Environmental Monitoring and Assessment (2006) **112:** 115–135
DOI: 10.1007/s10661-006-0767-4

EXTREME SPATIAL VARIABILITY AND UNPRECEDENTED METHYLMERCURY CONCENTRATIONS WITHIN A CONSTRUCTED WETLAND

D. G. RUMBOLD* and L. E. FINK
South Florida Water Management District, West Palm Beach, Florida, U.S.A.
*(*author for correspondence, e-mail: drumbol@sfwmd.gov)*

(Received 14 April 2004; accepted 10 January 2005)

Abstract. We began monitoring concentrations of both total mercury (THg) and methylmercury (MeHg) in surface water at Stormwater Treatment Area-2 (STA) on July 20, 2000. This 2602 hectare STA was constructed with three independent marshes to remove phosphorus from agricultural runoff and reduce eutrophication in the northern Everglades. However, there was concern that in doing so, STA-2 might inadvertently worsen the existing mercury problem in the Everglades. Accordingly, operating permits stipulated that flow-through operation of these treatment cells could not begin until concentrations of THg and MeHg in the interior marsh were not significantly greater than corresponding concentrations in the supply canal. Cells 2 and 3 quickly met the start-up criteria in the fall of 2000. In contrast, Cell 1 exhibited anomalously high MeHg concentrations in the fall of 2000 and 2001, and the summer of 2002. During the last such event, water-column concentrations in Cell 1 reached 32 ng THg/L and an unprecedented 20 ng MeHg/L. Tissue Hg in resident fishes reached levels as high as 430 ng/g in mosquitofish, *Gambusia holbrooki*, 930 ng/g in sunfish, *Lepomis spp.*, and 2000 ng/g in largemouth bass, *Micropterus salmoides*. Guided by results from the monitoring program, flow rate and water depth were managed as a means to alter sulfur biogeochemistry and, thereby, reduce *in situ* mercury methylation. This adaptive management strategy likely played a role in the decline in water-column concentrations of THg and MeHg in Cell 1 by late 2002 and the subsequent declines in tissue Hg levels in resident fishes. Cell 1 finally met formal start-up criteria on November 26, 2002.

Keywords: bioaccumulation, constructed wetland, mercury, methylmercury

1. Introduction

In 1994, the Florida legislature enacted the Everglades Forever Act (§373.4592, F.S.) that established long-term water quality goals for the restoration and protection of the Everglades. A crucial element of the plan was the construction of six wetlands, termed stormwater treatment areas (STAs), to reduce phosphorus loading in runoff from the Everglades Agricultural Area (EAA). These STAs were to be built on formerly cultivated lands within the EAA and total over 20,000 hectares. However, the Everglades was also known to have elevated mercury levels in fish (Ware *et al.,* 1990) and there was concern that in attempting to reduce downstream eutrophication, the restoration effort could inadvertently worsen the existing mercury problem (Mercury Technical Committee, 1991). This concern stemmed from

studies that showed flooded soils of new impoundments to be a source of inorganic mercury (Cox *et al*., 1979). Of even greater concern, other studies had shown wetlands to be an important site of mercury methylation (St. Louis *et al*., 1994; for review, see Rudd, 1995). Methylmercury (MeHg) is the more toxic and bioaccumulative form of mercury. Finally, there had already been reports of fish with elevated mercury levels in newly created reservoirs (Abernathy and Cumbie, 1977; Bodaly *et al*., 1984; Verdon *et al*., 1991).

To address these concerns, permits issued by the Florida Department of Environmental Protection (FDEP) to the South Florida Water Management District (SFWMD) for the construction and operation of these STAs required extensive monitoring of mercury levels in sediments, surface water and resident fishes. Although studies elsewhere continued to report both "first flush" of inorganic mercury and MeHg biomagnification leading to a so-called "reservoir effect" (Kelly *et al*., 1997; Paterson *et al*., 1998; Bodaly and Fudge, 1999), results from monitoring the prototype STA, the Everglades Nutrient Removal (ENR) Project, found no evidence of these problems (Watras, 1994; Miles and Fink, 1998). Quite the opposite, the ENR Project was found to be a net sink for both total mercury (THg) as well as MeHg. What is more, fish from the ENR Project had some of the lowest tissue Hg levels in south Florida (Miles and Fink, 1998; Fink, 2000). Nevertheless, monitoring programs at STAs constructed subsequently have revealed transitory spikes in MeHg production (Rumbold, 2004a). We report here on anomalous MeHg production encountered during the start-up of STA-2. We describe how the monitoring program was expanded to define the spatial extent of the anomalous condition and to guide appropriate adaptive management decision-making regarding short-term corrective actions and long-term operational optimization.

2. Materials and Methods

2.1. Study area

STA-2 is located in western Palm Beach County, Florida (Figure 1). To treat discharges from several basins, STA-2 was developed to provide a total effective treatment area of 2,602 hectares (Cell 1 is 805 hectares, Cells 2 and 3 are each 898 hectares). Inflows from S6 and G328 enter the supply canal and are conveyed southward to the inflow canal, which extends across the northern perimeter of the STA. A series of inflow culverts convey water from the inflow canal to the respective treatment cells. Water then flows southward through the treatment cells and eventually discharges into the discharge canal via culverts or gated spillways. Stage is recorded at the tailwater of inflow and headwater of outflow structures; rainfall is recorded at both S6 and S7 structures (Figure 2).

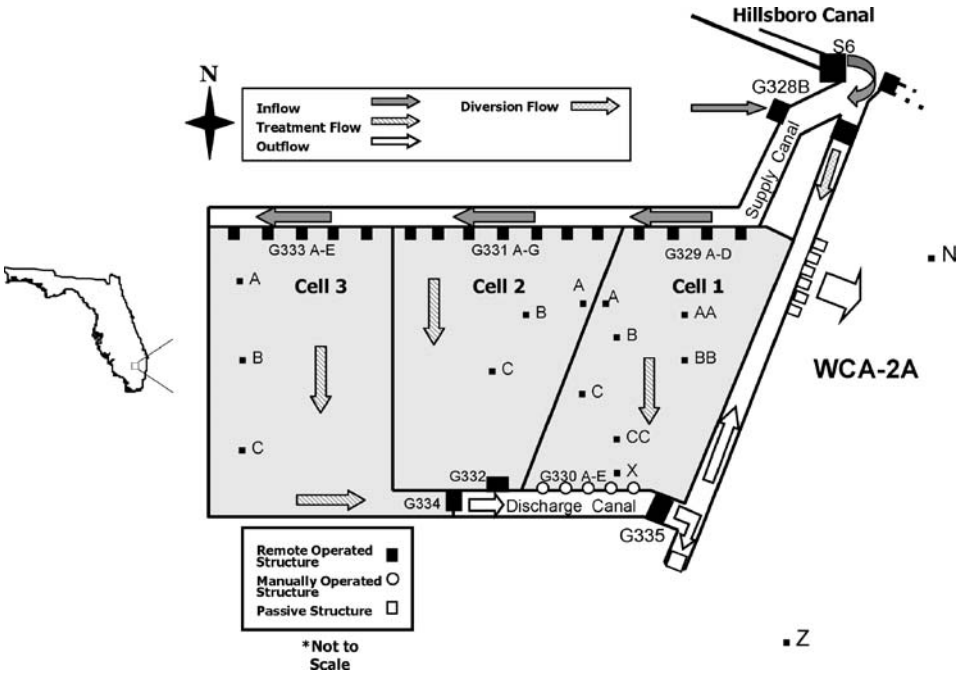

Figure 1. Map showing sampling sites in relation to structures and flows at Stormwater Treatment Area-2. Note, culverts were labeled (A, B, C etc.) from east to west.

Cell 1 discharges through five culverts, labeled G330-A through G330-E, situated along the southern perimeter levee, which extends the length of the discharge canal (Figure 1). Each culvert consists of a corrugated metal pipe with a weir box and removable slide gate. Discharge from Cell 2 is via a single gated spillway (G332) that is located at the cell's southeast corner and the far west end of the discharge canal (Figure 1). Discharge from Cell 3 is also via a gated spillway (G334) situated at the end of a short canal that conveys water from the cell to the west end of the discharge canal (Figure 1). Both G332 and G334 can be operated remotely. The STA-2 outflow pump station, G335, which also can be operated remotely, conveys water to the L-6 Borrow canal. Water in the L-6 canal travels north and then east into WCA 2 or, under high-flow conditions, south and east into WCA 2.

Portions of STA-2 were still being farmed immediately prior to construction. Cell 3 had about 30% in sugarcane and 45% in sod production. Cell 2 had about 10% in sod production (in the northwest corner). Farming had not occurred on the remaining portions of Cell 2 or on any part of Cell 1; instead this area had been maintained in natural condition as the Brown's Farm Wildlife Management Area.

Construction activities began in January 1998 and were completed in June 1999 (N. Larson, SFWMD, personal communication). At that time, inflow gates to Cells 1 and 2 were opened for a brief period then closed; Cell 3 inflow gates remained open

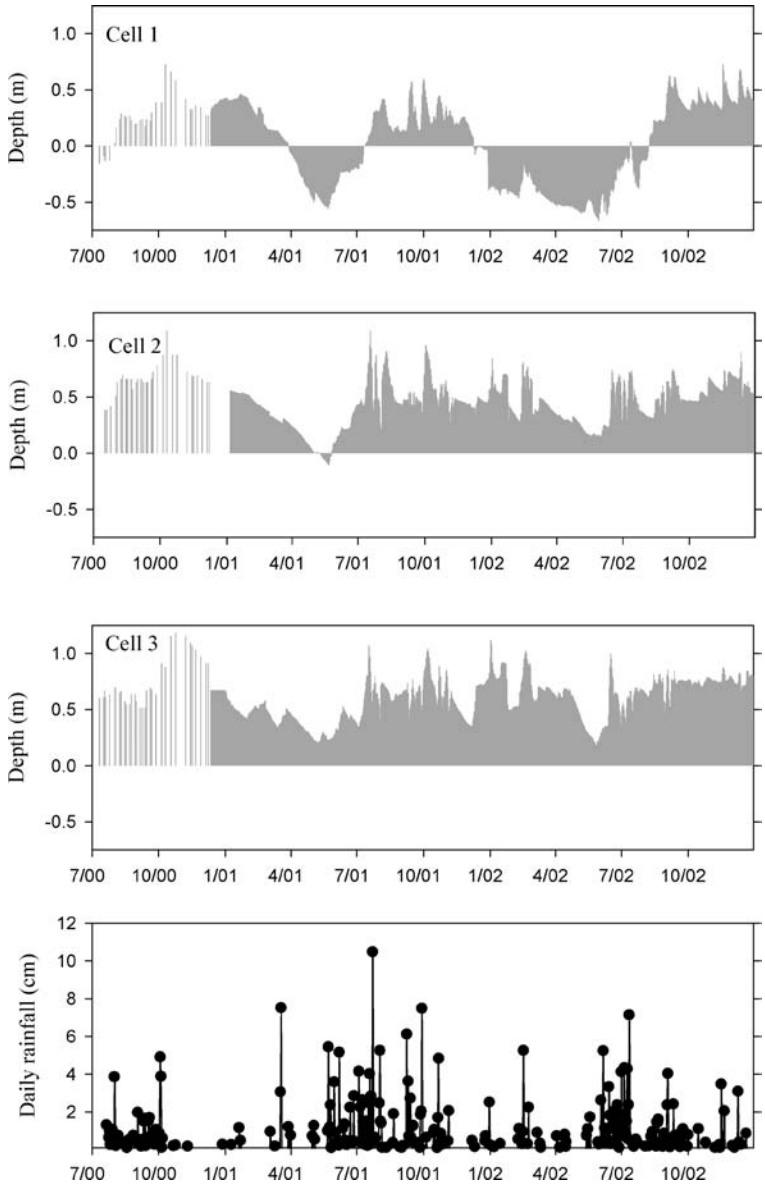

Figure 2. Average water depth within individual cells of STA-2 and average rainfall. Note, depth estimated from mean stage of tailwater at inflows and headwater at outflows minus average ground elevation (3.6 m in Cell 1, 3.1 m in Cell 2, and 2.9 m in Cell 3).

for several months including for the duration of Hurricane Irene (October 15, 1999). Although the inflow gates to Cells 1 and 2 were reopened briefly in December 1999–January 2000, the cells may have partially dried out during the 1999–2000 dry season.

2.2. Routine monitoring

FDEP issued STA-2's operating permit to SFWMD on September 29, 2000. The mercury monitoring plan included requirements for: (1) establishing a soil baseline for mercury (data not presented); (2) start-up monitoring to avoid first flush; (3) operational monitoring; (4) receiving waters monitoring (data not presented); (5) annual reporting; (6) adaptive management; and (7) a quality assurance project plan.

Start-up monitoring required biweekly collection of unfiltered water samples from both the supply canal and from each treatment cell's interior. As a special condition of the permit, flow-through operation could not begin until concentrations of both THg and MeHg in the interior of a treatment cell were no longer significantly greater than the corresponding concentrations in the supply canal. Upon satisfying those criteria, start-up sampling was to be discontinued to be replaced by operational monitoring.

Operational monitoring required quarterly sampling of unfiltered surface water from the supply and discharge canals for THg and MeHg analysis, semi-annual sampling of mosquitofish (*Gambusia holbrooki*) from the supply and discharge canals, and from each treatment cell's interior for THg analysis, annual sampling of sunfish (*Lepomis spp.*) and largemouth bass (*Micropterus salmoides*) at those same sites for THg analysis, and soil sampling for THg and MeHg every 3 years. Although efforts are made to locate sampling sites systematically within cells, marshes were densely vegetated and, consequently, final locations were determined by open water and habitat.

Monitoring of receiving waters, which is a requirement shared by all STA permits, required weekly collection and THg analysis of bulk rainfall at three sites, annual collection and THg analysis of mosquitofish, sunfish and bass at seven downstream marsh sites, and the annual collection and THg analysis of feathers from great egret (*Adrea alba*) nestlings at two downstream colonies (for details, see Rumbold, 2004b).

2.3. Sampling and analytical methods

Surface water sampling followed strict ultra-trace "clean hands – dirty hands" techniques (EPA Method 1669). At each location, two 500- or 1000 mL Teflon bottles were filled using a peristaltic pump and Teflon sampling train; one bottle was for THg determination and the other MeHg. All re-useable equipment and bottles coming into contact with sample water were pre-cleaned, i.e., acid-washed, in a mercury clean lab and transported to and from the sampling site in double zip-lock bags. Bottles and sampling equipment were triple rinsed with ambient water prior to sample collection. Off-the-shelf Meissner capsule filters (0.45 μm) were used in field filtration. Water samples were transported from the field and shipped overnight on blue ice to FDEP's Chemistry Laboratory (Tallahassee, Florida).

A grab sample of 100 to 250 mosquitofish was collected from each location using a dipnet and composited in a labeled zip-lock bag. Mosquitofish composites were later homogenized using a Polytron® tissue homogenizer and the homogenate stored in a clean scintillation vial. Twenty each of sunfish and largemouth bass were collected primarily via electroshocking methods. Whole sunfish were homogenized using a meat grinder. Largemouth bass were filleted and a section of muscle removed for analysis. Tissues were then frozen pending shipment to FDEP's Chemistry Laboratory for analysis.

Total mercury (e.g., Hg^0, Hg^I, and Hg^{II}, as well as organic mercury) determination in surface water followed EPA Method 1631 (Mercury in Water by Oxidation, Purge and Trap, and Cold Vapor Atomic Fluorescence Spectrometry) or a modification of Method 1631. The modification or alternate test procedure for THg analysis, employed during the first 10 months of the monitoring period, involved the substitution of aqua regia for bromine monochloride for the oxidation of mercury species to Hg^{II}. MeHg in surface water was determined by: (1) removing interference from dissolved organic carbon through the addition of 6 N HCl, citric buffer, and 1% diluted antifoaming reagent; (2) direct aqueous phase ethylation using sodium tetraethyl borate followed by purge-and-trap on a Tenex™ trap; and (3) thermal desorption of trapped material into an isothermal GC column for peak separation and quantification by Cold Vapor Atomic Fluorescence Spectrometry (CVAFS). THg determination in tissues followed either EPA Method 245.6 or, if at low level, a modification of Method 1631. In the latter, samples were first predigested in nitric acid, then oxidized with bromine monochloride. This was followed by purged and trap on dual gold traps and CVAFS.

2.4. Quality assurance

This monitoring program was conducted under SFWMD's quality assurance (QA) program and a project-specific quality assurance project plan (approved by FDEP) with stringent laboratory and field quality control requirements. Laboratory quality control (QC) samples included method blanks, lab-fortified blanks, matrix spikes, standard reference materials, laboratory duplicates and blind tissue splits. The FDEP laboratory was certified under the National Laboratory Accreditation Program (NELAP) and routinely ranked high in round-robins for both THg and MeHg analysis (for review, see Niu and Tintle, 2003). Field QC samples included trip blanks, field blanks, equipment blanks, both of pre-cleaned equipment at the start of sampling and of field-cleaned equipment at the end of sampling, container (e.g., vials and bags used to ship solids) and processing equipment blanks (e.g., blender, grinder, Polytron©, and cutting boards) and field duplicates. Annual assessments of this QA/QC program are reported elsewhere (Rumbold and Fink, 2002, 2003a; Rumbold, 2004b).

2.5. Statistical analysis

Water-column concentration at the interior site was compared to average concentration in the supply canal using a single sample, one-tailed Student's *t*-test, i.e., mean plus 95% upper confidence level (UCL). To partition or remove the confounding influence of age (i.e., length of exposure), mercury concentrations in largemouth bass were standardized to an expected mean concentration in 3-year-old fish (hereafter symbolized as EHg3) at a given site by regressing mercury against otolith age. This protocol (and estimated age) is consistent with the protocol used by the Florida Fish and Wildlife Conservation Commission to evaluate risk from harvesting legal-sized bass. Sunfish were not aged, so age normalization was not available; however, efforts were made to estimate a least square mean (LSM) Hg concentration based on the fish length. Where appropriate, analysis of covariance (ANCOVA), was used to evaluate spatial and temporal differences in mercury concentrations, with age (largemouth bass) or length (sunfish) as a covariate (SAS, SAS Institute Inc., Cary, NC). However, use of ANCOVA is predicated on several critical assumptions (for review, see ZAR, 1996). Regressions also require that collected samples exhibit a relatively wide range of the covariate. Where these assumptions were not met, standard analysis of variance (ANOVA) or Student's *t*-tests (SigmaStat, Jandel Corporation, San Rafael, CA) were used with covariates 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, they were used in ANCOVA or ANOVA. If they did not meet the assumptions, then raw data sets were evaluated using nonparametric tests, such as the Kruskal-Wallis ANOVA. If the multi-group null hypothesis was rejected, groups were compared using either Tukey HSD (honestly significant difference) or Dunn's method.

3. Results and Discussion

Biweekly start-up monitoring of surface water at STA-2 began on July 20, 2000 (Figure 3). Cell 3 satisfied the start-up criteria on September 14, 2000, when water-column concentrations of both THg and MeHg in the interior (1.6 ng THg/L and 0.045 ng MeHg/L) were not significantly greater than average concentrations in the supply canal (mean plus the 95% UCL; $1.34 + 0.46$ ng THg/L, $n = 5$ and $0.29 + 0.26$ ng MeHg/L, $n = 3$ unqualified values). During this same period, concentrations of both THg and MeHg fluctuated in Cells 1 and 2, but were consistently greater than concentrations in the supply canal (Figure 3). Water-column concentrations had fluctuated similarly in the other STAs, owing to their natural variability, even through a diurnal cycle (1.5–2 fold change in THg and 2–5 fold change in

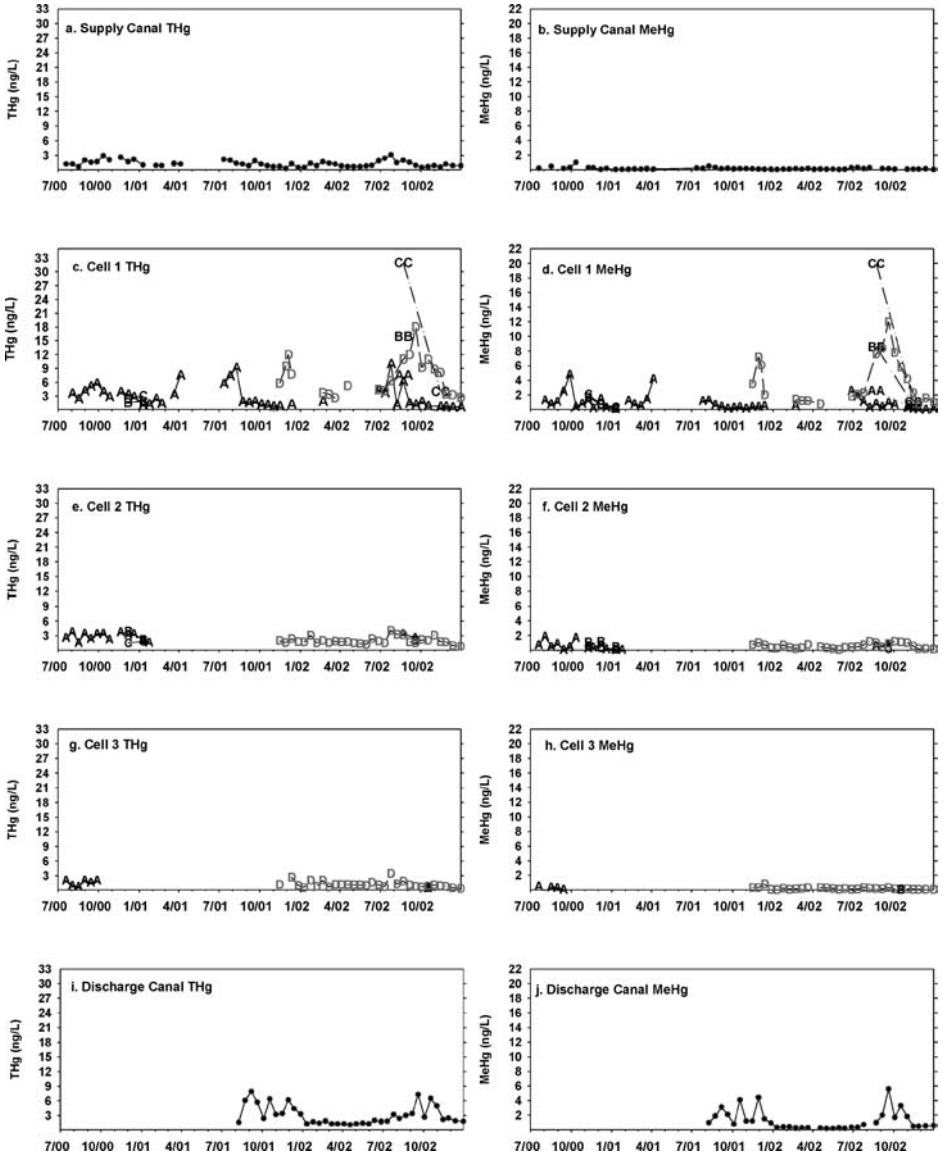

Figure 3. Time series of total mercury (THg) and methylmercury (MeHg) concentrations in unfiltered surface water collected at STA-2. Sampling sites within cells labeled alphabetically from north to south (for map, see Figure 1). Note, site D represents sampling immediately upstream of outflow structures in each cell, i.e., G330A in Cell 1, G332 in Cell 2 and, G334 in Cell 3.

MeHg; Krabbenhoft *et al*., 1998), rapid response to influx of new inorganic Hg or sulfate (Gilmour *et al*., 2004), as well as analytical variability.

The first indication of an anomalous situation in STA-2 came on September 28, 2000, when a sample from Cell 1 (i.e., site A) was found to contain

4.8 ng MeHg/L. A confirmed MeHg concentration of this magnitude had not previously been observed in any water sample collected in South Florida by SFWMD or the U.S. Geological Survey (D. Krabbenhoft, USGS, personal communication). The U.S. Environmental Protection Agency (USEPA) had encountered only two slightly higher MeHg values in 100s of samples of surface water collected in South Florida from 1995–1999 (D. Scheidt, USEPA, personal communication). Further, 4.8 ng MeHg/L represented 83% of the 5.8 ng THg/L present in the paired sample and this %MeHg was elevated relative to other areas (Southworth *et al*., 2002) and even relative to high values typically found in the Everglades (Hurley *et al*., 1998; Miles and Fink, 1998). MeHg concentration in the adjacent Cell 2 was only 10% of that detected in Cell 1 (Figure 3). Because the three cells shared a common supply canal (i.e., inflow loading), would be expected to receive equal rainfall (i.e., atmospheric wet deposition), and were located away from potential emission sources (i.e., that might produce a steep gradient in dry deposition), the extreme among-cell differences were somewhat puzzling.

On November 9, 2000, Cell 2 satisfied the start-up criteria when water-column concentrations of both THg and MeHg in the interior (2.0 ng THg/L and 0.24 ng MeHg/L) were not significantly greater than average concentrations in the supply canal ($1.68 + 0.39$ ng THg/L, $n = 9$ and $0.40 + 0.25$ ng MeHg/L, $n = 6$ unqualified values). A few samples were subsequently collected from Cell 2 prior to our receiving laboratory results of acceptable concentrations; some of these showed THg and MeHg to be at slightly greater concentration in Cell 2's interior than in the supply canal (Figure 3). This had also occurred following Cell 3 satisfying the start-up criteria and was expected given the inherent variability in environmental conditions, sampling and ultra-trace analyses.

Pursuant to negotiations with FDEP, the monitoring program was formally expanded in Cell 1 in late November 2000. Specifically, SFWMD agreed to begin collecting filtered samples and would increase the number of surface water collection sites in Cell 1 from one to three, thereby establishing a transect from north (starting at site A) to south along the flow gradient. A similar transect would also be sampled in Cell 2 as a reference. Additionally, mosquitofish collections, which SFWMD had voluntarily begun immediately after receipt of the laboratory results showing anomalously high water-column MeHg, were to continue on a monthly basis from Cell 1. Because mercury methylation occurs primarily in sediments and is known to be influenced by a number of different sediment parameters (e.g., total sulfur, total iron, etc.), SFWMD would also collect and analyze a second set of sediment cores from locations sampled during baseline collections. Results from sediment re-sampling did not explain the among-cell differences in water-column concentrations (Rumbold and Fink, 2002). As discussed in ensuing sections, the expanded, follow-up monitoring of surface water and fish would continue for 90 days.

Sampling along the transect in Cell 1 soon confirmed the elevated MeHg concentrations (Figure 3) and demonstrated a slight positive gradient in water-column

concentrations from north to south (i.e., from site A to C, Figure 1). A similar gradient was not observed in Cell 2. Filtered samples showed percent dissolved THg, operationally defined as passing through a 0.45 μm filter, ranged from 67% to 94% ($n = 10$ unqualified values); percent dissolved MeHg ranged from 46% to 196% ($n = 16$, median = 82%). The apparent reversal, i.e., filtered concentration greater than unfiltered, may have resulted from temporal variability in serially collected samples (unfiltered and then filtered), the analytical uncertainty at ultra-trace levels or a combination of both (the 196% dissolved MeHg was calculated from 0.107 ng MeHg/L and 0.21 ng MeHg/L in unfiltered and filtered samples, respectively). Regardless, there was no compelling evidence to suggest that the elevated MeHg concentrations were simply a result of suspended solids.

As expected, mercury bioaccumulation by mosquitofish tracked MeHg concentrations in surface water, both spatially and temporally (Figure 4). Mosquitofish were selected as one of the sentinel species because of its widespread occurrence in the Everglades, its ability to invade newly flooded areas, and because of its relatively small home range and short life span. The latter is important because the population is adjusting (tissue levels) to new conditions more rapidly than, say, a population of bass that are longer-lived. These characteristics make the mosquitofish a potentially excellent bioindicator of changes in environmental MeHg concentrations through time. Initial collections made in October 2000, showed tissue Hg level in Cell 1 mosquitofish to be 2.6–13 times greater than concentrations in mosquitofish from Cell 2 and the supply canal, respectively.

From September 2000 through March 2001, water-column concentrations of both THg and MeHg fluctuated in Cell 1, but did not exceed the previously observed concentration spike (Figure 3). At the same time, tissue Hg levels began to climb in Cell 1 mosquitofish, reaching 346 ng/g in fish at site C in March 2001 (Figure 4). A tissue Hg concentration of this magnitude had not previously been observed in mosquitofish collected by SFWMD. Furthermore, similar to water-column concentrations, a positive gradient was observed in tissue Hg levels in mosquitofish within Cell 1, i.e., levels increased from site A to site C (Figure 4).

Heavy rains fell on March 18 and 19, 2001, temporarily reversing a drawdown of Cell 1, which had begun in early 2001 due to a severe drought (Figure 2). Following the rains, THg increased dramatically in Cell 1 (Figure 3); MeHg concentration also increased in Cell 1 until it reached 4.2 ng/L on April 5, 2001. This sequence of a rainfall event followed by a spike in surface water THg concentration was repeated several times during the monitoring period (Figures 2 and 3). This was expected, given that bulk rainfall collected in South Florida has some of the highest THg concentrations in the National Atmospheric Deposition Program's (NADP) Mercury Deposition Network (annual, volume-weighted concentration in 2001 ranged from 13.1 to 13.9 ng THg/L at the three MDN stations in south Florida; Rumbold, 2004b). It was also not a surprise that the correlation between rainfall and water-column THg concentration would be confounded by among-cell differences in stage and inflows (i.e., to dilute THg in the rain). For instance, the shallower depth in Cell 1,

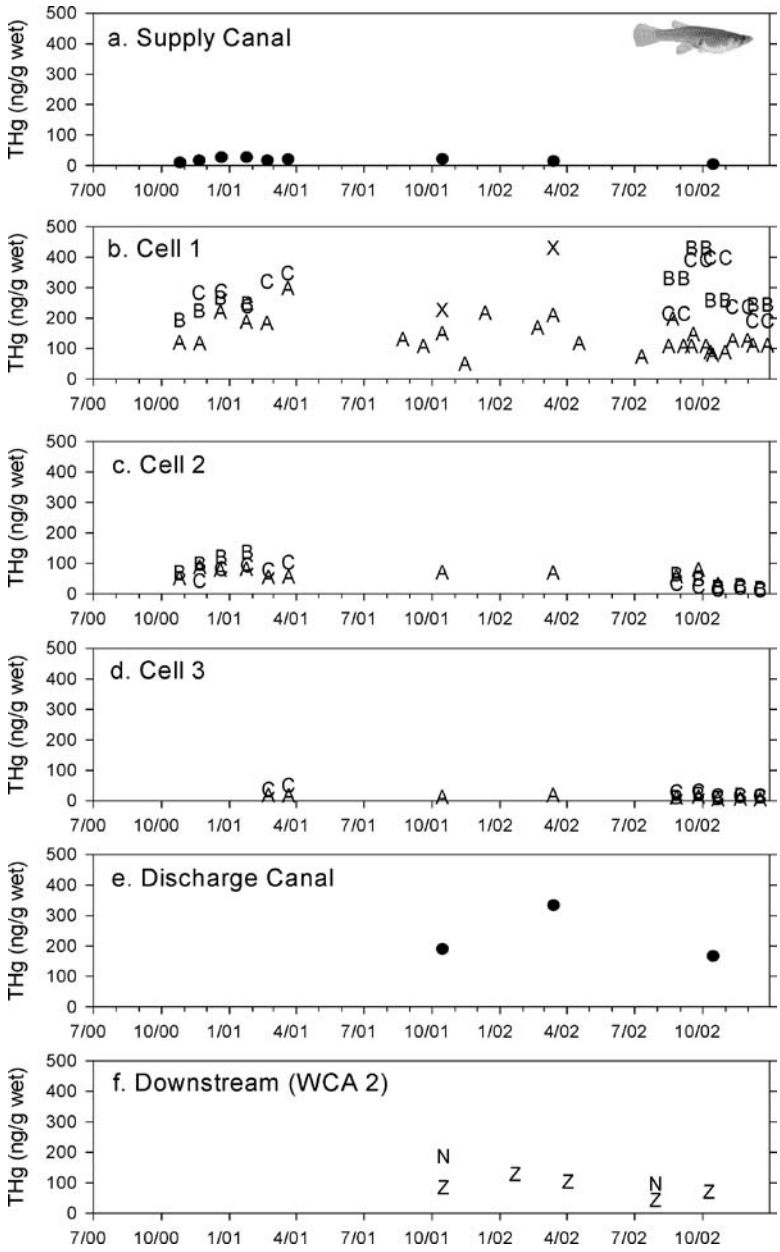

Figure 4. Time series of tissue Hg levels in composited mosquitofish collected within (along transects, labeled alphabetically from north to south) and downstream of STA-2. Note, in an attempt to locate more representative sites in Cell 1, the transect was shifted to the east, to sites AA, BB and CC (Figure 1), in 2002.

relative to the other cells (Figure 2), may have led to its March 2001 spike in THg (Figure 3). The shallower depth may have also allowed a greater amount of inorganic Hg in the rain to reach the sediments and, thus, sulfate-reducing bacteria, before being processed in the water column (e.g., reduction and efflux back to atmosphere, complexation with DOC, etc.). Following a short time lag, this may have resulted in the spike in MeHg concentration, relative to the other two cells (for response times, see Gilmour *et al*., 2004). Yet, THg and MeHg concentrations also fluctuated during periods of little or no rainfall. Thus, it is unlikely that among-cell differences in stage, inflow and dilution could alone account for the spikes. Among-cell differences were more likely attributable to, as yet, unmeasured differences in THg release from soils (Cox *et al*., 1979; Kelly *et al*., 1997), or in the way new Hg was assimilated within each cell.

In April and May 2001, Cells 1 and 2 went dry, respectively, due to a region-wide drought (Figure 2). Alternatively, Cell 3 received supplemental water deliveries to preserve submerged aquatic vegetation and, consequently, did not dry out. Following local rains, Cell 2 was reflooded in early June 2001; Cell 1 remained dry until mid July 2001 (Figure 2).

These results from the 90-day follow-up monitoring program were submitted to FDEP and later published on the internet for stakeholder review (Rumbold and Fink, 2002; Fink, 2002). At the same time, a request was made to FDEP for a modification of the permit to allow immediate start-up of flow-through operation of Cell 1 as an adaptive management strategy to alter its sulfur biogeochemistry and, thereby, disrupt MeHg production (for supporting documentation, see Fink, 2002). The basis for the request was that (1) shallow, standing water in Cell 1 was likely to be the worst case for MeHg production, bioaccumulation and, hence, ecological risk; and (2) flow-through operation and increased sulfate loading was more likely than not to foster conditions in pore water, e.g., redox conditions and sulfide concentrations, that would inhibit MeHg production (for discussion of sulfide inhibition see, Benoit *et al*., 1999). A similar, serendipitous condition was believed to be responsible for inhibiting MeHg production in the ENR Project (Fink, 2002). In August 2001, FDEP granted the permit modification to authorize Cell 1 flow-through operation without it first satisfying the start up criteria. The modified permit required 12 additional months of expanded monitoring followed by a formal ecological risk assessment (results of which are reported in Rumbold, in press). Required monitoring included the continuation of biweekly surface water collection from the supply canal, Cell 1, and the discharge canal; continued monthly mosquitofish collection from Cell 1, the addition of quarterly mosquitofish collection from two downstream sites (i.e., N and Z, Figure 1); and semiannual sunfish collection from Cell 1.

From August through December 2001, concentrations of both THg and MeHg decreased at site A in Cell 1 and remained relatively low (Figure 3). This was initially interpreted as a indication that THg processing from atmospheric inputs or sedimentary release had stabilized and, consequently, net MeHg production had decreased. However, it was difficult to reconcile the low concentrations observed in

Cell 1's interior with the dramatic fluctuations in those constituents occurring in the discharge canal (Figure 3). Either Cells 2 or 3 had become a significant source of both THg and MeHg or site A was not representative of Cell 1 discharge. Because of the configuration of the cell outflows, especially the arrangement of the un-gated culverts from Cell 1 along the length of the discharge canal, there was also a growing concern regarding the representativeness of samples collected from the discharge canal. The concern was that, depending on the sequence of events preceding sample collection (i.e., pump operation, gate opening and closing), surface water in the discharge canal might misrepresent the true concentrations and loads exported. For example, if the outflow gates from Cells 2 or 3 were closed (via a remote signal from the Operation Control Room) prior to, or immediately following shutdown of the G335 Pump, the increase in the stage of the discharge canal, which can be lowered by as much as 2 m during extreme pumping, would be from flow through the un-gated culverts G330A-E or seepage through the berm. Any sample collected prior to the next pump start-up would reflect only the conditions and events surrounding the shut down and stage increase and not concentrations or load exported under steady state pumping conditions. In late 2001, efforts were made to sample the discharge canal only under steady state flow, but this was quickly abandoned due to the inherent difficulties in attempting to time routine sample collection to coincide with irregular pump operation at this remote location. To identify the source of the elevated THg and MeHg in the discharge canal, monitoring was further expanded in November 2001 to include sampling upstream of outflow structures in each of the cells (i.e., G330A in Cell 1, G332 in Cell 2 and G334 in Cell 3; for locations, see Figure 1; designated as site D in each panel of Figure 3). The results from monitoring the individual outflows revealed extremely high concentrations of both THg and MeHg in the outflow of Cell 1, relative to the outflows of Cells 2 and 3, standing water in the discharge canal and at site A in Cell 1 (Figure 3). On December 5, 2001, THg concentration at Cell 1's outflow reached the State's Water Quality Standard of 12 ng THg/L. These results suggested that the dramatic fluctuations in concentrations observed in the discharge canal from August through December 2001 (Figure 3) were likely a result of a disproportionate amount of source water from Cell 1.

Mosquitofish collections resumed from Cell 1 in August 2001, a month following its reflooding. An additional collection site, X, located in a ditch or swale immediately upstream of Cell 1's outflow culverts, was added to the monitoring program in October 2001. This was done because of the observed positive gradient within the cell and because reconnaissance in Cell 1 indicated that large-bodied fish collections would likely be unsuccessful at site A due to poor habitat (i.e., lack of deeper, open water). It was preferred that mosquitofish were collected at the same site as the large-bodied fish. Tissue Hg levels in the mosquitofish from site X were found to be very similar and tracked levels observed in fish from the discharge canal (Figure 4). For the same reasons discussed earlier regarding surface water exchange between Cell 1 and the discharge canal, this prompted speculations that fish were moving through the G330 culverts.

In October 2001, sunfish and largemouth bass were collected from STA-2. Sunfish exhibited significant spatial patterns in tissue Hg (Kruskal-Wallis ANOVA on Ranks; $H = 44.554$, $df = 5$, $p < 0.001$) reminiscent of patterns observed in mosquitofish (Figure 5). Sunfish collected at site X in Cell 1, for example, contained the highest median tissue Hg concentration (155 ng/g), which differed significantly from levels in fish both from Cell 3 and from the supply canal (Dunn's Method, $p < 0.05$). Owing to short hydroperiod and poor habitat, attempts to collect large-bodied fish at downstream site, N, were unsuccessful. Sunfish from the other downstream site, Z, were noteworthy in that the sampled population was significantly smaller in total length than most other sunfish from STA-2 (ANOVA, $df = 5, 110$; $p < 0.001$; Tukey Test, $p < 0.05$). Because these downstream sunfish exhibited a significant size – tissue Hg concentration relationship (Linear regression; $df = 1, 18$; $F = 9.6$; $p = 0.006$), larger fish, if they had been collected, could have been expected to contain greater concentrations. Visual inspection of the size-concentration relationships at sites within the STA, none of which were statistically significant ($p > 0.05$), showed smaller, presumably younger, fish in Cell 1 and the discharge canal contained higher Hg levels than larger fish. This suggested that fish from Cell 1 and the discharge canal had been exposed to increased environmental MeHg concentrations recently. The age distribution of the bass was too narrow at all but one site to allow for ANCOVA. However, because median age of the bass (1.8 yrs) was similar at five of the six sites (statistical analysis excluded two bass caught at site Z), spatial variability in tissue Hg levels was evaluated using a Kruskal-Wallis One-Way ANOVA and was found to be significant ($h = 52.8$, $df = 4$, $p < 0.001$; Figure 6). The spatial pattern in tissue Hg levels in bass was similar to those observed in mosquitofish and sunfish (Figures 4 and 5). The discharge canal was the only site at which age distribution of the sampled bass was sufficient to establish a significant age-concentration relationship ($df = 1, 18$; $F = 24.4$; $p < 0.001$). The estimated mean tissue Hg concentration in a 3-year-old bass (EHg3) at this site was $1{,}148 \pm 158$ ng THg/g. None of the other STAs contained bass with tissue Hg levels of this magnitude (Rumbold and Fink, 2003). Further, only two of the monitoring sites in the receiving waters, both known "MeHg hotspots", had fish with comparable levels (Rumbold and Fink, 2003).

In late November 2001, SFWMD requested and was granted permission to dry out Cell 1. This request arose from a growing concern about water availability, with the onset of the dry season within the longer-term drought, to maintain the target stage called for in the modified permit. Although there would be a short-lived increase in exposure and risk to fish-eating wildlife when fish were concentrated during the dry out, this was deemed more desirable than allowing MeHg production and biomagnification to continue unabated. At the same time, SFWMD and FDEP also began developing a Memorandum of Agreement (MOA) that included provisions for more intensive monitoring of surface water, sediments, pore water, and vegetation by SFWMD upon reflooding of Cell 1. These additional data would be necessary for mass balance calculations, including a water budget that could account

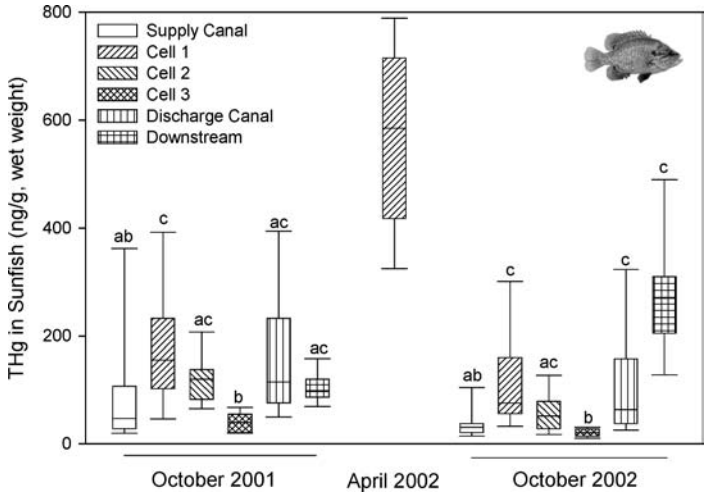

Figure 5. Tissue Hg levels in whole sunfish from STA-2 and downstream site. The boundary of the box represents the 25th and 75th percentile, the line within the box the median; whiskers above and below the box, the 90th and 10th percentile. Sites with similar letter designations during a given sampling event did not differ significantly.

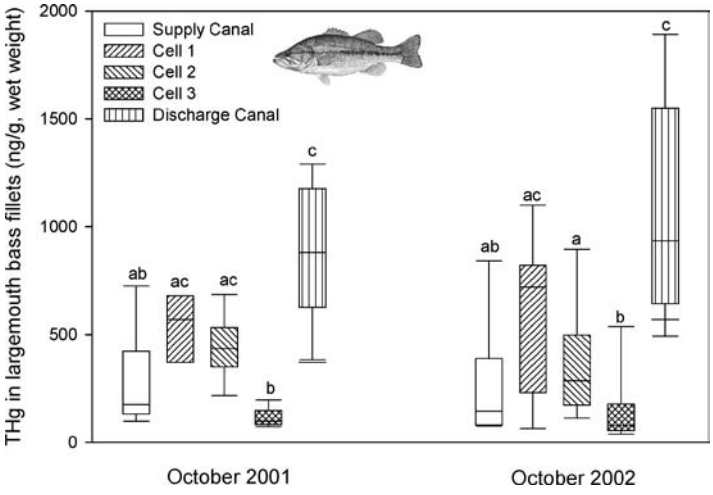

Figure 6. Mercury concentrations in fillets of largemouth bass collected at STA-2. Note, only two bass were collected at the downstream site, Z, both in 2001 (350 and 390 ng/g) and 2002 (560 and 580 ng/g). Sites with similar letter designations during a given sampling event did not differ significantly.

for ground water fluxes, which are too detailed and lengthy to report here (for results, see Fink *et al*., 2005). The MOA also called for process-level studies of this unintended, large-scale experiment by researchers with the U.S. Geological Survey and the Academy of Natural Sciences. The MOA was formally approved in February 2003. Process-level studies are not yet completed; however, preliminary results

showed elevated levels of sulfate and MeHg in sediment cores taken from Cell 1 that were dried and re-wet in the laboratory (C. Gilmour, Academy of Natural Sciences, personal communication). These results were consistent with findings from a 1999 field study on the effect that drought and muck fires had on mercury cycling in the Everglades (Krabbenhoft and Fink, 2001). The results of that study suggested sulfide pools in the sediments (e.g., organic sulfide, disulfides, and acid volatile sulfides) had oxidized during the drydown. Upon rewetting, the available sulfates stimulated sulfate-reducing bacteria and mercury methylation. This pulse of MeHg production continued until the sulfate was exhausted or until sulfide reached inhibitory concentrations. The findings from drying and re-wetting the cores also supported a growing suspicion that similar oxidation processes might be occurring in Cell 1 sediments, which should have otherwise been anoxic and reducing in nature.

From August 9 through November 29, 2001, average water depth in Cell 1 was 27.5 cm (Figure 2). However, closer inspection of stage data revealed periods of very shallow depth (i.e., as low as 5 cm) near Cell 1's outflow structures. The gradient in stage across the cell was a function of the timing and magnitude of inflow and outflow pump operation, as well as the capacity of the un-gated culverts. It was also a function of the flow rate across the cell that was impeded in certain areas by its gentle slope to the south (approximately 3 cm per km), topography and dense vegetation. The factors that produced the anomalous conditions within Cell 1 will not be fully understood until the results from all studies have been synthesized. However, the shallower conditions in the southern portion (i.e., bottom) of the cell were believed to have allowed sediments to partially oxidize and, as a result, were thought to have played a role (i.e., through sulfide oxidation) in the observed gradients in mercury methylation. Accordingly, to reduce the influence of outflow pump operation and prevent recurrence of the steep gradients in stage within the cell, weir boxes in front of the outflow culverts were reconfigured during the dry out to increase control elevation.

In April 2002, Cell 1 was almost entirely dry except for standing water in the small ditch located upstream of the outflow culverts. The tissue Hg level in mosquitofish collected from this ditch (i.e., site X) was at an unprecedented 430 ng/g (Figure 4). In accordance with the modified permit requiring semiannual sunfish collections, a net was pulled the length of the ditch and, surprisingly, twenty sunfish and one largemouth bass were captured. These sunfish contained a median tissue Hg concentration of 585 ng/g. This represented an almost 3 fold increase over levels observed in fish collected here in October 2001 (Figure 5); the difference in tissue Hg levels between sampling periods was statistically significant ($df = 34, t = -7.124$, $p < 0.001$). Moreover, average tissue concentration in these sunfish was twice the basin-wide average for sunfish collected at the downstream Everglades' sites at that time (Rumbold and Fink, 2003). A fillet from the bass, which was estimated to be 1.5 years old, contained 2000 ng Hg/g. It should be re-emphasized here that exposure and risk to fish-eating wildlife was negligible, or nearly so, because most of the treatment cell was dry.

In July 2002, Cell 1 began to reflood from direct rainfall and stormwater inflow (Figure 2). Almost immediately, water-column concentrations of both THg and MeHg began to increase in the cell (Figure 3). Over time, tissue Hg levels also increased in mosquitofish, particularly at sites CC and BB (Figure 4). Then, on August 22, 2002, results of sampling unfiltered surface water along the Cell 1 transect showed a steep positive gradient with concentrations of 32 ng THg/L and 20 ng MeHg/L at site CC (Figure 3). To our knowledge, this is the highest MeHg concentration reported in surface water (*cf.* 12.27 ng MeHg/L reported by Brigham *et al.*, 2002). Filtered samples collected immediately after the unfiltered samples contained 24 ng THg/L and 20 ng MeHg/L, suggesting 100% of the MeHg was in the dissolved form (and that 83% of the dissolved THg was in the methyl form). Concentrations of THg and MeHg were also extremely high immediately upstream of the Cell 1 outflow culvert (designated site D in Figure 3). On September 1, 2002, Cell 1 stage was sufficient to overflow the weirs and began discharging. Soon thereafter, concentrations of both THg and MeHg began to increase in the discharge canal (Figure 3).

In late August 2002, mercury levels also peaked in the Cell 1 mosquitofish, again reaching 430 ng/g (this time at site BB, Figure 4). In October 2002, the second annual collection of sunfish and bass occurred at STA-2. Although statistical analyses was somewhat hampered (i.e., despite various transformations the datasets were either not normally distributed or had unequal variances and, consequently, a two-way ANOVA was inappropriate for assessing between-year differences and location effect), the sunfish exhibited spatial patterns in tissue Hg that were very similar to 2001 (Kruskal-Wallis ANOVA on Ranks; $H = 63.6$, $df = 5$, $p < 0.001$; Dunn's Method, $p < 0.05$; Figure 5). In terms of temporal trends, interestingly, sunfish were larger at all sites in 2002, suggesting fish were older and, thus, had a longer period of exposure. Yet, tissue Hg levels were generally lower in 2002 than 2001 (Figure 5). However, caution must be exercised in making between-year comparisons in Cell 1 fish, because, despite a good-faith effort, electroshocking at site X failed to produce sunfish (or largemouth bass) in 2002. Consequently, fish were collected from another location near the inflows rather than the outflows. Hence, spatial variability may have obscured any temporal differences. Moreover, it is likely that these sunfish (and bass) collected immediately downstream of the inflow culverts had only recently dispersed into the re-flooded cell. Whatever the reasons, tissue Hg levels in sunfish from Cell 1 were only slightly elevated when compared to fish at other STAs (Rumbold, 2004a) or fish from downstream sites (Rumbold, 2004b). Between-year differences in tissue Hg levels were significant for fish from the supply canal (Mann-Whitney Rank sum test, $p = 0.03$), Cell 3 ($p = 0.001$), Cell 2 ($p < 0.001$), but not in fish from the discharge canal ($p = 0.12$). Sunfish at the downstream site, Z, were also much larger in 2002; average length was 97 ± 7 mm in 2001 ($n = 20$) and 155 ± 15 mm ($n = 13$) in 2002. Unlike fish from other sites, Hg levels were greater in sunfish at site Z in 2002. Because 2002 sunfish exhibited a significant size-concentration relationship (linear regression; $df = 1$,

11; $F = 6.03$; $p = 0.03$) similar to 2001 fish, ANCOVA was used and demonstrated significant between-year differences in Hg levels ($df = 1, 30$; $F = 4.9$; $p = 0.03$). However, these results must be viewed cautiously because the species of collected sunfish differed in 2001 (*Lepomis macrochirus* and *L. microlophus*) and 2002 (*L. macrochirus*, *L. gulosus*, *L. punctatus*) and species is known to be a significant factor in tissue Hg concentration (Rumbold, 2004b). When only the species with the largest sample size (*L. macrochirus*) was assessed, the data sets did not met the requirements for ANCOVA and, consequently, size as a co-variate could not be partitioned.

Largemouth bass collected in October 2002 also exhibited significant spatial patterns in tissue Hg (Kruskal-Wallis ANOVA on Ranks; $H = 46.6$, $df = 4$, $p < 0.001$) that were very similar to those observed in 2001 (Figure 6). Because sampled populations had a wider age distribution (except for site Z, where only two bass were collected) regressions were significant and EHg3 values were calculated as follows: 509 ± 184 ng/g (i.e., ± 95 CI) in the supply canal, 661 ± 196 ng/g in Cell 1, 384 ± 106 ng/g in Cell 2, 248 ± 117 ng/g in Cell 3 and, 1169 ± 233 ng/g in the discharge canal. However, caution must again be exercised when interpreting these values because datasets for tissue Hg in fish from supply canal, and Cells 2 and 3 were not normally distributed and, consequently, EHg3 values are considered tentative. Between-year differences in tissue Hg in bass from the discharge canal (met requirements for ANCOVA), were found not to be significant ($df = 1, 37$; $F = 0.01$; $p = 0.94$). Similar to sunfish, bass collected near the Cell 1 inflows had tissue Hg levels that were only slightly elevated when compared to fish in the supply canal, fish at other STAs (Rumbold, 2004a) or fish from many downstream sites (Rumbold, 2004b).

In the weeks following the August 22 spike, water-column concentrations of THg and, more importantly, MeHg declined at all sites within Cell 1, with concomitant decreases in the discharge canal (Figure 3). Likewise, tissue Hg levels began to decline in mosquitofish from both Cell 1 and the discharge canal (Figure 4). Cell 1 formally satisfied its start-up criteria on November 26, 2002, when water-column concentrations at the midpoint of the cell, site A (0.81 ng THg/L and 0.088 ng MeHg/L), were not significantly greater than average concentrations in the supply canal ($1.32 + 0.14$ ng THg/L, $n = 55$ and $0.16 + 0.04$ ng MeHg/L, $n = 52$ unqualified values).

4. Conclusions

The start-up monitoring program was successful in that it forewarned a potentially serious MeHg anomaly within STA-2 Cell 1. Subsequent expanded monitoring helped to guide appropriate adaptive management decision-making regarding short-term corrective actions and longer-term operational optimization. The temporary drying of Cell 1 succeeded in preventing tissue Hg levels in sunfish and bass from

peaking as high as might have been expected given the concentrations observed in lower trophic level mosquitofish. Adaptive management of flow and water depth in Cell 1, as a means to alter sediment redox chemistry and sulfur biogeochemistry, appears to have succeeded in reducing MeHg production. However, the precise biogeochemistry has yet to be confirmed. Although successful under this set of unique conditions, this adaptive management strategy would not be commonly available. Moreover, the outcome, in terms of sediment sulfide levels, may not be desirable under a different set of conditions. In view of that, efforts will continue to synthesize information gained from this monitoring program along with results from on-going research and modeling within STA-2 to identify other operational alternatives to prevent or minimize the occurrence of similar MeHg anomalies in wetlands or impoundments constructed in south Florida in the future.

Acknowledgments

This work could not have been accomplished without the efforts of the STA sampling team of Christy Bauer, Angela Drummond, James Galloway, Joseph Jean-Jacques, Mark Kromer, Kevin Nicholas, Sharon Niemczyk, Nicole Niemeyer, Karl Strayer, Brent Warner, along with various contractors. We would also like to acknowledge the exceptional work of all the chemists in FDEP's laboratory, but especially Thekkekalathil Chandrasekhar and Kerry Tate, under the able direction of the Laboratory Director, Timothy Fitzpatrick. We need to express our appreciation to Thomas Atkeson and Don Axelrad at FDEP, who ensured effective coordination between agencies as the study design evolved in response to new challenges and new information and who supported our subsequent request for CWA Section 319 Grant funding to offset some of the costs of the expanded monitoring. Finally, we thank two anonymous reviewers for their constructive and useful comments.

References

Abernathy, A. R. and Cumbie, P. M.: 1977, 'Mercury accumulation by largemouth bass (*Micropterus salmoides*) in recently impounded reservoirs', *Bull. Environ. Contam. Toxicol.* **17**, 595–602.

Bodaly, R. A. and Fudge, R. J. P.: 1999, 'Uptake of mercury by fish in an experimental boreal reservoir', *Arch. Environ. Contam. Toxicol.* **37**, 103–109.

Bodaly, R. A., Hecky, R. E. and Fudge, R. J. P.: 1984, 'Increases in fish mercury levels in lakes flooded by the Churchill River diversion, northern Manitoba', *Can. J. Fish. Aquat. Sci.* **41**, 682–691.

Benoit, J. M., Gilmour, C. C., Mason, R. P. and Heyes, A.: 1999, 'Sulfide controls on mercury speciation and bioavailability to methylating bacteria in sediment pore waters', *Env. Sci. Technol.* **33**, 951–957.

Brigham, M. E., Krabbenhoft, D. P., Olson, M. L. and Dewild, J. F.: 2002. 'Methylmercury in flood-control impoundments and natural waters of Northwestern Minnesota, 1997–1999', *Water Air Soil Pollut.* **138**, 61–78.

Cox, J. A., Carnahan, J., Dinunzio, J., McCoy, J. and Meister. J.: 1979, 'Source of mercury in new impoundments', *Bull. Environ. Contam. Toxicol.* **23**, 779.

Fink, L. E.: 2000, ENR Project Mercury Studies: 1994–1999. Appendix 7-5 in *2000 Everglades Consolidated Report*. South Florida Water Management District. West Palm Beach, FL. Available: http://www.sfwmd.gov/org/ema/everglades/index.html. (26 March 2004)

Fink, L. E.: 2002, The Effect of Dryout and Rewetting on Mercury Bioaccumulation. Appendix 2B-3 in *2002 Everglades Consolidated Report*. South Florida Water Management District. West Palm Beach, FL. Available: http://www.sfwmd.gov/org/ema/everglades/index.html. (26 March 2004)

Fink, L. E., King, J., Adak, P. and Matson, F.: 2005, STA-2 Mercury Special Studies Project Report. Appendix 2B-2 in *2005 South Florida Environmental Report – Volume 1*. South Florida Water Management District. West Palm Beach, FL.

Gilmour, C. C., Krabbenhoft, D. P. and Orem, W. O.: 2004, Mesocosm Studies to Quantify How Methymercury in the Everglades Responds to Changes in Mercury, Sulfur and Nutrient Loading. Appendix 2B-3 in *2004 Everglades Consolidated Report.* South Florida Water Management District, West Palm Beach, FL. Available: http://www.sfwmd.gov/org/ema/everglades/ index.html. (26 March 2004)

Hurley, J. P., Krabbenhoft, D. P., Cleckner, L. B., Olson, M. L., Aiken, G. R. and Rawlik, P. S., Jr.: 1998, 'System controls on the aqueous distribution of mercury in the northern Florida Everglades', *Biogeochemistry* **40**, 293–311.

Kelly, C. A., Rudd, J. W. M., Bodaly, R. A., Roulet, N. P., St. Louis, V. L., Heyes, A., Moore, T. R., Schiff, S., Aravena, A., Scott, K. J., Dyck, B., Harris, R., Warner, B. and Edwards. G.: 1997, 'Increases in fluxes of greenhouse gases and methylmercury following flooding of experimental reservoir', *Environ. Sci. Tech.* **31**, 1334–1344.

Krabbenhoft, D. P. and Fink, L. E.: 2001, The Effect of Dry Down and Natural Fires on Mercury Methylation in the Florida Everglades. Appendix 7–8 in *2001 Everglades Consolidated Report.* South Florida Water Management District, West Palm Beach, FL. Available: http://www.sfwmd.gov/org/ema/everglades/index.html. (26 March 2004)

Krabbenhoft, D. P., Hurley, J. P., Olson, M. L. and Cleckner, L. B.: 1998, 'Diel variability of mercury phase and species distributions in the Florida Everglades', *Biogeochemistry* **40**, 311–325.

Mercury Technical Committee: 1991, Interim report to the Florida Governor's Mercury in Fish and Wildlife Task Force and Florida Department of Environmental Regulation. Center for Biomedical and Toxicological Research, Florida State University, Tallahassee, 60 pp.

Miles, C. J. and Fink, L. E.: 1998, 'Monitoring and mass budget for mercury in the Everglades Nutrient Removal Project', *Arch. Environ. Contam. Toxicol.* **35**, 549–57.

Niu, X. and Tintle, A.: 2003, Statistical Analysis and Summary of the HgRR3 Mercury Round Robin Data. *Technical Report Prepared for the Florida Department of Environmental Protection*, Tallahassee, FL. Available online at: http://www.dep.state.fl.us/labs/everglades/index.htm (30 September 2004).

Paterson, M. J., Rudd, J. W. M. and St. Louis, V. L.: 1998, 'Increases in total and methylmercury in zooplankton following flooding of a peatland reservoir', *Environ. Sci. Technol.* **32**, 3868–3874.

Rudd, J. W. M.: 1995, 'Sources of methylmercury to freshwater aquatic ecosystems: A review', *Water Air Soil Pollut*. **80**, 697–713.

Rumbold, D. G.: 2004a, Annual Permit Compliance Monitoring Report for Mercury in Stormwater Treatment Areas. Appendix 4A-4 in *2004 Everglades Consolidated Report*. South Florida Water Management District. West Palm Beach, FL. Available: http://www.sfwmd.gov/org/ema/everglades/index.html. (26 March 2004)

Rumbold, D. G.: 2004b, Annual Permit Compliance Monitoring Report for Mercury in Downstream Receiving Waters of the Everglades Protection Area. Appendix 2B-5 in *2004 Everglades Consolidated Report*. South Florida Water Management District. West Palm Beach, FL. Available: http://www.sfwmd.gov/org/ema/everglades/index.html. (26 March 2004)

Rumbold, D. G.: *in press*, A probabilistic risk assessment of the effects of methylmercury on Great Egrets and Bald Eagles foraging at a constructed wetland in South Florida. *Human and Ecological Risk Assessment.* April 2005.

Rumbold, D. G. and Fink, L. E.: 2002, Report on Expanded Mercury Monitoring at Stormwater Treatment Area-2. Appendix 4A.6 in *2002 Everglades Consolidated Report*. South Florida Water Management District. West Palm Beach, FL. Available: http://www.sfwmd.gov/org/ema/everglades/index.html. (26 March 2004)

Rumbold, D. G. and Fink, L. E.: 2003a, Annual Permit Compliance Monitoring Report for Mercury in Downstream Receiving Waters of the Everglades Protection Area. Appendix 2B-3 in *2003 Everglades Consolidated Report*, South Florida Water Management District. West Palm Beach, FL. Available: http://www.sfwmd.gov/org/ema/everglades/index.html. (26 March 2004)

Rumbold, D. G. and Fink, L. E.: 2003b, Annual Permit Compliance Monitoring Report for Mercury in Stormwater Treatment Areas. Appendix 4A-4 in *2003 Everglades Consolidated Report*. South Florida Water Management District. West Palm Beach, FL. Available: http://www.sfwmd.gov/org/ema/everglades/index.html. (26 March 2004)

St. Louis, V. L., Rudd, J. W. M., Kelly, C. A., Beaty, K. G., Bloom, N. S. and Flett, R. J.: 1994, 'Importance of wetlands as sources of methylmercury to boreal forest ecosystems', *Can. J. Fish. Aquat. Sci.* **51**, 1065–1076.

Southworth, G. R., Peterson, M. J. and Bogle, M. A.: 2002, 'Effect of point source removal on mercury bioaccumulation in an industrial pond', *Chemosphere* **49**, 455–460.

Verdon, R., Brouard, D., Demers, C., Lalumiere, R., Laperle, M. and Schetagne, R.: 1991, 'Mercury evolution (1978–1988) in fishes of the La Grande Hydroelectric Complex, Quebec, Canada', *Water Air Soil Pollut.* **56**, 405–417.

Watras, C.: 1994, Letter Report Summarizing Mercury Field Training Exercise and Recommendations for Sampling Modification. Correspondence to Larry Fink, South Florida Water Management District. 19 February 1994.

Ware, F. J., Royals, H. and Lange, T.: 1990, 'Mercury contamination in Florida largemouth bass', *Proc. Annual Conference Southeast Assoc. Fish Wildlife Agencies* **44**, 5–12.

Zar, J. H.: 1996, *Biostatistical Analysis* 3rd ed., Prentice-Hall, Upper Saddle River, New Jersey.