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Spatial and temporal variations of mercury levels in Okefenokee invertebrates: Southeast Georgia

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This study measured mercury levels in invertebrates and found the highest levels in amphipods.

Abstract

Accumulation of mercury in wetland ecosystems has raised concerns about impacts on wetland food webs. This study measured concentrations of mercury in invertebrates of the Okefenokee Swamp in Georgia, focusing on levels in amphipods, odonates, and crayfish. We collected and analyzed total mercury levels in these invertebrates from 32 sampling stations across commonly occurring sub-habitats. Sampling was conducted in December, May, and August over a two-year period. The highest levels of mercury were detected in amphipods, with total mercury levels often in excess of 20 ppm. Bioaccumulation pathways of mercury in invertebrates of the Okefenokee are probably complex; despite being larger and higher in the food chain, levels in odonates and crayfish were much lower than in amphipods. Mercury levels in invertebrates varied temporally with the highest levels detected in May. There was a lack of spatial variation in mercury levels which is consistent with aerial deposition of mercury.
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Keywords: Mercury; Invertebrates; Bioaccumulation; Okefenokee Swamp

1. Introduction

Elemental mercury and methyl mercury are of particular concern for environmental safety and they often accumulate in wetland ecosystems (Moore et al., 1995; Rood, 1996; St. Louis et al., 1996; Heyes et al., 1998; Naimo et al., 2000). Elemental mercury poses little risk. However, the methylated form of mercury is a potent neurotoxin and poses serious problems to animals in many ecosystems (Morel et al., 1998). In wetlands, sulfate-reducing bacteria are key in mercury methylation and as a result methyl mercury is frequently produced at high rates (Morel et al., 1998; St. Louis et al., 1994).

The bioavailability of mercury in wetland aquatic environments appears to be dependent on water temperature, dissolved oxygen levels, and hydrology. High temperatures reduce dissolved oxygen levels, enhancing methylation because mercury bound to sediment is released into the water column when oxygen levels are low (Henry et al., 1995). Intermittent or shallow flooding can increase methyl mercury bioavailability to organisms since these fluctuations cause release of methyl mercury from sediment (Morel et al., 1998). For example, mercury levels were higher in fish collected from South Carolina wetlands that experienced frequent fluctuations in water levels than those that were deep and permanent (Snodgrass et al., 2000).

Although mercury is found in wetlands, the source of contamination is not easily determined. Both anthropogenic and natural processes are possible sources of bioavailable mercury. Some of the greatest contaminant loads of mercury detected from wetlands have been in the Florida Everglades, where peat and natural mineral deposits are possible sources of

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mercury contamination (Facemire et al., 1995; Rood, 1996; Vaithiyanathan et al., 1996). Possible external anthropogenic inputs of mercury into wetlands include burning of fossil fuels, medical waste incineration, agriculture, and mining (Rood et al., 1995). There is increasing evidence that anthropogenic emissions significantly increase mercury levels in precipitation (Rolfhus and Fitzgerald, 1995; Keeler et al., 1995). In precipitation, however, methyl mercury constitutes a small part of total mercury, typically <1% of total concentrations (Bloom and Watras, 1989; Downs et al., 1998). The mercury in precipitation becomes a problem if it accumulates where conditions suitable for methylation occur. Runoff, which can be the result of increased precipitation, also increases mercury in a wetland and a positive correlation has been found between sediment mercury concentrations and watershed area (Wiener et al., 1990).

Sediment is a major source of bioavailable methyl mercury (Lasorsa and Allen-Gil, 1995; Tremblay et al., 1996). Sediment mercury concentration and the uptake of mercury by organisms from sediment both vary with temperature, dissolved oxygen, depth, and pH (Rood, 1996; Downs et al., 1998). In lakes, for example, a decrease in pH enhances release of mercury from the sediment (French et al., 1999) making it readily available to invertebrates and fish. In addition, methyl mercury release is inversely proportional to dissolved oxygen (Henry et al., 1995), resulting in increased levels of bioavailable mercury.

The diets of wetland organisms may have more influence on bioaccumulation of mercury than direct exposure to mercury in the water column (Bhattacharya and Sarkar, 1996; Downs et al., 1998). Top aquatic predators depend either directly or indirectly on plants and invertebrates. Several wetland plants sequester mercury and various levels of accumulation have been recorded from plant tissues. Submersed species of aquatic plants can sequester high levels of mercury (Thompson-Roberts et al., 1999), and the bryophytes feather and *Sphagnum* moss sequester the highest levels of mercury of any plants recorded. On the other hand, many common wetland species such as the yellow water lily sequester minimal amounts of mercury in their tissues (Thompson-Roberts et al., 1999), and the leaves of most trees and shrubs have very low levels of mercury (Moore et al., 1995). Another factor for bioaccumulation from plant material is the presence of decaying aquatic vegetation. As plant tissues rich in mercury decompose under anoxic conditions, the methylation of inorganic mercury results in the release of methyl mercury (Heyes et al., 1998).

Mollusks and crayfish are frequently used as bioindicators of heavy metal contamination (Eisemann et al., 1997). However, relatively little research has been conducted on bioaccumulation of mercury in other invertebrates (Odin et al., 1995; Visman et al., 1995; Hall et al., 1998). Under laboratory conditions, amphipods efficiently bioaccumulated mercury from both algae and sediment (Lawrence and Mason, 2001). Bioaccumulation of mercury by invertebrates allows mercury to become available to organisms higher in the food chain such as fish and birds.

Wetland fish can bioaccumulate particularly high levels of methyl mercury (Bloom, 1992; Mason et al., 1994; Kannan et al., 1998; Wong et al., 1997; Hall et al., 1998). As a result, piscivorous birds are exposed to bioavailable mercury (Gariboldi et al., 1997). Bioaccumulation of mercury in large predators is of considerable concern. Much of the concern about mercury in the Florida Everglades was attributable to high levels of mercury detected in the liver of an endangered Florida panther (Roelke et al., 1991). Since alligators are long-lived wetland predators, there are also concerns about the potential to bioaccumulate mercury in their tissues (Khan and Tansel, 2000).

The Okefenokee Swamp is one of the largest freshwater wetlands in North America. It is approximately 3800 km^2 and provides habitat for a variety of organisms (Porter et al., 1999). The Okefenokee Swamp has many characteristics that could lead to mercury accumulation and bioavailability problems, including high water temperature, frequent anoxic conditions, low pH (<4.0), intermittent hydrology, peat deposits, and abundant *Sphagnum* mosses. The Georgia Department of Natural Resources has placed restrictions on the consumption of two species of fish from the Okefenokee (bowfin, *Amia calva* and flier, *Centrarchus* sp.) due to high levels of mercury. Invertebrates are directly or indirectly important in the diets of both fish, so questions have developed about the role invertebrates might play in the biomagnification of mercury through the food web of the Okefenokee Swamp. The objective of this project was to describe spatial and temporal variation of mercury levels in Okefenokee invertebrates.

2. Materials and methods

Thirty-two sites were chosen in the Okefenokee that were distributed across the range of hydrological units and vegetative communities present in the swamp. They included sites centered around Grand Prairie, Double Lakes, Durden Lake, Chase Prairie, Floyd's Prairie, and Billy's Lake (Fig. 1). At each site, sampling was stratified to include shrub, prairie (lily and/or sedge marsh), and cypress habitats. In addition, samples were collected in managed boat trails and any deepwater habitats that were present such as lakes, rivers, or canals. Sampling was conducted in December 1998, May 1999, August 1999, December 1999, May 2000, and August 2000. At each sampling location we collected amphipods (Crangonyctidae) as available for 30 min using sweep nets. Beginning in May 1999, we also collected Odonata nymphs (primarily Anisoptera), and beginning in December 1999, we added crayfish (Cambaridae) to the collection. Invertebrates were placed in plastic vials and transported on ice back to the laboratory, and then frozen. Since the analyses of the sediments indicated that mercury levels were low (<0.002 ppm) and these organisms do not ingest sediment we did not wait for the organisms to clear their guts.

All amphipods, odonates, or crayfish collected for individual samples were combined into pools and sent to Clemson Institute of Environmental Toxicology at Clemson University, SC, for analysis. Total mercury levels were determined using Atomic Absorption Spectrophotometery (AAS) as described by Waldrop (1999). Validation trials were conducted in conjunction with each sampling run and lower detection limits were 0.25 ppb.

Variations in mercury levels among locations, sub-habitats, sample dates, and organisms were assessed concurrently using 4-way ANOVA. However, because each location–date–sub-habitat–organism combination was not replicated, we could not test for statistical interactions among factors in that analysis. Thus, a series of separate 3-way ANOVA's were used to address variation within each of the six locations, within each of the six dates, within each of the five sub-habitats, and for each of the three organisms. When ANOVA's were significant, post hoc Tukey's tests were used to separate means. When variances were not equal, mercury levels were $\log(x+1)$ transformed prior to analysis. All statistical analyses were conducted using SAS version 8.

To determine if hydrological patterns influenced mercury levels, each of the thirty-two sites was ranked according to the number of times the site

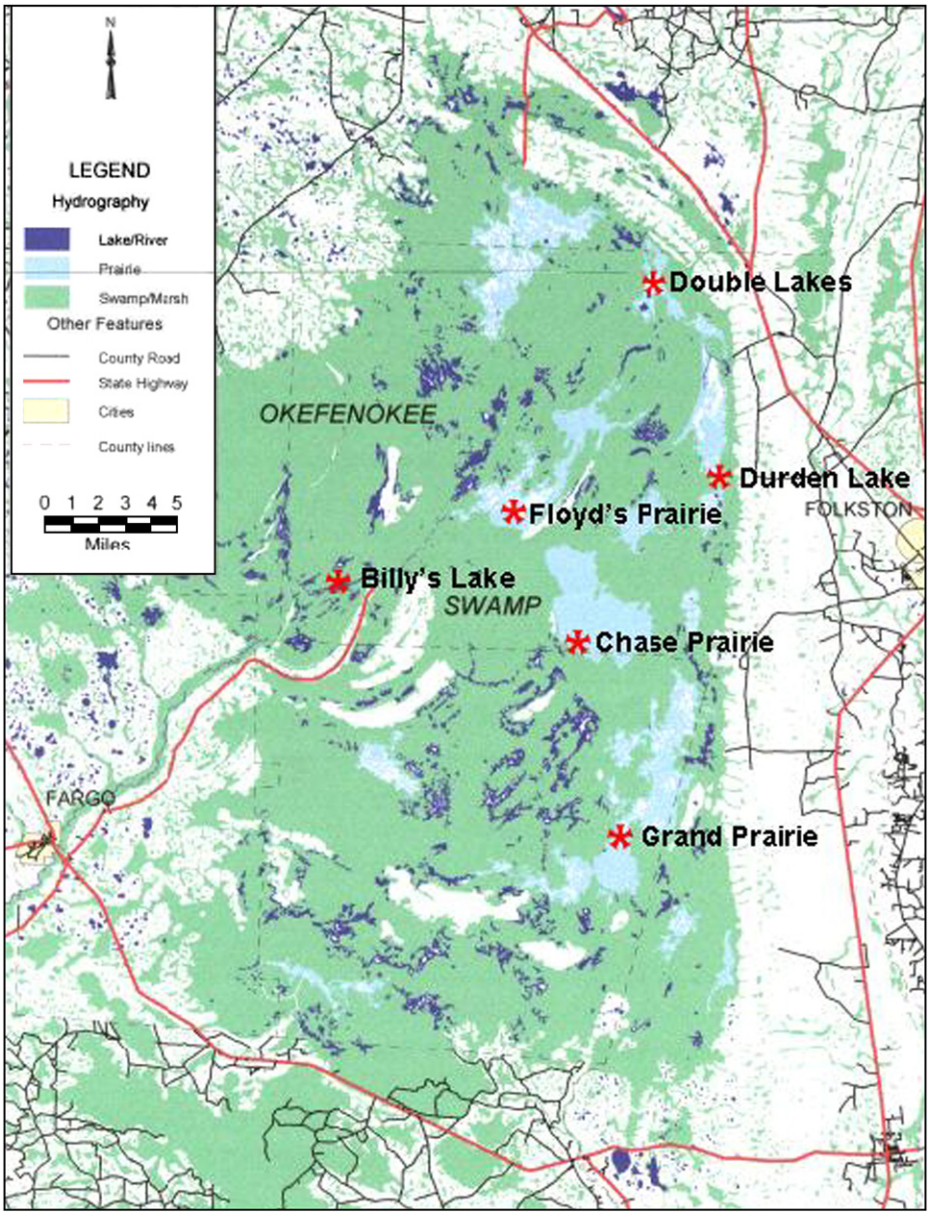

Fig. 1. Map of the Okefenokee Swamp showing the six locations (asterisks) where invertebrates were collected.

was flooded. For example, if a site was flooded once out of six sampling dates it received a rank of 1, if a site was flooded twice it received a rank of 2, and so on (with the highest rank being 6). The sites were then grouped into three categories. Sites that were classified 1, 2, or 3 were grouped together as ephemerally flooded, sites that were classified as 4 or 5 were grouped together as intermittently flooded, and sites that were classified as 6 were grouped as permanently flooded. Using amphipod data (the only organisms collected on all 6 dates), tissue mercury levels from sites that were flooded ephemerally, intermittently, or permanently were compared using 1-way ANOVA.

3. Results

Mercury levels varied dramatically among sample pools (averaged 1.6 ppm, but ranged from 0 to 86 ppm), and a 4-way ANOVA model concurrently addressing location, sub-habitat, sample date, and study organism accounted for 65.1% of this variation (Table 1). However, the kind of organism ($F_{2,184} = 93.07$, $P < 0.0001$) and the sample date ($F_{5,184} = 16.16$, $P < 0.0001$) were the only significant factors in the model, with the study organism and sample date accounting for 43.3% and 18.8% of variation, respectively.

Table 1
ANOVA table of mercury concentrations among locations, habitats, dates, and organisms

Source	DF	Type III SS	Mean square	F value	$P > F$
Location	5	2.4819	0.4963	1.67	0.1448
Sub-habitat	4	1.3239	0.3309	1.11	0.3527
Date	5	24.0676	4.8135	16.16	<0.0001
Organism	2	55.4481	27.7240	93.07	<0.0001
Error	184	54.8100			
Total	200	128.0000			

3.1. Variation among amphipods, odonates, and crayfish

Mercury concentrations were dramatically higher in amphipods than either odonates or crayfish (Fig. 2). When assessing only amphipods, as in the overall model, sample date was important ($F_{5,65} = 18.95$, $P < 0.0001$) and mercury concentrations peaked during the May 2000. In contrast, when assessing levels in odonates, levels of mercury peaked during December 1999, and for crayfish, mercury levels did not vary temporally. For both amphipods and crayfish, concentrations of mercury were similar among all six locations and all five sub-habitats (all $P > 0.05$). However, for odonates, mercury levels were marginally higher at Floyd's Prairie ($F_{5,78} = 1.67$, $P = 0.0311$), but did not differ among sub-habitats (all $P > 0.05$).

3.2. Variation among dates (12/1998, 5/1999, 8/1999, 12/1999, 5/2000, and 8/2000)

Overall, the highest levels of mercury occurred in the May 2000 (Fig. 3), which was largely driven by levels in amphipods (see above). When assessing patterns within individual sample dates, levels of mercury were higher in amphipods than odonates or crayfish for every sampling date except August 2000, when levels of mercury did not differ among organisms ($P = 0.7332$). Mercury levels in invertebrates were similar among all five sub-habitats for every date except May 2000, when significantly higher levels of mercury were present in samples from the boat trails ($F_{4,33} = 3.31$, $P = 0.0219$). For every sampling date, mercury levels in invertebrates were similar among all six locations.

3.3. Variation among individual locations (Grand Prairie, Chase Prairie, Durden Lake, Double Lakes, Floyd's Prairie, Billy's Lake) and sub-habitats (prairie, shrub, cypress, trails, and deepwater habitats)

Overall levels of mercury were similar among all six locations (Fig. 4; $F_{5,184} = 1.67, P = 0.1448$) and all five sub-habitats (Fig. 5; $F_{4,184} = 1.67, P = 0.3527$) (Table 1). Consistent with the overall analysis, mercury levels in amphipods were higher than in either odonates or crayfish at each of the six locations and at each of the five sub-habitats. Also, as evident overall, a peak in mercury concentration occurring during May 2000 was evident at the Grand Prairie, Chase Prairie, Durden Lake, and Billy's Lake locations (all $P < 0.05$). However, at the Double Lakes site, a peak was not evident in May 2000 and instead levels in December 1999 and August 1999 somewhat exceeded May 2000 levels. At the Floyd's Prairie location mercury levels were similar across sampling dates, although the site was dry in May 2000 and samples could not be collected. The peak in mercury levels during May 2000 occurred in cypress, shrub, and boat trail sub-habitats (all $P < 0.05$). At every sampling location, mercury concentrations in invertebrates were similar among all five sub-habitats, and in every sub-habitat, mercury levels were similar among all six locations (all $P > 0.05$).

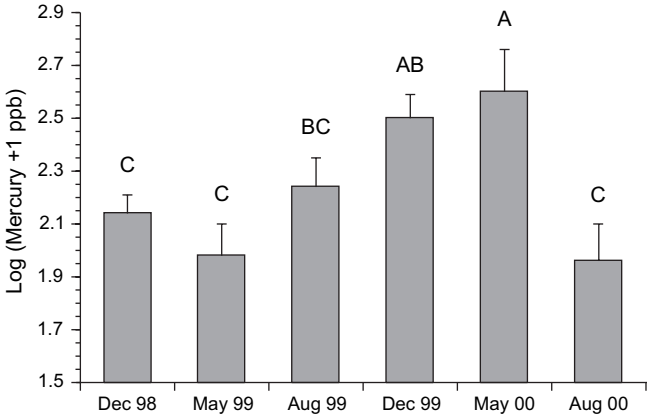

Fig. 3. Variation in total mercury concentrations in invertebrates among six sampling dates at the Okefenokee Swamp ($P < 0.001$). Bars indicated by the same letter are not significantly different ($P > 0.05$).

3.4. Mercury levels and hydroperiod

Over the two-year study period, the degree of habitat flooding varied greatly. December 1998 was the wettest month of

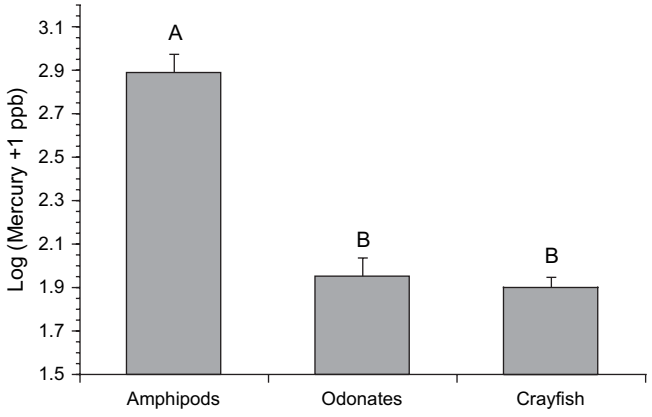

Fig. 2. Variation in total mercury concentrations among amphipods, odonates and crayfish collected from the Okefenokee Swamp ($P < 0.001$). Bars indicated by the same letter are not significantly different ($P > 0.05$).

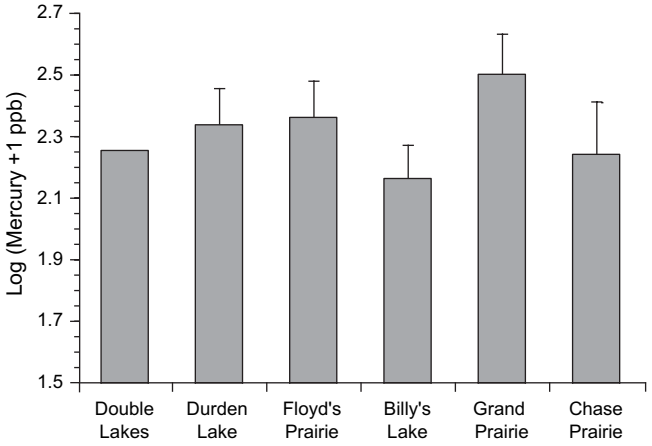

Fig. 4. Variation in total mercury concentrations in invertebrates among six locations at the Okefenokee Swamp ($P = 0.15$).

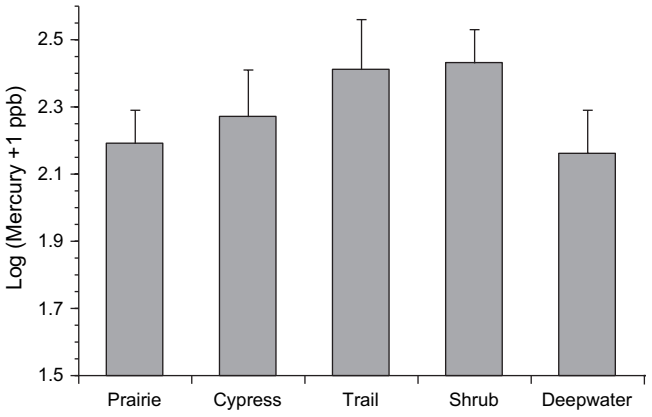

Fig. 5. Variation in total mercury concentrations in invertebrates among sub-habitats at the Okefenokee Swamp ($P = 0.35$).

the study when all 32 sample sites were flooded, while May 2000 was the driest month with only 17 of 32 sites being flooded (Fig. 6). Mercury levels (amphipods only) were highest in sites that were flooded for all six sampling dates (permanently flooded sites) ($P = 0.0117$; Fig. 6) as compared to sites that were flooded for <6 sampling dates (either intermittently or ephemerally flooded sites). We were concerned that this pattern developed because permanent water sites were some of the only locations flooded during the drought of May 2000, when overall levels of mercury peaked (Fig. 3). Therefore a second analysis was conducted using just the December 1998 sampling date when all sites were flooded. At that time mercury levels in amphipods were similar among ephemerally, intermittently, and permanently flooded sites ($P = 0.2928$).

4. Discussion

Levels of mercury detected in Okefenokee invertebrates seemed unusually high, even for wetlands. We frequently encountered mercury levels in excess of 20 ppm, and levels averaged 1.6 ppm. In comparison, mercury levels in invertebrates of the Florida Everglades averaged 0.3 ppm (Scheidt, 2000), and levels averaged 0.1 ppm in small depressional wetlands in South Carolina (Snodgrass et al., 2000). However, the higher than normal mercury levels detected in this study may not necessarily indicate a uniquely severe problem for the Okefenokee Swamp. The high levels occurred almost exclusively in amphipods, and these organisms are often not collected in other studies of mercury in wetland invertebrates.

Our analyses of how mercury in Okefenokee Swamp invertebrates was influenced by spatial (location, sub-habitat), temporal (season, annual), and taxonomic (organism) considerations suggested that, while no "hot spots" for mercury were detected, there were "hot times" and "hot organisms". The lack of spatial variation in mercury across the Okefenokee was consistent with aerial deposition of the material relatively even across the wetland (Fitzgerald et al., 1998).

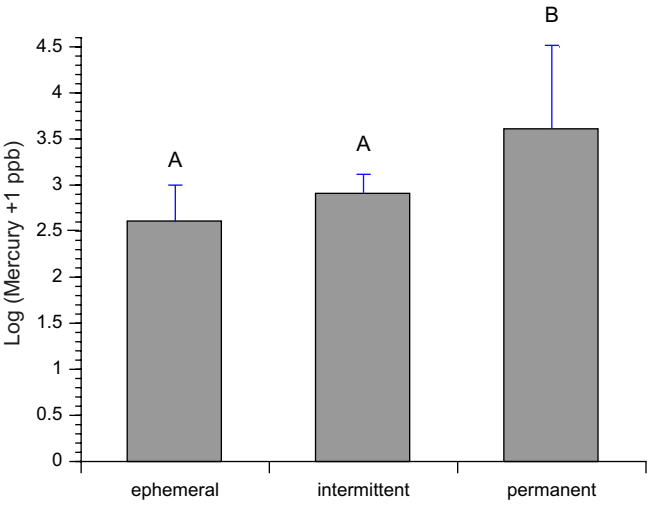

Fig. 6. Average mercury concentrations over two years in amphipods from ephemerally, intermittently, and permanently flooded sites of the Okefenokee Swamp ($P = 0.0117$). Bars indicated by the same letter are not significantly different ($P > 0.05$).

4.1. Variation in mercury levels among organisms

The most important source of variation of mercury in Okefenokee invertebrates was the *kind* of organism involved. Because organisms vary in terms of feeding habits, physiology, and habitat preferences, it was not unexpected to find that mercury levels varied among organisms. However, the finding that mercury levels in amphipods were much higher than in odonates or crayfish was surprising. Biomagnification of mercury in invertebrates seems most likely in predators such as odonates or large, long-lived organisms such as crayfish, rather than small, algivorous or detritivorous organisms like amphipods (Pennak, 1989). The concentrations of mercury in predatory insects have been reported to increase by a factor of 2–5 over that found in prey (Mason et al., 2000). Studies from lakes (Wong et al., 1997) indicate that levels of mercury in odonates (0.12 ppm) and amphipods (0.13 ppm) can be very similar. In the Okefenokee, average mercury levels in odonates were similar to lakes (0.18 ppm), but levels in amphipods (4.0 ppm) were much higher. Biomagnification of mercury by Okefenokee odonates may be similar to other habitats, but the relationship between mercury and amphipods is apparently unique.

Crayfish consume many of the same foods as amphipods, have similar habitats, and are larger and longer-lived than amphipods, yet mercury levels in these organisms (0.23 ppm) were lower than in amphipods. Studies elsewhere have shown that mercury levels in crayfish are usually lower than other co-existing predatory insects but higher than non-predatory insect groups (Mason et al., 2000). Mercury levels in crayfish in the Okefenokee were similar to levels in the Everglades (Scheidt, 2000). However, why levels in amphipods in the Okefenokee were greater than levels in crayfish remains unclear.

Possible reasons for high mercury levels in amphipods include the close association of amphipods with sediment or with mercury sequestering plants. Mercury is often bound to sediments and is released via methylation (Raldua and

Pedrocchi, 1996) or by drying and reflooding of habitats (Snodgrass et al., 2000; Warren et al., 2001). Sediment seems a likely source for mercury transfer to aquatic invertebrates, and as detritivores, amphipods probably frequent the sediment boundary to feed (Pennak, 1989). Studies have indicated that amphipods exposed to sediments spiked with methyl mercury showed an increased uptake of mercury as compared to those exposed to spiked pore water (Lawrence and Mason, 2001).

Biomagnification of mercury from plants or periphyton might also explain higher levels of mercury in amphipods. Mercury concentrations in periphyton and plant material are 3–10 times lower than those in invertebrates that feed on this material (Mason et al., 2000). Some of the highest levels of mercury have been documented from *Sphagnum* mosses, with low levels occurring in water lily (Cymerman and Kempers, 1995; Moore et al., 1995; Thompson-Roberts et al., 1999). We collected and analyzed plant material from the Durden Lake study area; mercury levels generally were low, but the highest levels (0.43 ppm) were collected from *Sphagnum* mosses (unpublished data). In the Okefenokee, we observed that amphipods were abundant in and around *Sphagnum* mosses, and this relationship might explain the higher levels of mercury in the amphipods.

Amphipods might be especially useful for detecting high levels of mercury because, as Sferra et al. (1999) reported, mercury toxicity in amphipods can exceed 4.1 ppm. Many organisms might die before accumulating such high levels. It follows that since amphipods are often a major source of food for fish they might be contributing to the high levels of mercury in Okefenokee Swamp fish.

4.2. Temporal variation in mercury levels

The second most important source of mercury variation in Okefenokee invertebrates was *when* the sample was collected. Levels, at least in amphipods, peaked in May 2000. However, that peak did not reflect a seasonal pattern because levels in the previous May (1999) were low (Fig. 3). Drought conditions might explain the high levels in May 2000. At that time, precipitation levels were very low and only 17 of the 32 sample sites were flooded. Drought conditions might increase bioavailable mercury because the increase in temperature can cause a decrease in dissolved oxygen, which in turn promotes the formation of methyl mercury (Morel et al., 1998). Both Snodgrass et al. (2000) and Hall et al. (1998) reported that levels of mercury in invertebrate increased after reflooding of a dried habitat. However, when many sites in the Okefenokee reflooded in August 2000, mercury levels in invertebrates declined rather than increased. In the Okefenokee, permanently flooded sites (lakes, canals, and boat trails) had somewhat higher mercury levels in amphipods than those habitats that were intermittently or ephemerally flooded (Fig. 6), which might suggest that mercury levels in Okefenokee invertebrates are less affected by reflooding cycles than elsewhere.

An unusually large fire occurred in the Okefenokee Swamp during the summer of 1999, and burning of plant material and peat can release mercury into the air (Lamontagne et al., 2000). Mercury levels in invertebrates increased gradually after that event, but levels of mercury did not peak until a year later in May 2000 (Fig. 3). In Canada, however, extensive wildfires did not increase levels of mercury in lake zooplankton or fish (Garcia and Carignan, 1999, 2000). Fire might have been involved in the peak for mercury in Okefenokee invertebrates of May 2000, but drought seems the more likely explanation.

4.3. Implications

Crayfish are the arthropods most commonly used as bioindicators of heavy metal contamination in wetlands; mollusks are often used in lakes and rivers, but they are rare in acidic wetlands. If we, however, had relied solely on crayfish to monitor mercury, we would have developed a very different picture of mercury distributions in the Okefenokee. An assortment of organisms, including amphipods, should probably be used for monitoring programs. Our study also suggests that it is important that monitoring programs address temporal variation. Again, if we had only sampled once, we would have developed a skewed perception of the mercury problem in the Okefenokee.

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