

This document was prepared in conjunction with work accomplished under Contract No. DE-AC09-96SR18500 with the U. S. Department of Energy.

DISCLAIMER

This report was prepared as an account of work sponsored by an agency of the United States Government. Neither the United States Government nor any agency thereof, nor any of their employees, makes any warranty, express or implied, or assumes any legal liability or responsibility for the accuracy, completeness, or usefulness of any information, apparatus, product or process disclosed, or represents that its use would not infringe privately owned rights. Reference herein to any specific commercial product, process or service by trade name, trademark, manufacturer, or otherwise does not necessarily constitute or imply its endorsement, recommendation, or favoring by the United States Government or any agency thereof. The views and opinions of authors expressed herein do not necessarily state or reflect those of the United States Government or any agency thereof.

This report has been reproduced directly from the best available copy.

**Available for sale to the public, in paper, from: U.S. Department of Commerce, National Technical Information Service, 5285 Port Royal Road, Springfield, VA 22161,
phone: (800) 553-6847,
fax: (703) 605-6900
email: orders@ntis.fedworld.gov
online ordering: <http://www.ntis.gov/help/index.asp>**

**Available electronically at <http://www.osti.gov/bridge>
Available for a processing fee to U.S. Department of Energy and its contractors, in paper, from: U.S. Department of Energy, Office of Scientific and Technical Information, P.O. Box 62, Oak Ridge, TN 37831-0062,
phone: (865)576-8401,
fax: (865)576-5728
email: reports@adonis.osti.gov**

Influence of Methylmercury from Tributary Streams on Mercury Levels in
Savannah River Asiatic Clams

M.H. Paller^{ad}, C.H. Jagoe^b, H. Bennett^c, H.A. Brant^b, J.A. Bowers^a

^a Environmental Analysis Section, Westinghouse Savannah River Company, Savannah River Site, Aiken, SC 29808, USA

^b Savannah River Ecology Laboratory, University of Georgia, Drawer E, Aiken, SC 29802 USA

^c Department of Biology, University of South Carolina at Aiken, Aiken, SC 29803, USA

^d Corresponding author: M.H. Paller, 803-725-5250 (phone), 803-725-7673 (fax), michael.paller@srs.gov, Building 773-42A, Savannah River Site, Aiken, SC 29808

Abstract

Average methylmercury levels in five Savannah River tributary streams sampled 11 times over two years (0.170 ng/l) were nearly twice as high as in the Savannah River (0.085 ng/l). Total mercury levels in the tributaries (2.98 ng/l) did not differ significantly from the river (2.59 ng/l). All of the tributaries drained extensive wetlands that would be expected to support comparatively high rates of methylation. Mercury concentrations in Asiatic clams (*Corbicula fluminea*) collected from the discharge plumes of Savannah River tributaries (average of 0.044 ug/g wet weight) were significantly ($P < 0.001$) higher than in Asiatic clams collected from the Savannah River upstream from the tributary mouths (average of 0.017 ug/g wet weight). These results indicate that streams draining wetlands into coastal plain rivers can create localized areas of elevated methylmercury with resulting increases in the mercury levels of river biota.

Key words: methylmercury, mercury, *Corbicula*, streams, wetlands, Savannah River

1. Introduction

Mercury levels in many aquatic ecosystems in the northern hemisphere are strongly influenced by the aerial deposition of mercury in the surrounding watershed

(Mierle 1990, Fitzgerald et al. 1991). Although much of this mercury may be retained in terrestrial soils (Nater and Grigal 1992), some is conveyed to surface and subsurface waters where it is bacterially converted to methylmercury (Gilmour et al. 1992). Wetland ecosystems are particularly efficient at producing methylmercury because they provide conditions that support methylating bacteria (Francis et al. 1998). Studies in northern lakes and rivers show that methylmercury produced in wetlands can be transported by surface flow to other water bodies where it contributes to total methylmercury loading (St. Louis 1994, Hurley et al. 1995, Hurley et al. 1998).

Many of the major rivers draining the coastal plain of South Carolina have fish consumption advisories as a result of mercury contamination. This region has substantial atmospheric mercury deposition (EPA 1997) and many wetlands that are drained by small tributaries that connect to larger rivers. The possible significance of methylmercury conveyed to rivers by such tributaries has not been investigated. A simple way of evaluating the influence of tributary mercury on river biota is to compare mercury levels in biota residing in the plumes where tributaries discharge into rivers with biota from other river reaches less strongly influenced by tributary discharge. Most studies of mercury levels in biota involve fish because of their importance in the human food chain, but the mobility of fish make them a poor choice for this type of study. Bivalves, on the other hand, are relatively sedentary, integrate bioavailable contaminant concentrations over time, and reflect contaminant transfer from water into the food web (Cope et al. 1999, Chase et al. 2001). Studies on the Asiatic clam, Corbicula fluminea, which has become widely established in United States rivers, indicate that mercury uptake at the whole organism level is directly proportional to mercury concentrations in the water column or sediment making Corbicula potentially useful for investigating localized differences in the bioavailability of mercury (Inza et al. 1997, Inza et al. 1998).

In this study, we measured aqueous methylmercury levels at a variety of locations in the Savannah River and several Savannah River tributaries. We also measured mercury levels in Corbicula fluminea collected from the discharge plumes of Savannah River tributary streams and from river locations not directly affected by tributary discharge. Our objectives were:

- 1) to compare aqueous methylmercury levels between the Savannah River and its tributaries as a means of assessing the potential significance of the tributaries as sources of methylmercury, and
- 2) to determine whether tributary methylmercury resulted in localized increases in tissue mercury levels in Corbicula fluminea residing in the Savannah River.

2. Materials and Methods

2.1 Study area

The Savannah River is formed by the confluence of the Tugaloo and Seneca Rivers in northeast Georgia and flows southeast through the Piedmont and Coastal Plain to the Atlantic Ocean. This study was conducted in the middle reach of the Savannah River between river kilometer (RK) 252 and RK 72. Part of this reach is bordered by the Savannah River Site (SRS), an 800 km² Department of Energy facility formerly used for the production of nuclear materials (Figure 1). The SRS is drained by several Savannah River tributaries including Upper Three Runs, Beaver Dam Creek, Four Mile Branch, Steel Creek, and Lower Three Runs which join the Savannah River at approximately RKs 253, 245, 242, 228, and 208, respectively. Two other tributaries of substantial size, Brier Creek and Ebenezer Creek, are located downstream of the SRS at RKs 157 and 72, respectively. Several of the SRS tributaries have industrial areas in their watersheds. The other tributaries drain largely rural and undeveloped lands where agriculture and forestry are the major land uses.

The Savannah River was over 50 m wide and often over 2 m deep within the study area. The tributary streams at their confluence with the Savannah River ranged from about 10 to 30 m wide and from about 0.5 to 2.0 m deep. River and stream banks were generally forested. Previous studies indicate that the portion of the Savannah River under study was characterized by dissolved oxygen concentrations averaging about seven to eight mg/l, slightly acidic pH (5.8-6.7), and average conductivities near 70 $\mu\text{S}/\text{cm}$ (Paller et al. 1985 and 1986). The tributary streams were acidic (average pH from 5.6 to 6.6), with conductivities ranging from 20-70 $\mu\text{S}/\text{cm}$, and with more variable and somewhat lower dissolved oxygen concentrations than the river (occasionally declining below 3.0 mg/l, Paller et al. 1986).

Approximately 99% of the mercury loading to the Savannah River watershed is from atmospheric deposition or the erosion of stream bank soils as opposed to industrial discharges or other point sources (EPA 2000). However, Four Mile Branch may have received mercury contamination from industrial seepage basins located near its headwaters, and a small tributary of Upper Three Runs received mercury from a groundwater air stripping facility approximately 6.5 km from its confluence with Upper Three Runs (Halverson et al. 2002). However, as described later, total and methylmercury levels in these streams were not exceptional compared with the other tributaries.

Land use maps of the Savannah River Site indicate that the five SRS tributaries drain extensive wetlands (Figure 1). The lower portions of Four Mile Branch, Beaver Dam Creek, and Steel Creek drain portions of a 3,900 ha floodplain swamp contiguous with the Savannah River. Each of these streams also drains an additional 600 to 900 ha of wetlands located along its stream corridor. Upper Three Runs and Lower Three Runs drain more extensive wetland corridors, each of which approximates 2,600 ha. Although wetland maps were not available for Briar and Ebenezer Creeks, exploration of the

lower reaches of these streams indicated that they were connected to extensive floodplain swamps.

2.2 Field and laboratory methods

Water samples for mercury analysis were collected from the mouths of the five SRS tributaries and from five locations in the Savannah River near (within 100 m of) the tributary mouths on: 8/28/00, 10/19/00, 11/7/00, 11/29/00, 12/5/00, 12/18/00, 4/17/01, 8/15/01, 2/25/02, 5/22/02, and 9/3/02 (Figure 1). Although the choice of these dates was affected by logistics, they spanned a sufficient range to provide a reasonable estimate of aqueous mercury levels in each habitat. River samples reflected mercury levels in the river since they were taken near the middle of the river and not in the tributary discharge plumes. Single samples were taken from each of the 10 locations on most dates, but triplicate samples were taken from each location on 12/18/00 and 4/17/0 for quality control purposes. Water samples were taken from the mouths of Brier Creek and Ebenezer Creek and in the Savannah River just upstream of Brier Creek and Ebenezer Creek on 7/2/02.

All water samples were collected from approximately 25 cm beneath the surface in areas with perceptible flow. Samples for total mercury and methylmercury were taken by dipping and filling a pre-cleaned, marked teflon bottle. Samples for dissolved mercury and methylmercury (taken only on 11/7/00, 11/29/00, 12/5/00, and 4/16/01) were collected by using a peristaltic pump and clean plastic tubing to force water through a 0.45 micron glass-fiber filter into pre-cleaned and marked teflon bottles (filters and bottles supplied by Frontier Geosciences). New filters were used for each sample, and the collecting tube was thoroughly flushed with ambient water before collecting each sample. All sampling was performed with clean sample handling techniques to prevent contamination (Bloom 1995, USEPA 2001). Samples were placed on ice and shipped the day of collection to a laboratory that specialized in ultra-low level total mercury and

methylmercury analysis (Frontier Geosciences) using cold vapor atomic fluorescence spectroscopy (USEPA 2001). All samples were analyzed for total and methylmercury. The coefficient of variation (standard deviation / mean x 100) of triplicate samples averaged 8.3% for methylmercury and 8.5% for total mercury indicating high analytical reliability.

Corbicula fluminea (hereafter referred to as Corbicula) were collected on 6/4-5/2002 from 12 locations, six of which were in the discharge plumes of Upper Three Runs, Four Mile Branch, Steel Creek, Lower Three Runs, Briar Creek, and Ebenezer Creek; and six of which were in the Savannah River just upstream of each creek mouth (Figure 1). Creek and river sample sites near UpperThree Runs, Four Mile Branch, Steel Creek, and Lower Three Runs corresponded with the sites at which aqueous mercury data were collected over 11 dates as previously described. The Brier Creek and Ebenezer Creek sites were at locations where aqueous mercury data were collected once. Corbicula were taken from creek mouths at their confluence with the Savannah River or just downstream within the creek discharge plumes (based on visual observation). Corbicula from the Savannah River were collected at least 50 m upstream from creek mouths and near the opposite river bank. Ten Corbicula were gathered from each site by collecting sediment with a cylindrical dredge and picking Corbicula from the sediment. The Corbicula were put in labeled, plastic zip-lock bags with approximately 1-2 l of water and returned to the laboratory where they were held in clean dechlorinated tap water for 12 hours to depurate. They were subsequently weighed (wet), measured with dial calipers (shell length at longest anterior-posterior length, width at point of greatest shell inflation, and height at umbones) and sacrificed. All soft tissues were removed, weighed, and individually frozen.

For mercury analysis, individual Corbicula were again weighed, freeze dried to a constant weight to determine moisture content, then homogenized using a Teflon pestle

in a 30mL polyethylene vial. Subsamples from each homogenized individual were analyzed for total mercury by thermal decomposition, gold amalgamation, and atomic absorption detection (EPA method 7473; DMA-80 Analyzer, Milestone, Inc., Monroe, CT). For QA/QC purposes, each group of ten samples included a replicate, blank, and certified reference material of known Mercury content (lobster hepatopancreas, dogfish liver, or dogfish muscle purchased from the National Research Council of Canada, Ottawa ON). If Mercury was detected in the blank, or differences between replicates exceeded 10%, or the value for the reference material was outside the certified limits, the entire group was re-analyzed. The detection limit was 0.22 ng/g. All Corbicula mercury concentrations were expressed on a wet-weight basis.

2.3 Data analysis

The methylmercury data collected from the five SRS tributaries and nearby river sample sites were analyzed using a mixed analysis of variance (ANOVA) model (Sokal and Rohlf 1995) with habitat (river vs. creek mouth) and sample date as treatment factors and sample site nested within habitat as a random factor. Data from Brier and Ebenezer Creeks, which were sampled only once, were excluded from this analysis. Triplicate samples collected on a single date were averaged to produce a single value. Wald-Wolfowitz runs tests above and below the median indicated a lack of significant serial correlation among data collected from each site over time, thereby demonstrating that the assumption of independence was met (Sokal and Rohlf 1995). The methylmercury data did not initially meet other assumptions of ANOVA, including homogeneity of variances among samples and normality of sample distributions (Sokal and Rohlf 1995). Since commonly employed transformations (e.g. log and square root) failed to rectify this problem, the variance stabilizing transformation was calculated from the slope of the log mean - log variance regression according to Taylor (1961):

$$X' = X^{1-(b/2)};$$

where X' is the transformed datum, X is the untransformed datum, and b is the slope of the log mean – log variance regression ($b = 3.7$ for the methylmercury data). Results are reported as arithmetic means.

Because the total mercury data failed to meet the assumptions of normality and homogeneity of variance even after transformation of the data (including Taylor's (1961) transformation), they were analyzed with the Scheirer-Ray-Hare test, a non-parametric analogue of the two-way ANOVA in which the data are ranked before analysis and the results compared to the chi-square distribution (Sokal and Rohlf 1995). Treatment factors were habitat and sample date. Results are presented as arithmetic means.

Corbicula tissue mercury data were analyzed by analysis of covariance (ANCOVA) with location (river vs. creek mouth) the categorical factor and Corbicula shell length the covariate. Shell length was included because Corbicula size and tissue mercury levels were related. The model also included a third factor, sample site nested within habitat, to determine if there were significant differences between individual sample sites. A preliminary ANCOVA that included interaction terms demonstrated that the relationship between Corbicula size and tissue mercury levels was not affected by habitat nor site, thus satisfying the requirement for homogeneity of slopes (Sokal and Rohlf 1995). Eight small Corbicula (shell length under 15.2 mm) had total mercury concentrations below the detection limit (0.22 ng/g). Because of the uncertainty associated with mercury levels in small Corbicula, all specimens under 15.2 mm in shell length (11 in total) were eliminated leaving a total sample size of 112. Corbicula tissue mercury data were \log_{10} transformed to satisfy statistical assumptions of normality and homogeneity of variance. Back transformed results were reported as geometric means and associated 95% confidence intervals.

All statistical results were considered significant at $P < 0.05$. Statistical analyses were conducted with Systat (SPSS 1997).

3.0 Results

ANOVA of the SRS tributary and river data indicated that the difference in aqueous methylmercury concentration between creek mouths and river was highly significant ($P < 0.001$) as were differences between individual sample sites within habitats ($P = 0.003$), differences between sample dates ($P < 0.001$), and the interaction between habitat and sample date ($P = 0.005$). The interaction was significant because the difference in methylmercury concentration between creek mouths and river was greater on some dates than others; however, methylmercury concentrations were at least slightly higher in the creek mouths on all sample dates (Figure 2). The average methylmercury concentration in the creek mouths (0.170 ng/l) was nearly twice as high as in the river (0.085 ng/l), and the creeks had higher percentages of methylmercury (average of 6.5%) than the river (average of 3.4%). The significance of sites within habitats reflected differences in methylmercury levels among creeks with the highest levels in Steel Creek and Lower Three Runs (Figure 3). Methylmercury levels measured on a single date in the mouths of Brier and Ebenezer Creeks (0.255 ng/l and 0.118 ng/l, respectively) were comparable to methylmercury levels in the SRS tributaries (Figure 3). There was a general correspondence between methylmercury levels in the river and creek mouths as indicated by a significant positive correlation between average methylmercury in each habitat over time ($r = 0.67$, $P = 0.026$, $n = 11$) (Figure 2).

Unlike the methylmercury ANOVA, the only significant term in the ANOVA for total mercury was the interaction between sample date and habitat ($P = 0.030$), indicating that total mercury concentrations were sometimes higher in the creek mouths and sometimes higher in the river (Figure 2). Average total mercury levels differed little between the creek mouths (2.98 ng/l) and the river (2.59 ng/l). An analysis based on all

of the water samples indicated that total mercury and methylmercury levels were significantly ($P < 0.001$) but moderately correlated ($r = 0.54$, $n = 109$).

An average of 72% of the methylmercury was dissolved (based on comparisons between filtered and unfiltered methylmercury data collected from the same locations on four dates). On an average basis, there was little difference between the tributaries and the river in this respect (means of 71.8% and 72.9%, respectively). However, there was considerable difference among tributaries with the percentage of dissolved methylmercury averaging 49% in Beaver Dam Creek, 65% in Upper Three Runs, 80% in Four Mile Branch, 90% in Lower Three Runs, and 71% in Steel Creek ($P < 0.001$, one-way ANOVA) (Figure 3). Somewhat less of the total mercury than the methylmercury was dissolved and, like methylmercury, there was little average difference in total mercury levels between the river (49.1%) and the tributaries (49.3%) but significant differences among individual tributaries ($P < 0.001$).

ANCOVA indicated that mercury levels in Corbicula were significantly related to Corbicula size ($P < 0.001$), that they differed significantly between river and creeks ($P < 0.001$), and that they differed significantly among sample sites within habitats ($P < 0.001$) (Figures 4 and 5). Geometric mean mercury levels normalized to an average shell length of 24.1 mm were 44 ng/g wet weight for the creek mouths and 17 ng/g wet weight for the river sites, indicating a 2.5-fold average difference between habitats. As indicated by the significant difference among sites within habitats, Corbicula mercury levels differed among creeks (Figure 5). Average Corbicula tissue mercury levels in the tributaries were not correlated with average aqueous methylmercury levels ($r = 0.22$, $n = 6$) or total mercury levels ($r = 0.13$, $n = 6$) in the tributaries.

4.0 Discussion

Total mercury concentrations in the Savannah River were within the range of concentrations measured in Wisconsin rivers draining wetland and forested watersheds

(Hurley et al. 1995). They can be contrasted with the much higher total mercury levels (1.3 ug/l) in a Tennessee stream that received mercury discharge from an industrial facility (Southworth et al. 1995). Except for rivers with significant point sources of mercury, most of the total mercury in rivers is derived from diffuse sources in the watershed (Balogh et al. 1998), especially the deposition of atmospheric mercury released by burning coal and other industrial activities that volatilize mercury (Lacerda and Fitzgerald 2001). In the Savannah River approximately 99% of the mercury load is atmospheric (EPA 2000) and possibly from the weathering of stream bank soils. Wetland watersheds are especially efficient at converting this mercury to methylmercury, possibly because they retain mercury well and provide conditions (low dissolved oxygen levels and high levels of organic matter) that favor the sulfate-reducing bacteria involved in methylation (Regnell 1994, Francis et al. 1998). As a result, methylmercury export is generally greater from wetland watersheds than from upland watersheds (St. Louis et al. 1994), and methylmercury levels are higher in rivers with a high proportion of wetlands in their watersheds (Hurley et al. 1995).

The Savannah River tributaries were characterized by significantly higher aqueous methylmercury concentrations than the Savannah River (Figure 3). Total mercury concentrations did not generally differ between the river and the tributaries (Figure 3), suggesting that differences in the amount of inorganic mercury for methylation were not responsible for the difference in methylmercury concentration between river and tributaries. The most likely reason for higher methylmercury levels in the tributaries was that the tributaries supported more favorable environments for methylation. All of the tributaries in this study drained extensive wetlands that would be expected to support comparatively high rates of methylation. The tributary water also tended to be relatively acidic, a condition often associated with increased methylation (Gilmour and Henry 1991).

Methylmercury concentrations and percentages of methylmercury varied among tributaries (Figure 3) suggesting variations in methylation efficiency among streams. It is unlikely that this was related to differences in total mercury. Beaver Dam Creek, the stream with the highest average total mercury concentration, had the lowest average methylmercury concentration. The two streams with the highest concentrations and percentages of methylmercury, Steel Creek and Lower Three Runs, had 400 to 1000 ha reservoirs in their upper reaches that may have directly contributed methylmercury or influenced instream methylation rates by affecting stream water chemistry (e.g., dissolved carbon levels).

Although methylmercury levels varied among tributaries, all tributaries were characterized by at least slightly higher average methylmercury levels than the river (Figure 3). The influence of this tributary methylmercury on the total methylmercury load in the Savannah River is difficult to determine. The temporal correlation between mercury levels in the creek mouths and the river suggests a direct linkage between the two that could result from transport of methylmercury to the Savannah River from the tributaries. However, it is impossible to exclude the alternative explanation that parallel patterns of methylmercury change in both habitats resulted from the action of unmeasured factors that affected both habitats similarly. Examination of USGS stream and river discharge records from 1990 to 2000 indicated that total discharge from the tributaries in this study (excluding Ebenezer Creek for which data were unavailable) composed about 13% of the discharge in the Savannah River, suggesting that mercury transported from them alone constituted a comparatively small proportion of the Savannah River mercury load. However, this comparison does not include the many other Savannah River tributaries not studied.

Although it is difficult to assess the cumulative impacts of tributary methylmercury on Savannah River biota as a whole, tributary discharge appeared to create localized

methylmercury “hotspots” that raised mercury levels in Corbicula residing within the tributary discharge plumes. Soft tissue mercury levels were about 2.5 times higher in Corbicula from the tributary discharge plumes than in Corbicula from the Savannah River upstream from the plumes. This difference was likely a direct consequence of exposure to elevated methylmercury levels in the tributary discharge. Laboratory studies demonstrate that Corbicula rapidly bioaccumulates methylmercury to a much greater extent than inorganic mercury, and that bioaccumulation is proportional to the methylmercury concentration in the water column or sediment (Inza et al. 1997).

Although mercury levels were consistently higher in Corbicula from the tributaries than in Corbicula from the river, tributary Corbicula mercury levels and tributary aqueous methylmercury levels were not significantly correlated. Corbicula are filter feeders (Inza et al. 1997) suggesting that their methylmercury intake may be more closely related to particulate (filterable) methylmercury than to dissolved methylmercury. Lower Three Runs, which had the highest Corbicula mercury levels, also had the highest proportion of filterable methylmercury in its discharge (Figure 3) lending support to this possibility. However, Corbicula can also act as deposit feeders, ingesting periphyton growing on the sediments, and they may be able to directly accumulate methylmercury from the water (Inza et al. 1997). Therefore, methylmercury uptake by Corbicula may not be determined solely by the methylmercury concentration in the water but also by the form of the methylmercury (particulate versus dissolved) and by the relative predominance of several possible modes of methylmercury uptake. Whatever the mode of uptake, methylmercury levels increased with Corbicula size suggesting that Corbicula accumulate mercury with age as fish do (Phillips et al. 1980).

The occurrence of relatively high levels of mercury in Corbicula from the creek mouths suggests the possibility that other organisms from these habitats may also have comparatively high mercury levels. This would likely be most evident in sedentary

organisms (primarily invertebrates) that are continuously exposed to tributary discharge but might also apply to fishes with limited movement such as sunfish that occupy relatively small home ranges (100 linear m, Gatz and Adams 1994). Furthermore, even relatively mobile biota located further upstream in the tributaries, where there is less possibility of movement to areas with lower methylmercury levels, might also have elevated mercury levels. From the standpoint of human exposure to methylmercury, which occurs primarily through the consumption of contaminated fish (EPA 1997), it is possible that consumption of fish caught in and near Savannah River tributaries could result in elevated methylmercury uptake.

The results of this study indicate that tributaries in the middle reaches of the Savannah River appear to be sources of elevated methylmercury that can result in localized increases in mercury levels in Corbicula and presumably other biota. Elevated methylmercury levels in the tributaries may be associated with the drainage of wetland habitats and water chemistry characteristics that favor methylmercury production. More intensive research will be needed to better understand methylmercury production in the tributaries, the impact of tributary discharge on methylmercury level in the mainstem river and river biota, and whether similar patterns of elevated tributary methylmercury levels occur in other river systems. Corbicula appear to be well suited for studying this phenomenon and, more generally, the effects of any point mercury inputs to aquatic systems because they are relatively stationary and accumulate mercury. However, the use of Corbicula within a restricted size range may facilitate comparisons among sites because Corbicula mercury levels are related to size.

Acknowledgements

We thank W.M. Fulmer and R.J. Roseberry for assistance with field collections. We also thank three anonymous reviewers for comments that improved this paper. This report was developed during U.S. Department of Energy Contract No. DE-AC09-96SR18500.

References

- Balogh S, Meyer M, Johnson K. Diffuse and point source mercury inputs to the Mississippi, Minnesota, and St. Croix Rivers. *The Science of the Total Environment* 1998;213:109-113.
- Bloom NS. Trace metals and ultra-clean sample handling. *Environ. Lab.* 1995;7:20.
- Chase ME, Jones SH, Hennigar P, Sowles J, Harding GCH, Freeman K, Wells PG, Krahforst C, Coombs K, Crawford R, Pederson J, Taylor D. Gulfwatch: monitoring spatial and temporal patterns of trace metal and organic contaminants in the Gulf of Maine (1991-1997) with the blue mussel, *Mytilus edulis L.* *Marine Pollution Bulletin* 2001;42:491-505.
- Cope WG, Bartsch MR, Rada RG, Balogh SJ, Rupprecht JE, Young RD, Johnson DK. Bioassessment of mercury, cadmium, polychlorinated biphenyls, and pesticides in the Upper Mississippi River with zebra mussels (*Dreissena polymorpha*). *Environmental Science and Technology* 1999;33:4385-4390.
- EPA (United States Environmental Protection Agency). Mercury study report to congress, volume III: fate and transport of mercury in the environment. EPA-452/R-97-005. Office of Air Quality Planning & Standards and Office of Research and Development, December 1997.
- EPA (United States Environmental Protection Agency). Total maximum daily load (TMDL) development for total mercury in the middle/lower Savannah River, GA. USEPA Region 4, Atlanta GA, 2000.
- Fitzgerald WF, Mason RP, Vandal GM. Atmospheric cycling and air-water exchange of mercury over mid-continental lacustrine regions. *Water, Air, & Soil Pollution* 1991;56:745-767.
- Francis DR, Jude DJ, Barres JA. Mercury distribution in the biota of a Great Lakes estuary: Old Woman Creek, Ohio. *J. Great Lakes Res.* 1998;24:595-607.

- Gatz AJ Jr., Adams SM. Patterns of movement of centrarchids in two warmwater streams in eastern Tennessee. *Ecol Freshwat Fish* 1994;3:35-48.
- Gilmour CC, Henry EA. Mercury methylation in aquatic systems affected by acid deposition. *Environ. Pollut.* 1991;71:131-169.
- Gilmour CC, Henry EA, Mitchell R. Sulfate stimulation of mercury methylation in freshwater sediments. *Environ. Sci. Technol.* 1992;26:2281-2287.
- Halverson NV, Bowers JA, Jackson DG, Paller MH, Martin FD. Aquatic mercury assessment interim data report: October 2000 – September 2001(U). Westinghouse Savannah River Company, Aiken, SC, 2002.
- Hurley JP, Benoit JM, Babiarz CL, Shafer MM, Andren AW, Sullivan JR, Hammond R, Webb DA. Influences of watershed characteristics on mercury levels in Wisconsin rivers. *Environmental Science and Technology* 1995;29:1867-1875.
- Hurley JP, Cowell SE, Shafer MM, Hughes PE. Tributary loading of mercury to Lake Michigan: importance of seasonal events and phase partitioning. *The Science of the Total Environment* 1998;213:129-137.
- Inza B, Ribeyre F, Maury-Brachet R, Boudou A. Tissue distribution of inorganic mercury, methylmercury and cadmium in the Asiatic clam (Corbicula fluminea) in relation to the contamination levels of the water column and sediment. *Chemosphere* 1997;35:2817-2836.
- Inza B, Ribeyre F, Boudou A. Dynamics of cadmium and mercury compounds (inorganic mercury or methylmercury): uptake and depuration in Corbicula fluminea. Effects of temperature and pH. *Aquatic Toxicology* 1998;43:273-285.
- Lacerda LD, Fitzgerald WF. Biogeochemistry of mercury in wetlands. *Wetlands Ecology and Management* 2001;9:291-293.

Mierle G. Aqueous inputs of mercury to Precambrian Shield lakes in Ontario.

Environmental Toxicology and Chemistry 1990;9:843-851.

Nater EA, Grigal DF. Regional trends in mercury distribution across the Great Lakes states, north central USA. Nature 1992;358:139-141.

Paller MH, O'Hara J, Osteen DV. Annual report on the Savannah River aquatic ecology program. September 1983 - August 1984. Vol. II. Ichthyoplankton. ECS-SR-18. Environmental and Chemical Sciences, Inc., Aiken, SC, 1985

Paller MH, Saul BM, Osteen DV. Distribution and abundance of ichthyoplankton in the mid-reaches of the Savannah River and selected tributaries. ECS-SR-27. Environmental & Chemical Sciences, Inc. Aiken, SC, 1986.

Phillips GR, Lenhart TE, Gregory RW. Relation between trophic position and mercury accumulation among fishes from the Tongue River Reservoir, Montana. Environmental Research 1980;22:73-80.

Regnell O. The effect of pH and dissolved oxygen levels on methylation and partitioning of mercury in freshwater model systems. Environmental Pollution 1994;84:7-13.

Sokal RR, Rohlf F.J. Biometry, 3rd edition. WH Freeman, New York, 1995.

Southworth GR, Turner RR, Peterson MJ, Bogle MA. Form of mercury in stream fish exposed to high concentrations of dissolved inorganic mercury. Chemosphere 1995;30:779-787.

SPSS. Systat 7.0. SPSS Inc., Chicago, 1997

St. Louis VL, Rudd JWM, Kelly CA, Beaty KG, Bloom NS, Flett RJ. Importance of wetlands as sources of methylmercury to boreal forest ecosystems. Canadian Journal of Fisheries and Aquatic Science 1994;51:1065-1076.

Taylor LR. Aggregation, variance and the mean. Nature 1961;189:732-735.

WSRC-MS-2003-00489
June 25, 2003

U.S. EPA. (Environmental Protection Agency). Guidance for Implementation and use of
EPA Method 1631 for the determination of low-level mercury (40 CFR part 136).
EPA 821-R-01-023. March 2001.

List of Figures

Figure 1. Map of the middle reaches of the Savannah River showing important tributary streams and the Savannah River Site (SRS). Circles represent areas where Water and Corbicula fluminea samples were collected from the tributary mouths and the Savannah River (see text for more details). Samples were also collected further downstream near Ebenezer Creek (not shown). Gray shading represents wetlands on the Savannah River Site.

Figure 2. Average aqueous methylmercury and total mercury levels for five Savannah River locations and five SRS Savannah River tributary creeks on 11 sample dates. Error bars represent standard errors.

Figure 3. Aqueous methylmercury and total mercury levels in the Savannah River (SR) and seven tributary creeks: Upper Three Runs (UTR), Beaver Dam Creek (BDC), Four Mile Branch (FMB), Steel Creek (SC), Lower Three Runs (LTR), Brier Creek (BC), and Ebenezer Creek (EC). With the exception of Brier and Ebenezer Creeks, each bar represents an average (with standard error) calculated from data collected over 11 sample dates. Brier and Ebenezer Creeks were sampled once. Percentages above the bars represent methylmercury expressed as a proportion of total mercury.

Figure 4. Relationship between mercury concentration and shell length in Corbicula fluminea collected from the Savannah River and several Savannah River tributaries. Regression equations are for \log_{10} transformed data.

Figure 5. Geometric mean mercury levels in Corbicula fluminea collected from six Savannah River tributaries (Upper Three Runs, UTR; Beaver Dam Creek, BDC; Four Mile Branch, FMB; Steel Creek, SC; Lower Three Runs, LTR; Brier Creek, BC; Ebenezer Creek, EC) and six Savannah River sites located just upstream from each tributary. Error bars represent 95% confidence intervals.









