Appendix 4-4: Annual Permit Compliance Monitoring Report for Mercury in the Stormwater Treatment Areas

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KEY FINDINGS AND OVERALL ASSESSMENT

This report summarizes data from compliance monitoring of mercury (Hg) storage, release, and bioaccumulation in Stormwater Treatment Areas (STAs) during Water Year 2004 (WY2004) (May 1, 2003 through April 30, 2004).

Key findings are as follows:

1. **All STAs:** There were no violations of the Florida Class III numerical water quality standard (WQS) of 12 nanograms (ng) of total mercury per liter (THg/L) during the reporting year at any of the STAs. As such, the project has met the requirements of Section 6.i of the Hg monitoring program of the referenced permits.

2. **STA-1W:** Stormwater Treatment Area 1 West (STA-1W) subsumed the Everglades Nutrient Removal (ENR) Project in April 1999; the ENR project had served as the prototype STA and had been in operation since 1994. After nine years of operation, this STA continued to have only low concentrations of both total mercury (THg) and methylmercury (MeHg) in surface water and consistently exhibited a negative percent change in both THg and MeHg (i.e., concentrations in the outflows were consistently lower than in the inflow). Furthermore, MeHg bioaccumulation in resident large-bodied fishes (e.g., sunfish and largemouth bass) has remained relatively constant over the monitoring period at levels lower than observed in fishes from the other STAs and the downstream Everglades. Levels of Hg in these fish would not appear to exceed Florida's 0.5 parts per million (ppm) threshold for a limited fish consumption advisory to protect human health (note that only the Florida Department of Health has the authority to make a formal determination and issue an advisory). Moreover, Hg levels in fish at this STA do not appear to pose a threat to fish-eating wildlife based on the U.S. Fish and Wildlife Service (USFWS) and the U.S. Environmental Protection Agency (USEPA) predator protection criteria.

3. **STA-1E**: Soil cores were collected from Stormwater Treatment Area 1 East (STA-1E) in March 2004, prior to initial flooding. Although the THg maxima was elevated compared to averages from other STAs, MeHg concentrations were relatively low. In particular, both the median absolute concentration of MeHg and the median %MeHg (i.e., percent of THg that is

in the methyl form) were significantly lower at STA-3/4, than cores taken at STA-2 prior to its start-up. As of April 30, 2004, start-up monitoring of Hg had not begun at STA-1E.

4. **STA-2:** Water column concentrations of both THg and MeHg have declined dramatically at STA-2 since its start-up in 2000; however, both constituents continue to be at greater concentration in the outflow than in the inflow. Tissue-Hg levels have also declined dramatically in mosquitofish from Cell 1 and the discharge canal, the two areas which previously had shown anomalously high bioaccumulation. These reductions in tissue-Hg levels in mosquitofish are expected to propagate up the food chain over time. As yet, Hg levels have not declined significantly in sunfish from either Cell 1 or the discharge canal. Alternatively, sunfish from Cell 2 have shown a progressive downward trend in Hg, with levels declining first in 2002, and then again in 2003. Largemouth bass caught in the discharge canal in 2003 were also found to contain significantly less Hg than fish collected from the discharge canal in either 2002 or 2001. However, Hg levels in these bass continue to exceed Florida's threshold for a limited fish consumption advisory to protect human health. In accordance with a special condition in the August 2001 permit modification, a probabilistic ecological risk assessment of Hg exposures was completed for STA-2 and submitted to the FDEP in March 2004. The assessment estimated the risks posed by MeHg to the great egret (*Ardea alba*) and the bald eagle (*Haliaeetus leucocephalus*). Based on 2003 data, the likelihood that MeHg exposures to birds foraging throughout STA-2 would exceed the effects threshold was low. By comparison, birds foraging exclusively from Cell 1 or the discharge canal were predicted to experience greater exposures and, as a consequence, were at some elevated risk. However, this worst case risk was comparable or lower than risk levels encountered in nearby Water Conservation Areas (WCAs) or the Everglades National Park (ENP or Park). Appendix 2B-1 of the *2005 South Florida Environmental Report – Volume I* (2005 SFER) discusses the results of the final year of the Mercury Special Studies (MSS) to more accurately characterize the anomalous Hg behavior of STA-2.

5. **STA-3/4:** Soil cores were collected from STA-3/4 in June 2003. Although sediment-THg concentrations were within the range observed at other STAs, levels of MeHg were elevated, particularly in cores from Cells 1B and 2B. As a consequence, %MeHg was also elevated in cores from STA-3/4; median %MeHg at STA-3/4 was second only to that observed at STA-2. While Cell 1 satisfied start-up Hg criteria in January 2004, both THg and MeHg remained at higher concentrations in the Cell 3 interior marsh than the two inflows. However, it should be noted that concentrations at STA-3/4 have never approached levels observed during the start-up of STA-2 Cell 1. Routine operational monitoring of Cell 1 was initiated in the first quarter of 2004. At that time, both THg and MeHg were at greater concentration in the outflow than in the inflow and were slightly elevated as compared to other STAs. Due to dry-season low-water conditions, expanded monitoring, pursuant to special conditions of the permit modification authorizing flow-through operation of Cell 3 (March 2004), was not fully implemented until June 2004, which was outside of the current reporting year. Resident fishes were not sampled from this STA in WY2004.

6. **STA-5:** Water column concentrations of both THg and MeHg remained low at STA-5 in WY2004, relative to spikes observed during the previous year. Whereas THg exhibited a negative percent change across the STA, MeHg was consistently at greater concentration in the outflow than in the inflow and thus exhibited a slight positive percent change for the year. Likewise, during the second half of the water year, mosquitofish near the outflows contained higher Hg levels than fish from the inflows. Despite good-faith efforts, the Florida Fish and Wildlife Conservation Commission (FWC), under contract to the South Florida Water Management District (District or SFWMD), was unable to fill sampling quotas for

large-bodied fish at STA-5 again in WY2004. Of the sunfish collected during the reporting year, Hg was found to be at significantly higher levels in fish from Cell 1B than in fish from the supply canal, but not in fish from the discharge canal. However, more importantly, Hg levels were found to have declined in sunfish from the discharge canal in WY2004 as compared to WY2002. In addition to hampering trend analysis, data gaps in Hg levels in large-bodied fishes also increased uncertainty in risk assessments. However, if the preferred prey fish are not present, as suggested by electrofishing results, then exposure cannot occur and thus no risk. Nonetheless, when available data on fish were evaluated against the USFWS and USEPA criteria, fish-eating wildlife foraging preferentially at STA-5 would not appear to be at an elevated risk from Hg exposure.

7. **STA-6:** Water column concentrations of both THg and MeHg remained at low concentrations at STA-6 in WY2004. Moreover, after almost six years of operation, concentrations of both THg and MeHg exhibited a negative percent change across the STA (i.e., in terms of annual and cumulative medians), suggesting that it was a net sink for both constituents. Alternatively, large-bodied fish (both sunfish and bass) from the discharge canal were again found to have higher Hg levels than fish from the supply canal (i.e., positive percent change). This spatial pattern at STA-6 has been consistent over the length of the monitoring period. Yet, resident fishes (e.g., mosquitofish, sunfish, bass) from the interior marshes contained less Hg than fish from either the supply or the discharge canals. Furthermore, when data on sunfish from Cells 3 and 5 were pooled, levels were found to be lower last year than in 2000 or 2001. Although temporal trend analysis of Hg levels in bass was hampered by data gaps in the period of record (for each of the two cells), bass from Cell 5 also showed a decline; fish collected in 2003 contained much less Hg than fish collected in 2001. Equally important, for the first time, bass collected from the discharge canal in 2003 contained less Hg than fish collected from the same canal in 1998. Therefore, it appears that Hg has declined in these fish and is now lower than baseline levels. However, it should be noted that these levels of Hg in the discharge canal bass population continue to exceed, albeit slightly, Florida's threshold for a limited fish consumption advisory to protect human health. In terms of the risk to fish-eating wildlife, only sunfish from the discharge canal exceeded either the USFWS or the USEPA criteria and, thus, it appears that the risk of Hg exposure to fish-eating wildlife foraging preferentially at STA-6 has declined.

INTRODUCTION

This is the annual permit compliance monitoring report for mercury (Hg) in Stormwater Treatment Areas (STAs). This report summarizes the Hg-related reporting requirements of the Florida Department of Environmental Protection (FDEP) Everglades Forever Act (EFA) permits [Chapter 373.4592, Florida Statutes (F.S.)], including permits for STA-1W, STA-2, STA-3/4, STA-5, and STA-6 (Nos., 503074709, 0126704, 192895, 0131842, and 2629183090, respectively). This report summarizes the results of monitoring in Water Year 2004 (WY2004) (May 1, 2003 through April 30, 2004). The results of Hg monitoring at sites downstream of the STAs in accordance with these permits, as well as non-Everglades Construction Project (non-ECP) discharge structures (see Permit No. 06,502590709; and marshes) will be reported separately in Appendix 2B-1 of the *2005 South Florida Environmental Report – Volume I* (2005 SFER).

This report consists of key findings and an overall assessment, an introduction and background, a summary of the Mercury Monitoring and Reporting Program, and monitoring results. The background section briefly summarizes previously identified concerns regarding possible impact of STA operation on South Florida's Hg problem. The following section summarizes sampling and reporting requirements of the Mercury Monitoring Program within the STAs. Monitoring results are summarized and discussed in two subsections: (1) results from pre-operational monitoring, and (2) results from STA operational monitoring. Recent results from the Mercury Monitoring and Reporting Program describe significant spatial distributions and, in some instances, between-year differences in Hg concentrations.

BACKGROUND

The STAs are constructed wetlands designed to remove phosphorus from stormwater runoff originating from upstream agricultural areas and Lake Okeechobee releases. The STAs are being built as part of the Everglades Construction Project (ECP) authorized under the Everglades Forever Act [EFA; Section 373.4592, Florida Statutes (F.S.)]. When completed, the ECP will include six large treatment marshes totaling about 47,000 acres.

Even prior to passage of the EFA, concerns were being raised that, in attempting to reduce downstream eutrophication, the restoration effort could inadvertently worsen the Hg problem known to be present in the Everglades (Ware et al., 1990; Mercury Technical Committee, 1991). This stemmed from studies in other areas that showed flooded soils in new impoundments to be a source of inorganic Hg (Cox et al., 1979). Of greater concern, studies had shown wetlands to be an important site of Hg methylation; methylmercury (MeHg) is the more bioaccumulative and toxic form of Hg (St. Louis et al., 1994; for review, see Rudd, 1995). Decomposition of flooded terrestrial vegetation and soil carbon in new reservoirs had been reported to stimulate the sulfate-reducing bacteria (SRB) that methylate inorganic Hg (Kelly et al., 1997; Paterson et al., 1998). Environments that favor methylation drive bioaccumulation. For example, Paterson et al. (1998) found that annual fluxes of MeHg increased 10 to 100 times through a zooplankton community after impoundment. Newly created reservoirs have also been found to contain fish with elevated Hg burdens (Abernathy and Cumbie, 1977; Bodaly et al., 1984; Bodaly and Fudge, 1999). This so-called "reservoir effect" can occasionally persist for several decades after initial flooding (Bodaly et al., 1984; Verdon et al., 1991; for review, see Fink et al., 1999). For instance, Verdon et al. (1991) reported that Hg levels in northern pike (*Esox lucius*) increased from 0.61 to 2.99 parts per million (ppm) and were still increasing nine years after initial flooding. Given these observations, Kelly et al. (1997) recently recommended that in siting a new reservoir 1) total land area flooded should be minimized and 2) flooding the wetlands, which contain larger quantities of organic carbon than the uplands, should be avoided.

However, applying these observations directly to the Everglades was problematic because most of these observations were made in deep water lakes or reservoirs in temperate regions. In a report to the SFWMD, Watras (1993) stated that "the boreal and temperate watersheds, wetlands and reservoirs studied to date are very different geologically, hydrologically, meteorologically and ecologically from the subtropical systems in the Everglades." He recommended monitoring and integrating mass balance and process-oriented studies to improve our understanding of how the system would behave. Such studies were initiated in 1994 with the start-up of the prototype STA, the Everglades Nutrient Removal (ENR) Project. Baseline collections at the ENR Project (funded by the SFWMD and others) found no evidence of MeHg spikes in either surface water (PTI, 1994 attributed to KBN, 1994a; Watras, 1993; 1994) or resident fishes (mosquitofish and largemouth bass; PTI, 1994 attributed to KBN, 1994b). During the first two years of operation, median concentrations of THg and MeHg in unfiltered surface water were reported to be 0.81 and 0.074 nanograms per liter (ng/L), respectively (Miles and Fink, 1998). These low levels persisted in later years; from January 1998–April 1999, median water column concentrations in the interior marsh (i.e., excluding inflows and outflows) were 0.81 ng THg/L and 0.04 ng MeHg/L (Rumbold and Fink, 2002b). Resident fishes also continued to have only low Hg levels: 8 to 75 nanograms per gram (ng/g) in mosquitofish, and 100 to 172 ng/g in three-year-old bass (Miles and Fink, 1998; SFWMD, 1999b; Lange et al., 1999). Finally, a mass balance assessment found the ENR Project to be a net sink for both THg and MeHg, removing approximately 70 percent of the inflow mass (Miles and Fink, 1998).

Nonetheless, to provide continuing assurance that the ECP does not exacerbate the Hg problem, construction and operating permits for the STAs, issued by the FDEP, require the SFWMD to monitor levels of THg and MeHg in various abiotic (e.g., water and sediment) and biotic (e.g., fish and bird tissues) media, both within the STA and the downstream receiving waters. Florida has a numerical water quality criterion for THg, 12 ng/L or parts per trillion (ppt), but does not have a numerical standard for MeHg [62-302.530, Florida Administrative Code (F.A.C.)]. However, a narrative standard (Line 62, 62-302.530, F.A.C.) states that substances in concentrations which injure, are chronically toxic, or produce adverse physiological or behavioral response in humans, plants, or animals shall not be present. The U.S. Environmental Protection Agency (USEPA), recognizing the limited utility of its recommended water quality criterion for the protection of human health, recently published guidance on a new criterion expressed not as a water column concentration of Hg, but as a concentration of Hg in fish tissue (0.3 mg/kg in fish tissue; USEPA, 2001); however, this guidance has yet to be adopted by rule in Florida.

SUMMARY OF THE MERCURY MONITORING AND REPORTING PROGRAM

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. This was submitted by the SFWMD to the FDEP, the USEPA, and the U. S. Army Corps of Engineers (USACE) 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 SFWMD's Quality Assurance Project Plan (QAPP) for the Mercury Monitoring and Reporting Program, which was approved on issuance of the permit by the FDEP. QAPP revisions were approved by the FDEP on June 7, 1999.

EVERGLADES MERCURY BASELINE MONITORING AND REPORTING REQUIREMENTS

Levels of THg and MeHg in the pre-operational soils of each of the STAs and various compartments (media) of the downstream receiving waters define the baseline condition from which to evaluate Hg-related changes, if any, brought about by the operation of the STAs. The pre-ECP Hg baseline conditions are defined in the Everglades Mercury Background Report, which summarizes all of the relevant Hg studies conducted in the Everglades through July 1997. This was during the construction, but prior to the operation of, the first STA. Originally prepared for submittal in February 1998, the report was revised to include the most recent data released by the USEPA and the U. S. Geological Survey (USGS) and was submitted in February 1999 (FTN Associates, 1999).

PRE-OPERATIONAL MONITORING AND REPORTING REQUIREMENTS

Prior to completion of construction and flooding of the soils of each STA, the District is required to collect 10-cm core samples of soil at six representative interior sites for THg and MeHg analyses. Prior to initiation of discharge, the District is also required to collect biweekly samples of supply canal and interior unfiltered water for THg and MeHg analyses. When concentrations at the interior sites are found not to be significantly greater than that of the supply canal, this information is reported to the permit-issuing authority, and the biweekly sampling can be discontinued. Discharge begins after all the start-up criteria are met.

OPERATIONAL MONITORING

Following approval for initiation of routine operation of an STA and, thereafter, the permits require that the following samples be collected at the specified frequencies and analyzed for specified analytes:

Water

On a quarterly basis, 500-ml unfiltered grab samples of water are collected in pre-cleaned bottles using the ultraclean technique at the supply canals and outflows of each STA. They are analyzed for MeHg and THg (this includes the sum of all Hg species in sample, e.g., Hg^0, Hg^I, and Hg^{II}, as well as organic Hg). THg results are compared with the Florida Class III water quality standard of 12 ng/L to ensure compliance. Outflow concentrations of both THg and MeHg are compared to concentrations at the supply canal.

Sediment

Triennially, sediment cores are collected at depth from 0 to 10 cm at six representative interior sites. Each depth-homogenized core is then analyzed for THg and MeHg.

Prey Fish

Semiannually, grab samples of between 100 and 250 mosquitofish (*Gambusia* sp.) are collected using a dip net at the supply canal sites, interior sites, and outflow sites of each STA. Individuals are composited from each size, and the homogenate is subsampled in quintuplicate. Each subsample is then analyzed for THg. On March 5, 2002, the FDEP approved a reduction in the number of replicate analyses of the homogenate from five to three (correspondence from F. Nearhoof, FDEP).

Top Predator Fish

Annually, 20 largemouth bass are collected primarily via electroshocking methods at representative supply canal and discharge canal sites and representative interior sites in each STA. The fish muscle (fillet) samples are analyzed for THg as an indicator of potential human exposure to Hg.

In 2000, the District began routine collection of sunfish at the same frequency, intensity (i.e., n = 20), and locations as largemouth bass. This permit revision fulfilled a USFWS recommendation (USFWS recommendation 9b in USACE Permit No. 199404532; for details, see correspondence to Bob Barron, USACE, dated July 13, 2000). Sunfish, analyzed as whole fish, also serve as a surrogate for attempts to monitor Hg in wading birds that do not nest in the STAs. (For details on the monitoring program tracking Hg in wading birds in downstream areas, see Appendix 2B-1 of this volume.) The addition of sunfish to the compliance monitoring program was approved by the FDEP on March 5, 2002 (correspondence from F. Nearhoof, FDEP).

It should be recognized that tissue concentrations in each of the three monitored fishes will reflect ambient MeHg levels, i.e., integrate exposure, as a function of a combination of factors including body size, age, rate of population turnover, and trophic position. Mosquitofish should respond rapidly to changing ambient MeHg concentrations due to their small size, lower trophic status, short life span, and rapid population turnover. Alternatively, owing to their specific life history characteristics, sunfish and bass should take a greater amount of time to respond, in terms of tissue concentrations, to changes in ambient MeHg availability. Most importantly, they represent exposure at higher trophic levels with a requisite time lag for trophic exchange. Furthermore, this focus on a three-year old bass, while appropriate to assess exposure to fishermen, complicates interpretation because its tissue concentration will reflect integration over a three-year period. The key is to use these species-related differences to better assess MeHg availability within the system.

It is important to also recognize that virtually all (> 85 percent) of the Hg in fish muscle tissues is in the methylated form (Grieb et al., 1990; Bloom, 1992). Therefore, the analysis of fish tissue for THg, which is a more straightforward and less costly procedure than for MeHg, can be interpreted as being equivalent to the analysis of MeHg. Further details regarding rationales for sampling scheme, procedures, and data reporting requirements are set forth in the Everglades Mercury Monitoring Plan revised in March 1999 (Appendix 1 of QAPP, June 7, 1999).

QUALITY ASSURANCE MEASURES

For a quality assurance/quality control (QA/QC) assessment of the District's Mercury Monitoring Program during WY2004, see Appendix 2B-1 of this volume.

STATISTICAL METHODS

The proper interpretation of residue levels in tissues can sometimes prove problematic due to the confounding influences of age or species of collected animals. For comparison, special procedures are used to normalize the data (Wren and MacCrimmon, 1986; Hakanson, 1980). To be consistent with the reporting protocol used by the FWC (Lange et al., 1998 and 1999), Hg concentrations in largemouth bass were standardized to an expected mean concentration in three year-old fish at a given site by regressing Hg against age (hereafter symbolized as EHg3). To adjust for the month of collection, otolith ages were first converted to decimal ages using protocols developed by Lange et al. (1999). Sunfish were not aged, so age normalization was not available. Instead, arithmetic means were reported. However, efforts were made to estimate a least square mean (LSM) Hg concentration based on the weight of the fish. Additionally, the distribution of the different species of *Lepomis* (warmouth, *L. gulosus*; spotted sunfish, *L. punctatus*; bluegill, *L. macrochirus*; and red ear sunfish, *L. microlophus*) that were collected during electroshocking was also considered (qualitatively) as a potential confounding influence on Hg concentrations prior to each comparison.

Where appropriate, analysis of covariance (ANCOVA), using the SAS General Linear Model (GLM) procedure, was used to evaluate spatial and temporal differences in Hg concentrations, with age (largemouth bass) or weight (sunfish) as a covariate. However, use of ANCOVA is predicated on several critical assumptions (for review see ZAR, 1996). These assumptions are that the (1) regressions are simple linear functions; (2) regressions are statistically significant (i.e., nonzero slopes); (3) covariate is a random, fixed variable; (4) both the dependent variable and residuals are independent and normally distributed; and (5) slopes of regressions are homogeneous (parallel) (i.e., no interactions).

Regressions also require that collected samples exhibit a relatively wide range of covariate, that is, that fish from a given site are not all the same age or weight. Where these assumptions were not met, ANCOVA was inappropriate. Instead, standard analysis of variance (ANOVA) or student's "t" tests (SigmaStat, Jandel Corporation, San Rafael, CA) were used. Possible covariates were considered separately and often qualitatively. The assumptions of normality and equal variance were tested by the Kolmorogov-Smirnov and Levene Median tests, respectively. Datasets that either lacked homogeneity of variance or departed from normal distribution were natural-log transformed and reanalyzed. If transformed data met the assumptions, then they were used in ANOVA. If they did not meet the assumptions, then raw data sets were evaluated using nonparametric tests, such as the Kruskal-Wallis ANOVA on ranks or the Mann-Whitney Rank sum test. If the multigroup null hypothesis was rejected, then groups were compared using either Tukey HSD (honestly significant difference) or Dunn's Method.

SITE DESCRIPTIONS

Site descriptions and operational plans for STA-1W, 2, 3/4, 5, and 6 are published elsewhere (SFWMD, 1997; 1998a; 1998b; 1999a; 2004); similar information on STA-1E was not available as of the date of this report. For maps of monitoring locations, see **Figures 1** through **6**.

STA1W MERCURY SAMPLING LOCATIONS

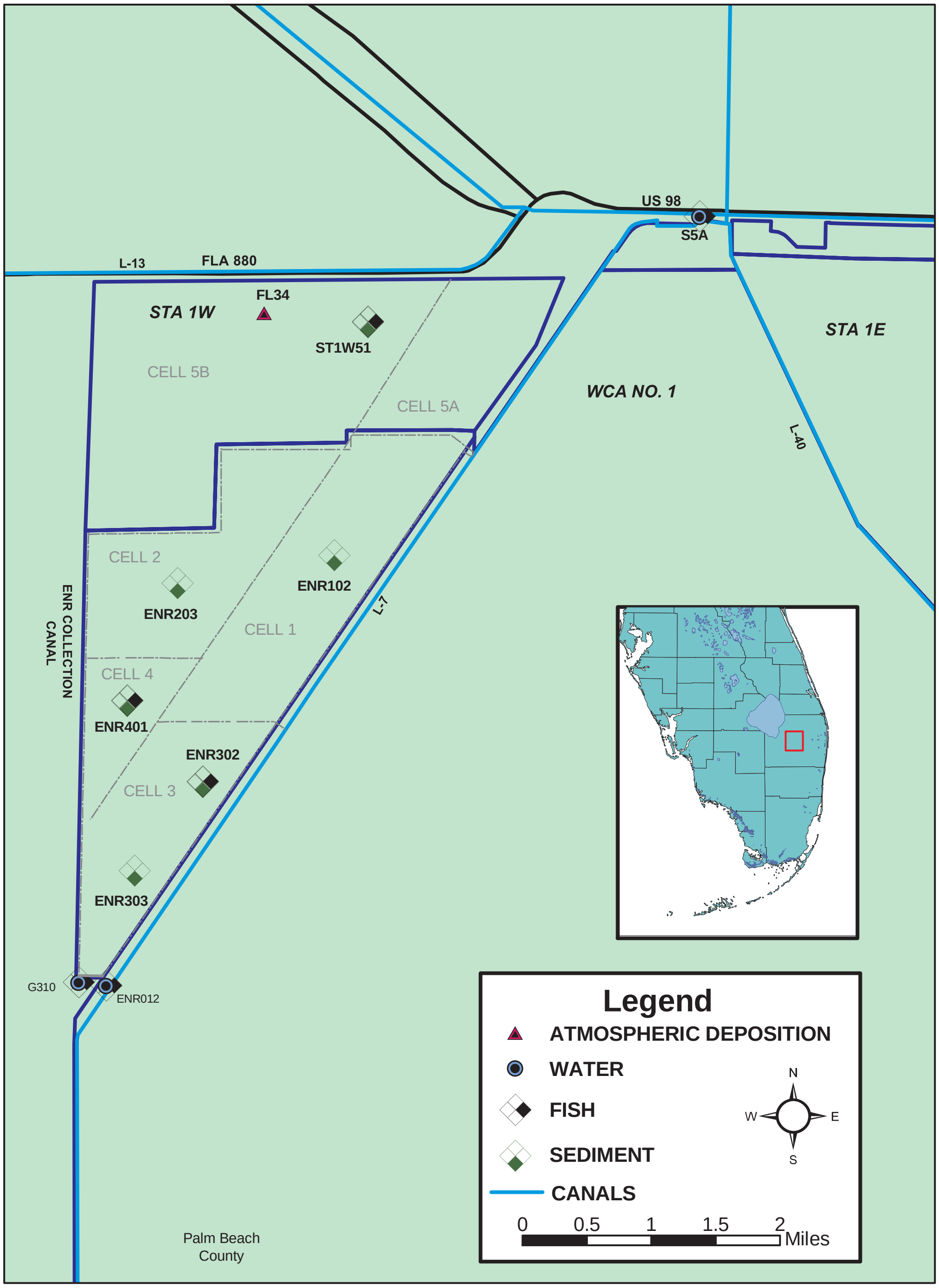

Figure 1. Stormwater Treatment Area 1 West (STA-1W) showing mercury monitoring sites.

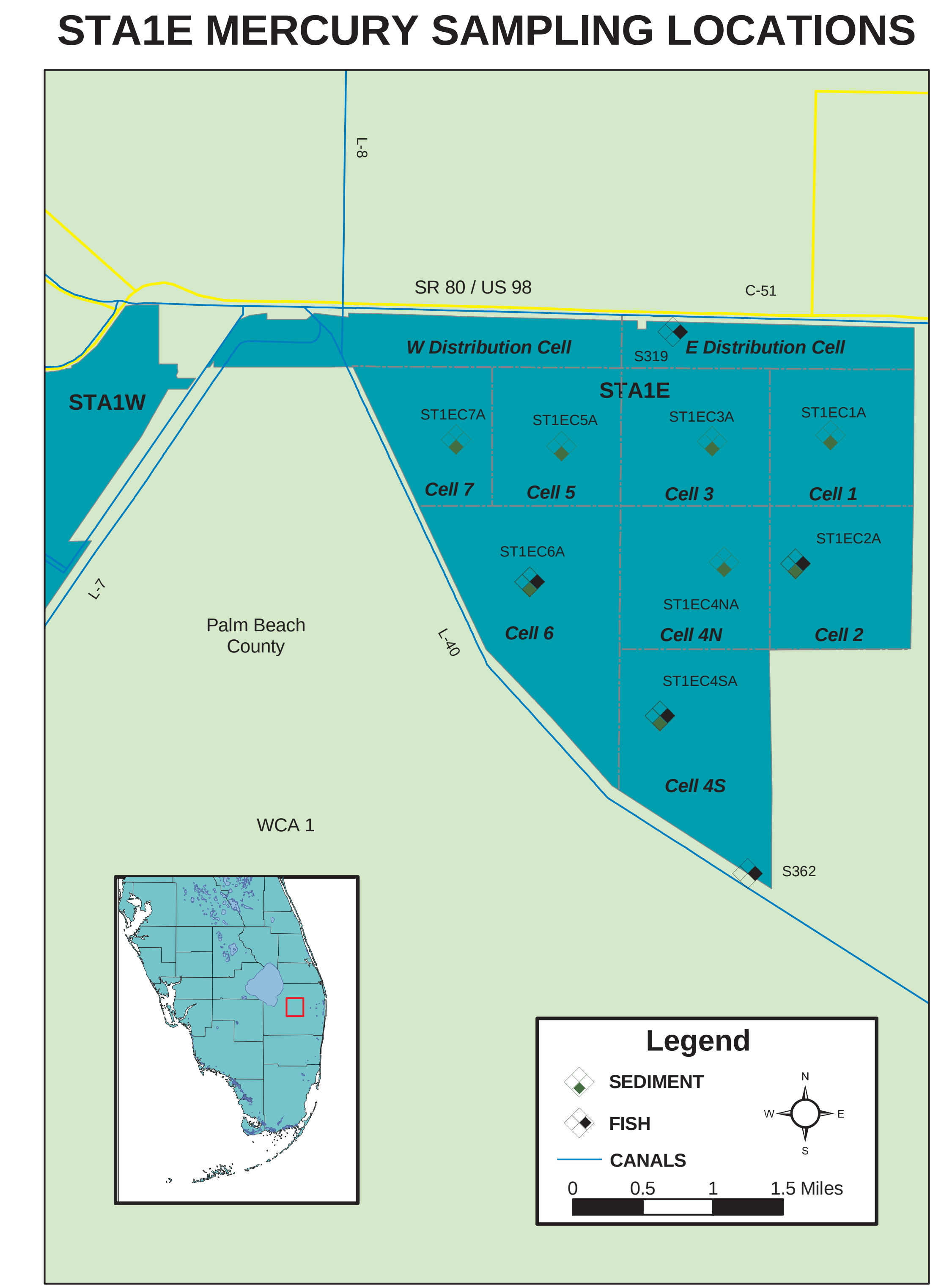

Figure 2. Map of Stormweatment Area 1 East (STA-1E) showing sediment collection sites and future fish collection sites.

STA2 MERCURY SAMPLING LOCATIONS

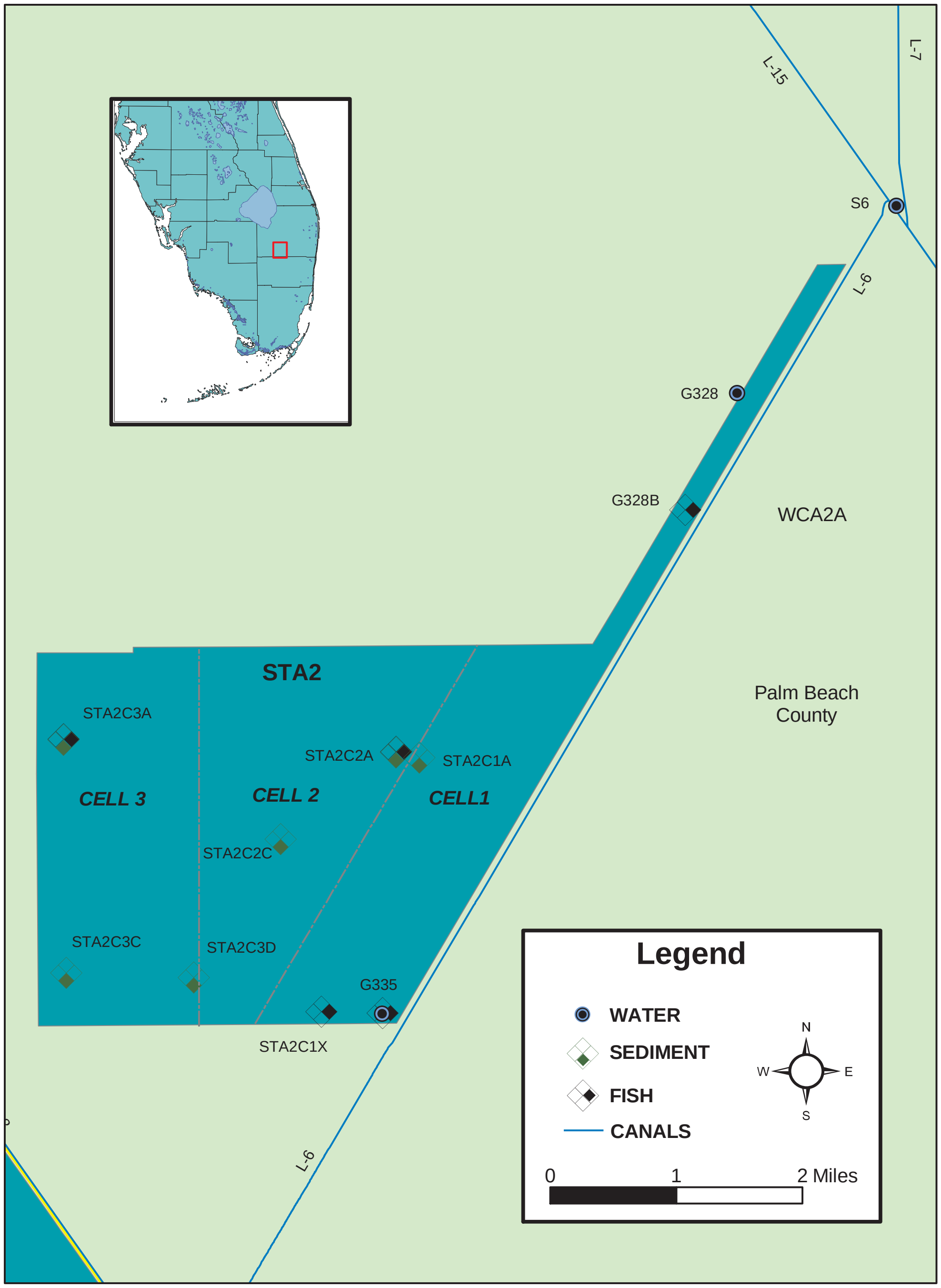

Figure 3. Map of STA-2 showing mercury monitoring sites.

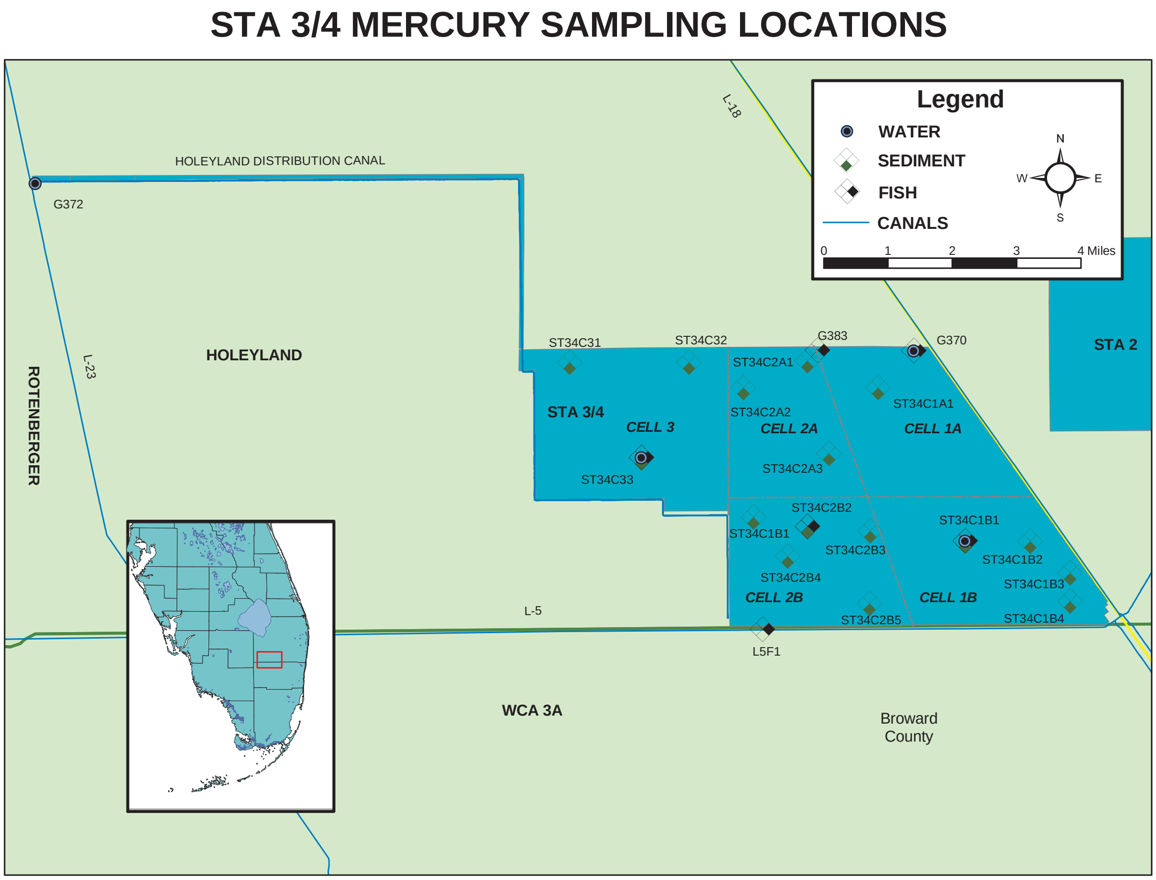

Figure 4. Map of STA-3/4 showing mercury monitoring sites.

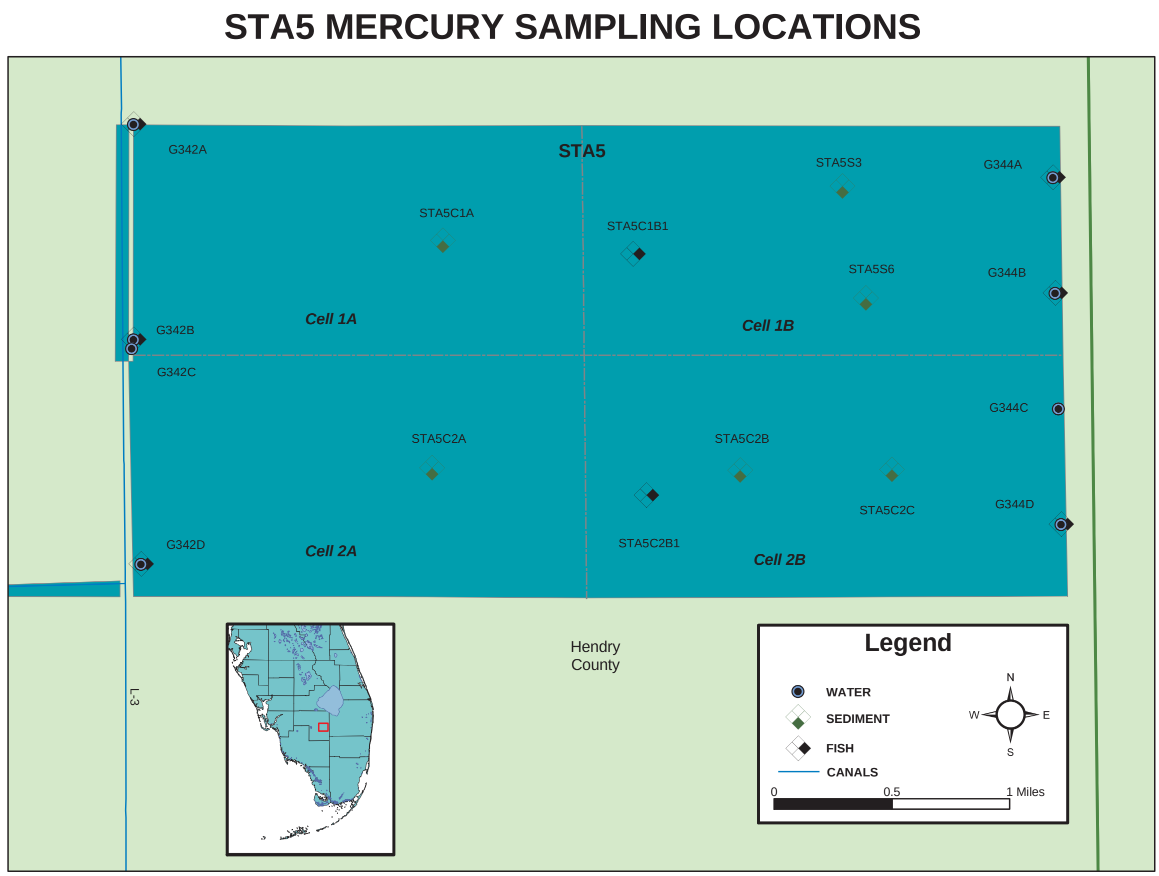

Figure 5. Map of STA-5 showing mercury monitoring sites.

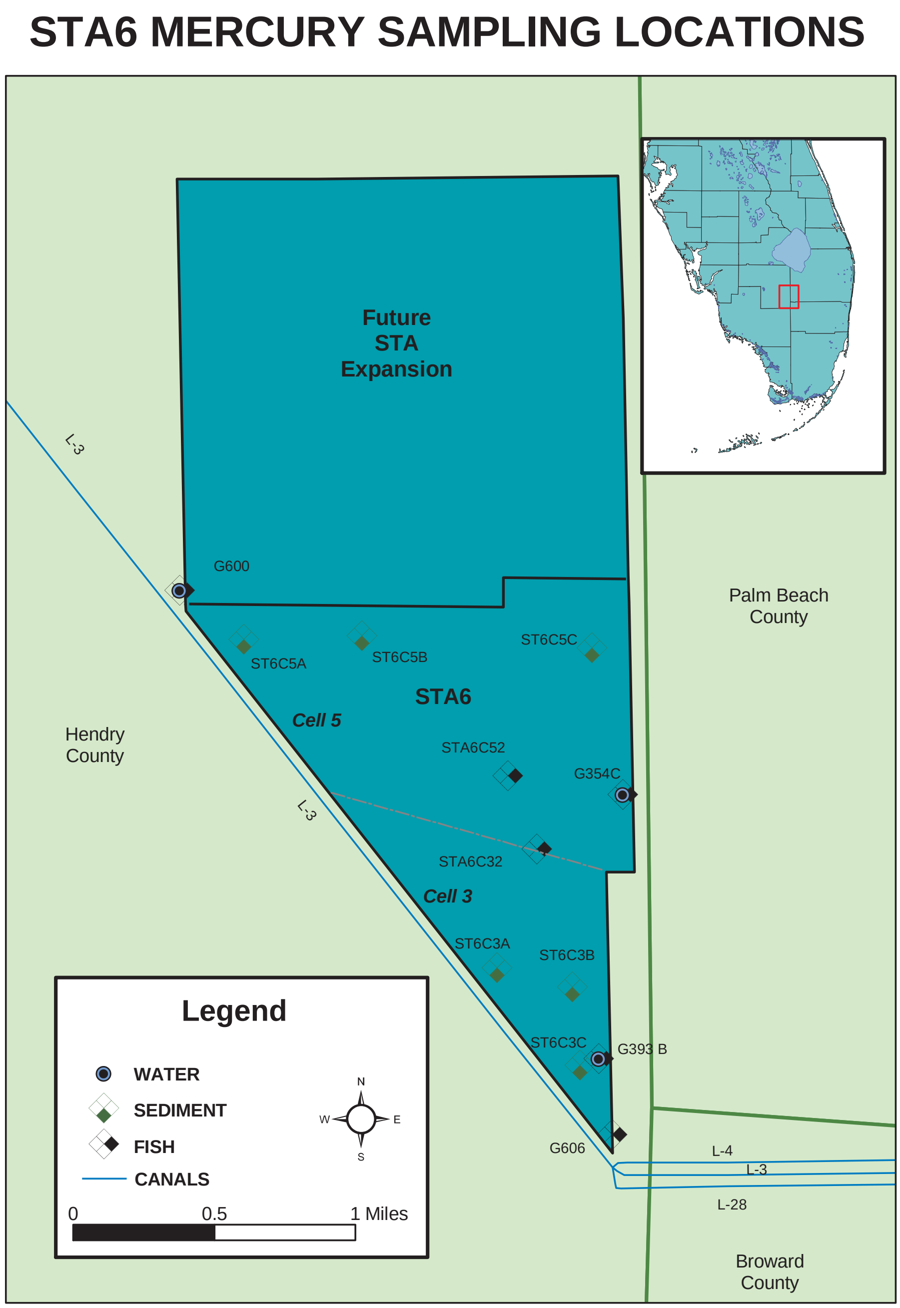

Figure 6. Map of STA-6 mercury monitoring sites.

MONITORING RESULTS

PRE-OPERATIONAL MONITORING

Results from pre-operational monitoring of STA-1W, 2, 5, and 6 have been reported previously (SFWMD, 1998c and 1999c; Rumbold and Rawlik, 2000; Rumbold et al., 2001a; Rumbold and Fink, 2002a; Rumbold and Fink, 2003a; Rumbold 2004a).

STA-3/4

Because of its large size relative to other STAs, 16 sediment cores (0 to 10 cm depth) were collected at STA-3/4 on June 19 and 20, 2003 (rather than six cores specified in the permit). Unlike the other STAs, it is notable that most of the cells of STA-3/4 contained standing water, to a depth ranging from 5 to 40 cm (Cells 1A, 2, 2B, and 3), when the cores were collected; only sites C1B4 and C2B2 were dry. Sediments were described as varying from peat, marsh vegetation (e.g., cattail, primrose), and gravel-clay mix to fine silts. Remnant sugar cane was documented in field notes at sites C2B2, C2B3, and C2B5. By comparison, most of Cell 1B remained in "Everglades" type condition prior to flooding (N. Larson, personal communication).

THg concentration in the cores ranged from 33.6 ng/g wet weight, in a core from site C2B4, to 100 ng/g wet, in a core from C2B3 (**Table 1**). These values are within the range observed in sediments from the other STAs (90% UCL = 100.1 ng THg/g). A one-way ANOVA revealed no significant differences in sediment THg concentrations among STAs (based on pre-start-up cores, $n = 42$; $df = 4, 37$; $f = 1.217$; $p = 0.32$; note that pre-start-up cores from STA-1W were not included in this analysis because they were collected from 0 to 15 cm. Furthermore, they were analyzed by a different laboratory that had a much higher Method Detection Limit (MDL) for MeHg and, consequently, results were below detection level, or BDL). MeHg concentrations in the cores ranged from 0.084 ng/g, in a core from site C1A1 to 3.03 ng/g, in a core from C2B4 (**Table 1**). Although concentration of MeHg was elevated in cores from STA-3/4, particularly cores from Cells 1B and 2B, relative to average values from other STAs (90% UCL = 1.71 ng/g), they did not exceed the maximum, 5.08 ng MeHg/g, previously observed in a core from STA-2 Cell 1. A Kruskal-Wallis one-way ANOVA on ranks revealed significant differences in MeHg levels among STAs ($df = 4$, $H = 14.7$, $p = 0.005$), with levels observed at STA-2 and STA-3/4 greater than STA-1E (Dunn's Method, $p < 0.05$; note that no other pairwise comparison of STAs was significant). Cores from STA-3/4 also had elevated %MeHg (percent of THg that is in the methyl form), ranging as high as 9.02 percent in a core from Cell 2B. Among-STA differences in %MeHg was significant ($df = 4$; $H = 10.7$; $p = 0.03$), with STA-2 levels significantly greater than STA-1E (no other pairwise comparison was significant); median %MeHg of STA-3/4 (1.1 percent) was second only to STA-2 (2.2 percent). There was no apparent pattern in terms of MeHg with sediment characteristics or pre-flood vegetation, i.e., both highest and lowest MeHg concentrations occurred in peat sediments with marsh vegetation.

In December 2003, start-up Hg monitoring began in flow-ways 1 and 3 of STA-3/4 (**Figure 7**); Cell 2 remained under construction through the end of the reporting year. Cell 1 satisfied start-up criteria for Hg in January 2004. While fluctuating over time, both THg and MeHg remained at higher concentrations in surface water at the interior marsh site of Cell 3 (i.e., site C33) than the two inflows (**Figure 7**); however, concentrations in Cell 3 never approached levels observed during start-up of STA-2 Cell. On March 12, 2004, the FDEP granted a permit modification authorizing flow through operation of this flow-way (see J. Brooks, FDEP letter dated March 12, 2004; modified in letter dated June 4, 2004).

Due to various potential, confounding influences, caution must be exercised when assessing the relationship between bulk sediment concentration (in cores from 0 to 10 cm depth) and subsequent water column concentrations (i.e., than the start-up of other STAs or relationships observed within downstream Everglades marshes). As one example, prior to and immediately following its impoundment, Cell 3 was reported to have received large quantities of groundwater from seepage/upwelling from the adjacent Holey Land Wildlife Management Area (due to large head differential; N. Larson, SFWMD, personal communication). As another example, unlike most other STAs, standing water was present at the time that cores were collected at most sites in STA-3/4.

STA-1E

Sediment cores (0 to 10 cm depth) were collected from eight sites within STA-1E on March 11, 2004. Cells were dry and soils were described as sandy or a mix of sand and muck with a mix of grass and weeds.

THg concentration in cores varied greatly and ranged from 3 ng/g wet weight, in a core from Cell 4NA, to 189 ng/g, in a core from Cell 2A (**Table 1**). MeHg concentration ranged from 0.007 ng/g, in a core from Cell 3A, to 0.4 ng/g, in a core from Cell 7A (**Table 1**). Although the THg maxima was elevated as compared to averages from other STAs (90% UCL = 100.1 ng THg/g), MeHg concentrations were relatively low (90% UCL = 1.71 ng MeHg/g). Furthermore, as stated previously, both the median absolute concentration of MeHg and the median %MeHg were significantly lower in cores from STA-1E than STA-2 (Dunn’s Method). As of April 30, 2004, start-up monitoring had not begun at STA-1E.

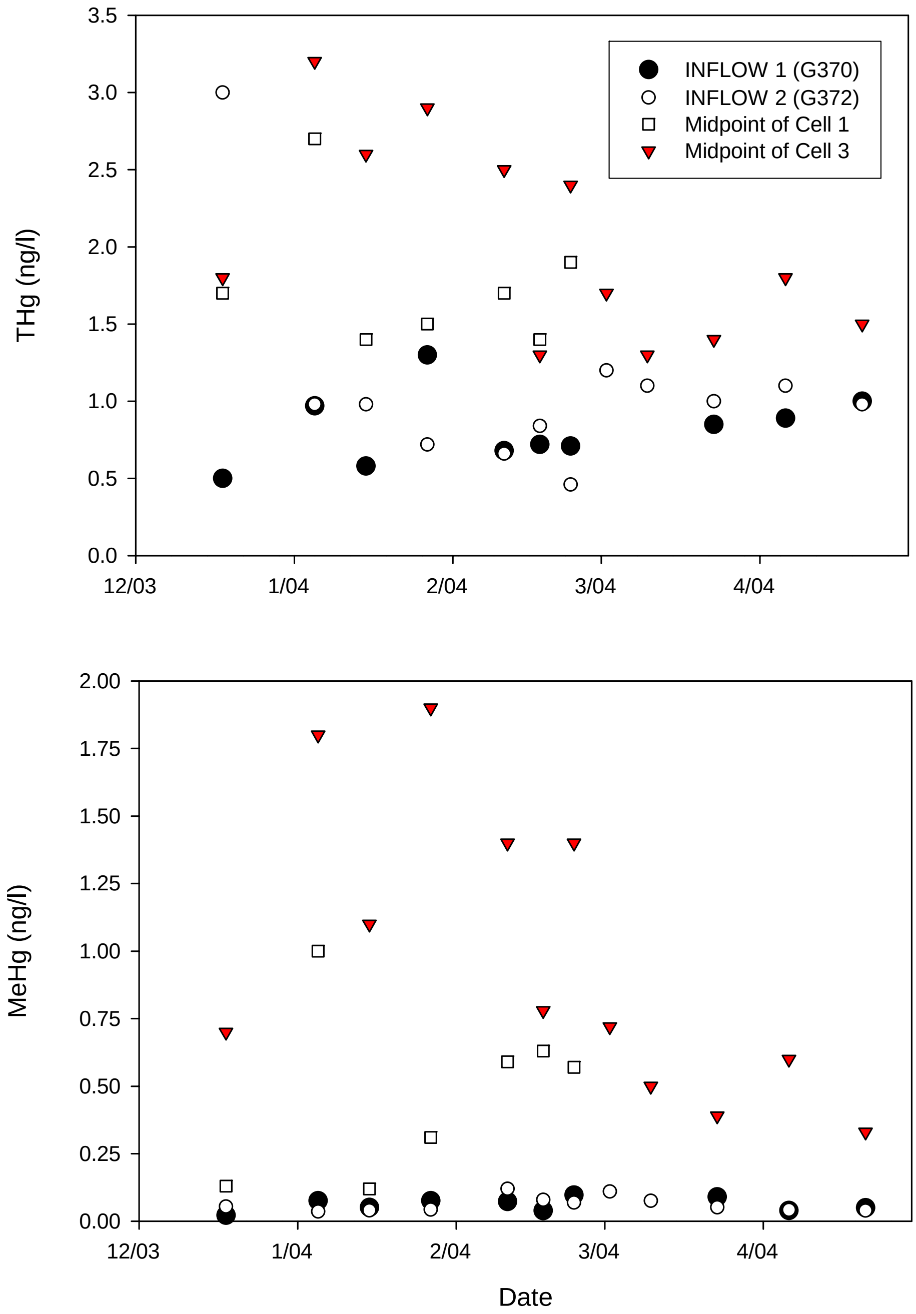

Figure 7. Concentrations of (a) THg and (b) MeHg (ng/L) in unfiltered surface water collected during start-up of STA-3/4.

Table 1. THg and MeHg concentrations (ng/g, dry weight) in STA soils (i.e., 10-cm depth composited).

STA	Year	Station	n	THg	remark*	MeHg	remark	%MeHg
STA-3/4	2004	Cell1A	1	57		0.08		0.15%
	2004	Cell1B	4	64		1.52		2.36%
	2004	Cell2A	3	60		0.28		0.46%
	2004	Cell2B	5	69		1.64		2.37%
	2004	Cell3	3	79		0.31		0.39%
	Mean			65.9	± 8.6	0.77	± 0.75	1.15%
STA-1E	2004	Cell1A	1	4		0.01		0.23%
	2004	Cell2A	1	189		0.13		0.07%
	2004	Cell3A	1	55		0.01		0.01%
	2004	Cell4NA	1	3		0.02		0.60%
	2004	Cell4SA	1	46		0.15		0.32%
	2004	Cell5A	1	54		0.14		0.25%
	2004	Cell6A	1	32		0.30		0.95%
	2004	Cell7A	1	55		0.40		0.73%
	Mean			54.7	± 58.4	0.14	± 0.14	0.39%

OPERATIONAL MONITORING

STA-1W

In 2000, STA-1W subsumed the ENR Project (Treatment Cells 1 through Cell 4, **Figure 1**), which had been in operation since 1994. STA-1W surface water passed start-up criteria during the week of January 17, 2000; flow-through operations began in early February 2000. Formal monitoring of Hg levels in STA-1W surface water began on February 16, 2000 (for discussion of results observed prior to WY2004, see Rumbold and Rawlik, 2000; Rumbold et al., 2001a; Rumbold and Fink, 2002a; Rumbold and Fink, 2003a; Rumbold, 2004a).

As shown in **Table 2** and **Figure 8**, concentrations of both THg and MeHg in surface water at the outflows of STA-1W remained low as compared to its inflow than other STAs; both constituents exhibiting a strong negative percent change across the cell (**Table 3**). Interestingly, THg concentration spiked in the inflow during the fourth quarter (9 ng THg/l; **Figure 8**); however, this was likely a result of suspended solids in the unfiltered samples (the analytical laboratory noted that the sample had a significant amount of suspended particulate matter).

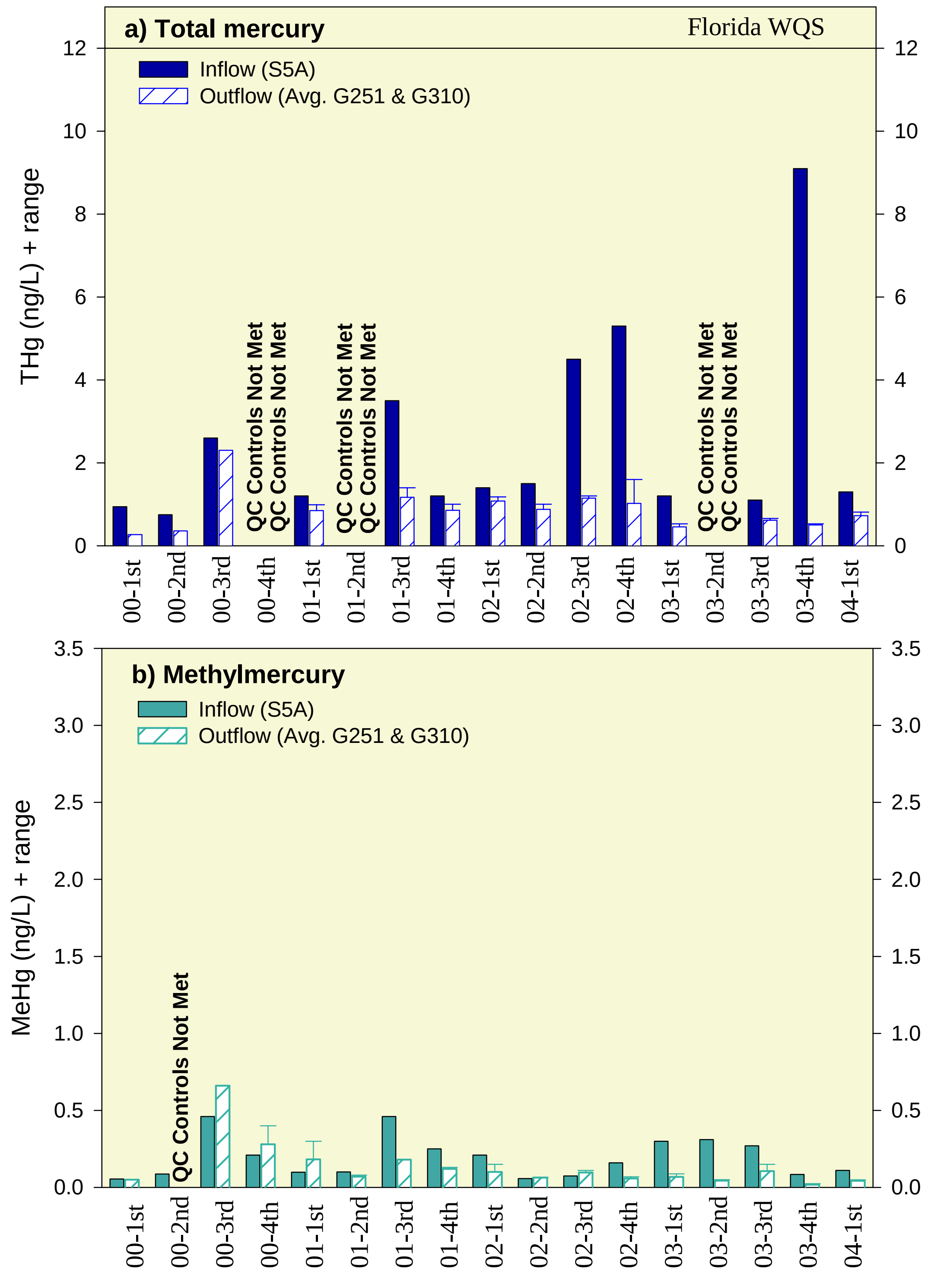

Figure 8. Concentrations of (a) THg and (b) MeHg (ng/L) in unfiltered surface water collected at STA-1W.

Table 2. Concentrations of THg and MeHg (ng/L) in surface water collected quarterly from the STAs.

STA	THg (ng/L)						MeHg (ng/L)				%MeHg	
	Yr-Quart	Inflow	remark*	Outflow	remark	THg WQS[a]	Inflow	remark	Outflow	remark	Inflow	Outflow
STA1-W[b]	03-2	3.4	J3	1.94	J3	<WQS	0.31		0.04		9%	2%
	03-3	1.1		0.62		<WQS	0.27		0.11		25%	17%
	03-4	9.1		0.51		<WQS	0.08		0.02		1%	3%
	04-1	1.3		0.73		<WQS	0.11		0.04		8%	6%
STA-2[c]	03-2	0.56		1.70		<WQS	0.16		0.88		29%	52%
	03-3	1.50		1.60		<WQS	0.13		0.78		9%	49%
	03-4	0.59		0.66		<WQS	0.08		0.24		13%	36%
	04-1	0.69		0.73		<WQS	0.07		0.08		10%	11%
STA-3/4[d]	04-1	0.92		2.4		<WQS	0.07		0.97		8%	40%
STA-5[e]	03-2	1.33		0.97		<WQS	0.22		0.27		17%	28%
	03-3	1.38		1.17		<WQS	0.18		0.49		13%	42%
	03-4	1.28		0.71		<WQS	0.12		0.14		10%	20%
	04-1	1.26		0.59		<WQS	0.16		0.17		13%	29%
STA-6[f]	03-Apr	0.70		0.81		<WQS	0.10		0.25		14%	31%
	03-May	1.30		1.19		<WQS	0.22		0.33		17%	28%
	03-Jun	1.30	J3	0.61	J3	<WQS	0.54		0.12			
	03-Jul	1.50		1.20		<WQS	0.43		0.16		29%	13%
	03-Aug	1.80		1.23		<WQS	0.26		0.07		14%	6%
	03-Oct	0.53		0.71		<WQS	0.20	J3	0.11	J3		
	04-Jan	0.61		0.63		<WQS	0.12		0.12		20%	19%

* For qualifier definitions, see FDEP Rule 62-160: J3" - estimated value, poor precision, “V” - analyte detected in both the sample and the associated method blank.

a. Class III water quality standard of 12 ng THg/L.

b. STA-1W has a single inflow and two outflows; the reported value for the latter represents mean of valid data (unqualified).

c. STA-2 has two inflows and a single outflow; the reported value for the former represents mean of valid data (unqualified).

d. STA-3/4 has two inflows; reported value is mean. Outflows are monitored from each of the three trains; reported value is mean of two outflows from Train 1 only.

e. STA-5 has four inflows and four outflows; reported value represents mean of valid data (unqualified).

f. STA-6 has one inflow and two outflows; reported value is mean. Sampling at STA-6 was expanded in July 2002 to include monthly sampling of water, and sampling at a second outflow culvert from Cell 5 (G-354A).

Table 3. Percent change in concentration of THg and MeHg in surface water across STAs (i.e., outflow-inflow/inflow).

STA	Quarter	THg	MeHg
STA-1W	03-2	**	-86%
	03-3	-44%	-61%
	03-4	-94%	-79%
	04-1	-44%	-61%
Annual median		-44%	-70%
Cumulative median		-44%	-52%
STA-2	03-2	204%	445%
	03-3	7%	500%
	03-4	12%	202%
	04-1	6%	17%
Annual median		7%	323%
Cumulative median		33%	349%
STA-3/4	04-1	159%	1276%
Annual median		159%	1276%
Cumulative median		159%	1276%
STA-5	03-2	-27%	21%
	03-3	-15%	177%
	03-4	-45%	14%
	04-1	-53%	7%
Annual median		-36%	18%
Cumulative median		-25%	7%
STA-6	03-Apr	15%	153%
	03-May	-9%	52%
	03-Jun	**	-78%
	03-Jul	-20%	-63%
	03-Aug	-31%	-71%
	03-Oct	34%	**
	04-Jan	3%	-2%
Annual median		-3%	-32%
Cumulative median		-14%	-14%

** Only valid (unqualified) data used in calculations; see **Table 1** for raw data and qualifiers.

Concentrations of THg in mosquitofish are summarized in **Table 4** and are graphically presented in **Figure 9**. Levels of Hg in mosquitofish from STA-1W were similar to, or have declined slightly, when compared to concentrations observed in fish collected previously from this area when it was operated as the ENR Project (see references above). Furthermore, Hg levels in STA-1W mosquitofish continue to be low compared to levels currently observed in fish from other areas of the Everglades (see Appendix 2B-1 of this volume). Similar to water column concentrations, mosquitofish consistently exhibited a negative percent change in tissue-Hg across STA-1W (**Table 4**). As discussed below, this pattern was also observed in sunfish and largemouth bass. Although, Hg levels in the mosquitofish have varied among sites in the past, levels in 2003 were very similar from multiple interior sites and from the two discharge canals (see ranges shown in **Figure 9**).

As is evident from **Table 5** and **Figure 9**, STA-1W sunfish continued to have Hg levels much lower than those observed in sunfish at the other STAs and locations within the Everglades (see Appendix 2B-1 of this Volume). Moreover, this pattern does not appear to be changing, i.e., there have been no obvious increases over time (**Figure 9)**. STA-1W sunfish did exhibit a significant spatial pattern (i.e., locational differences) in tissue-Hg in 2003 ($H = 38.302$, $df = 2$, $p = < 0.001$); post hoc pairwise comparisons (Dunn's Method) revealed fishes from both the interior marshes and the discharge canals ($p < 0.05$) contained less Hg than fish from the supply canal. Fishes from the former two areas did not differ significantly in Hg burdens ($p < 0.05$). However, as observed in the past, sunfish caught in the supply canal were much larger than fish from other areas in 2003 ($H = 27.376$, $df = 2$, $p = < 0.001$) and this (as an indication of age difference) may, in part, account for the higher Hg levels. Sunfish from Cell 5 continued to have higher levels when compared to sunfish from other cells; in 2003, this difference in tissue-Hg levels (between Cell 4 and Cell 5 fish; no sunfish were collected from Cell 3 in 2003) was significant ($F = 12.192$, $df = 1, 37$; $p = 0.001$). Although this does not appear to be size related (Cell 4 fish were larger), relatively fewer warmouth and spotted sunfish were collected in Cell 4 and this may account for the lower levels. Tissue-Hg levels did not differ between sunfish populations in the two discharge canals ($H = 0.514$, $df = 1$, $p = 0.473$). In terms of temporal trends, Hg levels in sunfish from the discharge canals (pooled) varied among years (pooled, $H = 28.347$, $df = 4$, $p = < 0.001$); 2001 levels were higher than levels in all other years ($p < 0.05$), but no other pairwise comparison among years was significant. Among-year differences were not significant for tissue-Hg levels in interior sunfish ($H = 5.277$, $df = 4$, $p = 0.260$).

Similar to sunfish, largemouth bass from STA-1W contained lower levels of Hg than bass from the other STAs (**Table 6)**. Moreover, STA-1W bass contained much lower Hg than fish from downstream sites in the WCAs (see Appendix 2B-1 of this volume). As with mosquitofish and sunfish, the bass exhibited a negative percent change in Hg levels across the STA (**Table 6**). Because of a statistically significant interaction between location and age ($F = 10.96$, $df = 2, 54$; $p < 0.001$), an ANCOVA could not be used to assess differences (in LSM Hg levels) between supply and discharge bass. For the same reason, an ANCOVA could also not be used to compare tissue-Hg levels in bass collected from the two discharge canals in 2003 ($F = 39.3$, $df = 1, 36$; $p < 0.001$).

In terms of temporal trends, age-adjusted Hg levels in bass from site ENR012 (one of the two discharge canals) did not differ among 1999, 2001 or 2003 (i.e., only three years which had a significant age-concentration relationship; ANCOVA, $F = 2.88, 2, 42$, $p = 0.07$). Temporal trends could not be assessed in bass from site G310 (i.e., other discharge canal) because the age-concentration relationship was not statistically significant in most years.

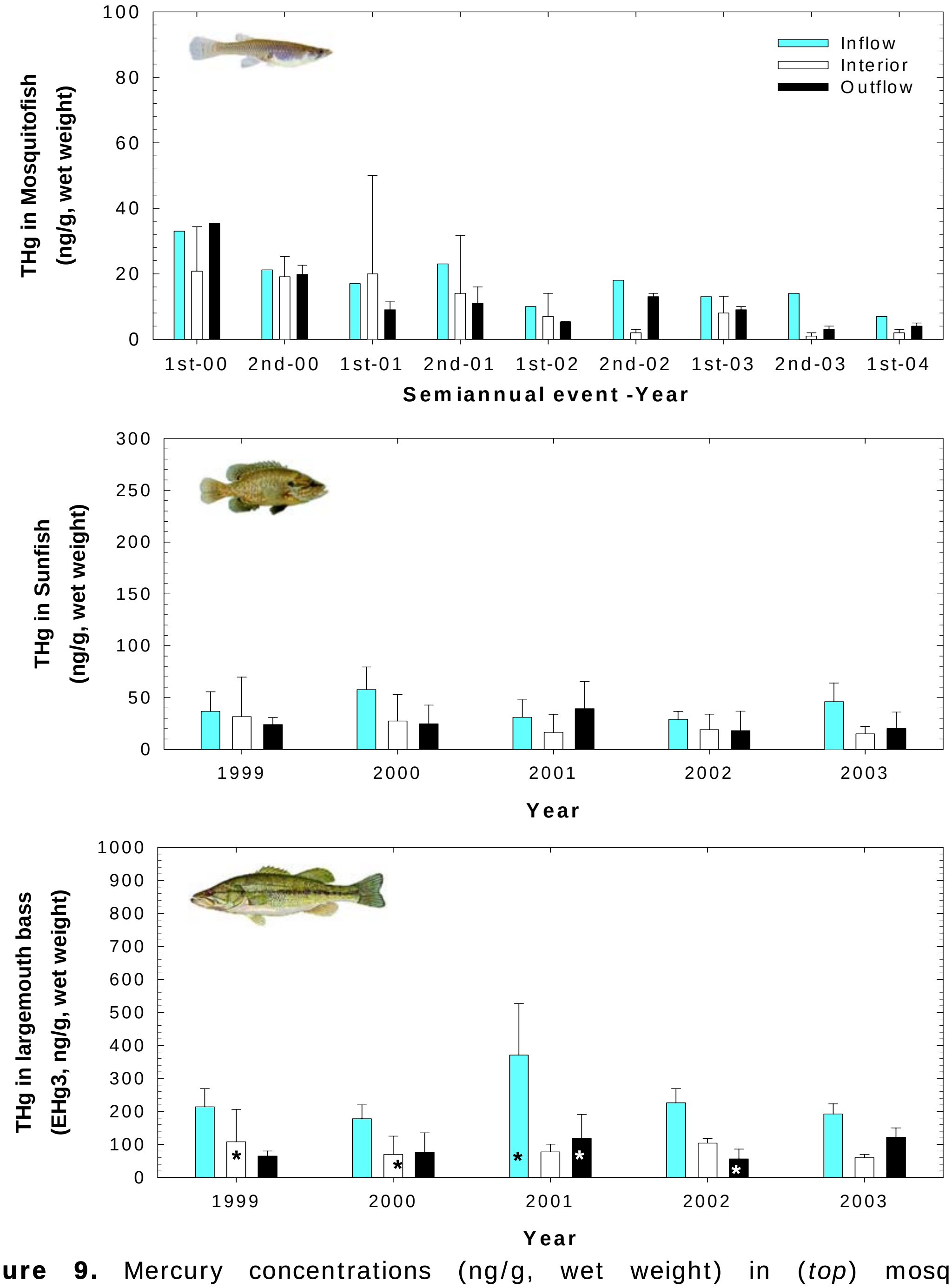

Figure 9. Mercury concentrations (ng/g, wet weight) in (*top*) mosquitofish composites (+ range), (*middle*) whole sunfish (± SD), and (*bottom*) fillets of largemouth bass (± 95% CI or, if arithmetic, SD) collected at STA-1W. Note that the latter are reported as the expected concentration in a three-year-old fish, EHg3, unless this could not be calculated (* for details, see **Table 5**), in which case, the arithmetic mean is reported.

Table 4. Concentration of THg (ng/g, wet weight) in mosquitofish composites collected semiannually from STAs.

STA	Half-year	Inflow Fish	Interior Fish	Outflow Fish	Percent Change[a]
STA 1W	2003-2	14	1 ±1	3 ±1	-79%
	2004-1	7	2 ±1	4 ±1	-43%
Annual mean		11	2	3	-73%
Cumulative mean		31 ±43	11 ±8	19±25	-39%
STA 2	2003-2	5	17 ±23[b]	8	60%
	2004-1	3	13 ±8	17	467%
Annual mean		4	15	13	225%
Cumulative mean		9 ±7	68 ±66	130 ±125	1344%
STA 5[c]	2003-2	13 ±3	3 ±1	13 ±7	0%
	2004-1	15 ±5	7 ±2	26 ±7	73%
Annual mean		14	5	19	36%
Cumulative mean		30 ±13	26 ±30	30 ±17	0%
STA 6	2003-2	21	4 ±3	5±2	-76%
	2004-1	23	9 ±5	16±12	-30%
Annual mean		22	7	10	-55%
Cumulative mean		27 ±19	17 ±14	39 ±24	44%

a - Percent change = outflow-inflow/inflow

b - Standard deviation is reported where multiple composites are collected from location (e.g., multiple inflows or outflows, multiple cells); range is reported where two sites are sampled; other values represent mean of three analyses of a single composite sample.

c - Per FDEP approval on March 5, 2002, collection locations were reduced from 4 to 2 for both the inflow and outflow of STA-5.

Table 5. Concentration of THg (ng/g, wet weight) in sunfish (*Lepomis* spp.) collected from STAs in 2003 (sample size in parentheses).

STA	Inflow Fish	Interior Fish	Outflow Fish	Percent Change[a]
STA-1W	46 ±18 (20)	15 ±7 (39[b])	20 ±16(39)	-57%
Cum. mean[C]	41	20	25	-39%
STA-2	103 ±104(20)	147 ±149 (87)	148 ±156(20)	44%
Cum. mean	83	150	143	72%
STA-5	53 ±28 (20)	79 ±31 (20 [b])	73 ±46 (20 [b])	38%
Cum. mean	67	109	99	48%
STA-6	62 ±20 (20)	41 ±48 (24[b])	117 ±138(20)	89%
Cum. mean	58	59	100	72%

a. Percent change = outflow-inflow/inflow

b. Where n > 20; multiple sites were sampled and pooled, i.e., multiple interior or outflows.

c. Grand mean of annual means; sunfish collected in 1999, prior to permit revision or STA operation (in the case of STA-5 and STA-1W) were included in the cumulative average.

Table 6. Standardized, EHg3 ± 95%, and arithmetic mean concentration (mean ± 1 SD, n; in parentheses) of THg (ng/g, wet weight) in fillets from largemouth bass collected at STAs in 2003.

STA	Inflow Fish	Interior Fish	Outflow Fish	Percent Change[a]	Consumption Advisory Exceeded[b]
STA-1W	192 ± 31 (195 ± 120, 20)	60 ± 10 (42 ± 22, 21[c])	122 ± 28 (66 ± 68, 40)	-66%	No
Cumulative mean[d]	255	72	76	-70%	
STA-2	218 ±64 (247 ± 218, 20)	NC (1) (303 ± 248, 74)	702 ± 149 (770 ± 357, 20)	212%	Yes
Cumulative mean	273	327	909	233%	
STA-5	NC (2) (92 ± 28, 20)	NC (1) (139 ± 22, 8)	NA NA	---	Unknown
Cumulative mean[d]	199	315	440	121%	
STA-6	266 ±21 (228 ± 121, 20)	152 ± 36 (53 ± 49, 15)	510 ± 72 (369 ± 126, 20)	62%	Yes
Cumulative mean	274	361	519	89%	

a - Percent change = outflow-inflow/inflow.

b - Florida limited consumption advisory threshold is 500 ng/g in three-year-old bass.

c - Where n > 20; multiple sites were sampled and pooled, i.e., multiple interior or outflows.

d - Bass collected in 1999 prior to operation of STA-5 and STA-1W were included in the cumulative average based on arithmetic mean.

NC = not calculated, where: (1) regression slope was not significantly different from 0, or (2) poor age distribution of collected fish.

NA = not available; no bass in sample.

Hg levels in fish tissues can also be put into perspective and evaluated for the exposure to fish-eating wildlife. The 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 the U.S. Congress, the 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). STA-1W mosquitofish, sunfish, and largemouth bass continue to have tissue-Hg levels well below both the USEPA and USFWS guidance level for predator protection. Therefore, unlike most other areas of the Everglades, fish-eating wildlife foraging preferentially at STA-1W would not appear to be at risk from Hg exposure.

STA-2

STA-2 Cells 2 and 3 met Hg start-up criteria on September 26, 2000 and November 9, 2000, respectively. Cell 1 did not meet start-up criteria until November 26, 2002. Operational monitoring of Hg at STA-2 began during the third quarter of 2001 following the completion of the S6 connection in May 2001. (For previous results of start-up and expanded Hg monitoring under modified permit, see Rumbold and Fink, 2003b; Rumbold 2004a; for results of expanded studies during WY2004, see Appendix 2B-1 of this volume).

Results from monitoring Hg concentrations in surface water at STA-2 are shown in **Tables 2** and **3**, and graphically presented in **Figure 10**. During the reporting year, THg concentration in the outflow of STA-2 did not exceed the Class III WQS of 12 ng/L. More importantly, water column concentrations of both MeHg, which has no numerical WQS, and THg have declined dramatically at STA-2 than previously observed peak levels. Nonetheless, both constituents continued to be found in greater concentration in the outflow than in the inflow. This resulted in a large positive percent change across the STA, especially for MeHg (**Table 3**).

Results from operational monitoring of Hg concentrations in STA-2 mosquitofish are summarized in **Table 4** and **Figure 11**. (Note that the results from different interior sites are graphed separately because of the degree of spatial variability known to occur in this STA.) Although spatial patterns in tissue-Hg have remained consistent over time, Hg levels have declined dramatically in mosquitofish from Cell 1 and the discharge canal (**Figure 11**). Because mosquitofish are small, short-lived fish, tissue-Hg levels are expected to change more quickly in their population in relation to changes in ambient conditions, than in larger older fish whose tissues reflect exposure over many years. Therefore, these reductions in tissue-Hg levels in mosquitofish are very encouraging and are expected to propagate up the food chain over time.

Spatial patterns in tissue-Hg in sunfish were nearly identical to those observed for mosquitofish (**Figure 11**), with relatively higher levels occurring in resident fish from the Cell 1 and discharge canal. Hg levels differed significantly among sunfish collected from different treatment marshes in 2003 ($H = 41.54$, $df = 3$, $p = < 0.001$). Although levels did not differ in fish from the two sites sampled within Cell 1 (sites C1X and C1A; $p > 0.05$; two sites were sampled because fish were collected at the C1X in 2001, and C1A in 2002), Cell 1 fish did contain higher levels than fish from either Cell 2 or Cell 3 ($p < 0.05$). By comparison, Hg levels did not differ between sunfish from Cell 2 and Cell 3 ($p > 0.05$). Although there were some differences in the species of *Lepomis* collected from the four interior sites, there was no clear evidence that any species-related effect skewed the observed Hg levels. Likewise, weight (as a surrogate of age) did not appear to be a confounding factor; interior sunfish did not vary significantly in weight ($H = 5.26$, $df = 3$, $p = 0.153$). When fish from the two sites within Cell 1 were pooled, they were also found to contain significantly greater Hg than sunfish from either the supply or the discharge

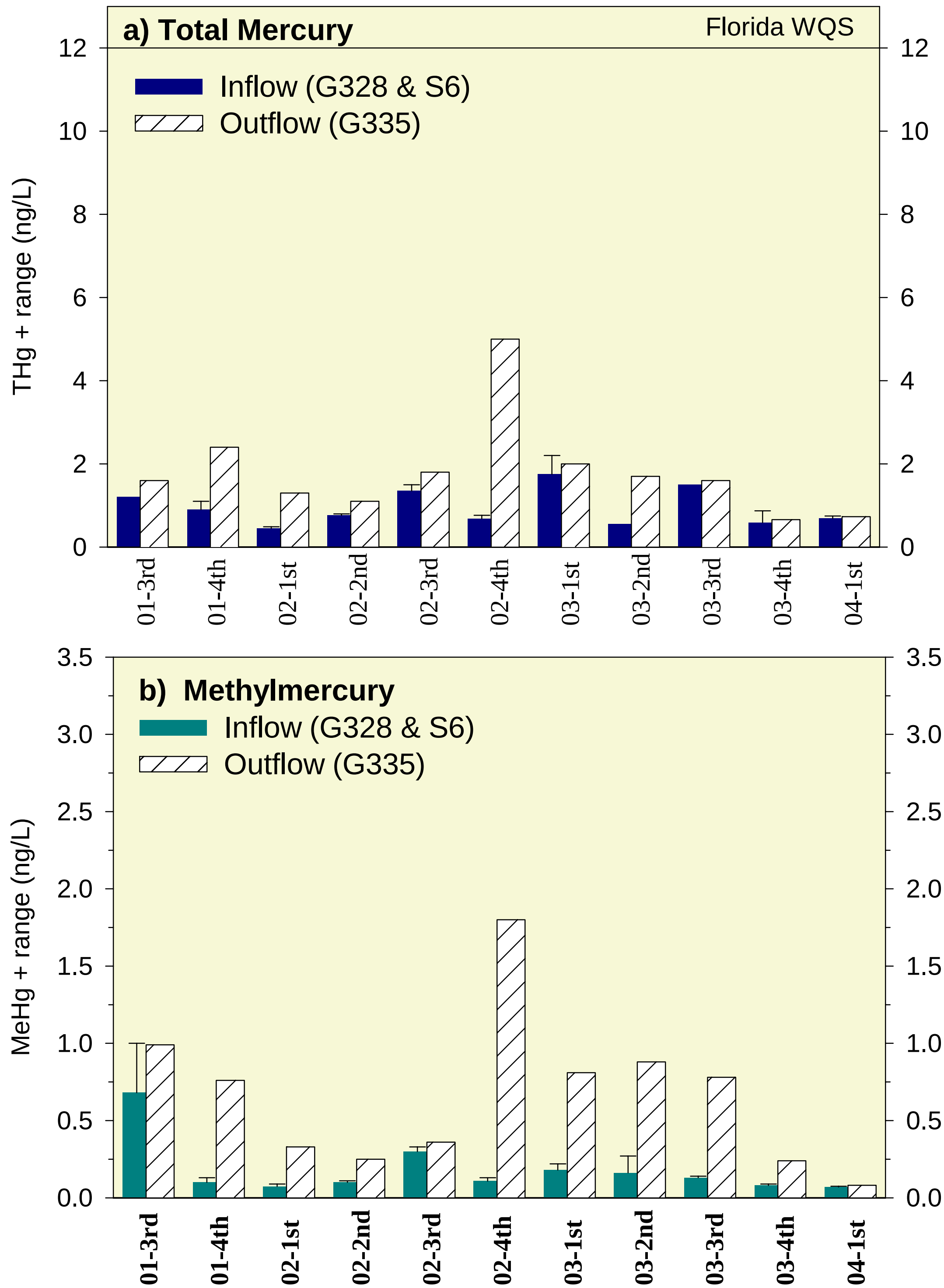

Figure 10. Concentrations of (a) THg and (b) MeHg (ng/L) in unfiltered surface water collected at STA-2.

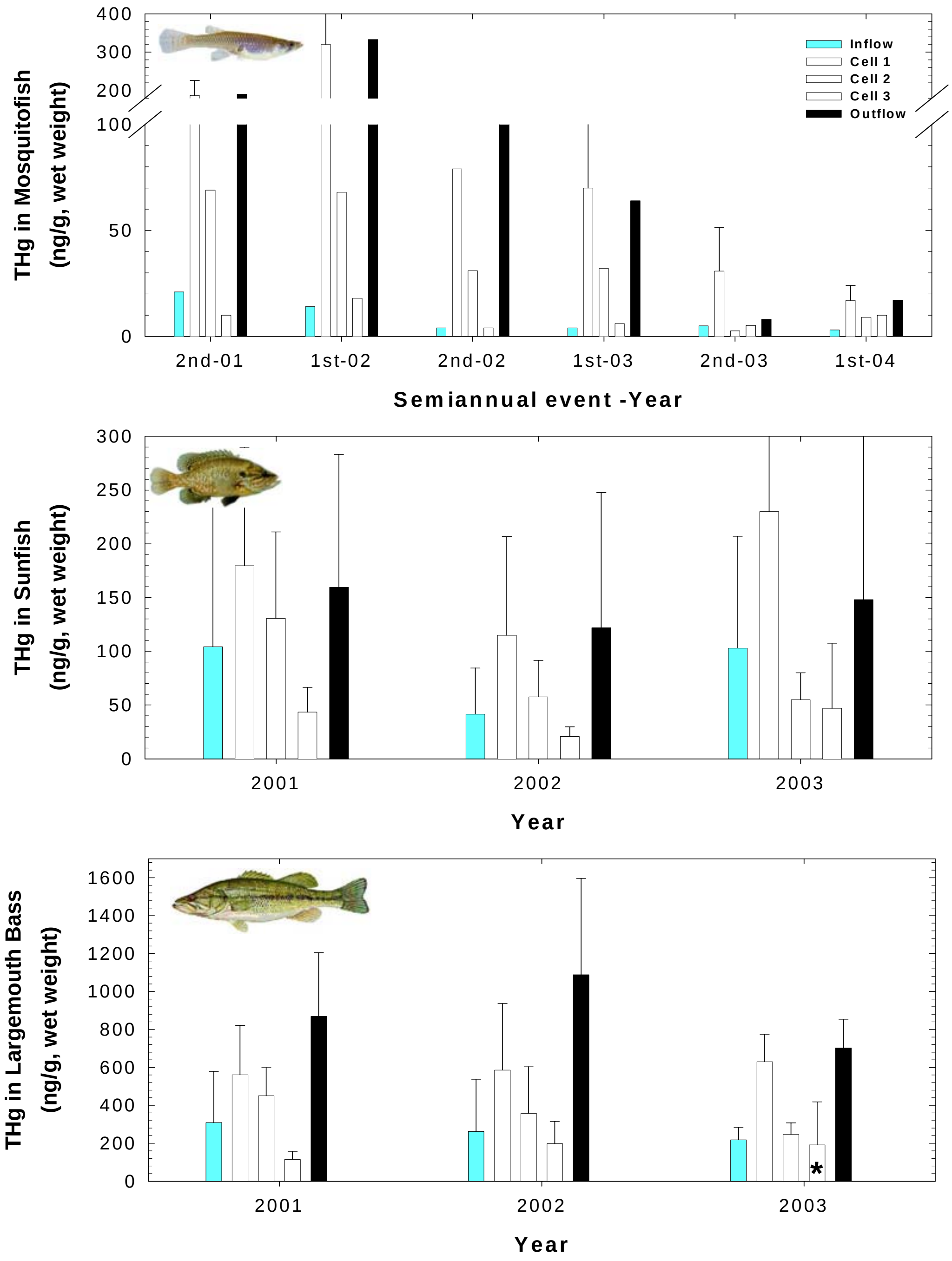

Figure 11. Mercury concentrations (ng/g, wet weight) in (*top*) mosquitofish composites (+ range), (*middle*) whole sunfish (± SD), and (*bottom*) fillets of largemouth bass [± 95-percent confidence interval (95% C.I.) or, if arithmetic, SD] collected at STA-2. Note: the latter are reported as the expected concentration in a three-year-old fish, EHg3, unless this could not be calculated (* for details, see **Table 5**), in which case, the arithmetic mean is reported.

canal (H = 14.401, df =2, p = < 0.001; Dunn's Method, p < 0.05). Alternatively, sunfish populations in the supply and discharge canals did not differ in Hg levels (p < 0.05).

As discussed above, fish (both sunfish and bass) have been collected from two different areas of Cell 1 in different years (i.e., after fish were not found at the primary site, a second site was sampled); therefore, temporal trend assessment must be done cautiously. Hg levels differed in 2002 and 2003 in sunfish from site C1A (F = 7.38, df = 1, 37, p = 0.01), with higher levels occurring in 2003. However, as reported last year, sunfish were thought to have recently invaded this site, which is located immediately downstream of inflow culverts, after the cell was reflooded in 2002. If these fish were indeed recent colonizers in 2002, then it is not surprising that after a year (in which the cell did not dry out) these fish would have higher levels. Alternatively, the levels of Hg did not differ from 2001–2003 in sunfish from site C1X (F = 2.3, df = 1, 34, p = 0.14). Likewise, Hg levels in sunfish inhabiting the discharge canal did not differ among years (H = 2.6, df = 2, p = 0.265). As noted in previous reports, it has been speculated that these fish also move freely through culverts (G330s), and in this case, between Cell 1 and the discharge canal. For the reasons noted above, any decline in Hg levels in sunfish from these two areas will likely lag behind declines observed in mosquitofish. Interestingly, sunfish in Cell 2 have shown a progressive downward trend in Hg, with levels declining first in 2002 and then again in 2003. Fish caught in 2002 and 2003 contained significantly lower Hg than fish collected 2001 (H = 24.05, df = 2, p = < 0.001; pairwise test both p < 0.05); however, the between-year difference for 2002 and 2003 was not statistically significant (p > 0.05).

Largemouth bass exhibited spatial patterns in Hg levels somewhat similar to those observed in mosquitofish and sunfish (**Table 6**, **Figure 11**). After adjustment for age (i.e., using LSM), bass from the discharge canal contained significantly more Hg than bass from the supply canal (ANCOVA, F = 33.4, df = 2, 37, p < 0.0001). Bass caught in the discharge canal in 2003 were also found to contain less Hg than fish collected in either 2002 or 2001 (ANCOVA, F = 9.63; df = 2, 56; p = 0.0003; Tukey Test, p < 0.0.01). Interestingly, like sunfish, levels of Hg were also lower in bass caught in Cell 2 in 2003 than 2002. Unlike the sunfish, the difference between years in Cell 2 bass was statistically significant (F = 5.71, df = 1, 37, p = 0.02). ANCOVA was not used to assess temporal trends in bass from either Cells 1 or 3 due to a lack of a significant age-concentration relationship and, in the case of Cell 1, previously discussed uncertainties regarding movements of fish following drydown and rewetting.

With regard to risks to fish-eating wildlife, a probabilistic ecological risk assessment was recently completed for STA-2 (Rumbold, 2004b; Rumbold, in prep.) and submitted to the FDEP (transmittal letter from R. Bearzotti, SFWMD, dated March 30, 2004) as a special condition for a permit modification. The assessment estimated the risks posed by MeHg to the great egret (*Ardea alba*) and the bald eagle (*Haliaeetus leucocephalus*). Exposure models were based on literature-derived life history parameters combined with site-specific MeHg concentrations in water, sediment, and fish. To assess risk, daily MeHg intake by females and cumulative MeHg consumed by nestlings were compared to literature-derived effects thresholds. The results indicated the likelihood that MeHg exposures to birds foraging throughout STA-2 would exceed the effects thresholds was low, based on 2003 data. By comparison, birds foraging exclusively from Cell 1 or the small discharge canal were predicted to experience greater exposures and, as a consequence, were at some elevated risk. However, this worst case risk was comparable or lower than risk levels encountered in nearby water conservation areas or the Everglades National Park (ENP or Park).

STA-3/4

As reported above, STA-3/4 Cell 1 satisfied start-up criteria for Hg in January 2004. Routine operational monitoring of this flow-way began during the first quarter of 2004. As shown in **Table 2**, water column concentrations of both THg and MeHg were slightly elevated than other STAs. Both constituents were at greater concentration in the outflow than the inflow (almost twofold for THg and over twelvefold for MeHg), resulting in a positive percent change across this STA (**Table 3**). However, at no time during the reporting year did THg concentrations exceed the Class III water quality standard of 12 ng/L. Due to dry season, low-water conditions (see R. Bearzotti, SFWMD, e-mail dated May 28, 2004) expanded monitoring pursuant to special conditions of the modified permit that authorized flow-through operation of Cell 3 was not fully implemented until June 2004, which was outside of the current reporting year.

Resident fishes were not sampled from this STA during WY2004.

STA-5

STA-5 met start-up criteria for Hg in September 1999, and routine operational monitoring began during the first quarter of 2000. However, because of drought conditions and the detection of high phosphorus concentrations at the outflows, STA-5 did not begin flow-through operation until July 2000. Results of monitoring prior to May 1, 2003 have been reported previously (Rumbold and Rawlik, 2000; Rumbold et al., 2001a; Rumbold and Fink, 2002a; Rumbold and Fink, 2003a; Rumbold, 2004a).

As shown in **Table 2** and **Figure 12**, water column concentrations of both THg and MeHg remained low at STA-5 during the water year relative to previously observed spikes. At no time during the reporting year did THg concentrations exceed the Class III WQS of 12 ng/L. Equally important, THg exhibited a negative percent change across the STA (**Table 3**). Alternatively, during the reporting year, MeHg concentration, which has no numerical WQS, was consistently higher in the outflow than in the inflow and thus exhibited a slight positive percent change across this STA for the year (**Table 3**).

Mosquitofish collected near the outflow and inflow culverts of STA-5 contained similar levels of Hg in the second half of 2003. This change occurred in the first half of 2004 when outflow mosquitofish were found to contain slightly more Hg than inflow mosquitofish (**Figure 13**). More importantly, mosquitofish from the interior marshes contained less Hg than fish from either the inflows or the outflows. Overall, tissue-Hg concentrations remained low compared to levels observed at this STA in 2000, or to levels currently found in mosquitofish inhabiting many downstream marshes or canals (see Appendix 2B-1 of this volume).

Similar to 2002, the FWC (under contract to the District to electroshock and collect large-bodied fishes for Hg monitoring) encountered difficulty in filling sample quotas at STA-5 during the reporting year. Only limited numbers of bass were found in Cell 1B, while neither sunfish nor bass were found in Cell 2B, and no bass were found in the discharge canal.

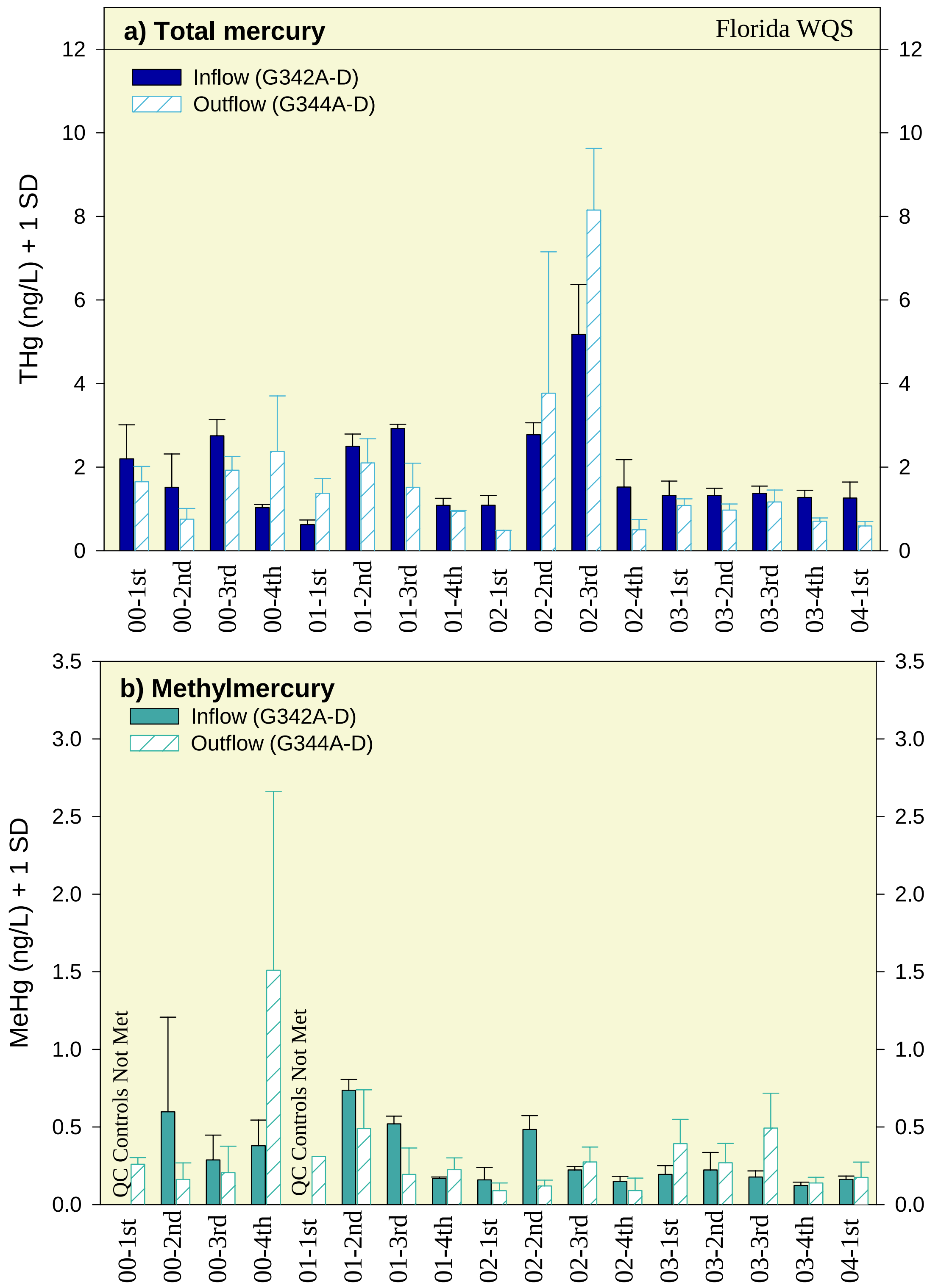

Figure 12. Concentrations of (a) THg and (b) MeHg (ng/L) in unfiltered surface water collected at STA-5.

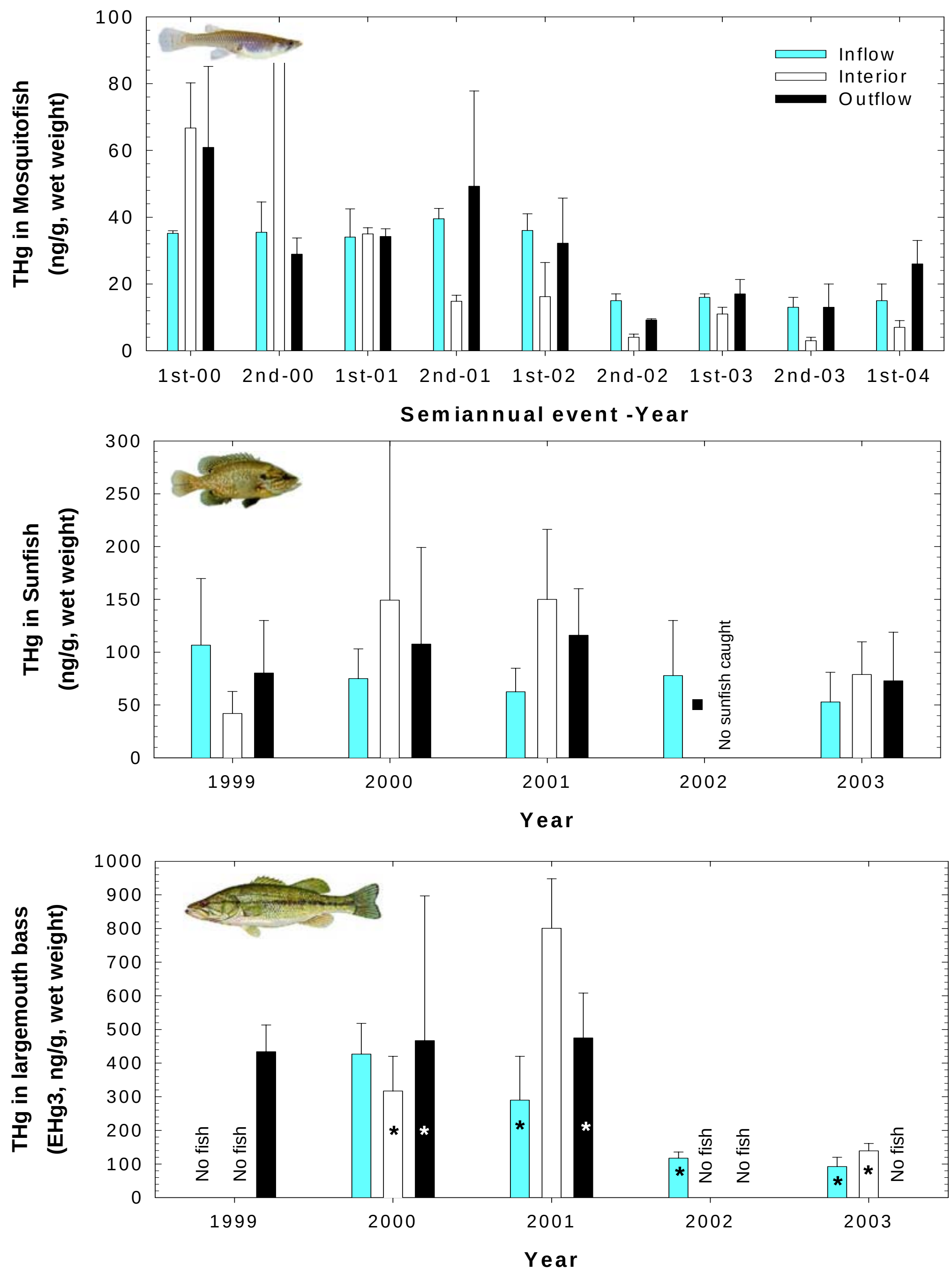

Figure 13. Mercury concentrations (ng/g, wet weight) in (*top*) mosquitofish composites (+ range), (*middle*) whole sunfish (± SD), and (*bottom*) fillets of largemouth bass (± 95% C.I. or, if arithmetic, SD) collected at STA-5. Note that the latter are reported as the expected concentration in a three-year-old fish, that EHg3, unless this could not be calculated (*for details, see **Table 4**), in which case the arithmetic mean is reported.

Hg levels in sunfish (**Table 5**, **Figure 13**) caught in 2003 differed among sites (H = 9.505, df = 2, p = 0.009), with fish from Cell 1B containing significantly higher levels than fish from the supply canal ($p < 0.05$; no other pairwise comparison was significant). However, the spatial patterns in tissue-Hg concentration may have been confounded by significantly larger (and possibly older) fish being caught in Cell 1B (F = 7.2, df = 2, 57, p = 0.002; Tukey post-hoc test, $p < 0.05$). It should be also noted that species of sunfish differed among sites, with fewer warmouth collected from the supply canal. Differences in age and exposure duration or trophic position among the four species of lepomids may account for observed differences in Hg levels.

Sunfish from the discharge canal exhibited interannual differences in Hg levels (H = 8.733, df = 3, p = 0.033), with 2003 levels lower than 2001 ($p < 0.5$, other pairwise comparisons not being significant). Interestingly, sunfish collected in the discharge canal in 2003 were also much larger than fish collected in 2001 (H = 8.733, df = 3, p = 0.033; Dunn's Method post hoc test). Thus, if other factors were equal, they should have contained more Hg, not less. As observed previously, tissue-Hg concentration in sunfish from Cell 1B differed among years (H = 76.502, df = 4, p = < 0.001), with higher levels in 2001 and 2000 than in 1999. Fish collected in 2003 did not differ from 2000 or 2001 fish, but did contain more Hg than 1999 fish.

Largemouth bass collected at STA-5 in 2003 were either limited in terms of age distribution (supply canal) or too few in numbers (Cell 1B) to establish a valid age-concentration relationship and, consequently, assessment of temporal trends must be done cautiously (**Table 6**). For example, the apparent reduction in Hg levels in bass from Cell 1B from 2001 (mean age = 1.8 yrs) to 2002 (1.0 yrs) and 2003 (1.2 yrs) (**Figure 13**) may have simply been a function of sampling different age cohorts between years.

In terms of the risk to fish-eating wildlife, mosquitofish from STA-5 contained Hg at concentrations lower than either the USFWS (100 ng/g), or USEPA criteria (77 ng/g). Sunfish from STA-5 contained levels approaching or just exceeding the USEPA criteria, but were less than the USFWS criteria. After adjusting the arithmetic mean Hg concentrations in fillets to whole-body concentrations (whole-body THg concentration = 0.69 x fillet THg; Lange et al., 1998: average of 63 ng/g and 96 ng/g in bass from the supply canal and Cell 1B, respectively), largemouth bass from STA-5 contained much less Hg than the USEPA predator protection criteria based on TL 4 fish (i.e., 346 ng/g). Therefore, fish-eating wildlife foraging preferentially at STA-5 would not appear to be at elevated risk from Hg exposure.

STA-6, Section 1

STA-6, Section 1 met start-up criteria for Hg in November 1997, and began operation in December 1997. Routine monitoring of Hg at STA-6 was initiated in the first calendar quarter of 1998. Results of monitoring prior to May 1, 2003 have been reported previously (SFWMD, 1998c and 1999c; Rumbold and Rawlik, 2000; Rumbold et al., 2001a; Rumbold and Fink, 2002a; Rumbold and Fink, 2003a; Rumbold, 2004a).

As previously reported (Rumbold, 2004a), THg and MeHg concentrations spiked in STA-6 outflows during the second quarter of 2002 (**Figure 14**). The monitoring program was immediately expanded to include monthly sampling of water to more fully understand the conditions surrounding the spikes. The monthly sampling continued into the current reporting year and ended August 2003, at which time sampling reverted back to quarterly schedules. Results summarized in **Table 2** (only results of quarterly samples are graphically presented in **Figure 14**), demonstrate that concentrations of both THg and MeHg, which had declined to more typical levels within a month following the initial spike (Rumbold, 2004a), remained low through to the end of the current water year. There was no period in which THg concentrations exceeded the Class III water quality standard of 12 ng/L. Furthermore, the annual median percent change across the STA remained negative for both THg and MeHg, suggesting STA-6 was a net sink for both constituents (**Table 3**).

Visual inspection of **Figure 15** suggests that Hg levels in mosquitofish from STA-6 during this reporting year increased slightly relative to the previous year; however, it important to note that this increase occurred first and was most pronounced in mosquitofish from the supply canal (**Table 4**). Mosquitofish from the interior marshes contained less Hg than fish collected adjacent to both the inflow and outflow culverts (**Figure 15**).

Similar to the mosquitofish, sunfish collected from the interior marshes of STA-6 contained less Hg in 2003 than fish from either the supply and discharge canals (**Table 5**, **Figure 15**, Kruskal-Wallis ANOVA on ranks; $H = 29.3$, $df = 2$, $p < 0.002$, Dunn's Method post-hoc test, $p < 0.05$). Because sunfish from the interior were also substantially larger than fish from both the supply and discharge canals that the difference in size between interior fish and discharge canal fish was significant ($p < 0.05$), size would not appear to be a confounding factor. Distributions in numbers of individuals from the four lepomid species were similar among sites. As observed over the previous four years, sunfish from the discharge canal contained greater Hg concentrations than fish from the supply canal (**Figure 15**), resulting in a positive percent change across this STA. However, this difference in Hg levels in fish from the supply and discharge canals was not statistically significant in 2003 (Dunn's Method post-hoc test, $p > 0.05$).

With regard to temporal trends, tissue-Hg levels differed among years in sunfish from the supply canal ($H = 30.632$, $df = 4$, $p < 0.001$); however, 2003 levels did not differ from other years in pairwise comparisons ($p > 0.05$; note that a two-way ANOVA for location and year was inappropriate because the data were not normally distributed). When data on sunfish from Cells 3 and 5 were pooled, Hg levels differed among years ($H = 37.181$, $df = 4$, $p = < 0.001$), with levels lower in 2003 than in 2000 or 2001 (Dunn's Method, $p < 0.05$). Because this assessment may have been confounded by differences in numbers of fish collected from the two cells over the monitoring period, the analysis was reiterated for Cell 5 fish only. Among year differences were again significant ($H = 24.74$, $df = 4$, $p = < 0.001$) with 2003 levels lower relative to 2001 and 2000 ($p < 0.05$), but not compared to 1999 or 2002 ($p > 0.05$). This is important because, as reported last year (Rumbold, 2004a), Cell 5 (which typically contained lower levels in most media as compared to Cell 3), showed peaks in water column concentrations of MeHg in 2002.

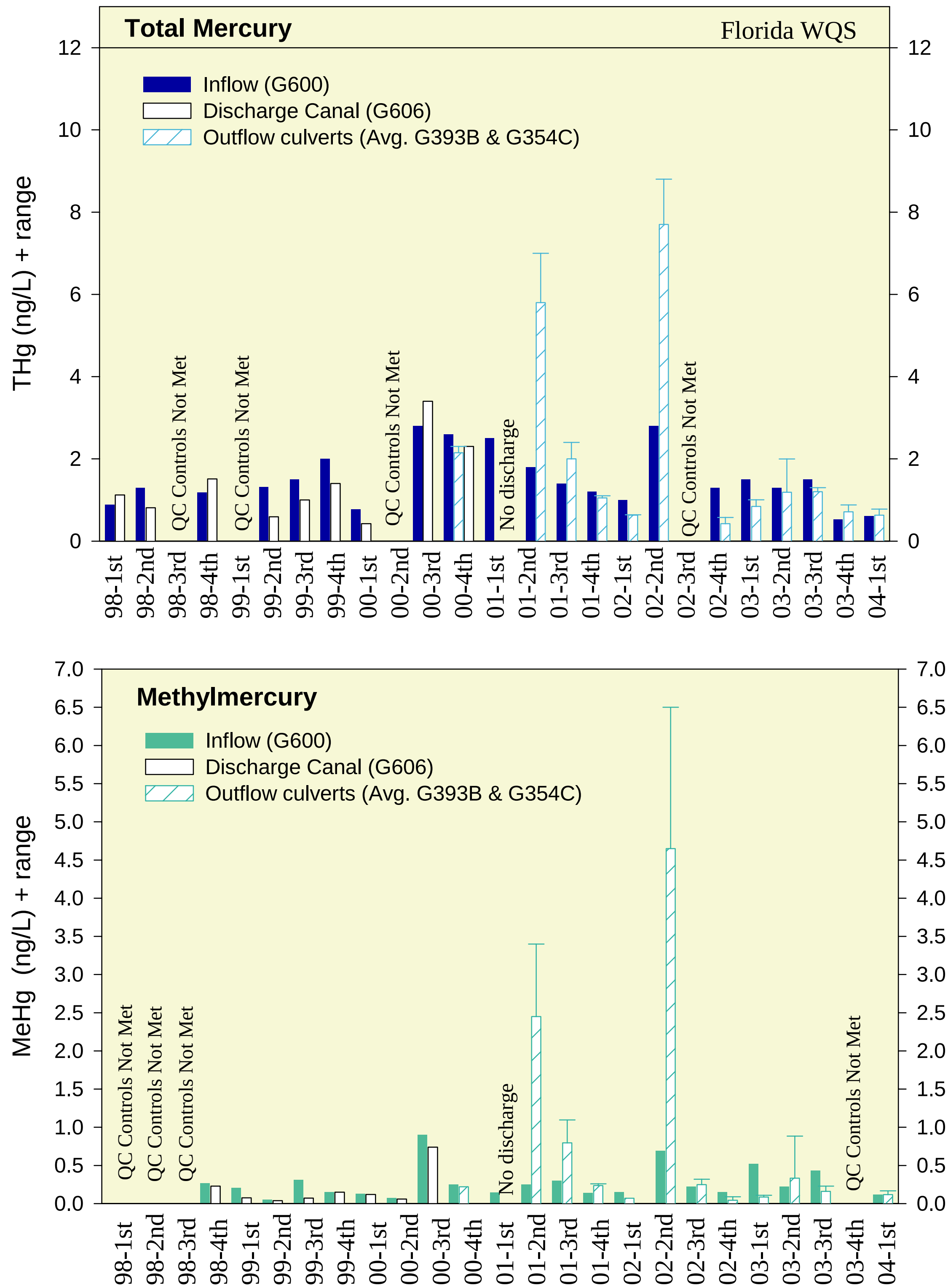

Figure 14. Concentrations of (a) THg and (b) MeHg (ng/L) in unfiltered surface water collected at STA-6.

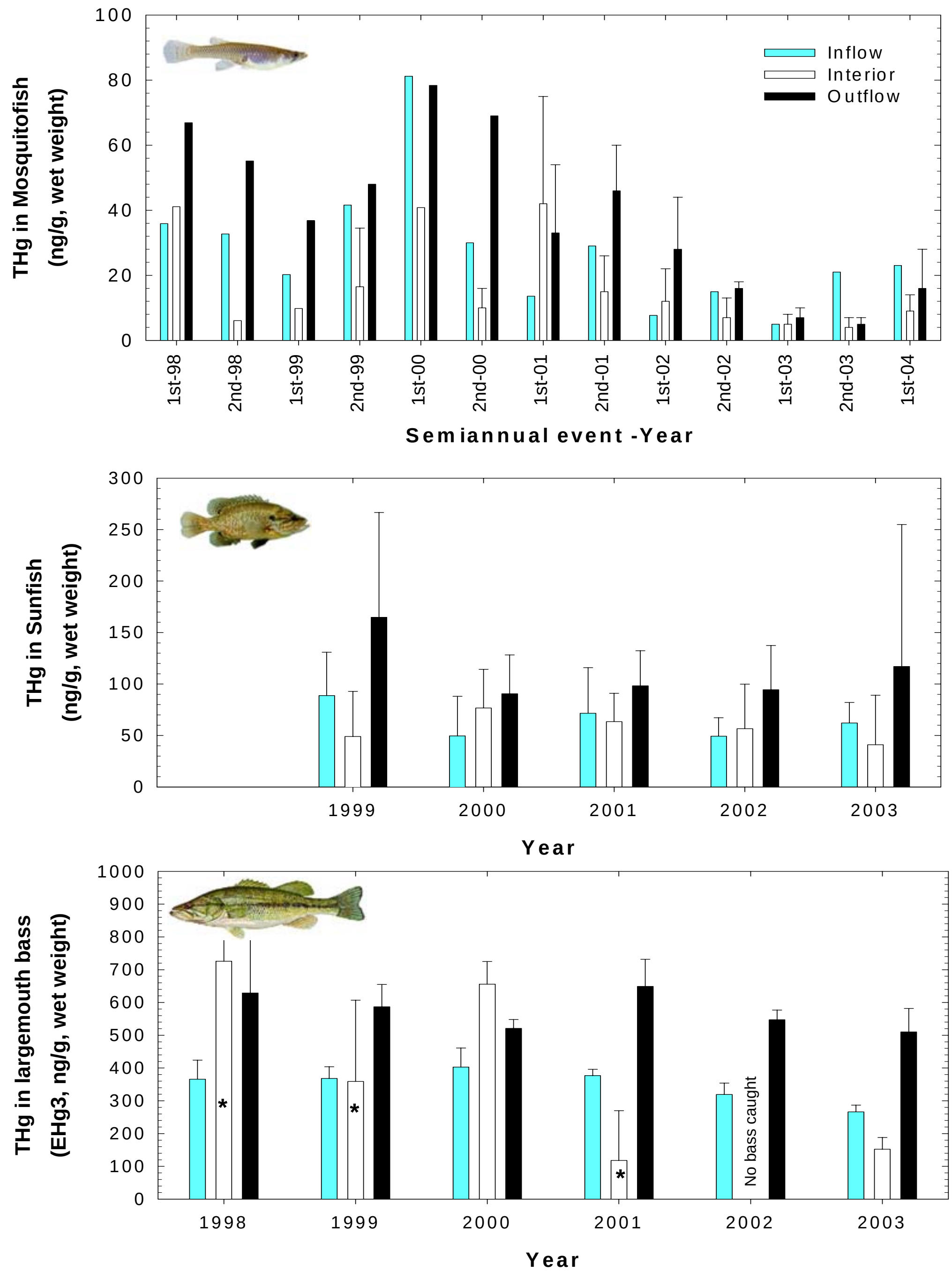

Figure 15. Mercury concentrations (ng/g, wet weight) in (*top*) mosquitofish composites (+ range), (*middle*) whole sunfish (± SD), and (*bottom*) fillets of largemouth bass (± 95% C.I. or, if arithmetic, SD) collected at STA-6. Note that the latter are reported as the expected concentration in a three-year-old fish, EHg3, unless this could not be calculated (* for details, see **Table 5**), in which case the arithmetic mean is reported.

Although Hg levels in sunfish from the discharge canal varied among years ($H = 10.386$, $df = 4$, $p = 0.034$), pairwise comparisons were not significant ($p < 0.05$).

Similar to the sunfish, largemouth bass collected from the STA-6 discharge canal contained greater Hg concentrations in fillets than fish from the supply canal, i.e., a positive percent change across this STA (**Table 6** and **Figure 15**). This pattern has been consistent over the six-year monitoring program. This difference in fillet-Hg concentration between the supply and discharge canals was shown to be statistically significant in 1998 and 2000 by ANCOVA, which can partition the effects of differences in age. Because of an interaction between the effects of age and location on Hg concentration, ANCOVA could not be used to assess among site differences in 1999, 2001, 2002 (see previous reports) or 2003 ($F = 12.2$, $df = 1.36$, $p = 0.001$). For the same reason, among-site differences, with a focus on bass from interior marshes, could not be assessed ($F = 8.8$; $df = 2, 49$; $p < 0.0005$); however, again similar to the sunfish, bass from Cell 5 contained much less mercury (EHg3) than bass from either the supply or discharge canals in 2003.

When temporal trends were assessed, bass from the discharge canal were found to differ among years in age-adjusted Hg levels ($F = 5.8$, $df = 5, 110$, $p < 0.001$). As reported previously, fillets from bass caught in the discharge canal contained less Hg in 2000, than (peak) levels observed in 2001 (Tukey Post-hoc pairwise test, $p = 0.03$); 2003 bass also contained less Hg than fish collected in 2001 ($p < 0.0002$). More importantly, bass caught in the discharge canal also contained less Hg in 2003 than fish collected in 1998 ($p < 0.001$). It is important to note that tissue burdens in bass collected in 1998, less than one year following the start-up of the STA, probably reflected conditions prior to its operation as an STA. Based on this, it appears that Hg levels in bass from the discharge canal have declined and are now lower than baseline levels. Unfortunately, temporal trend assessment of Hg levels in bass from the treatment marshes was difficult because, despite good faith efforts to collect fish from all locations, bass were collected only from Cell 3 in 1998 and 2000 and from Cell 5 in 1999, 2001, and 2003. As discussed in previous reports, the two cells differ significantly in ambient Hg levels and, thus, locational effects would likely confound any temporal trend assessment. When only bass from Cell 5 were assessed, fish caught in 2003 (53 ng/g, $n = 15$) contained much less Hg than fish collected in 2001 (118 ng/g, $n = 9$); sampled bass were of similar age (mean of 1.2 years old in 2003 and 1.1 years old in 2001).

In terms of the risk to fish-eating wildlife, mosquitofish from STA-6 contained Hg at concentrations that were lower than both the USFWS (100 ng/g) or USEPA criteria (77 ng/g). Hg levels in sunfish from the discharge canal exceeded the criteria of both agencies; however, levels in sunfish from the treatment marshes were below both criteria. Whole-body Hg concentrations in bass from STA-6 (157, 36, and 245 for supply, treatment marsh and discharge canal, respectively; estimated from arithmetic mean fillet concentrations using the empirical relationship derived by Lange et al., 1998: THg concentration = 0.69 x fillet) were also below the USEPA predator protection criteria based on TL 4 fish (i.e., 346 ng/g). Therefore, the risk of Hg exposure to fish-eating wildlife foraging preferentially at STA-6 appears to have declined, and based on these criteria may no longer represent a significant threat.

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