

# Appendix 2B-1: Annual Permit Compliance Monitoring Report for Mercury in Downstream Receiving Waters of the Everglades Protection Area

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## KEY FINDINGS AND OVERALL ASSESSMENT

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This report summarizes data from compliance monitoring of mercury influx and bioaccumulation in the downstream receiving waters of the Stormwater Treatment Areas (STAs) during the reporting year May 1, 2000 through April 30, 2001. Results from this monitoring program describe significant spatial distributions and, in some instances, between-year differences in mercury concentrations.

Key findings of this report are:

1. As observed previously, rainfall volumes and total mercury (THg) concentration increased in the late summer through early fall and consequently, atmospheric wet-deposition of THg also increased during these months (i.e., third and fourth quarters). When combined, among-site differences in rainfall and THg concentration resulted in significant spatial and temporal differences in THg deposition. In 2000, atmospheric wet deposition was slightly lower at the Everglades Nutrient Removal (ENR) Project and Everglades National Park (ENP) sites, but slightly higher at the Andytown site compared to previous annual averages. Seasonal Kendall analyses of the Mercury Deposition Network data sets revealed statistically significant downward trends in monthly median THg concentration in rain at ENP, monthly rainfall amounts at the ENR Project and monthly deposition of THg at Andytown.
2. Average concentrations (i.e., not volume-weighted) of THg and MeHg increased during the reporting year relative to long-term averages at nine of the 10 monitored Non-ECP water control structures. Percent of THg that was methylmercury (MeHg) was also higher at most structures. Increases in concentrations occurred primarily during the third and fourth quarters (i.e., for pooled sites), which was consistent with seasonal increases in atmospheric wet deposition. Nevertheless, seasonal Kendall analyses found no statistically significant trends in either THg or MeHg concentration at any of the sites. Moreover, there were no violations of the Florida Class III numerical Water Quality Standard (WQS) of 12 ngTHg/L during the reporting year.

3. Mosquitofish collected in September through October 2000 showed substantial decreases in tissue-Hg concentration, relative to 1999 levels, at all downstream sites. The basin-wide average concentration in 2000 (68 ng/g wet) was similar to 1998 levels, but significantly lower than peak levels observed in 1999. Between-year differences ranged from a 32-percent decrease in THg at the P33 site to a 98-percent decrease at the L5F1 site. In 2000, Mosquitofish at only four of the downstream sites had THg concentrations exceeding either the USFWS or USEPA criterion for the protection of piscivorous avian and mammalian wildlife. This is a dramatic reduction from the previous year, when mosquitofish from 100 percent of the sites exceeded both criteria.
4. Sunfishes caught at four of the 12 downstream sites in 2000 showed significant among-year variation in tissue-Hg concentration. Of these four sites, two of the sites (L38F1, CA3F2) showed a decrease, and two sites (Holey Land, CA2U3) showed an increase in THg levels in whole sunfish. Tissue-Hg concentration in sunfishes must be interpreted cautiously due to the confounding factors of species collected (i.e., species of *Lepomis* caught) and fish size (i.e., age surrogate), which might suggest erroneous trends or, worse, obscure real trends in Hg levels. Sunfish from all but one site contained THg concentrations exceeding one or both of the predator-protection criteria in 2000. This finding is significant because sunfishes represent the preferred prey item of many fish-eating species in the Everglades and, consequently, represent the best measure of potential upper trophic-level exposure to THg.
5. Of seven sites where data on THg in Largemouth bass fillets met all assumptions necessary for statistical analysis, two sites (CA2U3, L5F1) showed an increase among years, three sites (L39F1, LOX4, CA3F1) showed a decrease and two sites (Holey Land, L67F1) showed no among-year variation. Largemouth bass at 50 percent of the sites, mostly the southern sites, exceeded the USEPA's predator- protection guidance value for TL4 fish.
6. Therefore, based on USFWS and USEPA guidance values, it appears that Everglades populations of piscivorous avian and mammalian wildlife continue to be at risk of adverse effects from mercury exposure.
7. Great egret eggs collected from the L67 Colony in early 2001 had higher mean and maximum concentrations of THg compared to eggs collected in 2000, 1999 and 1993. However, the among-year differences in concentrations were not statistically significant. Alternatively, concentrations of THg in feathers of egret nestlings at the L67 Colony exhibited significant among-year variation, with 2001 levels greater than concentrations observed in either 2000 or 1999. Based on published reports and on new data from MeHg-injection studies on eggs from Florida wading birds, egrets at L67 and possibly elsewhere in Water Conservation Area (WCA) 3A continue to appear to be at some elevated risk of mercury toxicity.

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## INTRODUCTION

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This is the fourth annual permit compliance monitoring report for mercury in the downstream receiving waters of the Everglades Protection Area (EPA). This report summarizes the mercury-related reporting requirements of the Florida Department of Environmental Protection (FDEP, or Department) Everglades Forever Act Permits (EFA, Chapter 373.4592, F.S.). The latter includes permits for Non-Everglades Construction Project Discharge Structures, Stormwater Treatment Area (STA) 6, STA 5, STA 1W, and STA 2 (No. 06,502590709, 262918309, 0131842, FL0177962-001, 0126704). This report summarizes the results of monitoring in the reporting year ending April 30, 2001. This year, results of mercury monitoring within the STAs will be reported separately in **Appendix 4A-8**.

The Report consists of Key Findings and Overall Assessment, Introduction, Background, Summary of the Mercury Monitoring and Reporting Program, and Monitoring Results. The Background section briefly summarizes the operation of the STAs and discusses their possible impact on South Florida's mercury problem. The next section summarizes both sampling and reporting requirements of the Mercury Monitoring Program. Monitoring results are then summarized and discussed. Recent results from the Mercury Monitoring Program describe significant spatial distributions and, in some instances, among-year differences in mercury concentrations.

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## BACKGROUND

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The STAs are treatment marshes designed to remove nutrients from stormwater runoff originating from upstream agricultural areas. The STAs are being built as part of the Everglades Construction Project (ECP). When completed, the ECP will include six STAs totaling about 43,000 acres of constructed wetlands. The downstream receiving waters to be restored and protected by the ECP include the District's water management canals of the Central and Southern Florida (C&SF) Project and the interior marshes of the Everglades Protection Area, encompassing Water Conservation Areas (WCAs) 1, 2 and 3 and the Everglades National Park (ENP or Park).

Concerns were raised that in reducing downstream eutrophication this restoration effort might inadvertently worsen the Everglades mercury problem (FGMFWTF, 1991). Widespread elevated concentrations of mercury were first discovered in freshwater fish from the Florida Everglades in 1989 (Ware et al., 1990). Mercury is a persistent, bioaccumulative toxic pollutant. Consequently, mercury can build up in the food chain to levels harmful to human and ecosystem health. Based on the levels observed in 1989, state fish consumption advisories were issued for select species and locations (Florida Department of Health and Rehabilitative Services and Florida Game and Fresh Water Fish Commission, March 6, 1989). Subsequently, elevated concentrations of mercury have also been found in predators, such as raccoons, alligators, Florida panthers and wading birds (Fink et al., 1999).

To provide assurance that the ECP is not exacerbating the mercury problem, the South Florida Water Management District (District) monitors concentrations of total mercury (THg) and methylmercury (MeHg) in various abiotic (e.g., water and sediment) and biotic (e.g., fish and bird tissues) media within the STAs and downstream. Monitoring mercury concentrations in aquatic animals provides several advantages. First, MeHg occurs at much greater concentration in biota

relative to surrounding water, making chemical analysis more accurate and precise. Though detection levels of parts per trillion (ppt or ng/L) have been achieved for THg and MeHg in water, uncertainty boundaries can become large when ambient concentrations are very low, as is often the case in the Everglades. Second, organisms integrate exposure to MeHg over space and time. While surface water concentrations fluctuate on a daily, event and seasonal basis, because mosquitofish are a short-lived species they can be used to monitor short-term changes in environmental concentrations of mercury through time. In contrast, sunfish and largemouth bass are long-lived species and represent average conditions that occurred over previous years. Finally, the mercury concentration in aquatic biota is a true measure of MeHg bioavailability and results in a better indicator of possible exposure to fish-eating wildlife than the concentration of MeHg in water.

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## **SUMMARY OF THE MERCURY MONITORING AND REPORTING PROGRAM**

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The monitoring and reporting program summarized below is described in detail in the “Mercury Monitoring and Reporting Plan for the Everglades Construction Project, the Central and Southern Florida Project, and the Everglades Protection Area,” which was submitted by the District to the Florida Department of Environmental Protection, the U.S. Environmental Protection Agency and the U.S. Army Corps of Engineers in compliance with the requirements of the aforementioned permits. The details of the procedures to be used in ensuring the quality of and accountability for the data generated in this monitoring program are set forth in the District’s “Quality Assurance Project Plan (QAPP) for the Mercury Monitoring and Reporting Program,” which was approved upon issuance of the permit by the Florida Department of Environmental Protection (FDEP). QAPP revisions were approved by FDEP on June 7, 1999.

### **PRE-OPERATIONAL MONITORING AND REPORTING REQUIREMENTS**

Levels of THg and MeHg in various compartments (i.e., media) of the downstream receiving waters collected prior to the operation of the first STA define the baseline condition from which to evaluate the mercury-related changes, if any, brought about by the operation of the STAs. The pre-ECP mercury baseline conditions are defined in the Everglades Mercury Background Report, which summarized all the relevant mercury studies conducted in the Everglades through July 1997 during the construction, but prior to the operation of, the first STA. Originally prepared for submittal in February 1998, it has now been revised to include the most recent data released by the U.S. Environmental Protection Agency and U.S. Geological Survey and was submitted in February 1999 (FTN Associates, 1999).

### **OPERATIONAL MONITORING AND REPORTING REQUIREMENTS**

The downstream system is monitored to track changes in mercury concentrations over space and time in response to the changes in hydrology and water quality brought about by the ECP (for site locations, refer to **Figures 1 and 2**).

**Rain Water:** From 1992 to 1996, the District, Department, USEPA and a consortium of Southeastern U.S. power companies sponsored the Florida Atmospheric Mercury Study (FAMS). FAMS results, compared with monitoring of surface water inputs to the Everglades, showed that

>95 percent of the annual mercury budget came from rain, meaning the major source of mercury to the Everglades was from the air. Accordingly, the District continues to monitor atmospheric wet-deposition of THg to the Everglades by participating in the National Atmospheric Deposition Program's Mercury Deposition Network (MDN). Following MDN protocols, bulk rainfall was collected weekly, at the top of 48-foot towers located at the Everglades Nutrient Removal (ENR) Project, Andytown substation of Florida Power and Light (I-75/U.S.27) and Everglades National Park, and analyzed for THg.

**District Structures Surface Water:** Quarterly, unfiltered grab samples of water were collected using ultra-clean technique upstream of the following structures and analyzed for THg and MeHg: S-5A, S-10C, S-140, S-9, S-32, S-151, S-141, S-190/L-28 interceptor, S-334 and S-12D. These sites bracket the WCAs or are major points of inflow or outflow. Monitoring of these sites is intended to capture the effect of seasonal changes in the relative contributions of rainfall and stormwater runoff contributing to water quality entering the EPA.

**Preyfish:** Annually, a grab sample of between 100 and 250 mosquitofish (*Gambusia sp.*) were collected using a dipnet at 12 downstream interior marsh sites. The samples were homogenized, the homogenate was subsampled in quintuplicate and each subsample was analyzed for THg. This species was selected as a representative indicator of short-term, localized changes in water quality because of its small range, short lifespan and wide occurrence in the Everglades.

**Secondary Predator Fish:** Annually, 20 fish in the genus *Lepomis* (sunfish species) were collected at 12 downstream interior marsh sites and each whole fish analyzed for THg. Because of their widespread occurrence and because they are a preferred prey for a number of fish-eating species, sunfish (*Lepomis spp*) were selected as an indicator of the exposure to wading birds and other fish-eating wildlife.

**Top Predator Fish:** Annually, 20 largemouth bass (*Micropterus salmoides*) were collected, primarily via electroshocking methods, at 12 downstream interior marsh sites and the muscle analyzed for THg. Largemouth bass were selected as an indicator of potential human exposure and because this species has been monitored at several Everglades sites since 1989.

A total of 85 to 99 percent of the THg in fish is MeHg (Grieb et al., 1990; R. Jones, FIU, pers. comm., 1995; L. Cleckner, University of Wisconsin, pers. comm, 1996; SFWMD, unpublished data) and that the percentage generally increases with each successive trophic level (Watras, 1993). Therefore, the analysis of fish tissue for THg is interpreted as equivalent to the analysis of fish tissue for MeHg for purposes of this report.

**Feathers:** Annually, feathers from 20 great egret nestlings from two different nesting colonies within WCA-3A will be collected and analyzed for THg under appropriate state and federal permits (WX99076, MB007948-1). Because MeHg bioaccumulates in top predator fish, the organisms most highly exposed in the Everglades are the fish-eating birds, including the wading birds. This is a modification from the sampling scheme initially proposed, which would have involved collecting molted feathers from post-breeding adults as they lay at or in the immediate vicinity of nests or from STAs. This modified sampling design is more consistent with protocols used in the collection of background data (Frederick et al., 1997).

In addition to the monitoring program described above, in accordance with Condition 4.iv of the Mercury Monitoring Program, the District is required to "report changes in wading bird habitat and foraging patterns using data collected in ongoing studies conducted by the permittee and other agencies."

Further details regarding rationales for sampling scheme, procedures and data reporting requirements can be found in the Everglades Mercury Monitoring Plan revised March 1999 (Appendix 1 of QAPP, June 7, 1999).

## **QUALITY ASSURANCE MEASURES**

The following section is an assessment of the District's Mercury Monitoring Program during the reporting year May 1, 2000 through April 30, 2001, and where appropriate evaluates the quality of the data in terms of accuracy, precision and completeness. This assessment is based on data quality objectives contained in the District's "Quality Assurance Project Plan (QAPP) for the Mercury Monitoring and Reporting Program," which was approved upon issuance of the permit by the Florida Department of Environmental Protection (FDEP; revisions approved June 7, 1999).

Quality assurance (QA) and quality control (QC) are integral parts of all compliance monitoring programs, but especially when dealing with ultra-trace concentrations of analytes common in natural and man-modified environments. The QA program consists of two distinct, but related, activities: quality assurance and quality control. Quality assurance includes design, planning and management activities conducted prior to implementation of the project to ensure that the appropriate kinds and quantities of data will be collected. The goals of quality assurance are to ensure that: (1) standard collection, processing and analysis techniques will be applied consistently and correctly; (2) the number of lost, damaged and uncollected samples will be minimized; (3) the integrity of the data will be maintained and documented from sample collection to entry into the data record; (4) all data will be comparable; and (5) results can be reproduced.

QC activities are implemented during the data collection phase of the project to evaluate the effectiveness of the QA activities. QC activities ensure that measurement error and bias are identified, quantified and accounted for or eliminated, where practicable. QC activities include internal and external checks. Typical internal QC checks include repeated measurements, internal test samples, use of independent methods to verify findings, and use of standard reference materials. Typical external QC checks include exchanging samples among laboratories for reprocessing to test comparability of results, independent performance audits and periodic proficiency examinations. Because mercury-related degradation of water quality is being defined in this project relative to baseline data generated by one or more laboratories, data comparability is a primary concern. Comparability of reporting units and calculations, database management processes and interpretative procedures must be ensured if the overall goals of the project are to be realized.

### **Laboratory QA/QC**

Comparability of laboratory efforts were ensured through compliance with the requirements in U.S. EPA Methods 1631 Rev. B ("Mercury in Water by Oxidation, Purge and Trap, and Cold Vapor Atomic Fluorescence Spectrometry, 821/R-96-001), Draft Method 1630 (Methylmercury in Water and Tissues by Distillation, Extraction, Aqueous Phase Ethylation, Purge and Trap, Isothermal GC Separation, Cold Vapor Atomic Fluorescence Spectrometry 01A0007846 CD-98-1600 08/01/1998), Method 245.5 (Mercury in Sediment by Cold Vapor AAS; 600/4-79-020), Method 245.6 (Mercury in tissues by Cold Vapor AAS, 600/4-91-010) and Method 245.7 (Mercury-CVA Fluorescence spectrometry; CD-98-Stan 02/01/1999), which identify performance based standards and the appropriate levels of QA/QC. The District's QA program relies on state approved Laboratory Quality Assurance Plans for documentation of the individual

laboratory's QA efforts and success criteria. Neither laboratory QA programs nor results of laboratory QC checks will be discussed here in detail.

The primary laboratory (Florida Department of Environmental Protection Central Laboratory) has applied to the U.S. EPA for an alternate test procedure (ATP) from Method 1631 for ultra-trace THg determination. Until the ATP is approved, all results for THg determination in surface water generated by FDEP using the ATP should be interpreted as experimental. Pending said approval, FDEP has revised methods to be compliant with Method 1631.

## Field QC Samples

A total of 429 field QC (FQC) samples was collected with unfiltered surface water samples at STA-1W, STA-2, STA-5, STA-6 and Non-ECP structures during the reporting year (**Tables 1 and 2**). This represents 51 percent of the 837 samples collected overall. These FQC check samples identified several persistent problems that might impact the long-term monitoring program. One major problem was the frequent occurrence of target analytes, both THg and MeHg, in FQC blank samples, e.g., trip blanks (TB), equipment blanks (EB) and field blanks (FB). FQC blanks with analyte concentrations exceeding two times the method detection limit (MDL) can result in the qualification (invalidation) of samples and, consequently, create gaps in the data record. Potential sources of contamination include: (1) field contamination; (2) bottle contamination; (3) constituents leaching from bottles; (4) contamination during transit; or (5) contamination of the sample at the analytical laboratory. Evaluation of FQC blanks relies on the assumption that the deionized, distilled water (DDW) supplied by the analytical laboratories for preparing FQC blanks is initially free of the analyte. However, both the primary (FDEP) and the secondary (Frontier Geosciences, Inc.) laboratory have acknowledged the presence of ultratrace levels of mercury in their DDW and reagents, a persistent problem in most ultratrace mercury labs (averaging 0.2 ng/L; District teleconference with FDEP, FGS on February 28, 2001). Because MeHg is not a ubiquitous contaminant, as is elemental or inorganic Hg, a similar problem is not typically observed for MeHg. Nevertheless, as shown in **Table 1**, MeHg did occur frequently in FQC blanks during the reporting year. The Primary laboratory also acknowledged possible organic Hg contamination of their DDW when the system was inadvertently connected to a groundwater supply system for a short time during this reporting year. The laboratory qualified affected blanks accordingly. Further, because of differences in laboratory methods, principally the use of in-bottle digestion, the primary laboratory also acknowledged potential MeHg contamination of reused Teflon bottles.

Both laboratories have undertaken corrective actions, including, but not limited to, a petition for approval of an alternate test procedure to USEPA (see discussion above), additional internal monitoring of DDW systems and switching to disposable glass bottles. However, because data validation requires consistent and accurate evaluation of analyte concentrations in FQC blanks relative to concentrations in DDW, for which data are incomplete at this time, all QA/QC results of surface water collections during the reporting year are tentative and currently under review.

Another persistent problem in ultratrace mercury monitoring is the inherent variability of THg and MeHg concentrations in surface water. Precision is critical in routine monitoring programs, such as this one, that rely on a single sample to represent water quality at a given site. The relative percent difference (RPD) between serially collected field duplicates is a measure of the representativeness of single samples to describe conditions at the monitoring sites. Because a FD is often collected only once every ten or twenty samples, a single FD often represents the precision for an entire day of sample collections. Poor precision, as demonstrated by high RPD between duplicates (i.e., >25 percent RPD), may indicate poor sampling technique, improper

handling, poor laboratory performance or a heterogeneous sample matrix in a rapidly changing environment. The latter is key in ultratrace mercury monitoring in South Florida. At ultratrace levels, a single suspended particle in unfiltered surface water can dramatically influence concentrations of inorganic and organic mercury species. Consequently, THg and MeHg concentrations are very heterogeneous in unfiltered surface water samples, even when samples are collected within a short time of each other. Additionally, precision near the detection limit, such as with ultra-trace mercury, is often inherently poor due to instrument limitations. Consequently, higher RPDs are not unexpected.

Based on the replicability observed in FDs collected from the ENR Project and elsewhere over a four-year period, the District established a 40-percent RPD as an acceptance criterion for precision between serially collected FD. This criterion is critical because it also defines “significant” differences in this program, which, as mentioned earlier relies on a single grab sample. Based on this acceptance criterion for precision, concentrations at two sites (i.e., samples from inflow and outflow of an STA) or at the same site at different times would be considered “significantly” different only if they differed by a value greater than 40 percent RPD. As shown in **Table 2**, replicability of surface water FDs rarely exceeded the acceptance criteria, with average RPDs for THg-FD less than MeHg-FD. However, RPDs between unlabeled FD (i.e., laboratory-blind duplicates) were much higher than for labeled FDs. It is uncertain at this time whether the lower RPDs of labeled-FDs were a result of laboratory re-runs. If that is the case, then the RPD between labeled FDs reflect field precision (or imprecision), whereas the unlabeled FD (not having been reanalyzed) would reflect a combination of field imprecision and laboratory imprecision. Because other samples are not routinely reanalyzed, the latter estimate may be a more accurate estimate of the precision of the monitoring program.

### **Aliquot Variability and Representativeness of Mosquitofish Composite Sample to Describe Population**

To monitor spatial and temporal patterns in mercury residues in small-bodied fishes, between 100 and 250 individual mosquitofish are collected at various locations in the STAs and ECP and Non-ECP marshes. These 100-to-250 individuals are then composited for each site. Composite sampling can increase sensitivity (i.e., by increasing the amount of material available for analysis), reduce intersample variance effects and dramatically reduce analytical costs. However, there are disadvantages to composite sampling. Sub-sampling from a composite introduces uncertainty if homogenization is incomplete. Since 1999, the District has used a Polytron® homogenizer to produce the composited mosquitofish homogenate. The homogenate is then subsampled in quintuplicate and each subsample is analyzed for THg. The arithmetical average from these multiple analyses is then reported, thus partitioning out analytical and homogenate variability.

During the reporting year, a total of 98 composite samples of mosquitofish were collected and shipped to FDEP for analysis. Of the 98, 63 were subsampled and analyzed five times and 35 were subsampled only three times (three aliquots taken, where sample mass was insufficient). Mean relative standard deviation (RSD) in THg concentrations among aliquots was 5.2 percent (median=4.4 percent; maximum=19.6 percent) when composites were sub-sampled five times and 4.1 percent (median=3.5 percent; maximum=9.9 percent) when composites were subsampled three times. Based on the apparent degree of homogenization, as evidenced by the low RSD among aliquots, the District is currently revising its Standard Operation Procedures (SOP) to reduce subsampling of mosquitofish homogenates from five to three. Equipment blanks (EB)

collected as QC checks during tissue sample processing, i.e., rinsate from grinders or tissue homogenizer, contained less than 0.1 µg/L THg (n=8).

Another disadvantage to composite sampling is that the same amount of information is not generated as when samples are analyzed individually. Because samples are physical averaged, no variance estimate for the population is generated and, consequently, uncertainty is introduced regarding the representativeness of the sample in describing the population. This also hampers statistical comparisons. To assess the representativeness of composite samples, 10 FD mosquitofish composites were collected during the reporting year, i.e., a second set of 100-to-250 individuals was collected at the site and composited as a second sample. The relative percent difference (RPDs) between composite means ranged from 3 to 50 percent and averaged 16 percent (median 11 percent).

### **Inter-Laboratory Comparability**

To ensure further comparability (i.e., reproducibility) between this and other ongoing mercury sampling initiatives, split samples were submitted to the secondary laboratory (Frontier Geoscience, Inc.) for independent analysis of THg and MeHg. This laboratory also generated all the pre-ECP soil and water data for the STAs and the Non-ECP structures, respectively. However, the primary laboratory generated all the baseline fish data.

#### **Water**

Results from independent analyses of split-water samples collected during STA-2 startup (grab samples only) and at Non-ECP structures (n=22 samples, 5 percent of water samples collected) are summarized in **Figure 3**. Although the mean RPD between paired data was about 42 percent, and the maximum RPD was as high as 96 percent, the reported values were correlated ( $r=0.52$ ,  $p=0.01$ ) and a Wilcoxon Signed Rank Test found no statistically significant (consistent) bias in ultratrace THg determination in surface water (n=22,  $W=-37$ ,  $p=0.56$ ).

Likewise, reported ultratrace MeHg concentrations in surface water splits also exhibited variance from the expected 1-to-1 line (**Figure 3b**); the RPD between splits averaged 63 percent (median RPD was 49 percent, maximum RPD was 166 percent). Like THg, MeHg concentrations in split samples were correlated between laboratories ( $r=0.71$ ,  $p < 0.001$ ) and no consistent bias was observed in MeHg determination (Wilcoxon Signed Rank test,  $W=-54.0$ ,  $p=0.36$ ). Nevertheless, because of this analytical variability caution should be exercised in drawing conclusions based on results from a single sample. Instead, conclusions should be based on averages and running means.

#### **Fish**

Where sample mass was sufficient, splits of mosquitofish homogenate collected during STA-2 startup were sent to the secondary laboratory (FGS, Inc.) for independent analysis (n=19; 19.4 percent of mosquitofish collected and sent to FDEP). Results are graphically shown in **Figure 4**. Mean values of replicate analyses (i.e., of aliquots) of split samples were highly correlated ( $r=0.93$ ,  $p < 0.001$ ) and did not differ significantly between laboratories (paired t-test;  $df=18$ ,  $t=-1.028$ ,  $p=0.32$ ).

Split samples of 141 of the 900 large-bodied fishes (i.e., 15.7 percent of whole sunfish homogenates and fillets of Largemouth bass) collected during the reporting year were sent to the Secondary laboratory (FGS, Inc.) for independent analysis. As shown in **Figure 5**, the primary laboratory reported lower concentrations for fishes with mid-level THg, but higher concentrations for fishes with low-level THg relative to the secondary laboratory. Notice that this graph is

log:log, which may reduce the visual impression of scatter. While the paired values were moderately correlated ( $r=0.43$ ,  $p < 0.001$ ), the difference between laboratory splits was statistically significant (Wilcoxon Signed Rank Test,  $W=-4540$ ,  $p < 0.001$ ). The average absolute difference between splits was 0.17 mg/kg (median was 0.022 mg/kg). This represented a mean RPD between splits of 21 percent, with a maximum RPD of 175 percent. The latter was a case where the primary laboratory reported a value of 0.83 mg/kg, compared to 12.3 mg/kg reported by the secondary laboratory. A reanalysis of this sample (i.e., the same digestate) by the secondary laboratory resulted in 10.6 mg/kg; however, an analysis of a split sample of this fish (also sent to the secondary laboratory) resulted in 0.79 mg/kg. An analysis of a split sample from this fish by a third laboratory, Florida Fish & Wildlife Conservation Committee (FFWCC, which maintained custody of subsample), confirmed the value reported by the primary laboratory.

### **Bird Monitoring**

Splits of an egg sample ( $n=1$ , 10 percent of collected samples) and a feather sample ( $n=1$ ; 8 percent of collected samples) that were sent to both labs for analysis had RPDs of 0.24 percent and 33 percent, respectively.

## **STATISTICAL METHODS**

Temporal trends in atmospheric THg deposition and water column THg and MeHg concentrations were evaluated using the seasonal Kendall test, which is a generalization of the Mann-Kendall test for trend detection (Chemstat; Starpoint Software Inc., Cincinnati Ohio; Gilbert, 1987). It is applied to data sets exhibiting seasonality. This test may be used even though there are missing, tied or nondetect values. The validity of the test does not depend on the data being normally distributed. However, use of this analysis presupposes the presence of large multi-year, multi-season data sets. Some argue that five years is a minimum data set for proper use of both the test and standard statistical tables. Consequently, the application of this test, in the fourth year of the monitoring program, should be approached cautiously, and results should be viewed as approximations only.

As stated above, monitoring Hg concentrations in aquatic animals provides several advantages; however, interpretability of residue levels in animals can sometimes prove problematic due to confounding influences of age or species of the collected animal. For comparative purposes, special procedures are used to normalize the data. Standardization to size, age or lipid content is a common practice (Wren and MacCrimmon, 1986; Hakanson, 1980). To be consistent with the reporting protocol used by FFWCC (Lange et al., 1998; 1999), mercury concentrations in largemouth bass were standardized to an expected mean concentration in three-year-old fish at a given site by regressing mercury against age (Lange et al., 1999 and references therein). Note, to adjust for month of collection, otolith ages were first converted to decimal age using protocols developed by Lange et al., 1999. Sunfish were not aged and, consequently, age normalization was not available. Instead, arithmetic means were reported. However, efforts were made to estimate a least-squares mean (LSM) THg concentration based on weight of fish. Additionally, the distribution of the different species of lepomis (e.g., *L. gulosus* warmouth; *L. punctatus*, spotted sunfish; *L. macrochirus*, bluegill; *L. microlophus*, redear sunfish) collected during electroshocking was also considered, i.e., as a potential confounding influence on THg concentrations, prior to each comparison. To be consistent with the reporting protocol of Frederick et al. (1997) and Sepulveda et al., 1999, THg concentrations in nestling feathers were similarly standardized for each site and expressed as least-square means for a chick with a 7.1 cm bill.

Where appropriate, analysis of covariance (ANCOVA; SAS GLM procedure) was used to evaluate spatial and temporal differences in mercury concentrations, with age (largemouth bass), weight (sunfish) or bill size (egret nestlings) as a covariate. However, use of ANCOVA is predicated on several critical assumptions (ZAR, 1996), including: (1) that regressions are simple linear functions; (2) that regressions are statistically significant (i.e., non-zero slopes); (3) that the covariate is a random, fixed variable; (4) that both the dependent variable and residuals are independent and normally distributed; and (5) that slopes of regressions are homogeneous (parallel). Where these assumptions were not met, standard ANCOVAs or Student's t-tests (SigmaStat, Jandel Corporation, San Rafael, California) were used; possible covariates were considered separately. The assumptions of normality and equal variance were tested by the Kolmogorov-Smirnov and Levene Median tests, respectively. Data sets lacking homogeneity of variance or that departed from normal distribution were natural-log transformed and reanalyzed. If transformed data met the assumptions, they were used in ANCOVA. If not, raw data sets were evaluated using nonparametric Mann-Whitney Rank sum tests. If the multigroup null hypothesis was rejected, groups were compared using either Tukey HSD or Dunn's method.

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## MONITORING RESULTS

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### RAINFALL: NATIONAL ATMOSPHERIC DEPOSITION PROGRAM - MERCURY DEPOSITION NETWORK

On a weekly basis, samples of bulk rainfall were collected under the protocols of the National Atmospheric Deposition Program's Mercury Deposition Network (MDN) at the ENR Project, the Andytown substation, and at the Baird Research Center, Everglades National Park (for locations see **Figure 2**). For more information on MDN and to retrieve raw data, see <http://nadp.sws.uiuc.edu/mdn>.

As evident from **Table 3**, atmospheric deposition of THg to South Florida was highly variable both spatially and temporally. In general, results observed in 2000 were consistent with the seasonal trends observed during the Florida Atmospheric Mercury Study (FAMS, Guentzel, 1997). As shown in **Figure 6**, THg concentrations in precipitation were substantially higher during the summer months, possibly due to seasonal tall convective thunderstorms that can scavenge particulate Hg and water soluble reactive gaseous Hg (RGM) from the middle and upper troposphere. This is consistent with observations of Guentzel (1997) during the FAMS study. Because both THg concentration and rainfall volumes generally increase during the summer, the latter by a factor of 2-3, THg wet-deposition typically increases 5-8 fold during the wet season (**Figure 6**).

Although both weekly rainfall amounts and THg concentrations differed substantially among the three South Florida sites (**Table 3**), a Kruskal-Wallis ANOVAs on ranks revealed the differences were not statistically significant (rainfall:  $df=2$ ,  $H=4.5$ ,  $p=0.11$ ; THg concentration:  $df=2$ ,  $H=3.8$ ,  $p=0.15$ ; period of record: 1/1998 – 12/2000). Nevertheless, when combined, among-site differences in rainfall and concentration resulted in significant spatial differences in THg deposition (Kruskal-Wallis ANOVA on ranks;  $df=2$ ,  $H=7.98$ ,  $p=0.02$ ). Specifically, because it receives lower rainfall amounts with lessor concentrations of THg, the ENR Project receives significantly lower weekly atmospheric fluxes of THg (i.e., deposition) than does the ENP (Dunn's post-hoc test,  $p<0.05$ ). It should be noted that other pair-wise comparisons in deposition, e.g., ENR versus Andytown, ENP versus Andytown were not significant ( $p>0.05$ ).

THg concentrations in rainfall at the all three sites were greater than the median THg concentration reported for all North American MDN stations between 1995-2000 (9.7 ng/L; Sweet, 2001). Sweet (2001) reports that volume-weighted concentrations are lowest in New England and the Canadian Maritime provinces (6 ng/L) and highest in Florida.

Volume-weighted average concentrations of THg were greater than cumulative average concentrations at all three sites in 2000 (**Table 3**). At the same time, substantially less rainfall fell at the three sites in 2000 (**Table 3**). The combination of higher average concentration and lower rainfall were off-setting and resulted in slight declines in annual atmospheric wet-deposition of THg at ENR and ENP, but a slight increase at Andytown, as compared to their respective cumulative annual averages (**Table 3**). As summarized in **Table 3**, seasonal Kendall analyses of the MDN data sets revealed statistically significant negative (i.e., declining) trends in monthly median THg concentration in rain (ENP,  $p=0.05$ ), monthly rainfall amounts (ENR,  $p=0.04$ ), and monthly deposition of THg (Andytown,  $p=0.04$ ) (**Table 3**). However, as discussed previously, with only three to five years of data, these results should be viewed as preliminary. In the present study, for example, calculated Z-scores were substantially reduced at the site with the longest data set (i.e., ENP) by the relatively large amount of observed variance. A previous analysis of the pooled FAMs (1993-1996) and MDN (1996-1999) data sets by Pollman and Atkeson (2000) found no significant long-term temporal trend in wet deposition of THg to South Florida.

Collectively, the results reported here for wet-deposition of THg in comparison with monitoring of surface water at Non-ECP Structures (following section) continued to show that the major source of mercury to the Everglades is from the air. This is consistent with previous assessments by both FDEP (Atkeson, <http://www.dep.state.fl.us/labs/hg/flmercury.htm>) and U.S. EPA (USEPA, 1998). Dry deposition, which may exceed wet deposition by a factor of 2 (Keeler & Lindberg, 2001), likely adds significantly to the overall atmospheric input.

## SURFACE WATER AT NON-ECP STRUCTURES

**Table 4** and **Figure 7** summarize monitoring results of unfiltered THg and MeHg in surface water samples collected quarterly at Non-ECP structures (map of locations is shown in **Figure 1**). There are no baseline water concentration data generated by comparable analytical methods for any District structures prior to 1997. As in previous years, there were no exceedances of the Florida Class III WQS of 12 ng THg/L, at any of the structures monitored. The maximum THg concentration observed during the reporting year was 5.1 ng/L that occurred at S9 during the 4<sup>th</sup> quarter 2000 (**Figure 7**). The maximum MeHg concentration observed during the reporting year at a Non-ECP structure was 1.6 ng/L that occurred at S12D during the 4<sup>th</sup> quarter 2000. Note, currently, Florida has no WQS for MeHg. Average concentrations (i.e., not volume-weighted) over the last 4 quarters for both THg and MeHg were elevated, compared to site-specific cumulative averages, at all sites except S5A, (**Table 4**). Percent of THg that was MeHg was also higher at most sites over the last four quarters. A comparison of quarterly averages for the year against cumulative quarterly averages suggest that water concentrations of THg and MeHg increased primarily in the 3<sup>rd</sup> and 4<sup>th</sup> quarters of 2000 (**Table 4**). This increase over long-term seasonal trends is also graphically evident in **Figure 7**.

Nevertheless, seasonal Kendall analyses found no statistically significant trends in either THg or MeHg concentration at any of the sites. Calculated Z-scores, which were based on four seasons, i.e., quarterly samples, ranged from -1.1 for THg at S5A to +1.3 for MeHg at S10C (positive Z-score indicates increasing concentrations, whereas negative Z-score a decline). To test for an upward trend (one-tailed test) at the  $\alpha=0.05$  level, the null hypothesis of no trend would have been rejected if  $Z > Z_{0.95}$ , that is, if the absolute value of  $Z > 1.65$ . However, as discussed

above, with less than four years of data (large data gaps present where data were invalidated), these results should be viewed as preliminary.

## FISH FROM ECP AND NON-ECP INTERIOR MARSHES

Results from monitoring downstream interior marsh mosquitofish, sunfish and Largemouth bass are summarized in **Tables 5** through **7** (values for individual large-bodied fish are provided in **Attachment 1** at the end of this appendix). Fish are collected from a total of 12 downstream interior marsh sites (**Figure 1**). Where fish could not be collected after a good faith effort, collection sites defaulted to nearby canals where fish were more plentiful and the same source water was being sampled. These default sites are depicted in **Figure 8**. Mercury levels in Largemouth bass at three of these sites, LOX4 (WCA-1 GFC4), CA2U3 (WCA-2A U3), and CA3-15 (WCA-3A 15) were monitored by the FFWCC prior to initiation of the ECP (period of record extends back to 1993).

As discussed below, fishes collected in 2000 showed both spatial and temporal patterns in tissue mercury concentrations. In keeping with the primary objective of this monitoring program, the focus here will be on temporal changes in mercury concentration in fish tissues to assess possible adverse effects from the ECP and operation of the STAs. Nevertheless, spatial patterns of tissue mercury concentrations are important, particularly where there has been a variation from background conditions (i.e., pre-ECP conditions established by FFWCC). Therefore, spatial patterns will be reviewed in detail only where there has been change over time (i.e., interaction between treatment effects).

### Mosquitofish

THg concentrations in mosquitofish collected from marsh sites in 2000 ranged from 5 ng/g at L5F1 to 152 ng/g at P33 (**Table 5**). The basin-wide average concentration was 63 ng/g (**Table 5**, for locations see **Figure 9**), which represents a 68 percent decrease from the 1999 basin-wide average concentration. This between-year difference in mercury concentration in mosquitofish was statistically significant (ANOVA;  $df=2,35$ ;  $F=19.8$ ;  $p < 0.001$ ), with levels in 2000 similar to 1998 (Tukey Test,  $p=0.6$ ), but both lower than the peak levels observed in 1999 ( $p < 0.001$ ). In contrast to 1999, where mosquitofish at all sites showed significant increases in THg compared to 1998, in 2000, THg decreased in mosquitofish from all sites (**Table 5**, **Figure 9**). These decreases ranged from 32 percent at the P33 site to 98 percent at L5F1 site. The peak in THg concentrations in mosquitofish observed in 1999 was reportedly related to reflooding of marshes following a drydown (Krabbenhof and Fink, 2000). Mosquitofish are used to monitor short-term changes in environmental concentrations of mercury because they are short lived and, thus, the between-year variability in body burdens was not unexpected.

## Sunfish

THg concentrations in sunfish collected from marsh sites in 2000 ranged from 70 ng/g at L39F1 to 396 ng/g at L67F1 (**Table 6**). Sunfish also exhibited inter-annual differences in tissue mercury concentration, but the direction of change was variable among locations. Between-year percent change from 1999 to 2000 ranged from a 66 percent decrease in THg concentration in sunfish from the P33 marsh to a 100 percent increase in concentration in sunfish from the Holey Land WMA (**Table 6, Figure 10**). Despite these inter-annual variations at individual sites, the median concentrations of THg have not changed significantly in sunfish basin-wide over the three year period (Kruskal-Wallis ANOVA on ranks,  $H=0.11$ ,  $df=2$ ,  $p=0.95$ ; medians: 126 ng/g in 1998, 120 ng/g in 1999 and, 120 ng/g in 2000). The absence of a significant inter-annual difference in sunfish, where it occurred in mosquitofish, may be a result of a longer half-life of MeHg in sunfish and longer lifespan of sunfish, which results in a long-term integrated average concentration on the order of a year.

As discussed previously, attempts were made to use analysis of covariance (ANCOVA) to evaluate patterns of mercury concentrations in sunfish, *Lepomis spp.*, using weight as a covariate. However, use of ANCOVA was often inappropriate because weight–concentration relationships were inconsistent (i.e., slopes were either not significant or were not parallel each year). The lack of a strong concentration-size relationship likely resulted from interspecies differences (i.e., among the different *Lepomis* species) in growth and bioaccumulation factors. As shown in the previous reporting year (Rumbold et al., 2001), species was a significant factor in tissue mercury concentration in sunfishes caught in 2000 (ANOVA on ln-transformed data,  $df=3$ , 205;  $F=16.28$ ,  $p < 0.001$ ); THg concentrations in *L. punctatus* (Spotted sunfish, mean= $257 \pm 136$  ng/g)=*L. gulosus* (Warmouth, mean= $250 \pm 173$  ng/g) > *L. macrochirus* (Bluegill, mean= $177 \pm 162$  ng/g) > *L. microlophus* (Redear, mean  $110 \pm 77$  ng/g)(Tukey multiple comparison procedure). Interestingly, in the past, Warmouth (*L. gulosus*) was found to have significantly greater concentrations of THg than Spotted sunfish (*L. punctatus*; Rumbold et al., 2001a). However, Warmouth have shown a basin-wide trend in decreasing tissue-Hg ( $H=9.4$ ,  $df=2$ ,  $p=0.009$ ; median concentration in: 1998 was 397 ng/g, 1999 was 360 ng/g, 2000 was 170 ng/g), with 2000 levels significantly lower than 1998 levels (Dunn's post-hoc test,  $p < 0.05$ ). It should be noted that weights of Warmouth showed no significant among-year variation during this same period (ANOVA;  $df=2$ , 86;  $F=2.08$ ;  $p=0.132$ ) and, thus, the mercury trend does not appear to be related to any year-to-year biases in weight ( $\cong$  age) of the sampled population. Similar temporal patterns in tissue-Hg concentrations did not occur in Spotted sunfish ( $H=3.8$ ,  $df=2$ ,  $p=0.15$ ), Bluegill ( $H=4.7$ ,  $df=2$ ,  $p=0.09$ ), or Redear sunfish ( $H=4.6$ ,  $df=2$ ,  $p=0.1$ ).

During the first three years of the monitoring program there were occurrences of substantial among-year differences in species of *Lepomis* collected at individual sites. At CA3F2, for example, the proportion of collected sunfish that were Warmouth fell from 42 percent in 1998 to 35 percent in 1999 to 0 percent in 2000. Notice, the concurrent decline in THg concentrations in sunfish sampled from this site, which did not appear to be related to between-year differences in fish size (**Figure 10**). Similarly, the proportion of collected *Lepomis* that were Warmouth or Spotted sunfish also declined over the three year monitoring period at L39F1 and, as above, THg levels also declined; this despite a marked increase in size of fish collected in 1999 and again in 2000 (**Figure 10**). It is unknown whether the general decline in THg observed at the site resulted from a bias associated with collection of proportionately more Bluegill and Redear, which typically have low THg in their tissues relative to the other two species, or a function of reduced exposure. While there are statistical methods to address confounding factors, such as age or weight, addressing species differences is more problematic, particularly when it is one of two

possible confounding factors (i.e., weight, species or both). Statistical analyses of the sunfish data sets were also hampered or prevented because THg concentration, weights or both also often failed assumptions of normality and equal variance. Nonetheless, among-year differences in tissue-Hg and fish weights were assessed at each location using a one-way ANOVA (i.e., parametric tests on raw or transformed data or non-parametric tests, if assumptions were violated; **Figure 10**), with qualitative consideration of possible influences from among-year differences in collected species. At this point it might be worth noting that sunfish, while not the best species to evaluate spatial and temporal trends, provide the best measure of exposure and ecological risk because of their importance in the Everglades food web (i.e., predominate prey of many upper trophic level animals).

Sunfish were collected in sufficient numbers, in all three years, for a valid among-year assessment at ten sites (at P33, only three fish were collected in 1999, only one fish in 2000 and, consequently, trends were not tested). Of the ten sites, only four were found to have significant among-year differences in THg concentrations in sunfish, with two sites showing a decrease (L38F1, CA3F2) and two sites showing an increase (Holey Land, CA2U3). As reported last year, sunfish at L38F1 (WCA-2A) had lower THg concentrations in 1999 compared to 1998 (Tukey test,  $p < 0.05$ ); pair-wise comparisons with 2000 showed no significant change ( $p > 0.05$ ). This between-year difference in 1998-1999 was not attributable to differences in either fish size or species of collected fishes. By comparison, sunfish at CA3F2 had lower THg concentrations in 2000 as compared to both 1998 and 1999 ( $p < 0.05$ ). However, as discussed above, this apparent change in THg concentration may have been related to among-year differences in species of *Lepomis* collected at the site. Conversely, sunfish at CA2U3 (also within WCA-2A) contained significantly greater tissue concentrations of THg in 2000 as compared to 1999 sunfish (Tukey test,  $p < 0.05$ ) that, in turn, contained greater levels than in 1998 ( $p < 0.05$ ). Notice that this monotonic increase in THg occurred concurrently with a stepwise increase each year in fish size (**Figure 10**). Here again, ANCOVA was not appropriate because the concentration - weight relationship, while significant in 1998 ( $p = 0.02$ ), was not significant in either 1999 ( $p = 0.94$ ) or 2000 ( $p = 0.09$ ). Although we cannot dismiss the possibility that the observed increase in THg in CA2U3 sunfishes were simply a function of increased fish size, as will be discussed below, parallel increases in THg levels were also observed in CA2U3 bass collected during 1998-2000. Like sunfish at CA2U3, sunfish from the Holey Land WMA also showed a concomitant increase in both size and tissue-Hg level. While among-year differences in weight were not statistically significant ( $p = 0.06$ ), the  $p$  value (i.e.,  $0.05 < p < 0.1$ ) suggests a possible influence (i.e., treatment effect) of year on sunfish weight. The among-year difference in THg in Holey Land sunfish (Kruskal-Wallis ANOVA on ranks,  $H = 9.28$ ,  $df = 2$ ,  $p = 0.01$ ), was shown to stem from a between-year difference in 2000 versus 1998 (Dunn's Method,  $p < 0.05$ ; other pair-wise comparisons had  $p > 0.05$ ).

As reported in last year's report (Rumbold et al., 2001a), sunfish collected at L67F1 in 1999 contained some of the highest concentrations of mercury ever observed in Everglades *Lepomis*. A 45-g Bluegill (137 mm), for example, was found to have 3,300 ng THg/g (3.3 ppm), which is almost 5x greater than the next highest concentration previously, reported for this species. As shown in **Figure 10**, these high levels were not observed in 2000. Moreover, while visual inspection of the data in **Figure 10** suggests that arithmetic mean concentrations differed substantially among-years at L67F1, a non-parametric test on ranks revealed no significant difference in tissue-Hg among-years (Kruskal-Wallis ANOVA on ranks,  $H = 4.7$ ,  $df = 2$ ,  $p = 0.9$ ).

## Largemouth Bass

Similar to the lower trophic level fish, Largemouth bass exhibited significant patterns in tissue-Hg concentrations over both space and time. Seven monitoring sites had data sets meeting assumptions necessary for statistical analysis of tissue-Hg concentrations. Sample sizes at CA315, CA3F2 and P33 were insufficient to produce meaningful statistics. The L38F1 data set did not meet the criteria for ANCOVA; because bass differed in age among-years, an ANOVA was also inappropriate. Of the seven sites that were assessed statistically, two sites showed an increase among-years (CA2U3, L5F1), three sites showed a decrease (L39F1, LOX4, CA3F1) and two sites showed no among-year variation (Holey Land WMA, L67F1). Among-year variation ranged from a 61 percent decline in tissue-Hg from 1998 to 2000 in bass at CA3F1 to a 51 percent increase in THg in CA2U3 bass over the same period.

At the CA2U3 site, the among-year variation in standardized age(3) expected mercury concentration (EHg3) in bass was significant (ANCOVA;  $df=2,55$ ;  $F=18.64$ ,  $p<0.001$ ), with the estimated least square means (LSMs) for 1999 and 2000 greater than 1998 (Tukey HSD,  $p<0.00$ ). While the EHg3 was markedly higher in 2000, LSMs at CA2U3 did not differ between 1999 and 2000 ( $p>0.05$ ). Similarly, an ANCOVA revealed THg concentration in fillets of bass from L5F1 was greater in 2000 as compared to 1998 ( $df=1,37$ ;  $F=13.4$ ,  $p<0.001$ ; the 1999 data set was excluded from analysis due to failure to meet predicated assumptions for ANCOVA). The EHg3 increased at L5F1 by 39 percent from 1998 to 2000. These temporal patterns in THg concentration in the bass at CA2U3 and L5F1 were generally consistent with patterns observed in sunfish collected at the same site. As discussed above, while levels of THg in sunfish collected in 1998 and 2000 from L5F1 were not statistically different, the significant decline in weight of the 2000 sunfish (**Figure 11**) could have confounded interpretation of tissue-Hg. Interestingly, of the monitored sites, CA2U3 and L5F1 showed some of the highest percent increases in THg levels in sunfish in 1999 (i.e., as compared to 1998; +47 percent at CA2U3 and +23 percent at L5F1 Rumbold et al., 2001a).

Alternatively, bass collected at CA3F1 (i.e., the alternate for Non-ECP north site) in 2000 contained lower concentrations of THg than fishes collected in either 1999 or 1998 (Kruskal-Wallis ANOVA on ranks,  $H=21.01$ ,  $df=2$ ,  $p<0.001$ ; Dunn's test  $p<0.05$ ); note, ANOVA was a valid test because age did not differ significantly among-years. Similarly, ANOVA also revealed that bass collected at LOX4 contained lower concentrations of THg in 2000 and 1999 as compared to 1998 (Kruskal-Wallis ANOVA on ranks,  $H=12.4$ ,  $df=2$ ,  $p=0.002$ ; Dunn's test  $p<0.05$ ). While bass were not collected in 1998 at L39F1, despite the best efforts of FFWCC contractor, an ANOVA revealed that the difference in THg concentration in fish collected in 2000 (235 ng/g, **Table 7**) and 1999 (359 ng/g,  $n=10$ ) to be statistically important, if not significant ( $df=1,28$ ;  $F=4.1$ ;  $p=0.054$ ). These among-year variations in THg concentrations in Largemouth bass (i.e., decline) were consistent with general patterns observed in sunfish collected at CA3F1, LOX4 and L39F1; however, as discussed above, among-year variations in sunfish were not statistically significant. Nonetheless, as reported by Rumbold et al. (2001a) sunfish at these three sites showed some of the largest decreases in 1999 as compared to 1998 (i.e., as a possible prelude to changes translated up the food chain; -37 percent at CA3F1, -35 percent at LOX4; -26 percent at L39F1).

Bass from the Holey Land WMA (ANCOVA;  $df=2,55$ ;  $F=0.04$ ;  $p=0.96$ ), and L67F1 (ANCOVA;  $df=1,35$ ;  $F=0.96$ ;  $p=0.34$ ) showed no significant among-year variation in THg levels. While THg levels remained relatively stable in Holey Land bass, sunfish from the site did exhibit a slight but, statistically significant, increase in THg. However, as discussed above, Holey Land sunfish also showed a meaningful (i.e.,  $p=0.06$ ) increase in weight that could have confounded

the interpretation of THg. In 1999, sunfish from the Holey Land showed only a 5 percent increase in THg over 1998 levels (as compared to the  $\pm 27$  to 47 percent change in THg in sunfish from areas where bass subsequently showed statistically significant among-year variations). This trend of between-year differences in THg concentration in bass ( $\pm$ ) following a marked change in THg levels in sunfish ( $\pm$ ) during the previous year appeared to break down at L67F1. In 1999, THg increased in L67F1 sunfish by 86 percent over 1998 levels. Based on this, we would have predicted THg to significantly increase in L67F1 bass in 2000. However, as discussed above, while arithmetic mean concentrations (which are easily skewed by extreme values; see error bars in L67F1 sunfish collected in 1999, **Figure 10**), appeared to differ among-years at L67F1, median concentrations did not. Thus, patterns observed in THg levels in bass at L67F1 were consistent with the overall patterns observed in THg in sunfish from the same site, except the variability was dampened.

As stated previously, FFWCC has monitored mercury levels in Largemouth bass at several sites since 1993, i.e., prior to initiation of the ECP. Three of these sites, LOX4 (WCA-1 GFC4), CA2U3 (WCA-2A U3), and CA3-15 (WCA-3A 15) are co-located at sites where the District collects fish. Lange et al. (1999; 2000) report that standardized age (3) mercury concentrations have declined statistically at LOX4 and CA2U3 since 1996, with the most significant decreases occurring between 1996 and 1997 (**Figure 12**). However, as reported here, Lange and Richard (2001) report recent increases in EHg3 bass at a few sites, including CA2U3.

### **Predator Protection Criteria**

Levels of mercury in fish tissues can also be put into perspective and evaluated with regard to mercury risk to wildlife. The U.S. Fish and Wildlife Service (USFWS) has proposed a predator protection criterion of 100 ng/g THg in prey species (Eisler, 1987). More recently, in its "Mercury Study Report to Congress," USEPA proposed 77 and 346 ng/g for trophic level (TL) 3 and 4 fish, respectively, for the protection of piscivorous avian and mammalian wildlife (USEPA, 1997). In 2000, Mosquitofish, which are considered to be at TL 2-3 depending on age (Loftus et al., 1998), at only four of the downstream sites (i.e., 31 percent; **Table 5**) had THg concentrations exceeding either the USFWS or USEPA criterion. This is a dramatic reduction from the previous year when mosquitofish from 100 percent of the sites exceeded both criteria. By comparison, based on mean concentrations (**Table 6**), sunfish, which are at TL 3 (*L. gulosus* at TL 4; Loftus et al., 1998), at all but one contained THg concentrations exceeding one or both of the predator protection criteria in 2000 (sunfish from L39F1 did not). This finding is significant because, as noted above, sunfishes represent the preferred prey item of many fish-eating species in the Everglades and, consequently, represent the best measure of potential upper trophic level exposure to THg. After adjusting arithmetic mean THg concentrations in Largemouth bass filets (**Table 7**) to whole-body concentrations (whole-body THg concentration =  $0.69 \times$  fillet THg; Lange et al., 1998), bass at 50 percent of the sites, mostly the southern sites, also exceeded the guidance value for TL 4 fish. However, caution must be exercised in the latter assessment because Largemouth bass are considered to be at TL 5 (Loftus et al., 1998). Based on these guidance values, it appears that Everglades populations of piscivorous avian and mammalian wildlife continue to be at risk of adverse effects from mercury exposures.

### **WADING BIRD FEATHERS FROM ECP INTERIOR MARSHES**

Results from monitoring mercury levels in Great Egret nestlings are summarized in **Table 8**, and **Figures 13** and **14**. To evaluate temporal trends, results from the District wading bird monitoring program are compared to results of similar collections made by Frederick et al. (1997,

later published by Sepulveda et al., 1999) in 1994 and 1995. In accordance with USACOE permit 199404532 Condition 8b.2, these results were found to be representative of background mercury concentrations in Everglades wading birds (FTN Associates, 1999). The study by Frederick et al. (1997) involved monitoring THg in feathers of Great Egret (*Ardea albus*) nestlings at various Everglades colonies. The District's monitoring program focuses on two egret colonies, designated JW1 and L67, located in WCA-3A (**Figure 1**). These two colonies consistently showed the highest concentrations during background studies (Frederick et al., 1997, FTN Associates, 1999; Sepulveda et al., 1999).

Due to the continuing drought in South Florida, environmental conditions were not optimal for wading bird nesting in 2001 (personal communication, D. Gawlik, SFWMD; P. Frederick, UF). The JW1 colony was surveyed on March 26, 2001 by helicopter and on the ground and was found to contain no nesting birds. A survey out to a radius of 3.5 miles from the colony failed to locate any other nesting colonies. By comparison, when the L67 colony was surveyed a week earlier on March 15<sup>th</sup>, 2001, birds had nested and the majority of eggs had already hatched. Consequently, the data reported below is for the L67 colony only.

In 2001, feather-THg concentrations ranged from 3.8 to 14 µg/g dw (**Table 8**). However, THg concentration in nestling feathers is often dependent on duration of exposure and, thus, age of the bird. Accordingly, attempts were made to regress and standardize feather-Hg concentration for a nestling with a given bill length (i.e., age surrogate) using protocols established by Frederick et al. (1997). Because regressions of THg concentration on bill length were not significant for birds at the L67 colony in 1999-2001 (**Figure 13**), standardized concentrations were not calculated nor was ANCOVA used to assess between-year differences. However, because bill length of sampled nestlings did not vary significantly among years (ANOVA; df=2,40; F=1.28, p=0.29), among-year variability in feather-Hg was examined using a simple ANOVA and was found to be significant (df=2,40; F=13.8; p<0.001). Tukey post-hoc comparisons found 2001 levels to be greater than 2000 levels (p<0.001) and 1999 levels (p<0.001); 1999 did not differ from 2000 (p=0.85, **Figure 13**). Although comparisons to earlier surveys is complicated by the lack of standard feather-THg concentrations at L67, it is clear from **Table 8** that residue levels, although increasing in 2001, remain relatively low compared to 1994 and 1995. This conclusion is consistent with an independent assessment of trends in feather-THg in South Florida egret nestlings by Frederick and Spalding (2000).

In the past, in addition to collecting feather samples for compliance with the aforementioned federal and state permits, the District has also collected egret eggs to support an ecological risk assessment of MeHg (Rumbold, 2000) and to better assess spatial and temporal trends in wading bird exposure (Rumbold et al., in press). In response to a special request from FDEP (2/26/01 letter from Dr. Tom Atkeson, Mercury Coordinator for FDEP), the District continued to collect egret eggs in 2001.

Egret eggs collected from the L67 colony in 2001 had a mean egg THg concentration of 0.55 ±0.27 µg/g (fresh weight, n=10). While 2001 levels were slightly elevated compared to previous years (**Figure 14**), concentrations did not vary significantly among-years (i.e., 1999-2001; ANOVA; df=2,27; F=2.7; p=0.09).

Although egg concentration is thought to be the best predictor of MeHg risk to avian reproduction (Wolfe et al., 1998), embryonic sensitivity differs among species. To date, a critical egg concentration has not yet been determined for wading birds. However, Thompson (1996) has proposed generic benchmarks. Based on a literature review, with heavy emphasis on studies of Mallards, he concluded that adverse effects were unlikely to occur in birds at egg-THg concentrations less than 0.5 µg/g, but that toxic effects were probable at concentrations greater

than 2.0 µg/g; in between was a gray area characterized by great uncertainty in terms of probability of adverse effects. Notice that the mean THg concentration in egret eggs collected 2001 was just above Thompson's estimated NOAEL for *in ova* exposure.

However, results of a recent study may suggest that Thompson's benchmark underestimates risk to the egret eggs. As a special request from FDEP (2February 26, 2001 letter from Dr. Tom Atkeson, Mercury Coordinator for FDEP), the District assisted USGS during the reporting year in a study to reduce the uncertainty and to establish a critical egg concentration for various wading birds species. To assist USGS, the District collected 168 eggs of five species (47 Great Egret eggs, 29 Anhinga eggs, 58 White Ibis eggs, 21 Tricolor heron eggs and 13 Snowy Egret eggs) and shipped them live to USGS-Patuxent (Laurel, Maryland) where they were incubated after being injected with MeHg. Preliminary results from that study suggest that the embryos of some species of fish-eating birds may be more sensitive to MeHg than are the eggs of Mallards and that estimates of harmful levels of mercury in eggs, which have been based on reproductive trials with mallards in the lab, may have to be re-evaluated (Heinz et al., 2001).

Establishing a benchmark for critical feather-THg concentration has also been difficult because of observed or suspected interspecies differences in mercury sensitivity, particularly between piscivores and non-piscivores and between freshwater birds and seabirds. This is further complicated because, unlike MeHg in eggs, MeHg bonded to keratin and sequestered in feathers no longer represents a risk to the bird. Feather-THg concentration is used only as an indicator of MeHg level and possible risk in targeted organs. However, Bouton et al. (1999) and Spalding et al. (2000) recently reported results of a controlled dosing study of Great Egrets that combined feather analysis with toxicological observations. They dosed Great Egret juveniles with MeHg-containing gelatin capsules at 0.5 mg Hg/kg food (n=5) and found subtle behavioral changes and statistically significant differences in blood chemistry, liver biochemistry and weight index (Bouton et al., 1999; Frederick et al., 1979; Spalding et al., 2000). At five weeks, chicks in this dose group had 19 µg/g THg in feathers and showed a significant decline in packed cell volume (Spalding et al., 2000). Several recent studies report Florida waterbirds having feather-THg concentrations approaching or exceeding this value (Beyer et al., 1997; Frederick et al., 1997; Sepulveda et al., 1999). As already stated, the concentration of THg did not significantly increase with bill length (i.e., age surrogate) in birds at the L67 colony in 2001. If this lack of a concentration – age relationship holds, then concentrations in feathers would not be expected to increase significantly before 5 weeks and, thus, should not exceed the lowest observed adverse effect benchmark established by Spalding et al. (2000). However, concerns have arisen regarding the range of bill sizes of sampled birds and the adequacy of the sample number to fit and test the concentration-age regression at L67 (Spalding, personal communication). Thus, uncertainty remains regarding risk to the nestlings.

To place results of feather- and egg-THg monitoring into context with results from monitoring mercury in fish, it is important to realize sampling of birds and fish were not done at the same time. The most recent results reported here are for feathers and eggs collected during the 2001 nesting season (March – April), five to six months after the September-October 2000 fish collection. Proper interpretation of the data must consider this time lag. Because of dispersal patterns of the egrets immediately prior to nesting (see discussion above and, Rumbold et al., in press), the slight increase in egg-Hg concentration observed in the egrets in 2001 may be attributable to the increased levels observed in fishes in 1999 and 2000 at more northern locations (i.e., CA2U3, L5F1, Hole Land, L38F1). Alternatively, given that nestlings are feed prey items collected within about 9 km from nesting colonies (Bancroft et al., 1990; Bancroft et al., 1994; Smith, 1995), it is much more difficult to account for the significant increases in feather-Hg in nestlings at the L67 colony. With the exception of L67F1, which despite its name is actually

located about 37 km from the bird colony, monitoring sites surrounding the colony, including sites monitored by FFWCC, i.e., their site designated L67, have shown only declines in THg levels in fishes. Nonetheless, it is possible that: (1) conditions at these sites are not representative of fishes within the immediate vicinity of the colony; (2) THg levels have increased in fishes in WCA 3A, including fish at the monitoring sites, within the 5-6 months interval between fish collection and feather collection; or (3) the egrets shifted their diets increasing their diet-weighted intake even though average levels of THg in fishes declined.

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## **WADING BIRD HABITAT AND FORAGING PATTERNS**

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Various combinations of environmental characteristics determine the suitability of an area for foraging and nesting wading birds. Among others, these characteristics include water depth, vegetation density and, densities and size distribution of the preferred prey populations. These factors have been reviewed in previous reports (Rumbold and Rawlik, 2000). In accordance with Condition (4).iv of the Mercury Monitoring Program, the District conducted a literature search for both published and unpublished studies or monitoring programs that may show possible changes in wading bird habitat and foraging patterns within the Everglades basin during the reporting year. Studies and monitoring programs identified during this search are discussed below.

From January through June 2000, researchers for the USACOE carried out systematic reconnaissance flights for wading bird activity in the WCAs and Big Cypress National Preserve (Nelson and Theriot, 2000). The Holey Land WMA and the portion of STA 1W that was the ENR Project were also surveyed. Wading birds were enumerated along parallel transects with 2-km spacing. The SRF survey methodology estimates total numbers of birds on the marsh surface, which is composed of breeding birds out feeding, nonbreeding birds, and juvenile birds. In addition, water conditions were recorded during the survey, i.e., as wet, wet transitional, dry transitional or dry.

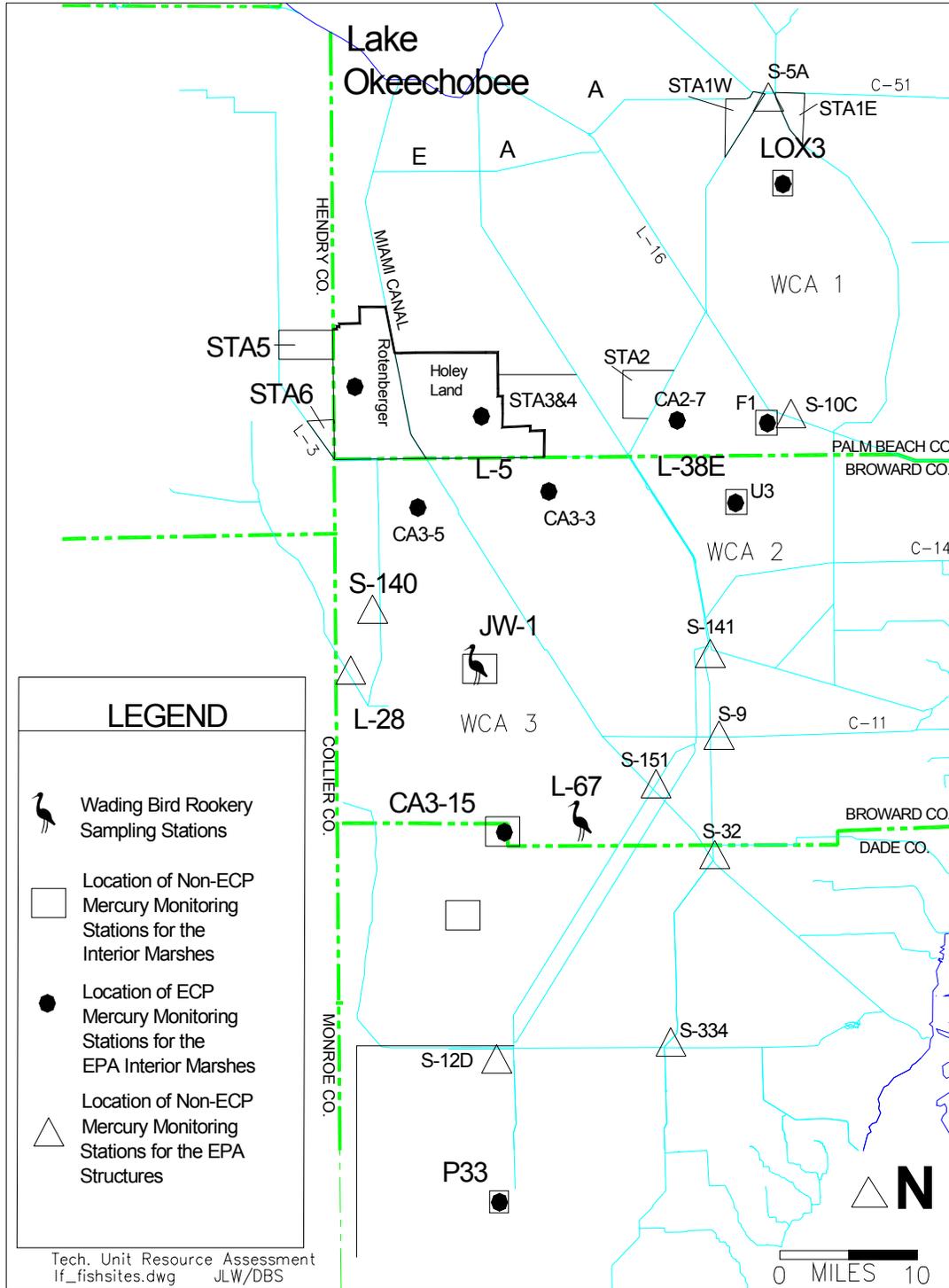
Results from SRFs carried out in 2000 showed higher numbers of birds in the basin from February-June compared to 1999, which had been elevated compared to 1998. Nelson and Theriot (2000) suggested that the pattern of rising water levels through the fall then declining to a minimum in spring allowed wading birds to take advantage of the drying conditions that concentrate prey and allowed for wading. Monthly counts ranged from 31,432 in January to a peak of 124,954 birds recorded in May; all species combined). The areas with the highest estimates of wading birds were WCA 3A for January to June. When the counts were normalized for area of each WCA, i.e., relative densities, WCA 1 and WCA 3A had the highest average monthly densities of wading birds. WCA 2B had the lowest average density. With regard to abundance at the ENR project, mean monthly number of birds was low in 2000 (42 birds) compared to previous years (1995:82 birds, 1996:174 birds, 1997:73 birds, 1998:23 birds; 1999:72 birds). The most abundant species at the ENR were Great Egrets and White Ibises. While total numbers of birds in the WCAs were much higher in 2000, spatial patterns were not dissimilar from that observed in during previous years (i.e., relative numbers of birds in the different WCAs). For example, estimated numbers of birds in WCA-2A increased from January (4,247) to February (8,046), but then dropped to just over 2,000 from March through May. In May, most of the birds within the Everglades basin were found either in WCA-1 (16,039) or WCA-3A (103,961).

In 2000, various individuals or agencies also made systematic aerial and ground surveys of nesting wading birds in South Florida (for a more detailed summary, see Gawlik, 2000). In 2000, the estimated number of wading bird nests (excluding cattle egrets) in South Florida was 39,480 (Gawlik, 2000). This represented a 40 percent increase over 1999, which had been one of the best nesting years in the past 10 years. The vast majority of nests were concentrated in WCAs, especially WCA 3A, as opposed to ENP or Florida Bay. In 2000, only 2,604 nesting attempts were recorded in WCA-1 (70 percent decrease from 1999; Thomas et al. in Gawlik, 2000). Survey personnel suspected that small flock size and low nesting efforts may have been a result of a lack of recovery in fish stocks from the previous year. Numbers of nests were further reduced following a large rain event in April 2000. By comparison, WCAs 2 and 3 combined were estimated to contain 29,728 nests, which represented a 30 percent increase over 1999 (Frederick et al. in Gawlik, 2000). As in previous years, the vast majority of nesting was concentrated in WCA-3A, with relatively little nesting in WCA-2A (only represented 4 percent of the nesting). While reproductive success was not monitored at individual nests, based on general observations at monitored colonies (i.e., maintenance of active nests, etc.), Frederick et al. (in Gawlik) concluded that nesting was largely successful throughout WCA 3 and 2. No wading bird colonies were detected again at the Holey Land and Rotenberger in 2000; probably due to the continuing dry condition at both. Likewise, nests were also not reported for any STA. While systematic ground surveys for nesting were not done in the STAs, any large nesting colony would likely have been observed during routine water quality sampling.

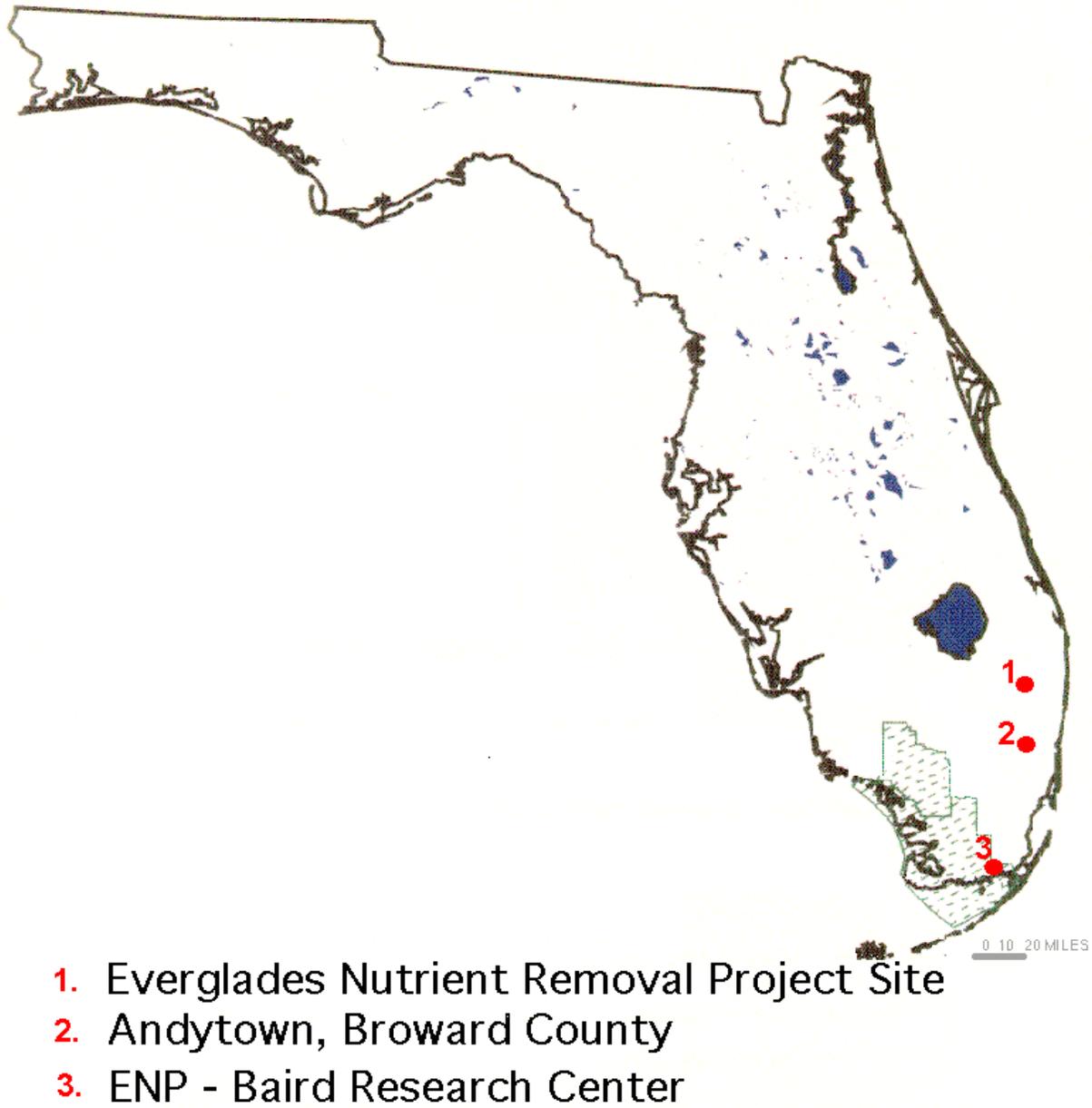
Based on a comparison of SRF results and nesting effort, Frederick et al. (2000) estimated 100 percent ("or more") of the birds in the WCAs nested in 2000. The authors acknowledged the problem in the estimate, i.e., greater than 100 percent nested, and possible observer bias that may have resulted in an underestimation of total breeding effort in the past. Interestingly, this follows recent debates regarding the proportion of birds that nest within the basin.

Gawlik (2000) suggested that a combination of three factors allowed for the increased nesting in 2000: increased number of birds in the Everglades basin (see results of SRF above) due to a drought in SE US, a very wet wet-season, and a rapid and prolonged drydown. Frederick et al. (2000) also cite hydrological factors, but also suggested that a reduction in mercury could have contributed to the increased reproductive effort and success documented in 2000. As discussed above, and elsewhere (Frederick and Spalding, 2000; Rumbold et al., 2001a; Rumbold et al., in press), mercury levels in Great Egrets declined dramatically in 1999 and 2000. It stands to reason that mercury levels have declined in other Everglades' species as well. However, there is a weakness in the theory offered by Frederick et al. (2000). The increases in nesting in 1999 and 2000 were primarily a result of an increase in nesting by White Ibises, which, owing to their lower trophic level diet, are not highly exposed to mercury. On the other hand, Great Egrets, which are more piscivorous and, thus, suffer greater exposure to mercury, have been nesting in large numbers, even meeting numeric-nesting targets set by the South Florida Ecosystem Restoration Task Force, prior to the decline in mercury. Unless, white ibises are more highly sensitive to mercury, a possibility that cannot be dismissed out of hand, the epidemiology seems inconsistent with the risk prediction.

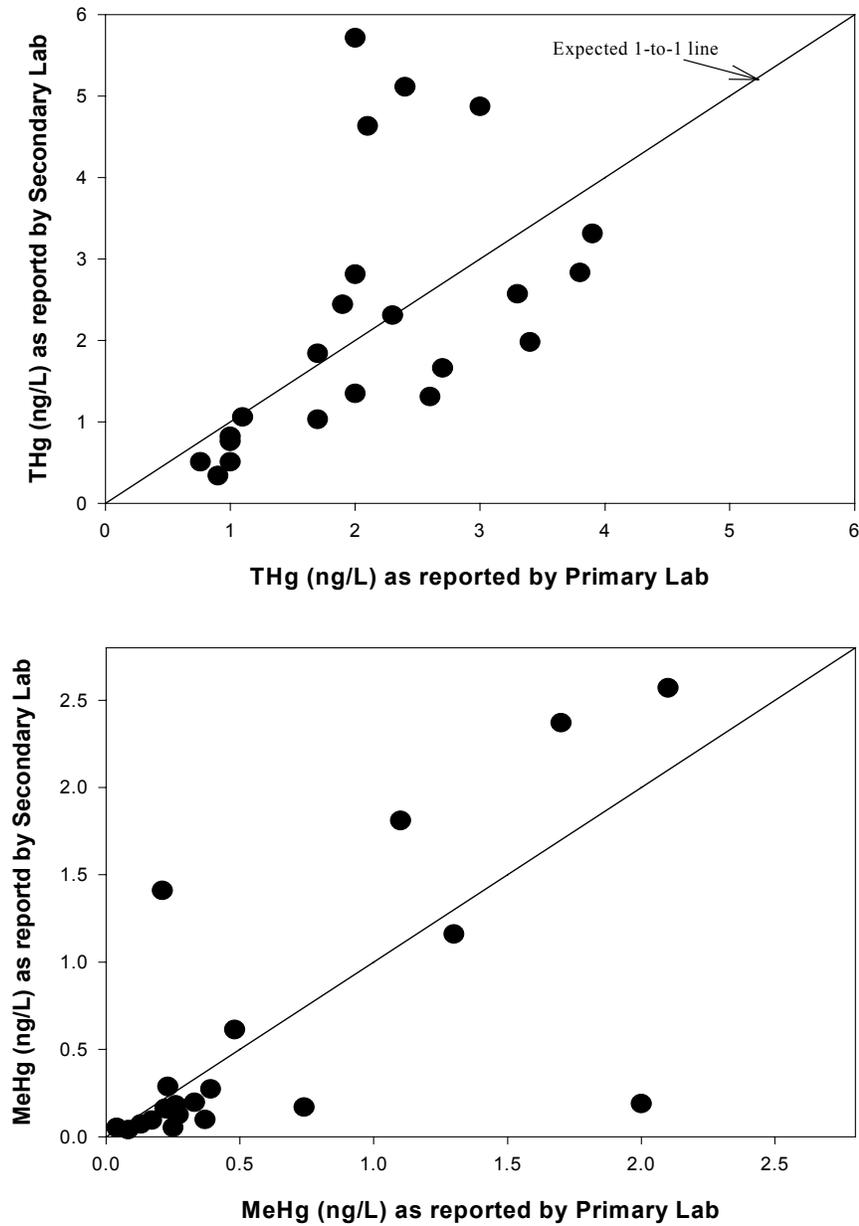
In summary, during this reporting year, the District is unaware of any evidence that would support any conclusion that wading bird foraging (or nesting) patterns have been significantly altered or impacted by construction or operation of the STAs or that such changes in foraging patterns would have led to an increased exposure to MeHg via consumption of MeHg-contaminated fish.



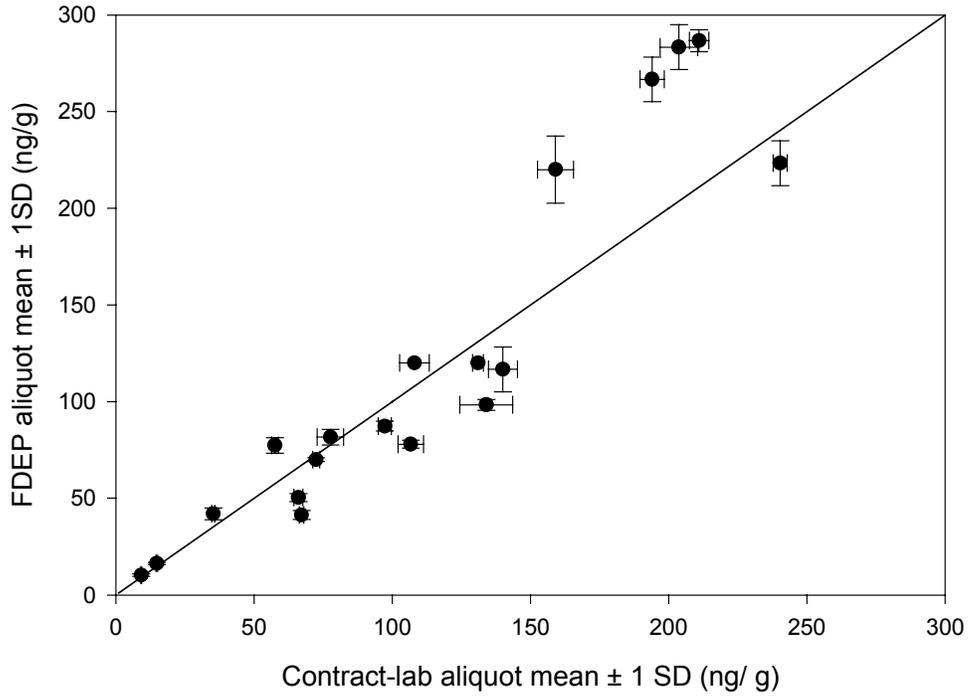
**Figure 1.** Downstream canal and interior marsh monitoring stations for water, fish and bird feathers



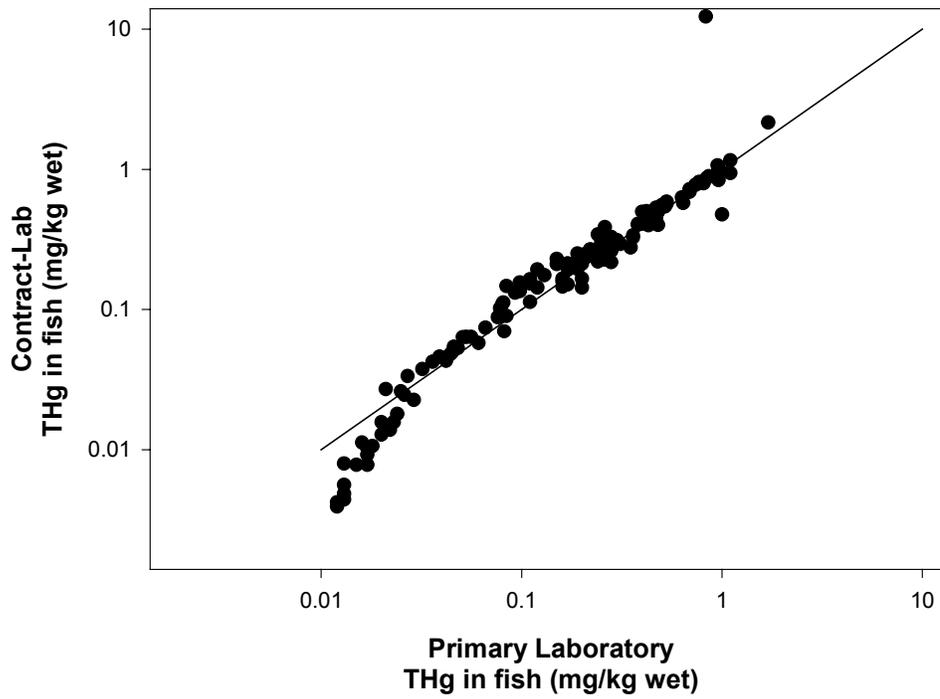
**Figure 2.** Mercury deposition network in South Florida



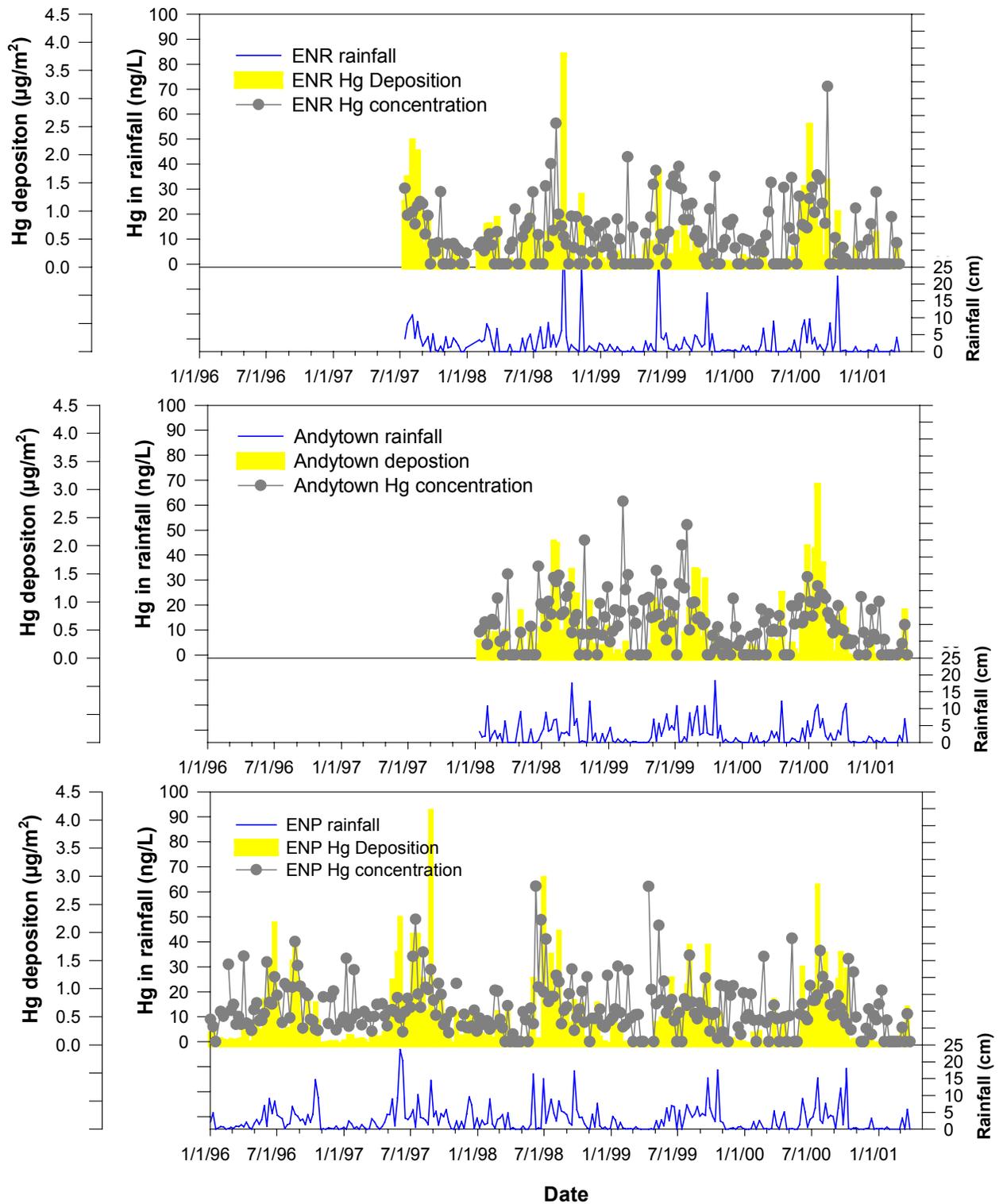
**Figure 3.** Inter-laboratory comparison for THg (a) and MeHg (b) determined in surface water splits from STA-2 and Non-ECP structures



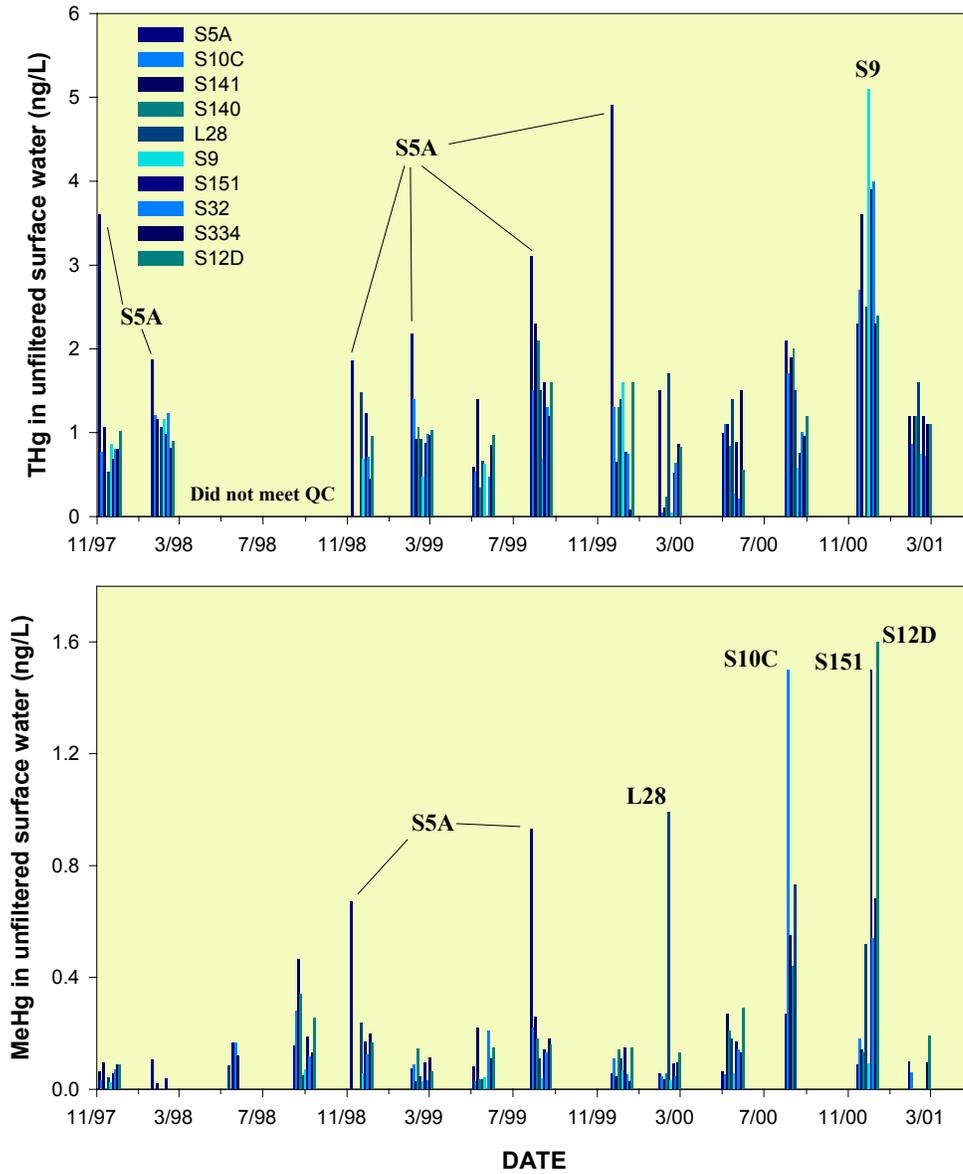
**Figure 4.** Interlaboratory comparison in determination of THg concentration in mosquitofish collected from STA-2



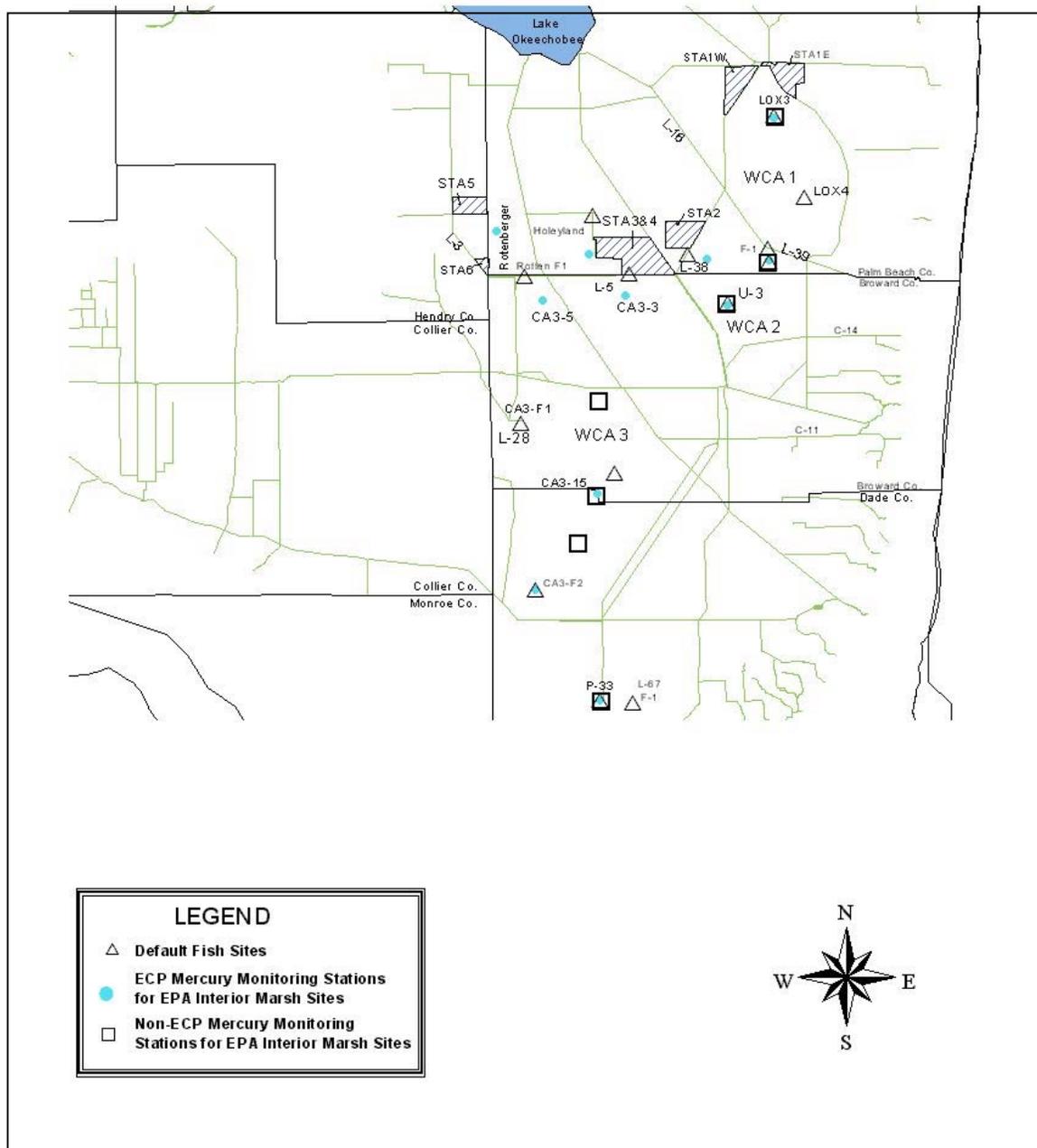
**Figure 5.** Inter-laboratory comparison in THg determination in large-bodied fishes (e.g., sunfish and Largemouth bass)



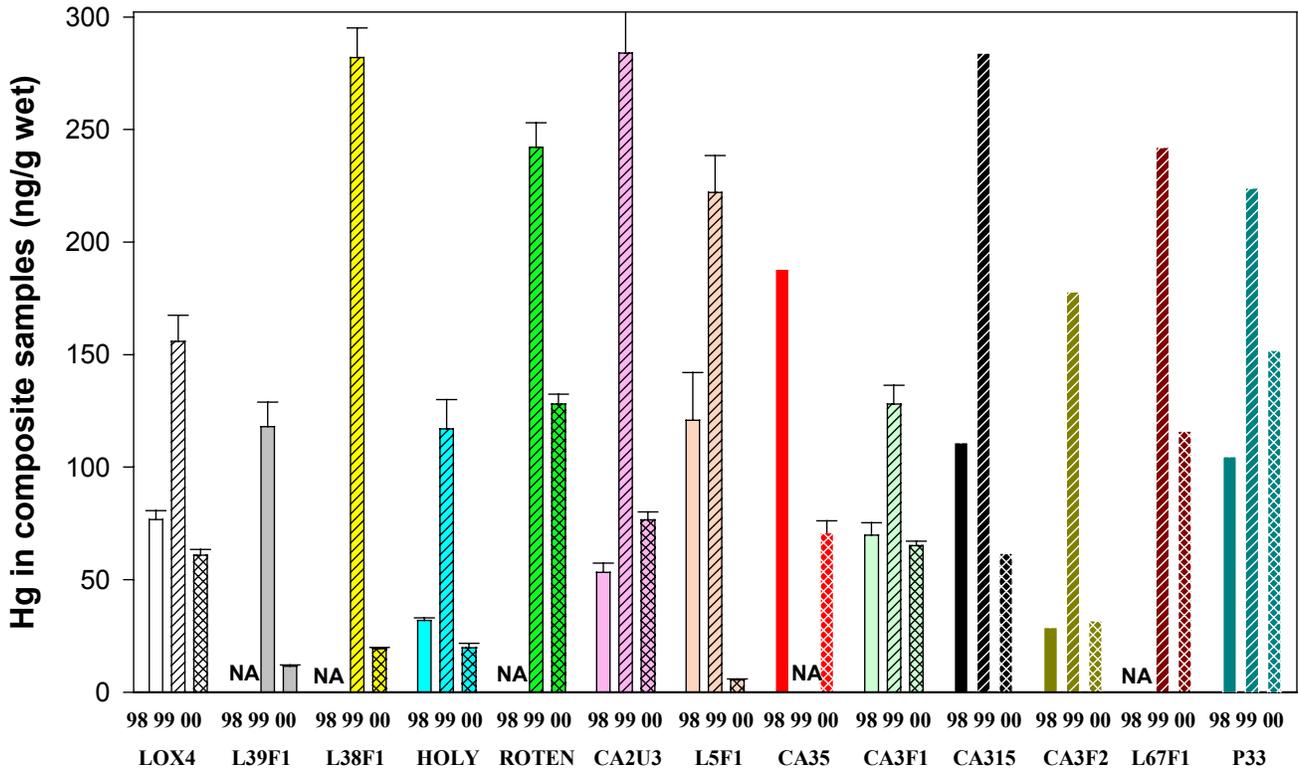
**Figure 6.** Time series of rainfall, rainfall Hg concentrations, and Hg rainfall deposition at MDN sites located at the ENR Project, Andytown and ENP Beard Research Center. All 2001 data, and 1998 – 2000 data for ENP should be considered preliminary.



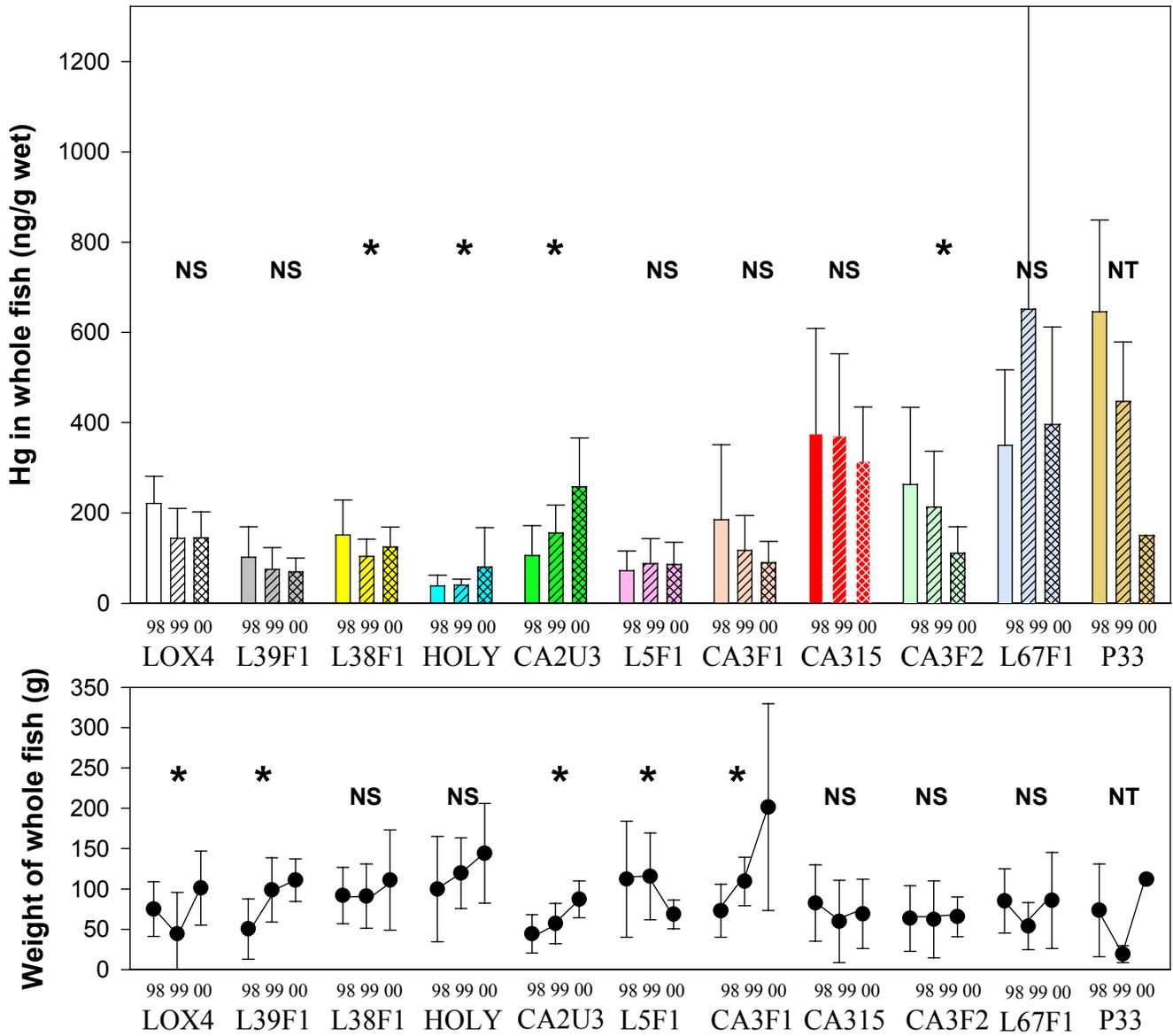
**Figure 7.** Concentrations of THg (top panel) and MeHg (bottom panel) in unfiltered surface waters at ten Non-ECP structures



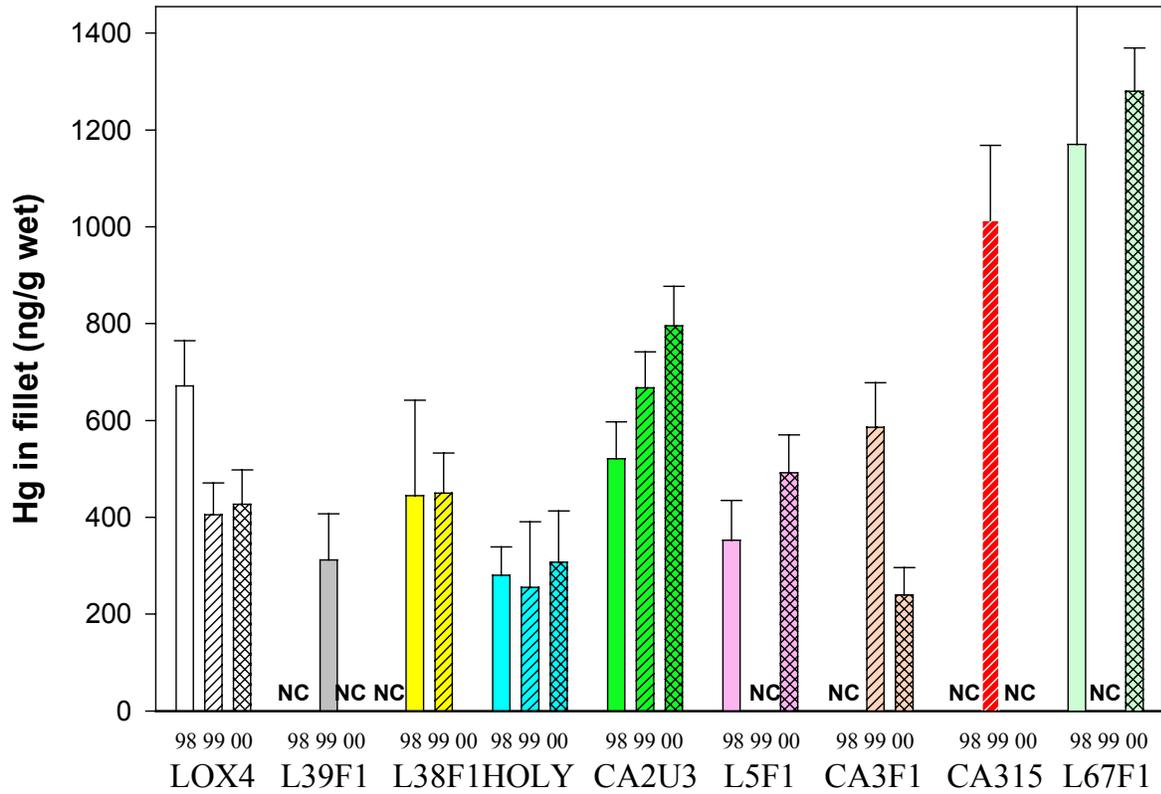
**Figure 8.** Default collection sites for large-bodied fish



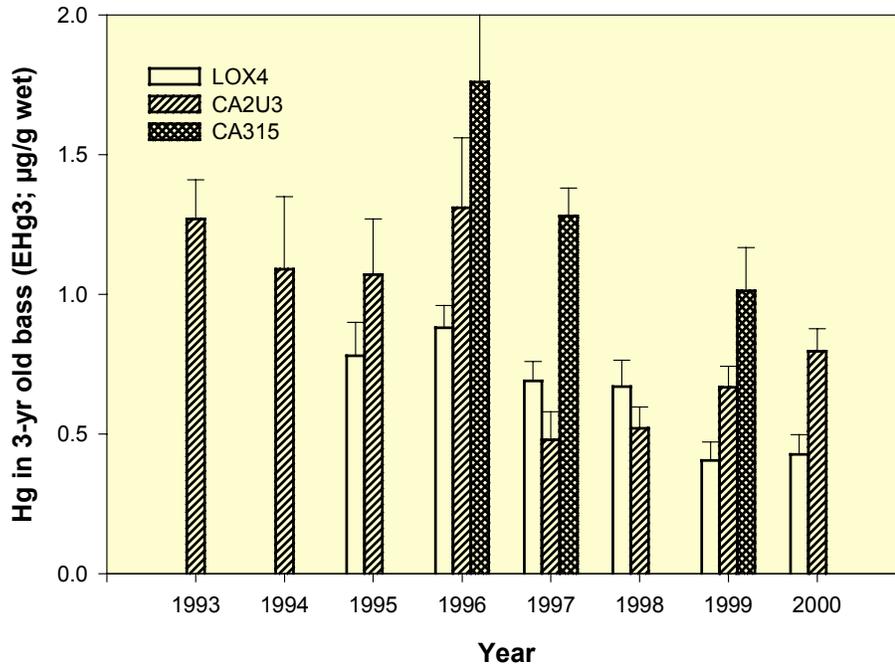
**Figure 9.** Mercury concentrations in mosquitofish (*Gambusia sp.*) collected at ECP and Non-ECP sites in 1998, 1999 and 2000. Not all sites sampled in all years (for details, refer to **Table 10**)



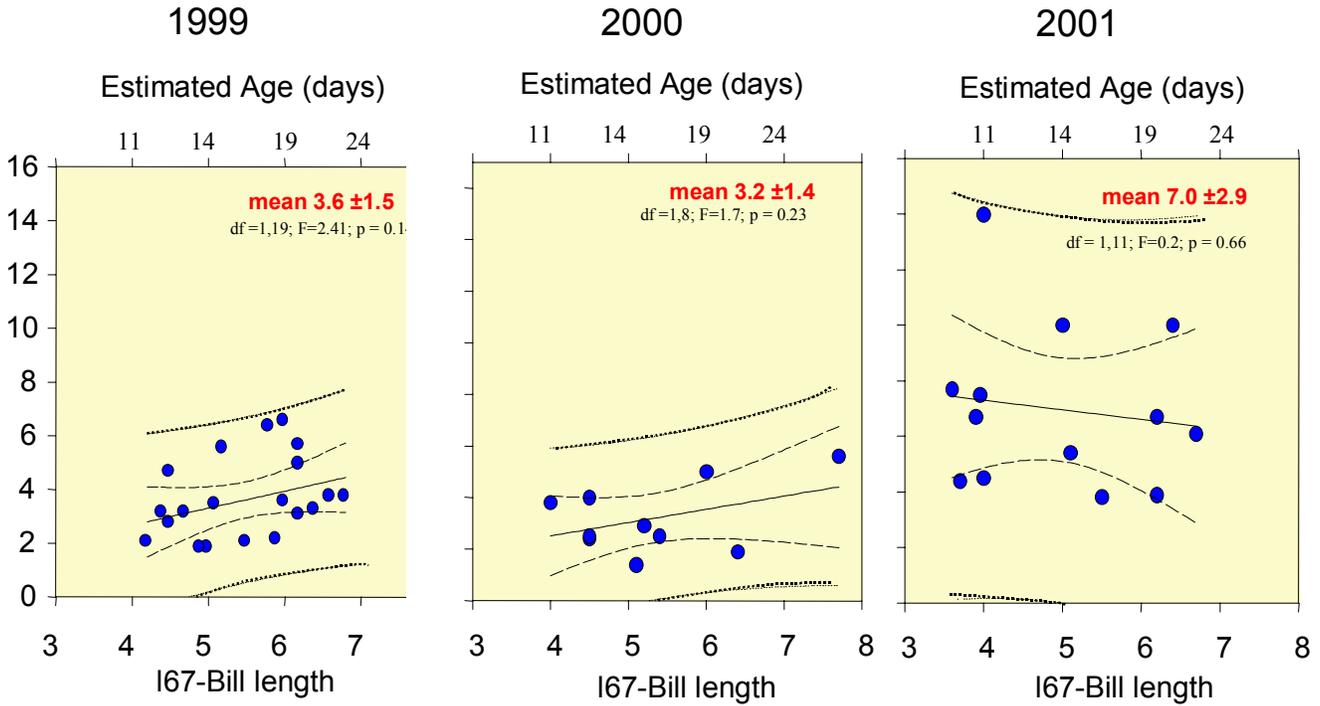
**Figure 10.** THg concentration (a) and weights (b) of whole sunfish (*Lepomis spp.*) collected at ECP and Non-ECP sites in 1998, 1999 and 2000; significant within-site among-year differences are designated by \*



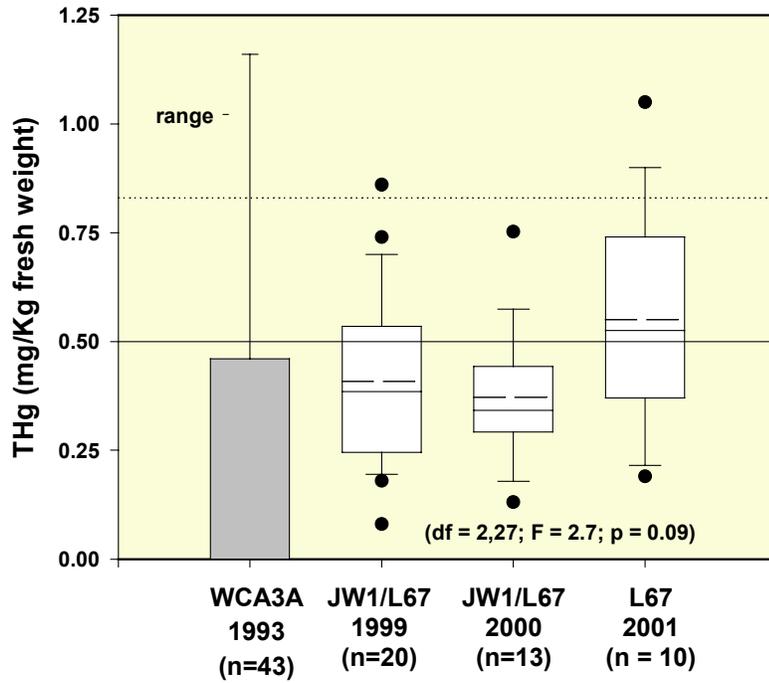
**Figure 11.** Standardized age(3) expected mercury concentration (EHg3) in Largemouth bass (*Micropterus salmoides*) collected at ECP and Non-ECP sites in 1998, 1999 and 2000; EHg3 not calculated (NC) where regressions were not significant or if age distribution was narrow



**Figure 12.** Time series of standardized age(3) expected mercury concentration (EHg3) in Largemouth bass (*Micropterus salmoides*) collected at long-term sites (data prior to 1998 from Lange et al., 1999; before 1997, CA2U3 also included bass collected nearby at the Glory Hole site)



**Figure 13.** Relationship between bill length, i.e., as an age surrogate, and THg concentration (mg/Kg) in egret nestling feathers from L67 colony; arithmetic mean and results of regressions are shown in each panel.



**Figure 14.** Boxplots of THg concentration in Great egret eggs collected from colonies within WCA-3A. Outliers that lie outside the 10<sup>th</sup> and 90<sup>th</sup> percentile shown as filled circles. Data from 1993 is from D. Day (USGS, BRD; pers. comm.) and is shown as vertical bar of mean + range.

**Table 1.** Frequency of occurrence and mean concentration (ng/L) of target analyte in field quality control (FQC) blanks collected with unfiltered surface water samples from STA1W, STA2, STA5, STA6, NON-ECP structures.

FQC*	THG						MeHg					
	n**	Collection frequency	n>MDL	ng/L	V <sup>‡</sup> flagged	% flagged	n**	Collection frequency	n>MDL	ng/L	V <sup>‡</sup> flagged	% flagged
TB	38	9%	8	0.65	6	16%	38	9%	7	0.12	6	16%
EB1	32	8%	8	0.35	3	9%	32	8%	8	0.07	5	16%
EB2	38	9%	6	0.35	5	13%	37	9%	5	0.09	3	8%
EB (unlabeled)	17	4%	3	0.16	1	6%	18	4%	5	0.07	4	22%
All EBs	87	21%	17	0.32	9	10%	87	21%	18	0.08	12	14%
FB	38	9%	6	0.27	2	5%	38	9%	4	0.21	4	11%

\*TB - trip blank, EB - equipment blank, EB1 - equipment blank collected at start of sampling, EB2 - equipment blank collected at the end of sampling, FB - field blank.

\*\* Total number (n) of unfiltered surface water samples collected under these 5 projects during the water-year was 420 THg and 417 MeHg.

‡ Indicates that the analyte was detected in the method blank.

**Table 2.** Relative percent difference (RPDs) between field duplicates as reported by primary laboratory

Analyte	Labeled FDs					Laboratory-blind FDs		
	N	Mean	Median	Max.	n*	Mean	Median	Max.
THg	33	6%	4%	31%	8	17%	15%	37%
MeHg	37	28%	17%	139%	8	62%	35%	168%

\* Frequency of blind FDs has been subsequently increased.

**Table 3.** Biweekly mean\* bulk rainfall THg concentration data (ng/L) from compliance sites of Mercury Deposition Network in reporting year ending April, 30 2001

Week ending	ENR (FL34)	Andytown (FL04)	ENP (FL11)
04/04/00	23.1	9.9	9.3
04/18/00	0.0	9.5	9.8
05/02/00	0.0	0.0	17.4
05/16/00	30.6	16.5	0.0
05/30/00	16.6	21.2	14.6
06/13/00	9.8	13.2	10.0
06/27/00	27.1	31.3	22.5
07/05/00	15.7	17.4	16.5
07/18/00	23.6	24.4	21.3
08/01/00	24.3	23.9	22.3
08/15/00	34.4	20.9	18.1
08/29/00	24.0	13.5	10.6
09/12/00	9.6	18.5	15.0
09/26/00	0.0	10.3	10.2
10/03/00	4.8	6.6	7.4
10/10/00	NC	NC	27.4
10/17/00	6.6	5.9	17.4
10/31/00	2.1	5.8	NC
11/14/00	0.0	NC	0.0
11/28/00	0.0	23.3	5.2
12/12/00	6.5	5.1	4.0
12/26/00	0.0	17.9	9.1
01/02/01	12.3	17.4	19.6
01/16/01	28.8	6.2	8.7
01/30/01	0.0	0.0	0.0
02/13/01	0.0	0.0	0.0
02/27/01	18.9	4.6	5.7
03/13/01	8.5	12.2	11.2
03/27/01	0.0	NC	0.0
Average concentration *			
2000 <sup>1</sup>	12.4 ng/L	15.8 ng/L	13.8 ng/L
Cumulative average**	11.5 ng/L	13.3 ng/L	13.1 ng/L
Seasonal Kendal <sup>2</sup>	-0.66 (p = 0.5)	-1.04 (p = 0.3)	-1.96 (p = 0.05)
Annual rainfall			
2000 <sup>1</sup>	103.9 cm	115.2 cm	143.5 cm
Cumulative average**	125.4 cm	134.6 cm	152.5 cm
Seasonal Kendal <sup>2</sup>	-2.02 (p = 0.04)	-1.56 (p = 0.12)	-0.39 (p = 0.7)
Annual deposition			
2000 <sup>1</sup>	12.8 $\mu\text{g m}^2 \text{yr}^{-1}$	18.1 $\mu\text{g m}^2 \text{yr}^{-1}$	19.8 $\mu\text{g m}^2 \text{yr}^{-1}$
Cumulative average <sup>1</sup>	13.2 $\mu\text{g m}^2 \text{yr}^{-1}$	17.9 $\mu\text{g m}^2 \text{yr}^{-1}$	20.0 $\mu\text{g m}^2 \text{yr}^{-1}$
Seasonal Kendal <sup>2</sup>	-1.0 (p = 0.3)	-2.08 (p = 0.04)	-0.78 (p = 0.4)

\* Volume-weighted mean concentration. \*\* Cumulative average based on 1998-00 for ENR, and Andytown, and 1996-99 ENP.

<sup>1</sup> January 1, 2000 – December 31, 2000.

<sup>1</sup> Annual average deposition based on 1996-00 for ENP and 1998-00 for ENR and Andytown.

<sup>2</sup> Seasonal Kendal Z-score based on monthly totals for rainfall and THg deposition, and monthly medians for THg concentration; reported p value is for a two-tailed test.

**Table 4.** Concentrations of total mercury (THg) and methylmercury (MeHg) in Non-ECP structure surface waters (units, ng/L)

Structure	Quarter	THg			MeHg		% MeHg
		ng/L	remark **	WQS*	ng/L	remark **	
<u>L28</u>	2nd Quarter	1.4		<WQS	0.180		13%
	3rd Quarter	1.5		<WQS	0.730		49%
	4th Quarter	2.5		<WQS	0.520		21%
	1st Quarter	1.6		<WQS	0.085	V	
	Average <sup>1</sup> last 4 qt. cumulative avg <sup>1</sup> .	1.75 1.35			0.48 0.26		27% 19%
<u>S10C</u>	2nd Quarter	1.1		<WQS	0.05	I	5%
	3rd Quarter	1.7		<WQS	1.50		88%
	4th Quarter	2.7		<WQS	0.18		7%
	1st Quarter	0.9		<WQS	0.06		7%
	Average last 4 qt. cumulative avg.	1.59 1.19			0.45 0.24		27% 23%
<u>S12D</u>	2nd Quarter	0.6		<WQS	0.29	A	52%
	3rd Quarter	1.2		<WQS	0.04	VI	
	4th Quarter	2.4		<WQS	1.60		67%
	1st Quarter	1.1		<WQS	0.19		17%
	Average last 4 qt. cumulative avg.	1.35 1.02			0.69 0.24		35% 21%
<u>S140</u>	2nd Quarter	0.8	A	<WQS	0.21	A	25%
	3rd Quarter	2.0		<WQS	0.44		22%
	4th Quarter	2.3	V	<WQS	0.13		
	1st Quarter	1.2		<WQS	0.12	V	
	Average last 4 qt. cumulative avg.	1.35 1.14			0.26 0.19		24% 17%
<u>S141</u>	2nd Quarter	1.1		<WQS	0.27		25%
	3rd Quarter	1.9		<WQS	0.55		29%
	4th Quarter	3.6		<WQS	0.14		4%
	1st Quarter	1.2		<WQS	0.11	V	
	Average last 4 qt. cumulative avg.	1.95 1.4			0.32 0.19		19% 14%
<u>S151</u>	2nd Quarter	0.89		<WQS	0.17		19%
	3rd Quarter	0.75		<WQS	0.095	V	
	4th Quarter	3.9		<WQS	1.5		38%
	1st Quarter	1.2	A	<WQS	0.14	V	
	Average last 4 qt. cumulative avg.	1.68 1.22			.84 0.25		29% 16%
<u>S32</u>	2nd Quarter	0.21	I	<WQS	0.14		67%
	3rd Quarter	1		<WQS	0.14	V	
	4th Quarter	4		<WQS	0.54		14%
	1st Quarter	0.72		<WQS	0.092	V	
	Average last 4 qt. cumulative avg.	1.48 1.1			.34 0.14		40% 20%

**Table 4 (Continued).** Concentrations of total mercury (THg) and methylmercury (MeHg) in Non-ECP structure surface waters (units, ng/L)

Structure	Quarter	THg		MeHg		% MeHg	
		ng/L	remark**	ng/L	remark**		
<u>S334</u>	2nd Quarter	1.5	A	<WQS	0.130	9%	
	3rd Quarter	0.96		<WQS	0.120	V	
	4th Quarter	2.3		<WQS	0.680	30%	
	1st Quarter	1.1		<WQS	0.096	A	
	Average last 4 qt.	1.46			0.302	16%	
	cumulative avg.	0.99			0.16	19%	
<u>S5A</u>	2nd Quarter	0.99		<WQS	0.064	6%	
	3rd Quarter	2.1		<WQS	0.270	13%	
	4th Quarter	2.3		<WQS	0.089	4%	
	1st Quarter	1.2	A	<WQS	0.099	8%	
	Average last 4 qt.	1.65			0.13	8%	
	Cumulative avg.	2.18			0.21	10%	
<u>S9</u>	2nd Quarter	0.27		<WQS	0.057	I	
	3rd Quarter	0.58		<WQS	0.054	VI	
	4th Quarter	5.10		<WQS	0.092	A	
	1st Quarter	0.74		<WQS	0.120	V	
	Average last 4 qt.	1.67			0.07	11%	
	Cumulative avg.	1.07			0.05	13%	
	Ann. avg <sup>1</sup> . 00-2	0.89	±0.4 (10) <sup>†</sup>		0.16	±0.1 (10)	24%
	Ann. avg. 00-3	1.37	±0.5 (10)		0.69	±0.5 (5)	40%
	Ann. avg. 00-4	3.20	±1.0 (9)		0.55	±0.6 (10)	21%
	Ann. avg. 01-1	1.09	±0.3 (10)		0.11	±0.1 (4)	10%
	Cum. avg <sup>1</sup> . 1 <sup>st</sup> Q	0.99	±0.4 (39)		0.11	±0.2 (27)	16%
	Cum. avg. 2 <sup>nd</sup> Q	0.81	±0.4 (19)		0.13	±0.1 (23)	20%
	Cum. avg. 3 <sup>rd</sup> Q	1.53	±0.6 (20)		0.32	±0.3 (25)	22%
	Cum. avg. 4 <sup>th</sup> Q	1.63	±1.3 (38)		0.22	±0.4 (39)	14%

\*Class III Water Quality Standard of 12 ng THg/L

\*\*For qualifier definitions, see FDEP rule 62-160: "A" - averaged value; "U" - undetected, value is the MDL; "I" - below PQL; "J" - estimated value, the reported value failed to meet established QC criteria; "J3" - estimated value, poor precision, "V" - analyte detected in both the sample and the associated method blank. Flagged values were not used in calculating averages.

<sup>1</sup> Averages were not volume-weighted.

<sup>†</sup> Value in parenthesis, i.e., (n), is number of unqualified values used to calculate mean ±1SD.

**Table 5.** Concentration of total mercury (THg) in mosquitofish composites (units ng/g wet weight) collected in 2000 from downstream sites. Value represents mean of 3-5 analyses

Location	Lat.	Long.	THg (ng/g)	Between-yr. change (%)	Cum. average
LOX3	26 35.750'N	80 21.330'W	NA		112
LOX4	26 27.750'N	80 17.950'W	61	-61%	98
CA2 F1	26 21.58'N	80 22.23'W	NA		40
CA2F1 Alt. (L39F1)	26 22.28'N	80 21.090'W	12	-90%	65
CA27 (Marsh)	26 22.07'N	80 30.67'W	NA		116
CA27 Alt. (L38F1)	26 20.09'N	80 32.15'W	19	-93%	151
Holey Land (North canal)	26 25.96'N	80 41.355'W	20	-83%	56
Rotenberger Alt. (RotenF1)	26 19.99'N	80 48.928'W	128	-47%	185
CA2U3	26 17.25'N	80 24.68'W	77	-73%	138
CA33	26 17.97'N	80 37.89'W	NA		
CA33 Alt (L5F1)	26 20.00'N	80 37.68'W	5	-98%	114
CA35	NA	NA	71		130
Non-ECP North (CA3F1; end of L-28)	26 05.502'N	80 49.192'W	65	-49%	88
CA315	26 00.305'N	80 38.927'W	62	-78%	152
Non ECP South (CA3F2)	25 48.748'N	80 47.629'W	32	-82%	79
P33	25 37.54'N	80 37.683'W	152	-32%	160
P33 Alt. (L67F1)	25 37.54'N	80 40.366'W	116	-52%	179
annual mean			63	-68%	114

NA = data not available due to the absence of fish at the site.

**Table 6.** Mean concentration ( $\pm$  1SD; ng/g wet weight) of total mercury (THg) in sunfish (*Lepomis spp.*) collected in 2000 from marshes within the EPA downstream of the STAs

Target location	Sampling Location	Lat. N	Long.W	Mean THg ng/g ( $\pm$ 1SD, n)	Between-yr. change (%)	Cum. Average
WCA1-LOX3	LOX4	26°27.75	80°17.95	145 ( $\pm$ 58, 20)	1%	170
WCA-2A F1	L39F1	26°21.580	80°22.230	70 ( $\pm$ 30, 20)	-7%	82
WCA-2A 2-7	L38F1	26°20.092	80°32.149	125 ( $\pm$ 44, 20)	20%	127
Holey Land	Holey Land	26°26.120	80°41.540	80 ( $\pm$ 88, 20)	100%	53
Rotenberger <sup>1</sup>				NA		
WCA-2A U3	CA2U3	26°17.250	80°26.680	258 ( $\pm$ 108, 20)	65%	173
WCA-3A 3	L5F1	26°20.004	80°37.683	86 ( $\pm$ 49, 20)	-2%	82
WCA-3A 5				218 ( $\pm$ 115, 8)		
Non-ECP North	CA3F1	26°05.502	80°49.192	89 ( $\pm$ 47, 20)	-24%	130
WCA-3A 15	CA315	26°00.305	80°38.927	314 ( $\pm$ 120, 20)	-15%	353
Non-ECP South	CA3F2	25°48.748	80°47.629	111 ( $\pm$ 58, 20)	-48%	196
ENP P33 Marsh	L67F1	25°37.540	80°40.366	396 ( $\pm$ 216, 20)	-39%	466
ENP P33 Marsh	P33 Marsh	25°37.541	80°37.683	150 ( $\pm$ NA, 1)	-66%	414

<sup>1</sup> Unable to collect 20 fish from each site.

NA = data not available due to the absence of fish at the site.

**Table 7.** Standardized (EHg3) and arithmetic mean concentrations of total mercury (THg) in largemouth bass fillets (ng/g wet weight) collected in 2000 from ECP and Non-ECP interior marsh sites

Target Location	Sampling Location	Lat. N	Long.W	EHg3 ± 95 <sup>th</sup> CI (mean ±1SD, n) ng/g wet	Consumption advisory exceeded*	Cum. mean <sup>2</sup>
CA1-LOX3	LOX4	26°27.75	80°17.95	427±71 (350±221, 20)	No	390
CA2-F1	L39F1	26°27.75	80°17.95	NC (1) (235±108, 20)		276
CA2-7	L38F1	26°20.092	80°32.149	NC (1) (413±186, 20)		516
Holeyland	HOLYBC	26°26.120	80°41.540	308±105 (462±236, 20)	No	420
Rotenberger <sup>1</sup>				NC (2) (NA, 0)		
CA2-U3	CA2U3	26°17.250	80°26.68	796±81 (809±282, 20)	Yes	592
CA3-3	L5F1	26°20.004	80°37.683	492 ±78 (486±257, 20)	No	448
CA3-5	CA3-5	26°17.970	80°51.480	NC (2) (990, 1)		990
Non-ECP North	CA3F1	26°05.502	80°49.192	240±56 (300±142, 20)	No	541
CA3-15	CA3-15	26°00.305	80°38.927	NC (2) (800±234, 3)	Likely	862
Non-ECP South	CA3F2	25°48.748	80°47.629	NC (2) (775±276, 2)	Likely	884
ENP-P33	ENP-P33	25°36.883	80°42.167	NC (2) (1,250 , 2)	Likely	1,250
ENP-P33	L67F1	25°37.540	80°40.366	1,280±89 (1,112±294, 18)	Yes	1,122

\* Florida limited fish consumption advisory threshold is 500 ng/g in 3-yr-old bass.

<sup>1</sup> Unable to collect fish from site.

<sup>2</sup> Cumulative arithmetic mean

NC - not calculated for: (1) insignificant slope or (2) if poor age distribution. NA - not available.

**Table 8.** Standardized (least square mean for a chick with a 7.1 cm bill) and arithmetic mean concentrations of THg ( $\mu\text{g/g dw}$ ) in growing scapular feathers collected annually from of great egret nestlings (2-3 weeks old) at JW1 and L67 colonies.

Colony	Least square mean $\pm$ 95 <sup>th</sup> CI (arithmetic mean $\pm$ 1SD, n)				
	1994 <sup>*1</sup>	1995 <sup>*</sup>	1999	2000	2001
JW1	21.12 $\pm$ 6.1 (25.0 $\pm$ 7.9, 9)	14.51 $\pm$ 3.31 (NA, 8)	7.18 $\pm$ 1.14 (4.0 $\pm$ 2.2, 13)	6.9 $\pm$ 1.3 (3.4 $\pm$ 1.9, 10)	Failed to initiate nesting
L67	16.29 $\pm$ 4.53 (NA, 27)	15.51 $\pm$ 6.16 (15.9 $\pm$ 6.16, 14)	NC (3.6 $\pm$ 1.5, 20)	NC (3.2 $\pm$ 1.4, 10)	NC (7.0 $\pm$ 2.9, 13)

\* Data from Frederick et al. (1997).

<sup>1</sup> Concentrations standardized to a bill length of 5.6 cm.

NC – not calculated where slope of regression was not significant ( $p > 0.05$ ).

Estimated mean age of sampled nestling, based on bill length, was 16 days in 1994, 24 days in 1995, 15 days in 1999, 16 days in 2000 and 15 days in 2001.

## ATTACHMENT 1. DATA ON INDIVIDUAL LARGE-BODIED FISH

The THg concentration (mg/Kg) and metadata for individual large-bodied fish collected in 2000 are provided in the table on the following pages.

<i>Location</i>	<i>Date</i>	<i>Sample ID</i>	<i>Species name</i>	<i>Age</i>	<i>Length (cm)</i>	<i>Weight (g)</i>	<i>THg (mg/Kg)</i>	<i>Remark</i>
CA315	07-Sep-00	900679	BLUE		159	71	0.5	
CA315	07-Sep-00	900680	BLUE		129	33	0.38	
CA315	07-Sep-00	900681	BLUE		132	28	0.19	
CA315	10-Oct-00	1000241	LMB	1	241	177	0.92	A
CA315	07-Sep-00	900554	LMB	1	245	198	0.95	
CA315	07-Sep-00	900555	LMB	0	172	70	0.53	
CA315	07-Sep-00	900664	RESU		145	63	0.26	A
CA315	07-Sep-00	900665	RESU		196	129	0.43	
CA315	07-Sep-00	900666	RESU		218	191	0.21	
CA315	07-Sep-00	900667	RESU		180	145	0.4	
CA315	07-Sep-00	900668	RESU		174	97	0.22	
CA315	07-Sep-00	900669	RESU		151	66	0.17	
CA315	07-Sep-00	900670	RESU		142	57	0.19	
CA315	07-Sep-00	900671	SPSU		165	63	0.42	
CA315	07-Sep-00	900672	SPSU		141	48	0.47	
CA315	07-Sep-00	900673	SPSU		128	45	0.24	
CA315	07-Sep-00	900674	SPSU		136	59	0.25	
CA315	07-Sep-00	900675	SPSU		129	49	0.26	A
CA315	07-Sep-00	900676	SPSU		109	28	0.25	
CA315	07-Sep-00	900677	SPSU		102	24	0.2	
CA315	07-Sep-00	900678	SPSU		122	38	0.25	
CA315	07-Sep-00	900682	WAR		166	93	0.47	
CA315	07-Sep-00	900683	WAR		140	56	0.53	
CA35	11-Oct-00	1000149	BLUE		74	8	0.12	
CA35	11-Oct-00	1000150	BLUE		74	8	0.13	
CA35	11-Oct-00	1000144	LMB	1	282	346	0.99	
CA35	11-Oct-00	1000145	RESU		181	128	0.24	
CA35	11-Oct-00	1000146	RESU		181	129	0.23	
CA35	11-Oct-00	1000148	SPSU		154	89	0.45	
CA35	11-Oct-00	1000147	WAR		159	110	0.3	
CA35	11-Oct-00	1000151	WAR		76	11	0.13	
CA35	11-Oct-00	1000152	WAR		63	6	0.14	
CA3F1	09-Oct-00	1000058	BLUE		190	136	0.1	
CA3F1	09-Oct-00	1000059	BLUE		173	100	0.09	
CA3F1	09-Oct-00	1000060	BLUE		158	78	0.089	
CA3F1	09-Oct-00	1000061	LMB	3	463	1452	0.41	A
CA3F1	09-Oct-00	1000062	LMB	5	518	2139	0.54	
CA3F1	09-Oct-00	1000063	LMB	3	380	759	0.41	
CA3F1	09-Oct-00	1000064	LMB	3	419	990	0.67	

<i>Location</i>	<i>Date</i>	<i>Sample ID</i>	<i>Species name</i>	<i>Age</i>	<i>Length (cm)</i>	<i>Weight (g)</i>	<i>THg (mg/Kg)</i>	<i>Remark</i>
CA3F1	09-Oct-00	1000065	LMB	3	431	1165	0.32	
CA3F1	09-Oct-00	1000066	LMB	3	349	610	0.27	
CA3F1	09-Oct-00	1000067	LMB	3	300	332	0.3	
CA3F1	09-Oct-00	1000068	LMB	2	361	615	0.15	
CA3F1	09-Oct-00	1000069	LMB	2	338	453	0.16	
CA3F1	09-Oct-00	1000070	LMB	3	333	546	0.21	
CA3F1	09-Oct-00	1000071	LMB	2	329	489	0.2	
CA3F1	09-Oct-00	1000072	LMB	3	369	675	0.41	
CA3F1	09-Oct-00	1000073	LMB	2	347	575	0.14	
CA3F1	09-Oct-00	1000074	LMB	3	321	428	0.34	
CA3F1	09-Oct-00	1000075	LMB	2	326	446	0.23	
CA3F1	09-Oct-00	1000076	LMB	2	331	421	0.17	
CA3F1	09-Oct-00	1000077	LMB	2	356	593	0.31	
CA3F1	09-Oct-00	1000078	LMB	3	337	467	0.3	
CA3F1	09-Oct-00	1000079	LMB	1	278	280	0.1	
CA3F1	09-Oct-00	1000080	LMB	4	337	569	0.36	
CA3F1	09-Oct-00	1000041	RESU		274	476	0.11	A
CA3F1	09-Oct-00	1000042	RESU		222	185	0.095	
CA3F1	09-Oct-00	1000043	RESU		218	196	0.095	
CA3F1	09-Oct-00	1000044	RESU		206	156	0.087	
CA3F1	09-Oct-00	1000045	RESU		216	182	0.047	
CA3F1	09-Oct-00	1000046	RESU		206	166	0.048	
CA3F1	09-Oct-00	1000047	RESU		182	114	0.12	
CA3F1	09-Oct-00	1000048	RESU		200	166	0.084	
CA3F1	09-Oct-00	1000049	RESU		221	213	0.032	
CA3F1	09-Oct-00	1000050	RESU		206	140	0.047	
CA3F1	09-Oct-00	1000051	RESU		218	204	0.029	
CA3F1	09-Oct-00	1000052	RESU		197	144	0.11	
CA3F1	09-Oct-00	1000053	RESU		142	52	0.17	
CA3F1	09-Oct-00	1000054	RESU		148	62	0.23	
CA3F1	09-Oct-00	1000055	RESU		252	400	0.065	
CA3F1	09-Oct-00	1000056	RESU		262	405	0.048	
CA3F1	09-Oct-00	1000057	RESU		271	454	0.093	
CA3F2	10-Oct-00	1000133	BLUE		148	68	0.24	
CA3F2	10-Oct-00	1000134	BLUE		168	93	0.11	
CA3F2	10-Oct-00	1000135	BLUE		135	48	0.062	
CA3F2	10-Oct-00	1000136	BLUE		128	40	0.14	
CA3F2	10-Oct-00	1000137	BLUE		148	66	0.11	
CA3F2	10-Oct-00	1000138	BLUE		129	44	0.13	
CA3F2	10-Oct-00	1000139	BLUE		125	35	0.18	
CA3F2	10-Oct-00	1000140	BLUE		114	27	0.083	
CA3F2	10-Oct-00	1000141	LMB	3	350	577	0.97	
CA3F2	10-Oct-00	1000142	LMB	1	230	154	0.58	A
CA3F2	10-Oct-00	1000121	RESU		174	105	0.12	A
CA3F2	10-Oct-00	1000122	RESU		170	89	0.13	
CA3F2	10-Oct-00	1000123	RESU		185	123	0.093	
CA3F2	10-Oct-00	1000124	RESU		168	82	0.055	
CA3F2	10-Oct-00	1000125	RESU		154	68	0.053	
CA3F2	10-Oct-00	1000126	RESU		149	61	0.04	
CA3F2	10-Oct-00	1000127	RESU		142	60	0.056	
CA3F2	10-Oct-00	1000128	RESU		140	47	0.056	

<i>Location</i>	<i>Date</i>	<i>Sample ID</i>	<i>Species name</i>	<i>Age</i>	<i>Length (cm)</i>	<i>Weight (g)</i>	<i>THg (mg/Kg)</i>	<i>Remark</i>
CA3F2	10-Oct-00	1000129	RESU		131	43	0.059	
CA3F2	10-Oct-00	1000130	SPSU		144	69	0.098	
CA3F2	10-Oct-00	1000131	SPSU		152	82	0.19	
CA3F2	10-Oct-00	1000132	SPSU		142	62	0.22	
HOLYBC	06-Sep-00	900253	BLUE		196	168	0.058	
HOLYBC	06-Sep-00	900254	BLUE		197	185	0.052	
HOLYBC	06-Sep-00	900255	BLUE		186	135	0.42	
HOLYBC	06-Sep-00	900256	BLUE		202	175	0.18	
HOLYBC	06-Sep-00	900257	BLUE		211	170	0.025	
HOLYBC	06-Sep-00	900258	BLUE		205	190	0.11	
HOLYBC	06-Sep-00	900259	BLUE		199	147	0.062	
HOLYBC	06-Sep-00	900260	BLUE		133	60	0.042	
HOLYBC	06-Sep-00	900261	BLUE		171	62	0.069	
HOLYBC	06-Sep-00	900262	BLUE		158	65	0.091	
HOLYBC	06-Sep-00	900263	BLUE		150	67	0.056	
HOLYBC	06-Sep-00	900439	LMB	8	511	2034	1.2	
HOLYBC	06-Sep-00	900440	LMB	9	413	1029	0.83	
HOLYBC	06-Sep-00	900441	LMB	3	354	592	0.27	
HOLYBC	06-Sep-00	900442	LMB	6	407	947	0.5	
HOLYBC	06-Sep-00	900443	LMB	2	355	642	0.19	
HOLYBC	06-Sep-00	900444	LMB	7	333	469	0.43	
HOLYBC	06-Sep-00	900445	LMB	6	351	517	0.53	
HOLYBC	06-Sep-00	900446	LMB	6	338	530	0.54	
HOLYBC	06-Sep-00	900447	LMB	3	327	448	0.32	
HOLYBC	06-Sep-00	900448	LMB	6	322	514	0.6	
HOLYBC	06-Sep-00	900449	LMB	3	324	453	0.27	
HOLYBC	06-Sep-00	900450	LMB	2	315	458	0.55	
HOLYBC	06-Sep-00	900451	LMB	3	335	491	0.38	A
HOLYBC	06-Sep-00	900452	LMB	3	325	469	0.39	
HOLYBC	06-Sep-00	900453	LMB	3	312	417	0.55	
HOLYBC	06-Sep-00	900454	LMB	3	318	444	0.19	
HOLYBC	06-Sep-00	900455	LMB	2	311	420	0.19	
HOLYBC	06-Sep-00	900456	LMB	3	311	336	0.43	
HOLYBC	06-Sep-00	900457	LMB	3	349	522	0.43	
HOLYBC	06-Sep-00	900458	LMB	2	303	419	0.46	
HOLYBC	06-Sep-00	900244	RESU		226	256	0.068	A
HOLYBC	06-Sep-00	900245	RESU		210	205	0.029	
HOLYBC	06-Sep-00	900246	RESU		201	190	0.026	
HOLYBC	06-Sep-00	900247	RESU		197	158	0.04	
HOLYBC	06-Sep-00	900248	RESU		211	202	0.078	
HOLYBC	06-Sep-00	900249	RESU		205	205	0.071	
HOLYBC	06-Sep-00	900250	RESU		179	123	0.039	
HOLYBC	06-Sep-00	900251	RESU		145	58	0.023	I
HOLYBC	06-Sep-00	900252	RESU		147	61	0.061	
L38F1	10-Oct-00	1000176	BLUE		170	85	0.17	
L38F1	10-Oct-00	1000177	BLUE		178	84	0.16	
L38F1	10-Oct-00	1000178	BLUE		156	73	0.19	
L38F1	10-Oct-00	1000179	BLUE		190	118	0.12	
L38F1	10-Oct-00	1000181	LMB	2	250	220	0.28	A
L38F1	10-Oct-00	1000182	LMB	1	260	223	0.3	
L38F1	10-Oct-00	1000183	LMB	2	274	237	0.7	

<i>Location</i>	<i>Date</i>	<i>Sample ID</i>	<i>Species name</i>	<i>Age</i>	<i>Length (cm)</i>	<i>Weight (g)</i>	<i>THg (mg/Kg)</i>	<i>Remark</i>
L38F1	10-Oct-00	1000184	LMB	2	292	344	0.27	
L38F1	10-Oct-00	1000185	LMB	3	356	494	0.48	
L38F1	10-Oct-00	1000186	LMB	2	272	289	0.27	
L38F1	10-Oct-00	1000187	LMB	2	285	276	0.71	
L38F1	10-Oct-00	1000188	LMB	2	300	344	0.29	
L38F1	10-Oct-00	1000189	LMB	2	279	247	0.59	
L38F1	10-Oct-00	1000190	LMB	3	270	229	0.37	
L38F1	10-Oct-00	1000191	LMB	3	278	212	0.26	
L38F1	10-Oct-00	1000192	LMB	2	246	170	0.33	
L38F1	10-Oct-00	1000193	LMB	2	253	189	0.37	
L38F1	10-Oct-00	1000194	LMB	2	282	242	0.37	
L38F1	10-Oct-00	1000195	LMB	2	277	178	0.92	
L38F1	10-Oct-00	1000196	LMB	1	232	160	0.34	
L38F1	10-Oct-00	1000197	LMB	1	244	172	0.5	
L38F1	10-Oct-00	1000198	LMB	1	245	169	0.24	
L38F1	10-Oct-00	1000199	LMB	1	242	157	0.42	
L38F1	10-Oct-00	1000200	LMB	2	248	165	0.25	
L38F1	10-Oct-00	1000161	RESU		162	76	0.086	A
L38F1	10-Oct-00	1000162	RESU		163	84	0.11	
L38F1	10-Oct-00	1000163	RESU		160	66	0.062	
L38F1	10-Oct-00	1000164	RESU		185	115	0.11	
L38F1	10-Oct-00	1000165	RESU		173	99	0.098	
L38F1	10-Oct-00	1000166	RESU		186	119	0.15	
L38F1	10-Oct-00	1000167	RESU		165	75	0.12	
L38F1	10-Oct-00	1000168	RESU		192	118	0.095	
L38F1	10-Oct-00	1000169	RESU		180	129	0.067	
L38F1	10-Oct-00	1000170	RESU		190	130	0.19	
L38F1	10-Oct-00	1000171	RESU		179	108	0.18	
L38F1	10-Oct-00	1000172	RESU		175	94	0.11	
L38F1	10-Oct-00	1000173	RESU		168	82	0.19	
L38F1	10-Oct-00	1000174	RESU		172	101	0.12	
L38F1	10-Oct-00	1000175	RESU		245	363	0.045	
L38F1	10-Oct-00	1000180	WAR		174	108	0.12	
L39F1	04-Sep-00	900029	BLUE		195	127	0.13	
L39F1	04-Sep-00	900030	BLUE		200	143	0.064	
L39F1	04-Sep-00	900031	BLUE		186	129	0.051	
L39F1	04-Sep-00	900032	BLUE		200	152	0.12	
L39F1	04-Sep-00	900033	BLUE		192	137	0.072	
L39F1	04-Sep-00	900034	BLUE		176	100	0.056	
L39F1	04-Sep-00	900035	BLUE		211	163	0.058	
L39F1	04-Sep-00	900036	BLUE		181	111	0.026	
L39F1	04-Sep-00	900037	BLUE		192	136	0.12	
L39F1	04-Sep-00	900038	BLUE		175	113	0.043	
L39F1	04-Sep-00	900039	BLUE		155	61	0.044	
L39F1	04-Sep-00	900040	BLUE		176	100	0.048	
L39F1	04-Sep-00	900271	LMB	4	450	1383	0.32	A
L39F1	04-Sep-00	900272	LMB	3	314	424	0.44	
L39F1	04-Sep-00	900273	LMB	3	329	486	0.096	
L39F1	04-Sep-00	900274	LMB	2	336	471	0.41	
L39F1	04-Sep-00	900275	LMB	2	288	367	0.31	
L39F1	04-Sep-00	900276	LMB	0	289	334	0.35	

<i>Location</i>	<i>Date</i>	<i>Sample ID</i>	<i>Species name</i>	<i>Age</i>	<i>Length (cm)</i>	<i>Weight (g)</i>	<i>THg (mg/Kg)</i>	<i>Remark</i>
L39F1	04-Sep-00	900277	LMB	2	293	366	0.15	
L39F1	04-Sep-00	900278	LMB	2	292	342	0.32	
L39F1	04-Sep-00	900279	LMB	1	284	306	0.2	
L39F1	04-Sep-00	900280	LMB	2	292	332	0.17	
L39F1	04-Sep-00	900281	LMB	1	280	304	0.25	
L39F1	04-Sep-00	900282	LMB	2	332	470	0.15	
L39F1	04-Sep-00	900283	LMB	1	250	206	0.24	
L39F1	04-Sep-00	900284	LMB	1	260	238	0.14	
L39F1	04-Sep-00	900285	LMB	2	276	279	0.4	
L39F1	04-Sep-00	900286	LMB	1	253	220	0.14	
L39F1	04-Sep-00	900287	LMB	2	261	214	0.17	
L39F1	04-Sep-00	900288	LMB	1	246	202	0.12	
L39F1	04-Sep-00	900289	LMB	1	238	186	0.21	
L39F1	04-Sep-00	900290	LMB	1	225	146	0.12	
L39F1	04-Sep-00	900021	RESU		175	98	0.035	A
L39F1	04-Sep-00	900022	RESU		168	83	0.08	
L39F1	04-Sep-00	900023	RESU		195	114	0.044	
L39F1	04-Sep-00	900024	RESU		168	83	0.059	
L39F1	04-Sep-00	900025	RESU		174	90	0.063	
L39F1	04-Sep-00	900026	RESU		179	97	0.084	
L39F1	04-Sep-00	900027	RESU		171	94	0.1	
L39F1	04-Sep-00	900028	WAR		159	88	0.1	
L5F1	06-Sep-00	900580	BLUE		140	50	0.054	
L5F1	06-Sep-00	900581	BLUE		152	61	0.045	
L5F1	06-Sep-00	900582	BLUE		152	61	0.073	
L5F1	06-Sep-00	900583	BLUE		150	56	0.041	
L5F1	06-Sep-00	900459	LMB	7	520	1558	1.4	A
L5F1	06-Sep-00	900460	LMB	3	379	693	0.39	
L5F1	06-Sep-00	900461	LMB	3	338	438	0.42	
L5F1	06-Sep-00	900462	LMB	2	340	448	0.4	
L5F1	06-Sep-00	900463	LMB	3	293	290	0.43	
L5F1	06-Sep-00	900464	LMB	3	295	272	0.75	
L5F1	06-Sep-00	900465	LMB	1	271	245	0.31	
L5F1	06-Sep-00	900466	LMB	3	291	296	0.47	
L5F1	06-Sep-00	900467	LMB	2	268	251	0.35	
L5F1	06-Sep-00	900468	LMB	2	277	249	0.33	
L5F1	06-Sep-00	900469	LMB	1	262	218	0.31	
L5F1	06-Sep-00	900470	LMB	3	280	238	0.41	
L5F1	06-Sep-00	900471	LMB	2	291	265	0.68	
L5F1	06-Sep-00	900472	LMB	0	244	180	0.33	
L5F1	06-Sep-00	900473	LMB	2	265	218	0.73	
L5F1	06-Sep-00	900474	LMB	2	257	228	0.45	
L5F1	06-Sep-00	900475	LMB	1	238	174	0.43	
L5F1	06-Sep-00	900476	LMB	1	237	172	0.57	
L5F1	06-Sep-00	900477	LMB	1	236	161	0.3	
L5F1	06-Sep-00	900478	LMB	1	227	147	0.26	
L5F1	06-Sep-00	900564	RESU		190	128	0.13	
L5F1	06-Sep-00	900565	RESU		147	59	0.023	I
L5F1	06-Sep-00	900566	RESU		155	62	0.028	A
L5F1	06-Sep-00	900567	RESU		171	91	0.035	
L5F1	06-Sep-00	900568	RESU		132	40	0.033	

<i>Location</i>	<i>Date</i>	<i>Sample ID</i>	<i>Species name</i>	<i>Age</i>	<i>Length (cm)</i>	<i>Weight (g)</i>	<i>THg (mg/Kg)</i>	<i>Remark</i>
L5F1	06-Sep-00	900569	RESU		160	65	0.08	
L5F1	06-Sep-00	900570	RESU		159	65	0.083	
L5F1	06-Sep-00	900571	RESU		172	82	0.16	
L5F1	06-Sep-00	900572	RESU		161	68	0.16	
L5F1	06-Sep-00	900573	SPSU		140	73	0.07	
L5F1	06-Sep-00	900574	SPSU		150	71	0.18	
L5F1	06-Sep-00	900575	SPSU		146	70	0.065	
L5F1	06-Sep-00	900576	SPSU		138	70	0.099	
L5F1	06-Sep-00	900577	SPSU		159	77	0.15	
L5F1	06-Sep-00	900578	WAR		149	66	0.1	
L5F1	06-Sep-00	900579	WAR		140	56	0.11	
L67F1	10-Oct-00	1000091	BLUE	2	168	91	0.44	
L67F1	10-Oct-00	1000092	BLUE	2	178	120	0.63	
L67F1	10-Oct-00	1000093	BLUE	2	171	109	0.26	
L67F1	10-Oct-00	1000094	BLUE	2	158	75	0.33	
L67F1	10-Oct-00	1000095	BLUE	2	152	67	0.54	
L67F1	10-Oct-00	1000096	BLUE	2	131	44	0.35	
L67F1	10-Oct-00	1000097	BLUE	2	134	47	0.95	
L67F1	10-Oct-00	1000098	BLUE	2	128	42	0.31	
L67F1	10-Oct-00	1000099	BLUE	2	122	33	0.17	
L67F1	10-Oct-00	1000100	BLUE	2	115	26	0.3	
L67F1	10-Oct-00	1000101	LMB	3	418	1158	1.4	A
L67F1	10-Oct-00	1000102	LMB	3	406	1002	1.6	
L67F1	10-Oct-00	1000103	LMB	4	450	1217	1.7	
L67F1	10-Oct-00	1000104	LMB	1	310	427	0.94	
L67F1	10-Oct-00	1000105	LMB	3	342	553	1.6	
L67F1	10-Oct-00	1000106	LMB	1	312	387	0.82	
L67F1	10-Oct-00	1000107	LMB	1	285	295	1	
L67F1	10-Oct-00	1000108	LMB	1	291	340	0.82	
L67F1	10-Oct-00	1000109	LMB	1	281	296	0.69	
L67F1	10-Oct-00	1000110	LMB	2	281	303	1.1	
L67F1	10-Oct-00	1000111	LMB	1	270	266	1.3	
L67F1	10-Oct-00	1000112	LMB	1	268	258	0.93	
L67F1	10-Oct-00	1000113	LMB	1	283	324	1	
L67F1	10-Oct-00	1000114	LMB	1	234	163	1.1	
L67F1	10-Oct-00	1000115	LMB	1	240	175	1.1	
L67F1	10-Oct-00	1000116	LMB	1	233	143	1.1	
L67F1	10-Oct-00	1000117	LMB	1	230	147	0.84	
L67F1	10-Oct-00	1000118	LMB	1	221	126	0.97	
L67F1	10-Oct-00	1000084	RESU	2	226	229	0.091	
L67F1	10-Oct-00	1000085	RESU	2	214	197	0.11	
L67F1	10-Oct-00	1000081	SPSU	2	125	40	0.52	
L67F1	10-Oct-00	1000082	SPSU	2	97	21	0.1	
L67F1	10-Oct-00	1000083	SPSU	2	104	21	0.34	
L67F1	10-Oct-00	1000086	WAR	2	194	184	0.58	
L67F1	10-Oct-00	1000087	WAR	2	151	85	0.56	A
L67F1	10-Oct-00	1000088	WAR	2	164	111	0.35	
L67F1	10-Oct-00	1000089	WAR	2	151	77	0.34	
L67F1	10-Oct-00	1000090	WAR	2	158	95	0.65	
LOX4	04-Sep-00	900015	BLUE		163	80	0.11	
WCA2U3	05-Sep-00	900048	BLUE		197	142	0.35	

<i>Location</i>	<i>Date</i>	<i>Sample ID</i>	<i>Species name</i>	<i>Age</i>	<i>Length (cm)</i>	<i>Weight (g)</i>	<i>THg (mg/Kg)</i>	<i>Remark</i>
WCA2U3	05-Sep-00	900049	BLUE		153	56	0.16	
WCA2U3	05-Sep-00	900050	BLUE		193	135	0.24	
WCA2U3	05-Sep-00	900051	BLUE		188	90	0.28	
WCA2U3	05-Sep-00	900052	BLUE		168	74	0.21	
WCA2U3	05-Sep-00	900534	LMB	8	526	1790	1.7	

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