the detailed procedure used (usually in first rate journals)

Chapter 4. General Characteristics of the Water Regime I find the oxygen levels in the canals and the marshes especially alarming. Some reference to the very poor light penetration would be useful. This would also support the observation that the chlorophyll levels seem to be lower than that expected from the total phosphorus concentration. It appears to be a light limited system and some comments on euphotic zone depth of light penetration to total depth would be appropriate. It is my opinion that the dark color of the water is a principal factor in mercury contamination of the ecosystem (see below). Since turbidity values are provided and light penetration is related to turbidity a correlation at selected sights between extinction coefficient for photosynthetically active radiation or the 1% light level and turbidity could be generated and used to provide the depth of euphotic zone. In the marshes, the DO values are higher. Somewhere, the percent saturation at 26-35 degrees should be provided. In ENP for example, the oxygen values must be near saturation. The implication of the second paragraph on page 4-9 is significant. Diel patterns of oxygen can influence microbial activity. For example, at night are levels sufficiently low to cause a diel pattern in methyl mercury production?

Response: We did not measure Secchi depth or light penetration. A comment has been added that, based on general observation, the canals are dark water systems. The percent saturation of DO at 26 to 35° C was also added to the text.

In the second paragraph of the Synthesis section (page 4-10) it reads "specific conductivity patterns provide an indication of. . ." add and DOC after conductivity. As will be discussed below the impact of EAA in adding DOC to the system is contributing to the mercury problem and the patterns of DOC can be traced throughout the everglades system.

Response: The sentence has been modified.

Prior to recent human influences on the Everglades ecosystem, water was supplied mostly from rainwater (and the buffering capacity and conductivity would be low). During these conditions the DOC levels were also likely quite low. With the changes in hydrology and water diversion, water with much higher calcium and buffering capacity was injected into the system. This may be significant.

In the synthesis section some discussion of the % saturation of oxygen at these high temperatures would be useful. The standard of 5 mg/L is interesting. What is the % saturation at 35 degrees? Most people work with water in the 20's not the 30's.

Response: % DO saturation has been added to the text.

The statement "that biogeochemical reaction rates might be expected to be relatively rapid in these systems" is not clear.

Response: The sentence has been modified to state "that because reaction rates are temperature dependent, biogeochemical reaction rates in the warm tropical Florida systems are expected to be more rapid than in temperate systems."

Chapter 5. Habitat. I have no comments.

Chapter 6. Soils

Page 6-2 (top of page) Other factors which contribute to subsidence of soils include the influence of adding water with substantial calcium in it. This well buffered water can alter the pH of the system (see above). For example, a bog has a very low pH but if well hard water is added or the water level is reduced the pH can be altered and microbial activity (even in the absence of added nutrients) can be turned on with the net result of a substantial loss in soil substrate. DO WE HAVE ANY RELIABLE HISTORICAL INFORMATION ON THE pH OF EVERGLADES WETLANDS? I think that this is vital information. Could it be that the pH in the peat regions was in fact much lower during the 1930-1950 period? This may also be a factor in the loss of sediment from the marshes discussed at the end of this chapter.

Response: The investigators are unaware of any such information.

On page 6-4 near the top it states that "maximum soil loss calculate (added a d for past tense) over the last 50 years indicate (add s) that the portion of WCA3 north of Alligator Alley has lost over 0.9 m (3 feet) of peat. I find this astounding and this must be a factor in the overall degradation of the Everglades.

Response: Agreed.

On page 6-7 pH is discussed but with the exception of the 5.8 value in WCA1 all are near or above 7. Clearly, some historical values would shed light on the discussion above.

Response: Agreed, but the investigators are unaware of any such historical information.

The heading for Figure 6.6 is not clear. Is this a plot of Log Bulk Density or Bulk Density plotted on a log scale. I think it is the former so the heading should be log bulk density.

Response: This is Log (Bulk Density). The heading has been clarified.

Chapter 7. Nutrients

Knowing that (line 3,4) that the marsh hydrology was precipitation driven is a key part of the story. During this time the pH was likely less well buffered and was likely lower than it is today. Certainly, the water was clearer and this has a profound influence on the mercury problem. This will be discussed more fully below

I would suggest replacing the word "drastically" (page 7-3 near the middle) with something like "rapidly".

Response: Change made.

On page 7-5 near the middle of the page, there is a statement that the TP decreases exponentially". Be careful that someone does not fit the data and find that it is not an exponential. It does decrease rapidly.

Response: The decrease is exponential, but the sentence will be modified to state that there is a rapid decline in TP, approaching an exponential decline.

On page 7-6 the TP "hot spots" are discussed. These are especially prevalent during the dry season. Could it be that the soils become sufficiently to cause internal loading?

Response: We do not have the data to determine this, but it is an interesting hypothesis that could be tested.

The total nitrogen (TN) concentrations in Table 7.4 are no doubt correct but the ratio of TN:TP to infer phosphorus deficiency should be viewed with caution (Page 8-5). The ratio of DON:DOC is about 0.1. This means that much of the DON in some areas would be dissolved organic nitrogen which is quite refractory and is not utilized by plants or microorganisms to any great extent.

Response: So noted.

Chapter 8. Mercury

At the bottom of page 8-1 perhaps there should be some discussion of the influence of drainage of peat soils. Historically, the rain fell on flooded soils and would run off without moving through the soils. Later, when soils were drained, rain would run through the soils and carry easily dissolved forms.

Response: This idea will be added to the text.

On page 8-3, it states "Hg being washed out by rainfall". It is the Hg^{2+} form that is soluble in rainwater. The statement about gaseous Hg coming into contact with the water requires that it is being oxidized after it equilibrates with the water phase. We have some evidence that this might occur in some areas but is there evidence that it occurs here?

Response: See Krabbenhoft et al. 1998.

I divided the reported deposition rates of total mercury of 40-50 kg/yr, measured by Landing et al., by the area (7600 sq km) and got a deposition rate of 6 $\mu g\ m^{-2}\ y^{-1}$ or by 5000 sq km and got a rate 10 $\mu g\ m^{-2}\ y^{-1}$. These values are consistent with other reliable values and some reference to other deposition rates should be made. It indicates there is nothing unusual about the mercury deposition rates to the Everglades. In fact, it is just like in our remote lakes in Ontario.

Please note the following. We have conducted experiments in a wide range of waters from Florida to the high Arctic and found that photoreduction of mercury (usually Hg^{2+}) results in the formation of Hg^0 or elemental mercury which due to its high Henry's Law Constant and low water solubility will volatilize from the water back into the air (one paper by Amyot et al enclosed). This rate has been estimated to be at least as high as the atmospheric deposition rate in clear waters. Here the water is dark there is little or no photoreduction. It may be that the color of the water in the WCAs inhibits photoreduction and the transport of Hg° back into the atmosphere. There are a number of references that color and mercury loading are correlated. In addition, the most significant predictor of fish mercury contamination in Ontario lakes is color. The bottom line is that in colored waters, more comes in and less gets out. This almost certainly is one of the critical factors which has caused the mercury contamination to occur in the Everglades.

Response: The variation of color in Everglades water is large. For the most part the canals have more highly colored water than the marsh which is mostly clear. Although there is no doubt that this flux is occurring it is difficult to determine how much color may be affecting the mercury flux across the marsh.

Page 8-8 at the top. The "exploratory analyses' should be better defined.

Response: A discussion of the statistical analyses conducted to determine the critical variables will be expanded in the text.

Pages 8-12 and further. There is some redundance from previous sections e.g., Fig. 7.3.

Response: Redundancy will be minimized. However, because of the report length, it is anticipated some readers will only read selected sections. Some redundancy is retained to summarize results from previous sections.

On page 8-13 the percent mercury as MeHg is give as 45%. This is extremely high. Without looking it up, I think that values are generally near 1% so what we have here is remarkable. Note that they axis of Fig. 8.20 is given as the Ratio. This should be percent.

Response: The percentage of MeHg is probably the highest ever report for any system. Fig. 8.20 will be corrected.

Synthesis on page 8-21. While mercury loading was dominated by atmospheric deposition the rate expressed in $\mu g/m^2/yr$ was not unusual. Furthermore, we have seen photochemical reduction rates at levels which would send this amount back into the atmosphere again (see above). This likely cannot occur in the Everglades due to the dark color.

Response: It most likely does occur since most of the marsh water is clear, and relatively shallow.

The patterns with latitude are especially interesting. It seems that there is partitioning of methyl mercury between the DOC perhaps or other dissolved ligands such as some of the sulfide or thiol compounds that causes a reduction in the bioaccumulation factor. As such, it is there but not completely available to the organisms. Perhaps there is also a biodilution and this illustrates the importance of the diet studies that were so well done. Alternatively, the MeHg partitioned to the DOC becomes available later on. These questions illustrate the need for more studies on the interrelationships between the forms of mercury and DOC. It is also known that MeHg can also be photodegraded. This almost certainly is inhibited in brown waters.

Response: These thoughts will be used to expand on the discussion in the text. Other than the canals and parts of the marsh with excessive phosphorus loading the water is extremely clear.

Volume II. Appendices. I have no comments

Comment of proposed solution
From the comments made above, I think that there should be studies to find ways to further reduce phosphorus leaving EAA. In addition, after reading this report there must be controls on DOC export from EAA. If the waters were clean, atmospheric deposition of mercury could easily be balanced by photoreduction (especially under the strong Florida sun). We would be pleased to make some of these measurements. It would seem that if the high DOC could be eliminated by some trick (which may even reduce phosphorus) the problems would not be as severe.

Response: We agree with your comments regarding phosphorus and DOC, however, photoreduction doesn't seem to have the effect observed in other systems. Without reducing the atmospheric wet deposition of mercury from local sources balancing the mercury flux in the system will remain problematic.

SFWMD

FAX MEMORANDUM

To: Dan Scheidt, US EPA

From: Christopher McVoy, HSM, SFWMD

Subject: Review of So. FL Ecosystem Assessment, Vol. I Technical Report

Date: 9 July, 1998

CC: Jerry Stober

Dear Dan:

Thank you for the opportunity to review this presentation of your project. This synoptic, system-wide study is a fine piece of work. The soil change information is striking and clearly very important. Nicely backed up with the organic matter and bulk density data.

I've made a few suggestions, most minor. Overall looks good. I would strongly encourage you to publish the graphs and maybe even the raw data in a scientific journal.

Christopher McVoy, Ph.D.

P. 6-1 "High rockland . ." Rockland was present only from about half way between New River and Miami River on south; to the north was all sands . . .

Response: Agreed.

- Stephens (1956): should probably read "once contained the largest body," rather than "contains"

Response: Agreed.

- "depth up to 6 m probably mean elevation of 20-21 feet; the pre-drainage peat thickness was about 3-4 m max.

Response: Agreed.

P. 6-2 Stephens (1984): Stephens would very likely suggest emphasizing process (6) oxidation, and (5) burning. These are irreversible, and are the dominant processes.

Response: Agreed.

- Add following refs: Shih, S.F., J.W. Mishoe, J.W. Jones, and D.L. Myers. D1979. Subsidence related to land use in Everglades agricultural areas. Trans. Amer. Soc. Agric. Eng. 22(3):560-563.

  Shih, S.F., E.H. Stewart, L.H. Alleo, Jr., and J.W. Hilliard. 1979. Variability of depth to bedrock in Everglades organic soil. Soil Crop Sci Soc ???. 38:66-71.

- "public Everglades" suggest using different name to emphasize not only ownership, but the land use difference form EAA, i.e., even the so-called wildlife habitat areas have lost soil under govt/public stewardship. Strongly agree on "poorly documented"!! Your work fills a key gap.

Response: Agreed.

- "unimpeded surf. ." Suggest emphasizing here the combined effect: (1) eliminated northern inflow from Lake O. overflow; (2) lowered water depths due to major canals; and (3) lack of impoundments to keep water from "just running out." Combination is important to emphasize as the pre-drainage system also had "unimpeded flow," but because of sufficient northern input, and no drainage canals, was able to keep peats wet.

Response: Agreed.

P. 6-3 "A geostat rep . . ." (1) make more specific - kriging, nearest neighbor, etc. If kriging was used, might not be bad idea to include a map of the estimated uncertainty/error. (2) suggest one sentence recapping method of measurement here; mention includes both organic and mineral soils.

Response: Agreed.

- Table 6.1 Very nice summary. Suggest 2 signif. digits only on % O.M.; use leading zero on s.d.'s for b.d.

Response: Agreed.

- "diff in peat thick . . ." need to give methodology here - how the difference map was calculated/created.

Response: Agreed.

- "Soil volumes . . ." this is separate topic - suggest moving to end of 6.2.1 section.

Response: Agreed.

- "Davis . . reports . ." If memory serves, Davis did give some methods - might even quote him. Should also emphasize that he had no. of years on foot, boat, in field in So Fl, and wrote whole monograph on peat deposits. In other words, he is a credible and highly qualified source . . .

  Then, as separate issue, take up point of how to deal with his 2 foot intervals.

Response: Agreed.

Pg. 6-4 "hi or lo extreme" replace "extreme" with "threshold"

Response: Agreed.

- "Max soil loss calc" "calculated"

Response: Agreed.

- "Up to 1.5 m (5 ft)" replace "3 to 5 feet" (there is a slight difference).

Response: Agreed.

- Question An alternative approach to the min max business would be to take the mean value and add a range: Ex: 3 to 5 ft class becomes 4 plus or minus 1 ft. This might avoid the lightning rod for criticism set out to be discussing the maximum change in thickness; nobody can argue with a mean.

Response: Noted.

- northeast S. Slough Capitalize (or not) consistently.

Response: Agreed.

- "53% of volume..." Suggest presenting loss percentages first as depth %s, rather than volume %s -- more direct. In later paragraph, come back to volume.

Response: Agreed.

- "system soil volume..." "system" is not clear; make more specific.

Response: Agreed.

- Table 6-2 (1) Soil volumes - max of 2 signific. figures; three not justified by methods. (2) suggest 1 signific. fig. For Min/max change (%) cols. Shouldn't plus signs be neg.? --loss of peat? Didn't understand first row: "-3.0 (4%)" - why different than other rows?

Response: (1) Agreed. (2) Some calculations do show an increase in soil thickness.

- Percent O. M. Need to state sample methods here -- how deep were cores? Maybe how % OM determined (or refer on this to Methods Chapter). Make "geostat. represent." more specific.

Response: Agreed.

P. 6-5 "0.76% to 97.5%" replace: "less than 1% to 98%"

Response: Agreed.

- "Are highly inorganic" replace: "primarily mineral".

Response: Agreed.

- "Mean of 92.0 ±6.8%" Replace: "92 ±7%" (similar on subsequent %s)

Response: Agreed.

- Bulk Density again, more specific on "geostat. display", maybe "kriged map."

Response: Agreed.

- on correlation w/OM suggest restricting sample to the >40% (39%) portion of your sample, which would cover the areas known as the organic soils, then recalculating the regression, might try linear-linear too.

Response: Noted. Linear regression was performed. Plot was much more scattered.

P. 6-6. Soil redox: can't comment knowledgeably on this.

Response: Noted.

- Transects: could map with location of transects be included? perhaps also some comment on the graphs and their relation to what was seen in the field? (Particularly for thickness and O.M.)

Response: Map of transects is included as Figure 2.1.

Figs:

- Fig. 6.1 legend recommend clarifying that the 1946 is thickness of peat soils only; 1996 is soil thickness, whether peat or mineral or mixed...

Response: Agreed.

- Fig. 6.2 (1) may be misleading to separate off WCA3A N of A. Alley -- this was only a partial hydrologic separation - 3A N generally seems to be too dry rather than too wet... (2) What happened to ENP along Tamiami Trail - funny gap?

Response: (1) Agreed. The purpose was to divide the system into geographic areas for data analysis and interpretation. The figure is not intended to infer a hydrologic separation.

(2) Agreed. Gap is inadvertent, map will be corrected.