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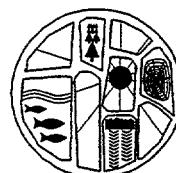
## ENVIRONMENTAL SCIENCES DIVISION

**Report on the Biological Monitoring  
Program at  
Paducah Gaseous Diffusion Plant  
January–December 1997**

L. A. Kszos  
M. J. Peterson  
M. G. Ryon  
J. G. Smith  
G. R. Southworth

Environmental Sciences Division  
Publication No. 4756

March 1998



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January-December 1997**

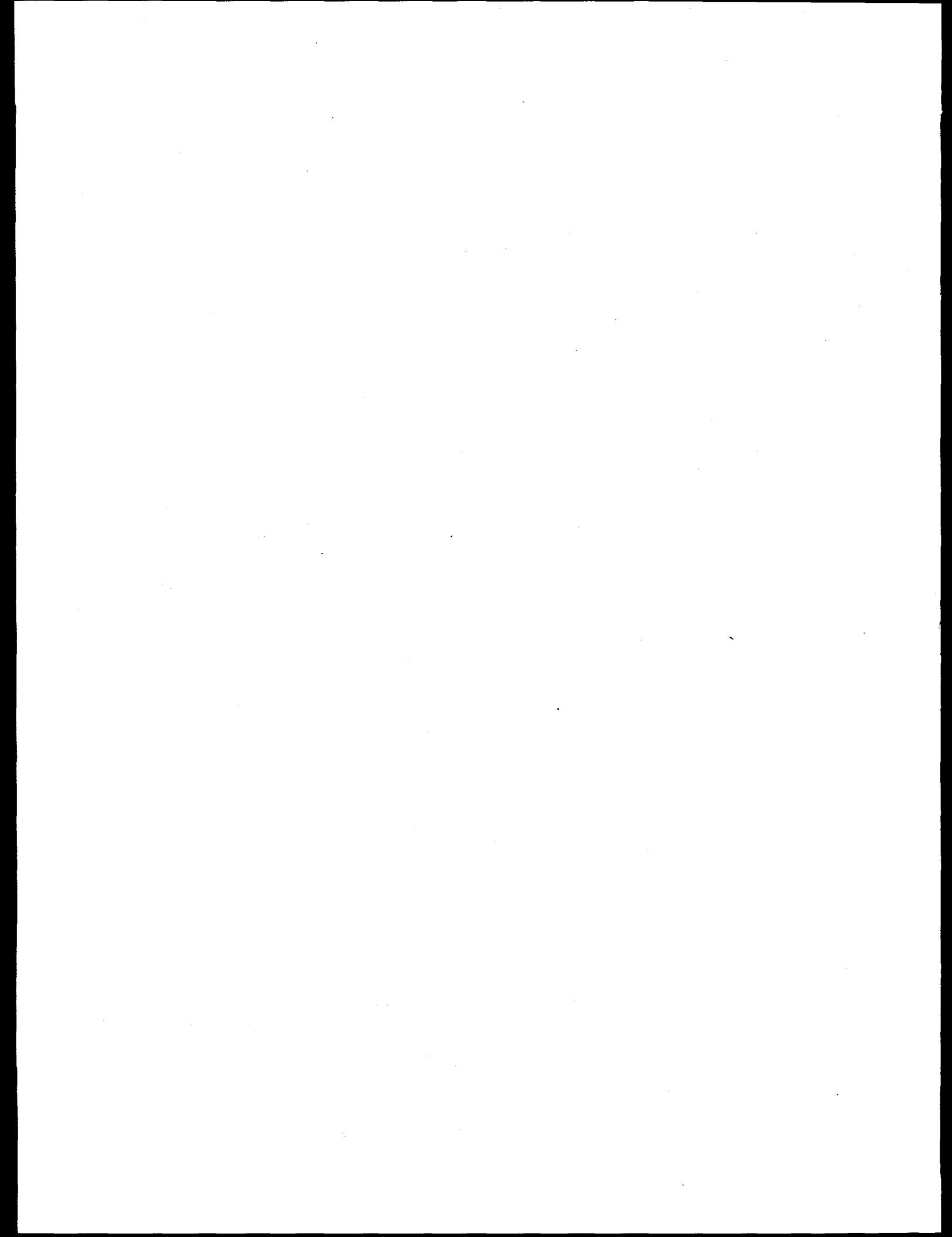
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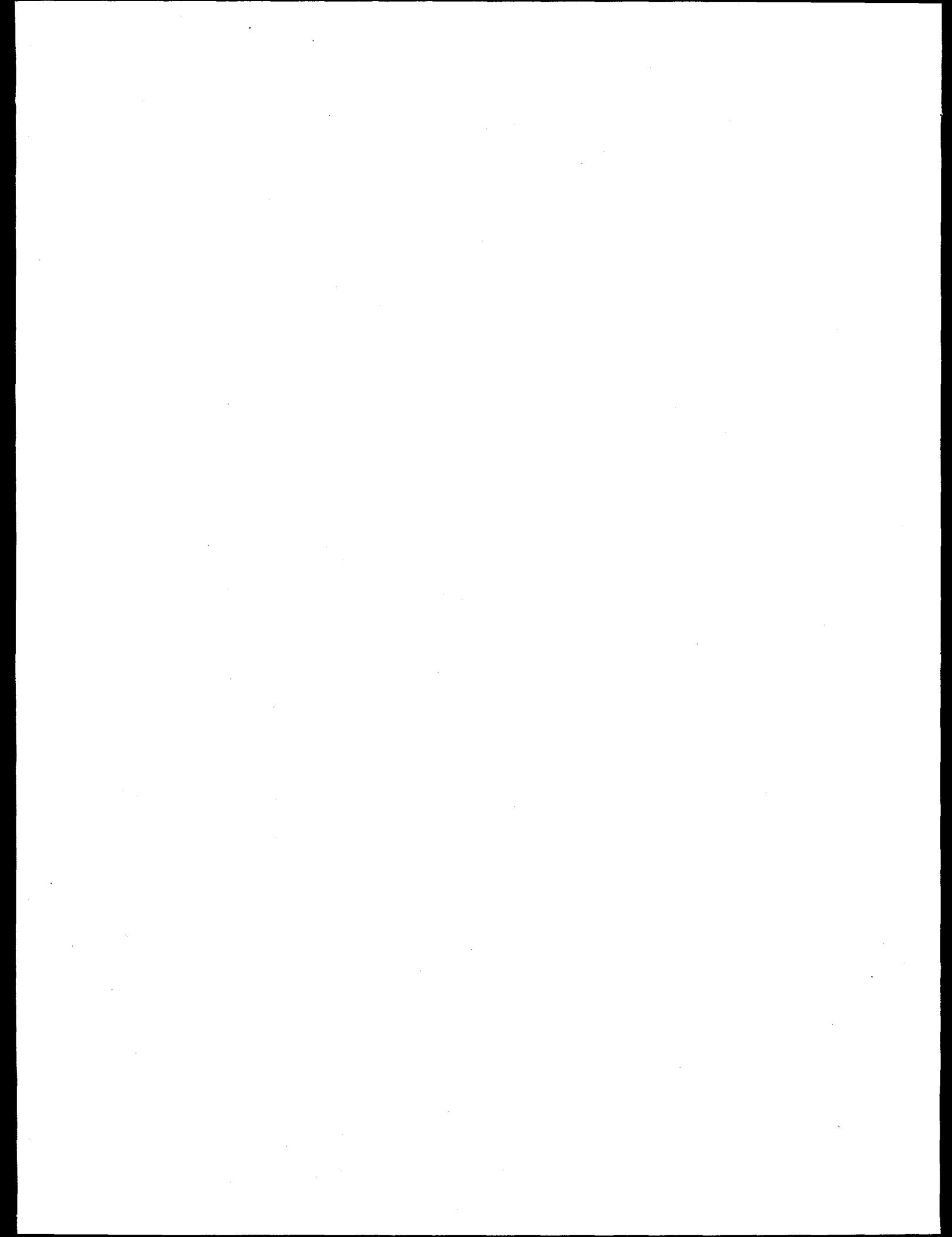
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## ACRONYMS

|          |   |
|----------|---|
| ANOVA    | analysis of variance  |
| BMAP     | Biological Monitoring and Abatement Program                 |
| BMP      | Biological Monitoring Program                               |
| BBK      | Big Bayou Creek kilometer                                   |
| DCBP     | decachlorobiphenyl  |
| DOE      | U.S. Department of Energy                                   |
| ESD      | Environmental Sciences Division                             |
| EPA      | U.S. Environmental Protection Agency                        |
| EPT      | Ephemeroptera, Plecoptera, Trichoptera                      |
| FDA      | U.S. Department of Agriculture Food and Drug Administration |
| GC/ECD   | gas chromatography/electron capture detection               |
| GLM      | general linear model  |
| HINDS CR | Hinds Creek   |
| IC       | inhibition concentration                                    |
| KDOW     | Kentucky Division of Water                                  |
| KPDES    | Kentucky Pollutant Discharge Elimination System             |
| LOAEL    | Lowest observable adverse effect level                      |
| LMES     | Lockheed Martin Energy Systems, Inc.                        |
| LMUS     | Lockheed Martin Utility Systems, Inc.                       |
| LUK      | Little Bayou Creek kilometer                                |
| MAK      | Massac Creek kilometer                                      |
| MS-222   | tricaine methanesulfonate                                   |
| NOAEL    | No observable adverse effect level                          |
| NCBP     | National Contaminant Biomonitoring Program                  |
| NOEC     | no-observed-effect concentration                            |
| NPDES    | National Pollutant Discharge Elimination System             |
| ORNL     | Oak Ridge National Laboratory                               |
| PCB      | polychlorinated biphenyl                                    |
| PGDP     | Paducah Gaseous Diffusion Plant                             |
| QA       | quality assurance   |
| RGA      | regional gravel aquifer                                     |
| RCW      | recirculating cooling water                                 |
| SAS      | statistical analysis system                                 |
| SD       | standard deviation  |
| SE       | standard error  |
| TRC      | total residual chlorine                                     |
| TU       | toxicity unit(s)  |
| TUc      | chronic toxicity unit(s)                                    |
| USEC     | United States Enrichment Corporation                        |
| USFWS    | U.S. Fish and Wildlife Service                              |

Biological Monitoring Program

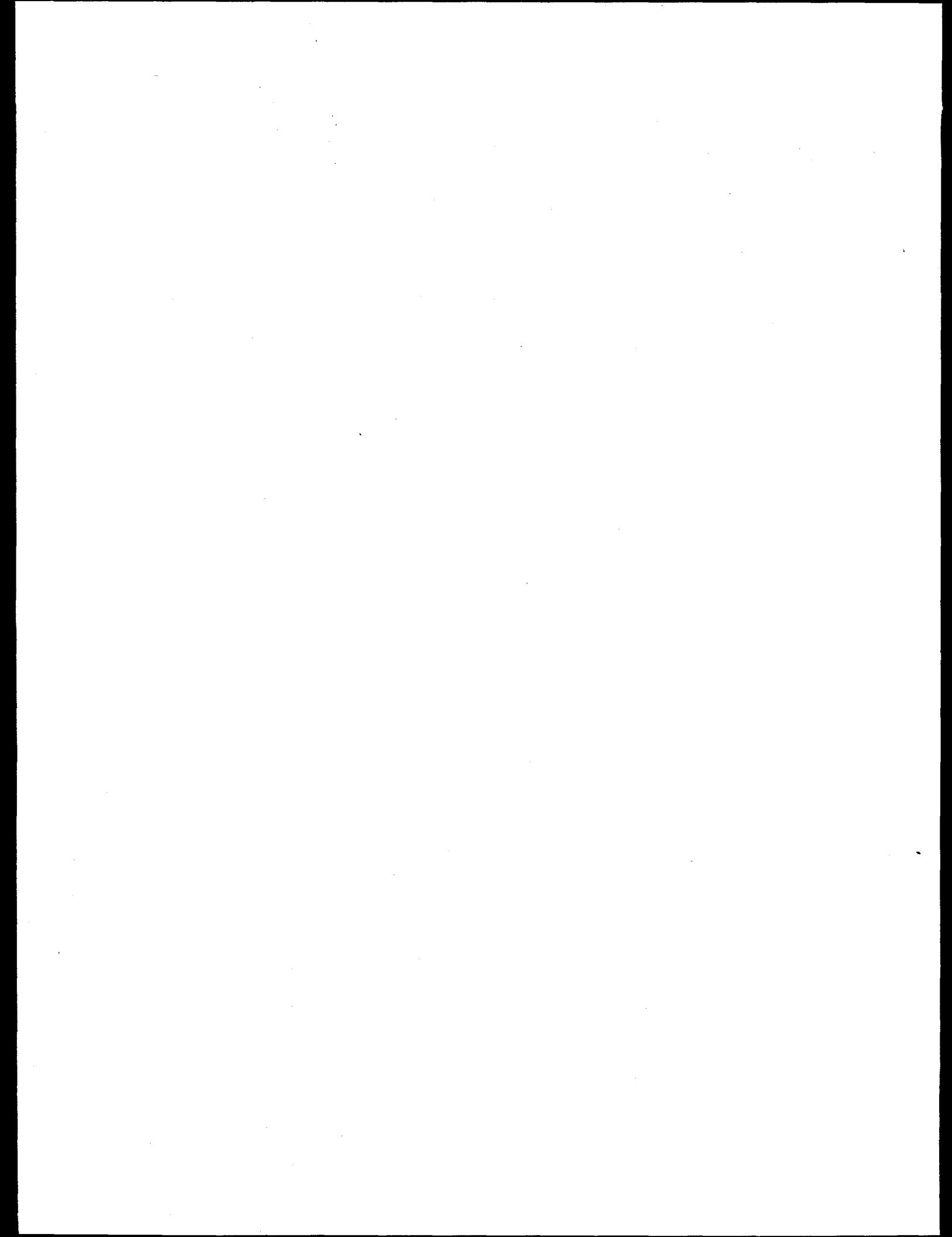
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USGS      U.S. Geological Service  
WKWMA    West Kentucky Wildlife Management Area

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## EXECUTIVE SUMMARY

On September 24, 1987, the Kentucky Natural Resources and Environmental Protection Cabinet issued an Agreed Order that required the development of a Biological Monitoring Program (BMP) for the Paducah Gaseous Diffusion Plant (PGDP). The PGDP BMP was conducted by the University of Kentucky between 1987 and 1991 and by staff of the Environmental Sciences Division (ESD) at Oak Ridge National Laboratory (ORNL) from 1991 to present. The goals of BMP are to (1) demonstrate that the effluent limitations established for PGDP protect and maintain the use of Little Bayou and Big Bayou creeks for growth and propagation of fish and other aquatic life, (2) characterize potential environmental impacts, and (3) document the effects of pollution abatement facilities on stream. In September 1992, a renewed Kentucky Pollutant Discharge Elimination System (KPDES) permit was issued to PGDP. The renewed permit required toxicity monitoring of continuous and intermittent outfalls on a quarterly basis. On April 6, 1996, an Agreed Order between the U.S. Department of Energy (DOE), United States Enrichment Corporation (USEC), and the Kentucky Division of Water (KDOW) was signed, which settled issues involving a challenge to the KPDES permit. The Agreed Order lists the requirements for limits on copper, cadmium, chromium, lead, nickel, zinc, temperature, phosphorous, pH, and chronic toxicity. A BMP is not currently required in either the Agreed Order or the renewed permit; however, biological monitoring of DOE facilities at PGDP is required under DOE Order 5400.1, General Environmental Protection Program.

In February 1998, draft KPDES permits were issued to the Department of Energy and USEC for PGDP. The renewed DOE permit requires chronic toxicity monitoring of one continuous outfall and acute toxicity monitoring of three intermittent outfalls on a quarterly basis. A watershed monitoring program must be developed within ninety days of the effective date of the renewed permit.

The BMP for PGDP consists of three major tasks: (1) effluent toxicity monitoring, (2) bioaccumulation studies, and (3) ecological surveys of stream communities (i.e., fish). This report focuses on ESD activities occurring from January 1997 to December 1997, although activities conducted outside this time period are included as appropriate.

### Study Area

The PGDP is owned by DOE. Production facilities are leased to the USEC and are managed by Lockheed Martin Utility Systems, Inc. (LMUS). The environmental restoration and waste management activities are managed by Lockheed Martin Energy Systems, Inc. (LMES). Construction of the plant was completed in 1954, although production began in 1952. PGDP is an active uranium enrichment facility consisting of a diffusion cascade and extensive support facilities. Support facilities include a steam plant, four electrical switchyards, four sets of cooling towers, a chemical cleaning and decontamination facility, water and wastewater treatment plants, a chromium reduction facility, and maintenance and laboratory facilities.

PGDP is located in the western part of the Ohio River basin. Surface drainage from PGDP enters Big Bayou Creek and Little Bayou Creek, two small tributaries to the Ohio River. Big Bayou Creek is a perennial stream with a drainage basin extending from ~4 km south of PGDP to the Ohio River. Part of its 14.5-km course flows along the western boundary of the plant. Little Bayou Creek originates in the Western Kentucky Wildlife Management Area and flows for 10.5 km north toward the Ohio River; its course includes part of the eastern boundary of PGDP. Four continuously flowing outfalls (001, 006, 008, and 009) discharge to Big Bayou Creek. Outfalls 002, 010, 011, and 012 are combined at the C617 pond and discharged via Outfall 010 into Little Bayou Creek. Effluent from Outfalls 013, 015, 016, 017, and 018 regularly discharge into Big Bayou and Little Bayou creeks when it rains.

Three sites on Big Bayou Creek—Big Bayou Creek kilometer (BBK) 12.5, BBK 10.0, and BBK 9.1—one site on Little Bayou Creek, Little Bayou Creek kilometer (LUK) 7.2; and one off-site reference station on Massac Creek, Massac Creek kilometer (MAK) 13.8, were routinely sampled to assess the ecological health of the stream. Two additional sites (LUK 9.0, and LUK 4.3) were sampled as part of the bioaccumulation monitoring task. Fish community sampling and bioaccumulation sampling were conducted twice annually in the spring and fall. KPDES outfalls evaluated for effluent toxicity in 1997 included 001, 006, 008, 009, 010, 013, 015, 016, 017, and 018.

### **Toxicity Monitoring**

*Ceriodaphnia dubia*<sup>1</sup> and fathead minnow toxicity tests of effluents from the continuously flowing outfalls (001, 006, 008, 009, and 010) and the intermittently flowing outfalls (013, 015, 016, 017, and 018) were conducted quarterly as required by the KPDES permit. Tests of effluent from Outfall 001 were conducted using *C. dubia* and fathead minnows. Tests of all other effluents were conducted using only fathead minnows. The 25% inhibition concentrations (IC25: that concentration causing a 25% reduction in fathead minnow growth or *Ceriodaphnia* survival compared with the control) were determined for each test. The chronic toxicity unit rating ( $TU_c = 100/IC_{25}$ ) is required as a compliance endpoint in the renewed permit. The higher the  $TU_c$ , the more toxic an effluent. Because Little Bayou and Big Bayou creeks have been determined to have a low flow of zero, a  $TU_c \geq 1.0$  would be considered a noncompliance (for the continuously flowing outfalls) and an indicator of potential instream toxicity. This report summarizes the toxicity test results for 1997.

During 1997, effluent from Outfall 001 exceeded the permit limit ( $TU_c \geq 1.0$ ) in August with a  $TU_c = 8.34$ . This is the first occurrence of a fathead minnow test with a  $TU_c \geq 1.0$  for Outfall 001 since testing began in October 1991. The confirmatory test conducted in September resulted in a  $TU_c < 1.0$ , demonstrating that the effluent was no longer toxic. The  $TU_c$ s for outfalls 006, 008, 009, and 010 were less than 1.0 for all tests conducted in 1997.

Toxicity tests of the intermittent outfalls were conducted in January, April, July, and December. The only cases of  $TU_c \geq 1.0$  were for the fathead minnow tests of Outfall 015 in July and Outfall 016 in April. The  $TU_c$  for Outfall 015 in July was 2.74. This is the first

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<sup>1</sup>*Ceriodaphnia dubia*, commonly known as the waterflea, is a small crustacean commonly accepted as a standard test organism.

case of a TUC  $\geq 1.0$  for Outfall 15 since November 1994. The subsequent test of Outfall 015 in December resulted in TUC  $< 1.0$ . The TUC for Outfall 016 in April was 19.61. Similar to Outfall 015, this is the first case of a TUC  $\geq 1.0$  for Outfall 15 since November 1994. The subsequent tests of Outfall 016 in July and December resulted in TUCs  $< 1.0$ . Total suspended solids and flow rate were quite variable between tests as would be expected due to the fact that flow of the intermittent outfalls is rainfall-dependant. The TUC = 19.61 for Outfall 016 occurred during the test period that also had the highest total suspended solids (TSS), suggesting that for this outfall, toxicity may be related to TSS or a contaminant related to TSS. A level of 2.74 chronic toxicity units for Outfall 015 did not correspond to the highest TSS for this outfall, but did correspond to the highest flow rate.

In December 1996, a bioavailability study was initiated to develop alternative metal limits for cadmium, chromium, copper, lead, nickel, and zinc. As stipulated in the Agreed Order, DOE/USEC must demonstrate to the satisfaction of the Cabinet that a more appropriate analytical technique or criteria is available, one that provides a better measurement of levels of metals present that would be toxic to aquatic life. Phase I of the study developed alternative metal limits for continuously discharging outfalls. A report detailing the results of Phase I has been submitted to the KDOW for comment. The overall objectives of Phase I were to

- evaluate the toxicity of continuous outfalls (001, 008, 009, and 010) at PGDP,
- determine the mean ratio of dissolved to total recoverable metal for Cd, Cr, Cu, Pb, Ni, and Zn in the continuous outfalls,
- determine whether the concentration of TR metal discharged causes toxicity to fathead minnows and/or *C. dubia*, and
- determine alternative metal limits for each metal of concern (Cd, Cr, Cu, Pb, Ni, and Zn).

### Bioaccumulation

Bioaccumulation monitoring conducted to date as part of the BMP identified PCB contamination in fish in Little Bayou Creek, and to a lesser extent, Big Bayou Creek, as primary concerns. Mercury concentrations in fish in Big Bayou Creek were found to be higher in fish downstream from PGDP discharges than in fish from an upstream site. The main objective of the 1996-97 bioaccumulation monitoring was to evaluate spatial and temporal changes in PCB contamination in fish from Little Bayou Creek. Monitoring for mercury and PCBs in fish from Big Bayou Creek was restricted to spotted bass. Longear sunfish were collected for PCB and mercury analysis from Little Bayou Creek in October 1996, April 1996 and October 1997. Spotted bass were collected from Big Bayou Creek in October 1996 and October 1997. Massac Creek in McCracken County, Kentucky, and Hinds Creek, Anderson County, Tennessee, were used as reference sites, providing data on background concentrations at uncontaminated sites and samples for use as analytical controls.

Mean PCB concentrations in sunfish from Little Bayou Creek were higher than in fish from reference sites on all sampling dates. On two of three dates, highest mean PCB concentrations were found in fish from the middle site on Little Bayou Creek, with an abrupt decrease in average concentration at the downstream site. Previously, mean PCB concentrations in sunfish from Little Bayou Creek had always been highest at the uppermost site nearest PGDP discharges, with a progressive decrease at the two downstream sites. The change in the downstream pattern of PCB accumulation in sunfish may indicate that chronic

PCB discharges from the PGDP facility are becoming less predominant relative to in-stream sources in determining levels of contamination in fish. The trend of decreasing PCB contamination over time in sunfish in Little Bayou Creek continued, with PCB concentrations in sunfish at the uppermost Little Bayou Creek site averaging less than 0.4  $\mu\text{g/g}$  in fall 1997 versus nearly 2  $\mu\text{g/g}$  in 1992. However, the rate of decrease over this period appears to have slowed over the past several years.

Mean mercury concentrations in bass from Big Bayou Creek and Little Bayou Creek in 1996 were typical of previous years. Low mercury concentrations in 1997 were a consequence of our inability to obtain larger specimens, and do not represent a temporal change. Mercury concentrations in bass from Big Bayou Creek appear to be typical of uncontaminated streams in the Paducah vicinity. Analysis of water samples from Big Bayou Creek above and below PGDP discharges, supported by site specific mercury studies in the East Fork Poplar Creek BMAP, found total mercury concentrations in the creek to be slightly higher downstream from PGDP, but well within the range of natural background concentrations for streams.

The continued low levels of PCB contamination in fish in Little Bayou Creek provides evidence of effective controls and remediation of sources within PGDP. Continued monitoring will help assess whether additional controls are needed.

### **Fish Community Monitoring**

Quantitative sampling of the fish community was conducted at three sites in Big Bayou Creek, one site in Little Bayou Creek, and at one offsite reference station (Massac Creek) during March and September 1997. Data on the fish communities of Big Bayou Creek and Little Bayou Creek downstream of PGDP were compared to data from reference sites located on Big Bayou Creek above PGDP and on Massac Creek. These comparisons indicated a slight but noticeable degradation in the communities downstream of PGDP. Effects on the fish community were greatest just downstream from PGDP at BBK 10.0. The fish community at this site had a low mean and total species richness. However, slight improvements of the fish community were noted in 1997 with one sensitive species, benthic insectivores, and a darter species taken at BBK 10.0. The lower species richness, compared with reference sites, may be a result of thermal impacts associated with outfalls (see Roy et al. 1996). Although the temperatures may not be lethal, they could produce avoidance of the areas of Big Bayou Creek near the plant outfalls. Compared with earlier sampling, BBK 10.0 demonstrated a rebound in spring productivity. Overall, the fish community at BBK 10.0 has demonstrated shortcomings in several evaluation metrics, but also has some indications of recent improvements.

The fish community at BBK 9.1 showed signs of impact but at less severe levels than at BBK 10.0 and less severe than earlier sampling at this site. Mean and total species richness were at very high levels, actually surpassing the levels at MAK 13.8. The number of sucker species and abundance of benthic insectivores also increased compared with 1996 samples. As with BBK 10.0, productivity estimates continued to improve from past years. These trends indicate a lessening of impacts on recruitment success for the fish community at BBK 9.1.

The fish community at LUK 7.2 was similar to that at the BBK 12.5 reference site. The mean species richness values were similar to those of the reference site and had rebounded substantially from a low point in fall 1994. Biomass also remained at high levels, but density declined to new lows for this site. Unlike conditions in Big Bayou Creek sites, productivity did not increase in 1997. Generally, the conditions at LUK 7.2 indicate only minor impacts

associated with PGDP operations, but the decline in densities should be closely monitored as it could be indicative of more substantial long-term impacts.

Monitoring of the fish communities associated with PGDP streams indicated some depressed conditions but did not specifically identify causative agents. The impacts were limited to sites closest to the plant, which suggests that PGDP discharges (e.g., high temperatures or increases in sedimentation) may be the cause.

## 1. INTRODUCTION

*L. A. Kszos*

On September 24, 1987, the Commonwealth of Kentucky Natural Resources and Environmental Protection Cabinet issued an Agreed Order that required the development of a Biological Monitoring Program (BMP) for the Paducah Gaseous Diffusion Plant (PGDP). A plan for the biological monitoring of the receiving streams (Little Bayou Creek and Big Bayou Creek) was prepared by the University of Kentucky, reviewed by staff at PGDP and Oak Ridge National Laboratory (ORNL), and submitted by the U.S. Department of Energy (DOE) to the Kentucky Division of Water (KDOW) for approval. The PGDP BMP was implemented in 1987 and consisted of ecological surveys, toxicity monitoring of effluents and receiving streams, evaluation of bioaccumulation of trace contaminants in biota, and supplemental chemical characterization of effluents. The PGDP BMP was patterned after plans that were implemented in 1985 for the Oak Ridge Y-12 Plant (Loar et al. 1989) and in 1986 for ORNL (Loar et al. 1991) and the Oak Ridge Gaseous Diffusion Plant (presently the East Tennessee Technology Park, Kszos et al. 1993). Because research staff from the Environmental Sciences Division (ESD) at ORNL were experienced in biological monitoring, they served as reviewers and advisers throughout the planning and implementation of the PGDP BMP. Data resulting from BMP conducted by the University of Kentucky were presented in a 3-year report issued in December 1990 (Birge et al. 1990) and a progress report issued in December 1991 (Birge et al. 1992).

Beginning in fall 1991, ESD added data collection and report preparation to its responsibilities for the PGDP BMP. The BMP has been continued because it has proven to be extremely valuable in (1) identifying those effluents with the potential for adversely affecting instream fauna, (2) assessing the ecological health of receiving streams, and (3) guiding plans for remediation and protecting human health. For example, BMP has documented the improved health of the streams in the vicinity of PGDP. The continued documentation of ecological recovery and improvement of water quality may be used to develop appropriate chemical limits and monitoring requirements. BMP has shown that (1) contaminants bioaccumulate to a significant degree in aquatic species and (2) the fish communities in Big Bayou Creek have been negatively impacted. Continued biological monitoring will assess the degree to which abatement actions ecologically benefit Big Bayou Creek and Little Bayou Creek. Data from continued monitoring can also be used to evaluate the need for additional remediation and to assess the impact of inadvertent spills or fish kills. Furthermore, BMP results can be used to educate the public about PGDP's commitment to environmental protection.

In September 1992, a renewed KPDES permit was issued to PGDP. The renewed permit requires toxicity monitoring of continuous and intermittent outfalls on a quarterly basis. On April 6, 1996, an Agreed Order between DOE, USEC, and the KDOW was signed which settled issues involving a challenge to the KPDES permit. The Agreed Order lists the requirements for limits on copper, cadmium, chromium, lead, nickel, zinc, temperature, phosphorous, pH, and chronic toxicity. A BMP is not required in either the Agreed Order or the renewed permit; however, biological monitoring of the DOE facilities at PGDP is required

**1-2 -- Biological Monitoring Program**

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under DOE Order 5400.1, General Environmental Protection Program. Data collected under BMP will also be used to support three studies in the Agreed Order: (1) temperature variability and instream effects of elevated temperature from PGDP outfalls, (2) development of site-specific metal limits for outfalls, and (3) instream monitoring for pH in Big Bayou and Little Bayou creeks.

In February 1998, draft KPDES permits were issued to the Department of Energy and USEC for PGDP. The renewed DOE permit requires chronic toxicity monitoring of one continuous outfall (Outfall 001) and acute toxicity monitoring of three intermittent outfalls (outfalls 015, 017, and 018) on a quarterly basis. A watershed monitoring program will be developed within ninety days of the effective date of the renewed permit.

The BMP for PGDP consists of three major tasks: (1) effluent toxicity monitoring, (2) bioaccumulation studies, and (3) ecological surveys of benthic macroinvertebrate communities and fish. With the exception of the benthic macroinvertebrate community surveys, this report focuses on activities from January to December 1997. Activities conducted outside this time period, particularly historical data used to describe trends, are also included as appropriate. The results of the benthic macroinvertebrate surveys were summarized in the 1997 semi-annual report (Appendix A).

## 2. DESCRIPTION OF STUDY AREA<sup>2</sup>

*L. A. Kszos*

### 2.1 SITE DESCRIPTION

The Paducah Gaseous Diffusion Plant is located in western Kentucky and owned by the United States Department of Energy (DOE). Construction of the plant was completed in 1954, although production began in 1952. PGDP is an active uranium enrichment facility consisting of a diffusion cascade and extensive support facilities (Jones et al. 1997). The uranium enrichment gaseous diffusion process involves more than 1800 stages with operations housed in 5 buildings covering ~300 ha. Including support facilities, the plant has ~30 permanent buildings located on a 1386-ha site (Jones et al. 1997). Support facilities include a steam plant, four electrical switchyards, four sets of cooling towers, a chemical cleaning and decontamination facility, water and wastewater treatment plants, a chromium reduction facility, and maintenance and laboratory facilities. Several inactive facilities are also located on the site. Currently, the Paducah cascade processes are being used for the enrichment of uranium up to 2%  $^{235}\text{U}$ . Most of the uranium produced is used for national defense and commercial reactors in the United States and abroad. In July 1993, DOE leased the plant production operations facilities, which are managed by Lockheed Martin Utility Systems, Inc. (LMUS), to the United States Enrichment Corporation (USEC). Under this lease, USEC has assumed responsibility for compliance activities directly associated with uranium enrichment operations. DOE maintains responsibility for the environmental restoration and waste management activities through its management contractor, Lockheed Martin Energy Systems, Inc. (LMES).

#### 2.1.1 Land Use

The area surrounding PGDP is mostly rural, with residences and farms surrounding the plant. Immediately adjacent to PGDP is the West Kentucky Wildlife Management Area (WKWMA), 850 ha of managed habitat either deeded or leased to the Commonwealth of Kentucky.

The population within a 80-km radius of the plant is about 300,500 people. The unincorporated communities of Grahamville and Heath are within 2–3 km, east of the facility. The largest cities in the region are Paducah, Kentucky, and Cape Girardeau, Missouri, located about 16 and 64 air km away respectively (U.S. Department of Commerce 1991).

For information on the geohydrology of the region, see D'Appolonia 1983; GeoTrans 1990; TERRAN 1990; CH2M Hill 1991; Kszos 1994a, 1994b; and Jones et al. 1997.

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<sup>2</sup>Sections 2.1 and 2.2 contain large excerpts from Jones et al. 1997. Paducah Site 1996 Annual Environmental Report. KY/EM-206. Lockheed Martin Energy Systems, Inc., Kevil, Kentucky.

### **2.1.2 Surface Water**

The PGDP is located in the western part of the Ohio River basin. The confluence of the Ohio River with the Tennessee River is ~24 km upstream of the site, and the confluence of the Ohio River with the Mississippi River is ~90 km downstream of the site. Surface drainage from PGDP is two small tributaries of the Ohio River, Big Bayou Creek and Little Bayou Creek (Fig. 2.1). These streams meet ~4.8 km north of the site and discharge to the Ohio River at kilometer 1524 (Fig. 2.2), which is ~56 km upstream of the confluence of the Ohio and Mississippi rivers. The PGDP is located on a local drainage divide; surface flow is east-northeast toward Little Bayou Creek and west-northwest toward Big Bayou Creek. Big Bayou Creek is a perennial stream with a drainage basin extending from ~4 km south of PGDP to the Ohio River; part of its 14.5-km course flows along the western boundary of the plant. Little Bayou Creek originates in the WKWMA and flows for 10.5 km north toward the Ohio River; its course includes part of the eastern boundary of the plant. The watershed areas for Big Bayou Creek and Little Bayou Creek are about 4819 and 2428 ha respectively. These streams exhibit widely fluctuating discharge characteristics that are closely tied to local precipitation and facility effluent discharge rates. Natural runoff makes up a small portion of the flow, and, during dry weather, effluents from PGDP operations can constitute about 85% of the normal base flow in Big Bayou Creek and 100% in Little Bayou Creek. During the dry season which extends from summer to early fall, no-flow conditions may occur in the upper section of Little Bayou Creek.

Precipitation in the region averages about 120 cm per year. Precipitation was 127.58 cm in 1997 with the highest rainfall occurring in May (Table 2.1). There were four major storms ( $\geq 5$  cm in 24–48 hours): one in April, one in June, and two in May. Daily rainfall data for 1997 are provided in Appendix B. See Kszos et al. (1994b, 1995, 1996, 1997) and Kszos (1996) for information on precipitation during 1992–96. The lower Bayou drainage has low to moderate gradient, and the lower reaches are within the flood plain of the Ohio River. The drainage basin is included in ecoregion 72 (Interior River Lowland) of the contiguous United States (Omernik 1987). Vegetation is a mosaic of forest, woodland, pasture, and cropland.

The majority of effluents at PGDP consist of once-through cooling water, although a variety of effluents (uranium-contaminated as well as noncontaminated) result from activities associated with uranium precipitation and facility-cleaning operations. Conventional liquid discharges such as domestic sewage, steam-plant wastewaters, and coal-pile runoff also occur. Routine monitoring activities provide data to quantify total discharges to surface water in order to demonstrate compliance with federal, state, and DOE requirements. Monitoring also assists with evaluating the effectiveness of effluent treatment and control programs.

## **2.2 WATER QUALITY AND PGDP EFFLUENTS**

The Clean Water Act is currently administered for PGDP by the Kentucky Division of Water (KDOW) through the KPDES Wastewater Discharge Permitting Program. The current sitewide KPDES permit (No. KY0004049) became effective on November 1, 1992. PGDP adjudicated the portions of the permit that contained unattainable effluent limits and implemented the portions of the permit not under adjudication (Jones et al. 1997). On April 6, 1996, an Agreed Order between DOE, USEC, and the KDOW was signed that settled issues involving a challenge to the KPDES permit. The Agreed Order stays the limits for

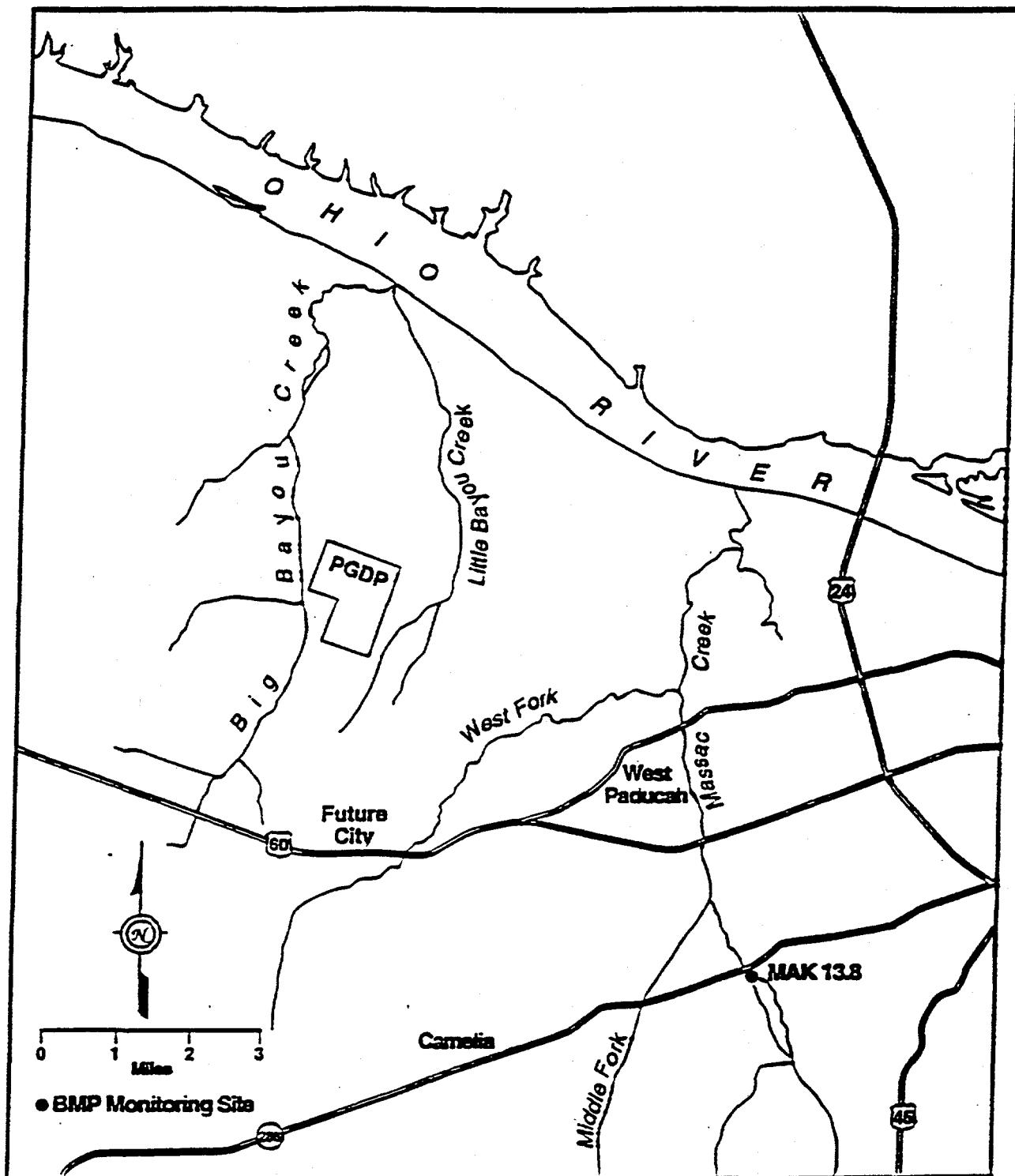
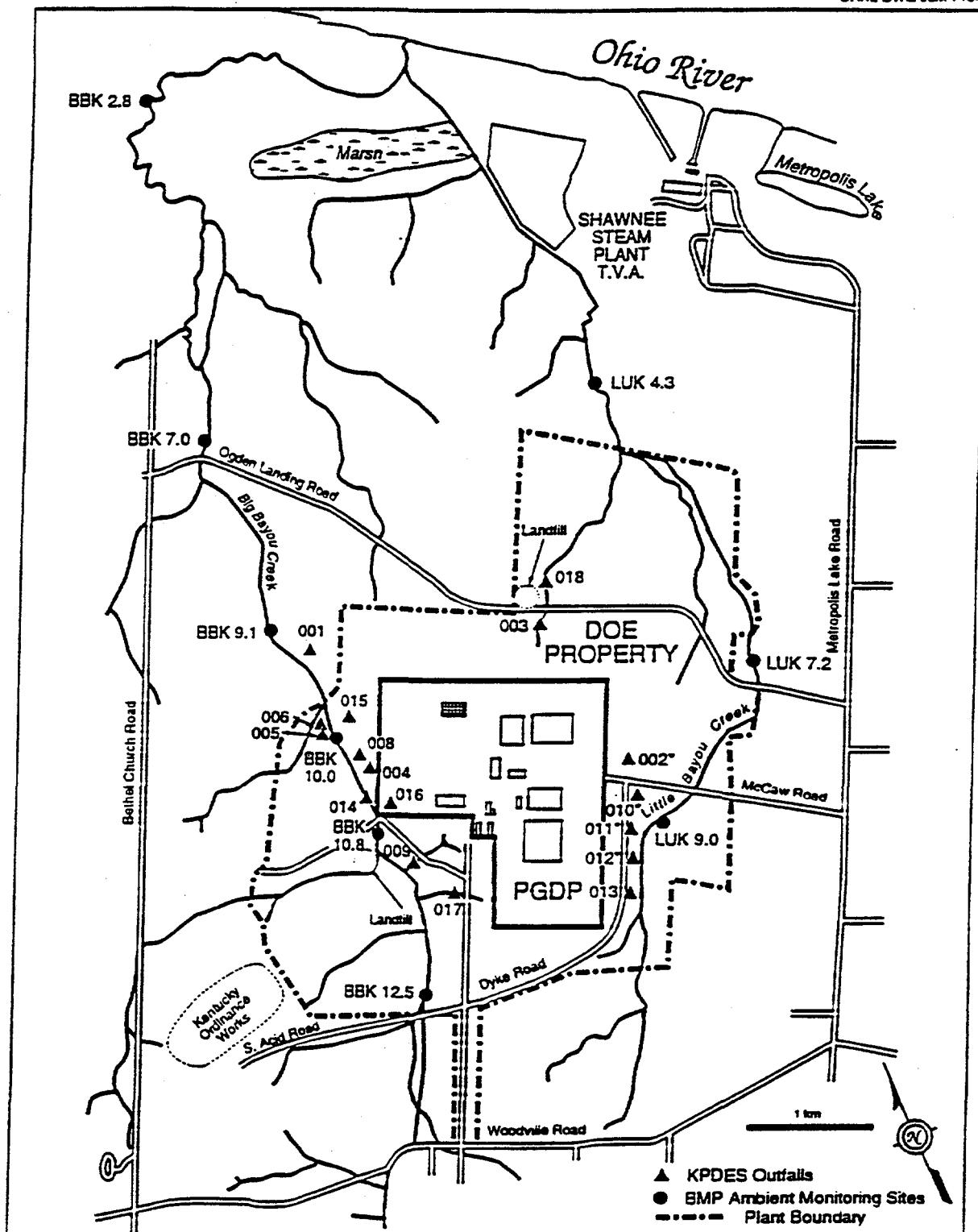


Fig. 2.1. Map of Paducah Gaseous Diffusion Plant (PGDP) in relation to the geographic region. The reference site for PGDP biological monitoring activities is located on Massac Creek at kilometer (MAK) 13.8.



\*Combined at C517 pond and discharged through 011/010

Fig. 2.2. Location of Biological Monitoring Program (BMP) sites and Kentucky Pollutant Discharge Elimination System (KPDES) permitted outfalls for the Paducah Gaseous Diffusion Plant (PGDP). BBK = Big Bayou Creek kilometer; LUK = Little Bayou Creek kilometer; T.V.A. = Tennessee Valley Authority; DOE = U.S. Department of Energy.

**Table 2.1 Summary of rainfall during 1997 at Barkey Regional Airport, Paducah, Kentucky**

| Month     | Total (cm) |
|-----------|------------|
| January   | 6.65       |
| February  | 13.51      |
| March     | 18.85      |
| April     | 12.78      |
| May       | 20.19      |
| June      | 16.74      |
| July      | 7.06       |
| August    | 7.26       |
| September | 6.12       |
| October   | 7.39       |
| November  | 5.33       |
| December  | 5.69       |
| Total     | 127.58     |

*Source:* Midwestern Climate Center, Champaign, IL, Station ID156110, Barkley Regional Airport, Paducah National Weather Service.

temperature, phosphorus, pH, cadmium, copper, chromium, lead, nickel, and zinc. In addition to the site wide KPDES permit, DOE also has a KPDES permit for a landfill outfall (K001).

Monitoring of individual outfalls and the landfill outfall is conducted in accordance with the KPDES Permit. Table 2.2 lists the outfalls in the site wide permit and their contributing processes; Fig. 2.2 shows the location of the outfalls. Eight of the 17 outfalls discharge continuously to the receiving streams. Outfalls 001, 006, 008, and 009 discharge continuously to Big Bayou Creek; outfalls 002, 010, 011, and 012 are combined at the C-617 pond and discharge through Outfall 010 continuously to Little Bayou Creek. After PCBs were detected in sediments from Outfall 011 in June 1994, the combined C-617 lagoon discharge was diverted on a full-time basis to Outfall 010. Outfall 011 has been a stormwater outfall since the change (C. C. Travis, USEC, Environmental Waste Management Division, Environmental Compliance Department, personal communication).

Table 2.2. Kentucky Pollutant Discharge Elimination System (KPDES) permitted outfalls at Paducah Gaseous Diffusion Plant

| Location <sup>a</sup> | Discharge source  | Flow <sup>b</sup> | Contributing processes   |
|-----------------------|---|-------------------|--|
| 001                   | C-616, C-600, C-400, C-410, C-635, C-335, C-337, C-535, C-537, C-746-A, C-747-A, C-635-6  | 8.8±1.7           | Recirculating cooling water blowdown treatment effluent, coal-pile runoff, once-through cooling water, surface runoff, roof and floor drains, treated uranium solutions, sink drains, discharge from the Northwest Plume Pump and Treat Facility |
| 002                   | C-360, C-637, C-337-A   | 1.6±3.9           | Once through cooling water, roof and floor drains, sink drains, extended aeration sewage treatment system  |
| 003                   | North edge of plant   | NM <sup>c</sup>   | Storm overflow of north/south diversion ditch discharges   |
| 004                   | C-615 sewage treatment plant, C-710, C-728, C-750, C-100, C-620, C-400  | 1.2±0.2           | Domestic sewage, laboratory sink drains, motor cleaning, garage drains, laundry, machine coolant treatment filtrate, condensate blowdown, once-through cooling water   |
| 005                   | C-611 primary sludge lagoon   | NM <sup>c</sup>   | Water treatment plant sludge, sand filter backwash, laboratory sink drains   |
| 006                   | C-611 secondary lagoon  | 3.3±0.8           | Water treatment plant sludge, sand filter backwash, laboratory sink drains from Outfall 005  |
| 007                   | Although outfall is still listed on the permit, the only discharge is storm water runoff, which has no monitoring requirements or limitations     | NM <sup>c</sup>   |  |
| 008                   | C-743, C-742, C-741, C-723, C-721, C-728, C-729, C-400, C-420, C-410, C-727, C-411, C-331, C-310, C-724, C-744, C-600, C-405, C-409, C-631, C-720 | 2.8±2.1           | Surface drainage, roof and floor drains, once-through cooling water, paint shop discharge, condensate, instrument shop cleaning area, metal-cleaning rinse water, sink drains  |
| 009                   | C-810, C-811, C-331, C-333, C-310, C-100, C-102, C-101, C-212, C-200, C-300, C-320, C-302, C-750, C-710, C-720                                    | 1.5±2.5           | Surface drainage, roof and floor drains, condensate, once-through cooling water, sink drains   |
| 010                   | C-531, C-331  | 2.3±0.7           | Switchyard runoff, roof and floor drains, condensate, sink drains  |
| 011                   | C-340, C-533, C-532, C-315, C-333, C-331  | 0.3±0.4           | Once-through cooling water, roof and floor drains, switchyard runoff, condensate, sink drains  |
| 012                   | C-633, C-533, C-333-A   | 4.1±12.1          | Roof, floor, and sink drains, condensate, surface runoff, extended aeration sewage treatment system  |
| 013                   | Southeast corner of the plant   | 3.4±7.0           | Surface runoff   |
| 014                   | C-611 U-shaped sludge lagoon  | NM <sup>c</sup>   | Sand filter backwash, sanitary water   |
| 015                   | West central plant areas  | 1.0±1.3           | Surface runoff   |
| 016                   | Southwest corner of the plant   | 0.2±0.3           | Surface runoff   |
| 017                   | Extreme south area of the plant   | 1.4±3.2           | Surface runoff   |
| 018                   | Landfill at north of plant  | 6.4± 10.8         | Surface runoff   |

<sup>a</sup>Numerical indicates outfall designation. Locations also identified in Fig. 2.2 of this report.

<sup>b</sup>Mean discharge in millions of liters per day ± 1 standard deviation. NA = not available. Mean value based on KPDES measurements for 1995.

<sup>c</sup>NM = Not monitored

*Note:* This table was taken from Kornegay et al. 1994 (Paducah Gaseous Diffusion Plant Environmental Report for 1993. ES/ESH-53. Oak Ridge National Laboratory, Oak Ridge, Tennessee)

### 2.3 DESCRIPTION OF STUDY SITES

Three study sites on Big Bayou Creek (Fig. 2.2), Big Bayou Creek kilometer (BBK) 12.5, BBK 10.0, and BBK 9.1; one site on Little Bayou Creek (Fig. 2.2), Little Bayou Creek kilometer (LUK) 7.2; and one off-site reference station on Massac Creek (Fig. 2.1), Massac Creek kilometer (MAK) 13.8, were routinely sampled to assess the ecological health of the stream. Prior to ORNL's initiation of the instream monitoring task for the PGDP BMP, a site selection study was conducted in 1990 (Kszos et al. 1994a). Qualitative sampling of the fish community at many of these sites was conducted in 1996 (Ryon 1997). Two additional sites (LUK 9.0, and LUK 4.3; Fig 2.2) were sampled as part of the bioaccumulation monitoring task; Massac Creek (MAK 13.8) served as a local source of uncontaminated fish in 1997. A more detailed description of the sampling locations for the bioaccumulation monitoring task is provided in Sect. 4. A summary of the site locations is given in Table 2.3. Biological monitoring activities conducted during 1997 are outlined in Table 2.4. Toxicity monitoring was conducted quarterly. Fish and benthic macroinvertebrates, community and bioaccumulation sampling were conducted twice annually (in the spring and fall). KPDES outfalls at which effluents were evaluated for toxicity during 1997 included 001, 006, 008, 009, 010, 013, 015, 016, 017, and 018.

**Table 2.3. Locations and names of sampling sites included in Paducah Gaseous Diffusion Plant Biological Monitoring Program 1997**

| Current site name <sup>a</sup> | Location <sup>b</sup>                                  |
|--------------------------------|--|
| <b>Big Bayou Creek</b>         |  |
| BBK 12.5 <sup>c</sup>          | ~200 m downstream of bridge on South Acid Road         |
| BBK 10.8                       | ~5 m upstream of Waterworks Road                       |
| BBK 10.0                       | ~50 m upstream of Outfall 006                          |
| BBK 9.1                        | ~25 m upstream of flume at gaging station at Bobo Road |
| <b>Little Bayou Creek</b>      |  |
| LUK 9.0                        | ~25 m downstream of Outfall 010                        |
| LUK 7.2                        | ~110 m downstream of bridge on Route 358               |
| LUK 4.3                        | ~500 m downstream of Outfall 018                       |
| <b>Massac Creek</b>            |  |
| MAK 13.8 <sup>c</sup>          | ~40 m upstream of bridge on Route 62, 10 km SE of PGDP |

<sup>a</sup>Site names are based on stream name and distance of the site from the mouth of the stream. For example, Big Bayou Creek Kilometer (BBK) 9.1 is located 9.1 km upstream of the mouth; LUK = Little Bayou Creek kilometer; and MAK = Massac Creek kilometer.

<sup>b</sup>Locations are based on approximate distances from a major landmark (e.g., bridge or outfall) to the bottom of the reach.

<sup>c</sup>Reference site.

**2-8 — Biological Monitoring Program**

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**Table 2.4. Sampling schedule for the four components of the Biological Monitoring Program at Paducah Gaseous Diffusion Plant, January–December 1997**

| Month | Toxicity monitoring | Bioaccumulation | Fishes | Benthic macroinvertebrates |
|-------|---------------------|-----------------|--------|----------------------------|
| Jan.  |                     |                 |        |                            |
| Feb.  |                     |                 |        |                            |
| Mar.  | X                   |                 | X      | X                          |
| Apr.  |                     |                 |        |                            |
| May   | X                   | X               |        |                            |
| Jun.  |                     |                 |        |                            |
| Jul.  |                     |                 |        |                            |
| Aug.  | X                   |                 |        |                            |
| Sept. | X <sup>a</sup>      |                 | X      | X                          |
| Oct.  |                     | X               |        |                            |
| Nov.  | X <sup>a</sup>      |                 |        |                            |
| Dec.  | X                   |                 |        |                            |

<sup>a</sup>Outfall 001 only.

### 3. TOXICITY MONITORING

*L. A. Kszos*

The toxicity monitoring task for BMP measures the toxicity of effluents as required by the KPDES permit. Until 1996, ambient water toxicity was monitored at four sites in Big Bayou Creek, one site in Little Bayou Creek, and one reference site in Massac Creek. The ambient monitoring was eliminated from BMP because there was no consistent evidence of chronic toxicity in water from the ambient locations, no correlation of reductions in fathead minnow survival or growth at the continuously flowing outfalls with reductions in fathead minnow survival or growth at ambient locations, and no significant change in the water chemistry of the ambient sites or outfalls (Kszos 1996b).

#### 3.1 INTRODUCTION

The ESD Toxicology Laboratory at ORNL began evaluating the toxicity of continuous and intermittent outfalls at PGDP in October 1991. As required by a draft Agreed Order, *Ceriodaphnia* and fathead minnow tests of the continuous and intermittent outfalls were conducted quarterly. In September 1992, a renewed KPDES permit was issued to PGDP. Under the requirements of this permit, *Ceriodaphnia* and fathead minnow tests were continued on a quarterly basis. As required, the test methods used are the Cladoceran (*C. dubia*) 3-brood, Survival and Reproduction Test (hereinafter referred to as the *Ceriodaphnia* test) and the Fathead Minnow (*Pimephales promelas*) 7-d, Larval Survival and Growth Test (hereinafter referred to as the fathead minnow test; Lewis et al. 1994). After May 1995, toxicity tests of continuously flowing outfalls 006, 008, 009, and 010 were conducted with fathead minnow larvae because they were shown to be the more sensitive species. Tests of continuously flowing Outfall 001 continued with *Ceriodaphnia* and fathead minnow larvae. After January 1996, tests of intermittently flowing outfalls 013, 015, 016, 017, and 018 were reduced to the more sensitive species (fathead minnow larvae).

#### 3.2 MATERIALS AND METHODS

Toxicity tests of effluents from the continuously flowing outfalls (001, 006, 008, 009, and 010) and the intermittently flowing outfalls (013, 015, 016, 017, and 018) were conducted according to the schedule shown in Table 3.1. This report summarizes the toxicity test results for all tests conducted during 1997. Toxicity test results from 1991 to 1996 are summarized in Kszos (1997).

Samples from the continuously flowing outfalls were collected by personnel from ESD and transported to a nearby offsite laboratory at the Paducah Community College. The tests were conducted using three, 24-h time-dependant composite samples collected over the 6 or 7 d test period. The intermittently flowing outfalls were rainfall dependent; thus, tests were conducted using one grab sample. Samples from the intermittently flowing outfalls were

Table 3.1. Summary of toxicity test dates for continuous and intermittent outfalls, 1997

| Outfall                      | Test date      | Species  |
|------------------------------|----------------|--|
| <i>Continuous outfalls</i>   |                |  |
| 001, 006, 009                | March 7-14     | Fathead minnow (all)<br><i>Ceriodaphnia</i> (001 only) |
| 008, 010                     | March 11-18    | Fathead minnow   |
| 001, 006, 008, 009, 010      | May 14-21      | Fathead minnow (all)<br><i>Ceriodaphnia</i> (001 only) |
| 001, 006, 008, 009, 010      | August 13-20   | Fathead minnow (all)<br><i>Ceriodaphnia</i> (001 only) |
| 001                          | September 4-11 | Fathead minnow   |
| 001                          | November 6-13  | <i>Ceriodaphnia</i>                                    |
| 001, 006, 008, 009, 010      | December 3-10  | Fathead minnow   |
| <i>Intermittent outfalls</i> |                |  |
| 013, 015, 016, 017, 018      | January 7-14   | Fathead minnow   |
| 013, 015, 016, 017, 018      | April 8-15     | Fathead minnow   |
| 013, 015, 016, 017, 018      | July 10-17     | Fathead minnow   |
| 013, 015, 016, 017, 018      | December 2-9   | Fathead minnow   |

collected by personnel from PGDP, refrigerated, and shipped to ESD using 24-h delivery. Tests were initiated the same day the samples were received. All samples were collected and delivered using established chain-of-custody procedures (Kszos et al. 1996c). Time of collection, water temperature, and arrival time in the laboratory were recorded.

The effluents were evaluated for toxicity using the Ceriodaphnia test (EPA method 1002.0) and the fathead minnow test (EPA method 1000.0) (Lewis et al. 1994). The *Ceriodaphnia* and fathead minnow tests were static-renewal tests, meaning that test water was replaced daily for 6 or 7 consecutive days. The fathead minnow test consisted of four replicates per test concentration with ten animals per replicate. Each day before the water was replaced, the number of surviving larvae was recorded. At the end of 7 d, the larvae were dried and weighed to obtain an estimate of growth. The *Ceriodaphnia* test consisted of ten replicates per test concentration with one animal per replicate. Each day the animals were transferred from a beaker containing old test solution and placed in a beaker containing fresh test solution. At this time, survival and the number of offspring produced were recorded. A control consisting of dilute mineral water augmented with trace metals was included with each test. On each fresh sample, subsamples of each effluent were routinely analyzed for pH, conductivity, alkalinity, and water hardness (Kszos et al. 1996c). The concentration of total suspended solids (TSS) was measured on each sample from the intermittent outfalls using a standard method (APHA 1989).

A linear interpolation method (Norberg-King 1993) was used to determine the 25% inhibition concentration (IC25, that concentration causing a 25% reduction in fathead minnow growth or *Ceriodaphnia* reproduction compared to a control). A computer program [A Linear Interpolation Method for Sublethal Toxicity: Inhibition Concentration (ICp) Approach, version 2.0] distributed by the EPA (Environmental Research Laboratory, Duluth, Minnesota) was used for the calculation. The chronic toxicity unit (TUC = 100/IC25) is required as a compliance endpoint in the renewed permit (September 1992 to present). The higher the TUC, the more toxic an effluent. Because Little Bayou and Big Bayou creeks have been determined to have a low flow of zero, a TUC > 1.0 for the continuously flowing effluents would be considered a noncompliance and an indicator of potential instream toxicity.

### 3.3 RESULTS AND DISCUSSION

A summary of the TUCs for all toxicity tests of effluent from continuously flowing outfalls 001, 006, 008, 009, and 010 conducted during 1997 are provided in Table 3.2. Mean survival and growth of fathead minnows and survival and mean reproduction of *Ceriodaphnia* for each outfall and test during 1997 are provided in Appendix C. During 1997, effluent from Outfall 001 exceeded the permit limit (TUC  $\geq$  1.0) in August with a TUC of 8.34. This is the first occurrence of a fathead minnow test with a TUC  $\geq$  1.0 for Outfall 001 since testing began in October 1991 (Kszos 1996a,b). The confirmatory test conducted in September resulted in a TUC < 1.0, demonstrating that the effluent was no longer toxic. The TUCs for outfalls 006, 008, 009, and 010 were less than 1.0 for all tests conducted in 1997.

A summary of the TUCs for all toxicity tests of effluent from intermittently flowing outfalls 013, 015, 017, and 018 conducted during 1997 is provided in Table 3.2. Mean survival and growth of fathead minnows for each outfall and test during 1997 are provided in Appendix C. Although PGDP does not have a compliance limit for the intermittent outfalls, TUC  $\geq$  1.0 was used as a benchmark. During 1997, the only cases of TUC  $\geq$  1.0 were for the fathead minnow tests of Outfall 015 in July and Outfall 016 in April. The TUC for Outfall 015 in July was 2.74. This is the first case of a TUC  $\geq$  1.0 for Outfall 15 since November 1994. The subsequent test of Outfall 015 in December resulted in TUC < 1.0. The TUC for Outfall 016 in April was 19.61. Similar to Outfall 015, this is the first case of a TUC  $\geq$  1.0 for Outfall 15 since November 1994. The subsequent tests of Outfall 016 in July and December resulted in TUCs < 1.0.

The concentration of TSS in the intermittent effluent samples and the flow rate of the effluents are given in Table 3.3. TSS and flow rate were quite variable between tests as would be expected due to the fact that flow is rainfall-dependant. The TUC of 19.61 for Outfall 016 occurred during the test period that also had the highest TSS, suggesting that for this outfall, toxicity may be related to TSS or a contaminant related to TSS. The TUC of 2.74 for Outfall 015 did not correspond to the highest TSS for this outfall but did correspond to the highest flow rate.

Table 3.2. Summary of toxicity test results for continuous and intermittent outfalls, 1997

| Outfall                      | Test date | Chronic toxicity units (TUc) <sup>a</sup> |                     |
|------------------------------|-----------|---|---------------------|
|                              |           | Fathead minnow                            | <i>Ceriodaphnia</i> |
| <i>Continuous outfalls</i>   |           |   |                     |
| 001                          | March     | <1  | <1                  |
|                              | May       | <1  | <1                  |
|                              | August    | 8.34                                      | <1                  |
|                              | September | <1  | NT <sup>b</sup>     |
|                              | November  | NT  | <1                  |
|                              | December  | <1  | NT                  |
| 006                          | March     | <1  | NA <sup>c</sup>     |
|                              | May       | <1  | NA                  |
|                              | August    | <1  | NA                  |
|                              | December  | <1  | NA                  |
| 008                          | March     | <1  | NA                  |
|                              | May       | <1  | NA                  |
|                              | August    | <1  | NA                  |
|                              | December  | <1  | NA                  |
| 009                          | March     | <1  | NA                  |
|                              | May       | <1  | NA                  |
|                              | August    | <1  | NA                  |
|                              | December  | <1  | NA                  |
| 010                          | March     | <1  | NA                  |
|                              | May       | <1  | NA                  |
|                              | August    | <1  | NA                  |
|                              | December  | <1  | NA                  |
| <i>Intermittent Outfalls</i> |           |   |                     |
| 013                          | January   | <1  | NA                  |
|                              | April     | <1  | NA                  |

Table 3.2 (continued)

| Outfall | Test date | Chronic toxicity units (TUC) <sup>a</sup> |                     |
|---------|-----------|---|---------------------|
|         |           | Fathead minnow                            | <i>Ceriodaphnia</i> |
| 015     | July      | <1  | NA                  |
|         | December  | <1  | NA                  |
|         | January   | <1  | NA                  |
|         | April     | <1  | NA                  |
|         | July      | 2.74                                      | NA                  |
|         | December  | <1  | NA                  |
| 016     | January   | <1  | NA                  |
|         | April     | 19.61                                     | NA                  |
|         | July      | <1  | NA                  |
|         | December  | <1  | NA                  |
| 017     | January   | <1  | NA                  |
|         | April     | <1  | NA                  |
|         | July      | <1  | NA                  |
|         | December  | <1  | NA                  |
| 018     | January   | <1  | NA                  |
|         | April     | <1  | NA                  |
|         | July      | <1  | NA                  |
|         | December  | <1  | NA                  |
|         | January   | <1  | NA                  |

<sup>a</sup>Chronic toxicity unit = 100/IC25; IC25 = the concentration causing a 25% reduction in fathead minnow growth or *Ceriodaphnia* reproduction. IC = inhibition concentration.

<sup>b</sup>NT = not tested.

<sup>c</sup>NA = not applicable; test not required.

### 3.4 BIOAVAILABILITY STUDY

Water quality criteria (WQC) established by the U.S. Environmental Protection Agency (EPA) to protect aquatic life are estimates of the highest concentration of a pollutant that can be present while still adequately protecting species in an aquatic community. National ambient WQC were derived from laboratory toxicity tests. However, the bioavailability and/or toxicity of most metals is strongly affected by factors such as the types and concentrations of dissolved and particulate organic matter in the water, pH, alkalinity, hardness, temperature, and metal-binding dissolved constituents, such as sulfide. These factors are not routinely incorporated

**Table 3.3. Concentration of total suspended solids and flow rate for the intermittent outfalls, 1997**

| Outfall | Date     | Total suspended solids<br>(mg/L) | Flow<br>(L/sec) |
|---------|----------|----------------------------------|-----------------|
| 013     | January  | 79                               | 3.81            |
|         | April    | 11                               | 2.67            |
|         | July     | 37                               | 9.29            |
|         | December | 2                                | 3.50            |
| 015     | January  | 50                               | 0.39            |
|         | April    | 12                               | 0.53            |
|         | July     | 21                               | 0.66            |
|         | December | 20                               | 0.31            |
| 016     | January  | 8                                | 0.04            |
|         | April    | 33                               | 0.04            |
|         | July     | 5                                | 0.22            |
|         | December | 8                                | 0.09            |
| 017     | January  | 3                                | 0.31            |
|         | April    | 16                               | 0.74            |
|         | July     | 29                               | 8.28            |
|         | December | 4                                | 0.92            |
| 018     | January  | 30                               | 0.57            |
|         | April    | 37                               | 2.40            |
|         | July     | 13                               | 7.80            |
|         | December | 32                               | 0.74            |

into present ambient WQC; therefore, the WQC may be underprotective or overprotective of aquatic biota.

In May 1996, the KDOW issued *Procedures to Facilitate Alternative Metal Limits* (KDOW 1996). The procedure requires demonstration, through chemical-specific analyses and toxicity testing, that an effluent is not toxic due to the presence of the metal in question. By means of the KDOW method, the amount of total recoverable metal measured in the effluent is adjusted by the dissolved metal:total recoverable metal (DM:TRM) ratio. A DM:TRM ratio is derived for each metal and at each outfall. The result is the calculated total recoverable metal (TRM) concentration, which can be reported in lieu of the measured TRM concentration for a particular metal. This calculated concentration may then be reported for determining compliance with the TRM permit limits. Based on the Agreed Order signed April 5, 1996,

DOE and USEC may use the KDOW method to attempt to develop limits for cadmium, copper, lead, nickel, zinc and chromium that are alternatives to the metal limits proposed in the KPDES permit. Using the method developed by the KDOW, biomonitoring results and chemical data will be used to meet the objectives of the study:

- evaluate the toxicity of continuous outfalls (001, 008, 009, and 010) and intermittent outfalls (003, 013, 015, 016, 017, and 018) at PGDP;
- determine the mean ratio of dissolved to TR metal for Cd, Cr, Cu, Pb, Ni, and Zn in the continuous and intermittent outfalls;
- determine whether the concentration of TR metal discharged causes toxicity to fathead minnows and/or *Ceriodaphnia*; and
- determine alternative metal limits for each metal of concern (Cd, Cr, Cu, Pb, Ni, and Zn).

Sampling and analysis for this study began in December 1996 and are fully described in Phipps and Kszos (1996). Two phases of the study are planned. Phase I developed alternative metal limits for continuously discharging outfalls. A report detailing the results of Phase I has been submitted to the KDOW for comment. Phase II will develop alternative metal limits for intermittently discharging outfalls. If prior to implementation of the schedules identified in the study plan (Phipps and Kszos 1996), KDOW issues to PGDP a new KPDES permit that includes metals limits, and such limits are not challenged by PGDP, then all activities scheduled to be completed in Phase II will be canceled and PGDP will meet the limits established in the new KPDES permit.

## 4. BIOACCUMULATION

*M. J. Peterson and G. R. Southworth*

### 4.1 INTRODUCTION

Bioaccumulation monitoring conducted as part of the BMP at PGDP has identified PCB contamination in fish in Big Bayou Creek and Little Bayou Creek as a major concern (Kszos 1996a,b, 1997). Mercury concentrations in fish from Big Bayou Creek were also found to be higher in fish collected downstream from PGDP discharges than in fish from an upstream site (Kszos 1996a,b, 1997). Concentrations of various other metals and organics in filets of fish from Big Bayou Creek and Little Bayou Creek were well below levels of concern for human consumption.

The primary objective of the 1996-97 bioaccumulation monitoring was to evaluate spatial and temporal changes in PCB contamination in longear sunfish (*Lepomis megalotis*) from Little Bayou Creek. PCB contamination in sunfish in Big Bayou Creek had declined to near background levels over the 1992-95 period, and monitoring in this stream was consequently reduced to a single site immediately downstream from the lowermost PGDP discharge to Big Bayou Creek. Similarly, mercury monitoring was conducted only at that site in Big Bayou Creek. Because Big Bayou Creek is capable of supporting a limited sport fishery for larger game fish, spotted bass (*Micropterus punctulatus*) were analyzed for mercury and PCBs to evaluate the maximum concentrations likely in fish near the PGDP.

Whole-body samples of small sunfish and minnows were collected in May 1997 while conducting the routine sunfish filet monitoring. The primary objective of this effort was to provide whole-body fish data that could be used in an initial assessment of the potential risks to terrestrial piscivores (e.g., kingfishers, mink) that may eat contaminated fish from waters near the PGDP. The focus of the evaluation was on the contaminants of most concern (i.e., PCBs and metals) in Little Bayou Creek and Big Bayou Creek. A brief overview of the forage fish sampling results is provided herein; for more detailed information, see Peterson (1997) and Sample (1997).

### 4.2 MATERIALS AND METHODS

Because sunfish are short-lived and have small home ranges, they represent recent contaminant exposure at the site of collection and are thus ideal monitoring tools for evaluating spatial and temporal trends in contamination. Collections of sunfish were restricted whenever possible to fish of a size large enough to be taken by sport fisherman in order to minimize effects of covariance between size and contaminant concentrations and to provide data directly applicable to assessing risks to people who might eat fish from these creeks. In general, high fish densities enabled the collection of 6 to 8 specimens of sunfish > 30 g at all sites except the upper Little Bayou Creek sites.

All fish were collected by backpack electrofishing. Longear sunfish were collected at PGDP sites on October 24, 1996, May 7-8, 1997, and October 28, 1997, as part of routine twice yearly monitoring of PCB concentrations in this species. Longear sunfish were collected for PCB analysis at three sites on Little Bayou Creek, LUK 9.0, LUK 7.2 and LUK 4.3 (Fig. 2.2). Spotted bass were collected from BBK 9.1 in the fall sampling periods for mercury analysis, and in fall 1996 for PCB analysis. Hinds Creek in Anderson County,

Tennessee, served as a source of uncontaminated reference fish for the 1996 collection, and Massac Creek (at MAK 13.8) served as a local source of uncontaminated fish for the 1997 sampling. Forage fish (central stoneroller, *Campostoma anomalum* and small longear sunfish) were collected from sites in Little Bayou Creek and Big Bayou Creek during the May 1997 sampling. Small longear sunfish were collected from four sites in the Little Bayou Creek drainage (Outfall 010 ditch, lower half; LUK 9.0; LUK 7.2; and LUK 4.3), one site on Big Bayou Creek (BBK 9.1), and one offsite reference station on Massac Creek (MAK 13.8). Stonerollers were collected only at sites where they were common (LUK 7.2, BBK 9.1, and MAK 13.8).

For filet analysis, each fish was individually tagged with a unique four-digit tag wired to the lower jaw and placed on ice in a labeled ice chest. Fish were held on ice overnight and processed within 48 hours. Each fish was weighed and measured, then filleted, scaled, and rinsed in process tap water. Samples of sunfish for specific analyses were excised, wrapped in heavy duty aluminum foil, labeled, and frozen in a standard freezer at 15°C. For larger fish (bass), filets were wrapped and labeled as were sunfish samples, but at a later date the frozen filets were partially thawed, cut into 2- to 4-cm pieces, and homogenized in a stainless steel blender. A 25-g sample of the ground tissue was wrapped in heavy duty aluminum foil, labeled, frozen, and submitted to LMES Analytical Chemistry Organization for PCB and mercury analyses. Any remaining tissue from filets of sunfish or larger fish was wrapped in foil, labeled, and placed in the freezer for short-term archival storage. Forage fish were grouped into three subgroups each containing ten fish. Individuals in each subgroup were weighed and measured, and the sample of ten fish was then homogenized in a stainless steel blender, packaged in aluminum foil, and frozen for delivery to the analytical laboratory.

PCB analyses were conducted using Soxhlet extraction techniques according to SW-846 Method 3540 and analysis by capillary column gas chromatography using SW-846 Method 8080 (EPA 1986). Fish were analyzed for total mercury by cold vapor atomic absorption spectrophotometry following digestion in  $\text{HNO}_3/\text{H}_2\text{SO}_4$  (EPA 1991, procedure 245.6). Metals were analyzed using inductively coupled plasma mass spectrometry (ICP/MS) according to EPA procedure 200.8 (EPA 1991).

Quality assurance was evaluated by a combination of blind duplicate analyses, analysis of biological reference standards and uncontaminated fish, and determination of recoveries of analyte spikes to uncontaminated fish. SAS software and procedures were used to calculate the mean, standard error, and standard deviation of mercury and PCB concentrations in fish at each site (SAS 1985 a,b). Samples were processed according to project-specific standardized technical procedures developed for the Biological Monitoring and Abatement Program to ensure quality and integrity (QAP-X-90-ES-065, Rev. 1: Biological Monitoring and Abatement Program Quality Assurance Plan, Bioaccumulation Monitoring Aquatic).

## **4.3 RESULTS AND DISCUSSION**

### **4.3.1 PCBs**

PCBs were detected in sunfish from all sites in Little Bayou Creek on all sampling dates (Table 4.1). PCBs were not detected in sunfish from reference sites in Tennessee (Hinds Creek) or Kentucky (Massac Creek). Average concentrations were well below the FDA limit (2  $\mu\text{g/g}$  wet wt) at all sites, and no individual fish exceeded that guideline.

**Table 4.1. Mean concentration of PCBs ( $\mu\text{g/g}$ , wet weight) in filets of fish from streams near the Paducah Gaseous Diffusion Plant October 1996, May 1997, and October 1997**

| Site <sup>a</sup>            | Species           | Mean <sup>b</sup> | SE    | Range      | n |
|------------------------------|-------------------|-------------------|-------|------------|---|
| <b>October 1996</b>          |                   |                   |       |            |   |
| BBK 9.1                      | Spotted bass      | 0.45              | 0.07  | 0.30-0.58  | 4 |
| LUK 9.0                      | Longear sunfish   | 0.64              | 0.13  | 0.35-1.19  | 6 |
| LUK 7.2                      | Longear sunfish   | 0.72              | 0.07  | 0.48-0.93  | 6 |
| LUK 4.3                      | Longear sunfish   | 0.13              | 0.06  | <0.01-0.32 | 5 |
| Reference<br>(Hinds Cr, TN)  | Redbreast sunfish | <0.01             |       |            | 4 |
| <b>May 1997</b>              |                   |                   |       |            |   |
| LUK 9.0                      | Longear sunfish   | 0.62              | 0.04  | 0.47-0.78  | 6 |
| LUK 7.2                      | Longear sunfish   | 0.48              | 0.12  | 0.22-0.85  | 6 |
| LUK 4.3                      | Longear sunfish   | 0.12              | 0.04  | <0.01-0.27 | 5 |
| Reference<br>(Massac Cr, KY) | Longear sunfish   | <0.01             |       |            | 4 |
| <b>October 1997</b>          |                   |                   |       |            |   |
| BBK 9.1                      | Spotted bass      | 0.07              | <0.01 | 0.06-0.07  | 4 |
| LUK 9.0                      | Longear sunfish   | 0.37              | 0.10  | 0.13-0.66  | 6 |
| LUK 7.2                      | Longear sunfish   | 0.48              | 0.15  | 0.12-1.11  | 6 |
| LUK 4.3                      | Longear sunfish   | 0.06              | 0.01  | <0.01-0.12 | 6 |
| Reference<br>(Massac Cr, KY) | Longear sunfish   | <0.01             |       |            | 4 |

<sup>a</sup>BBK = Big Bayou Creek kilometer; LUK = Little Bayou Creek kilometer.

<sup>b</sup>Value of  $\frac{1}{2}$  the detection limit was used in calculating means for samples

#### 4.3.1.1 Spatial trends

**Fall 1996.** Results of PCB analyses of sunfish collected from Little Bayou Creek from October 1996 to October 1997 are presented in Table 4.1 and Appendix D. PCB concentrations in sunfish collected October 1996 in Little Bayou Creek continued to exhibit a pronounced decrease with distance downstream from PGDP (Fig. 4.1). Concentrations were highest at LUK 9.0 and LUK 7.2 and decreased dramatically at LUK 4.3. Unlike the case in previous monitoring, the highest average concentration was not found at LUK 9.0, but rather

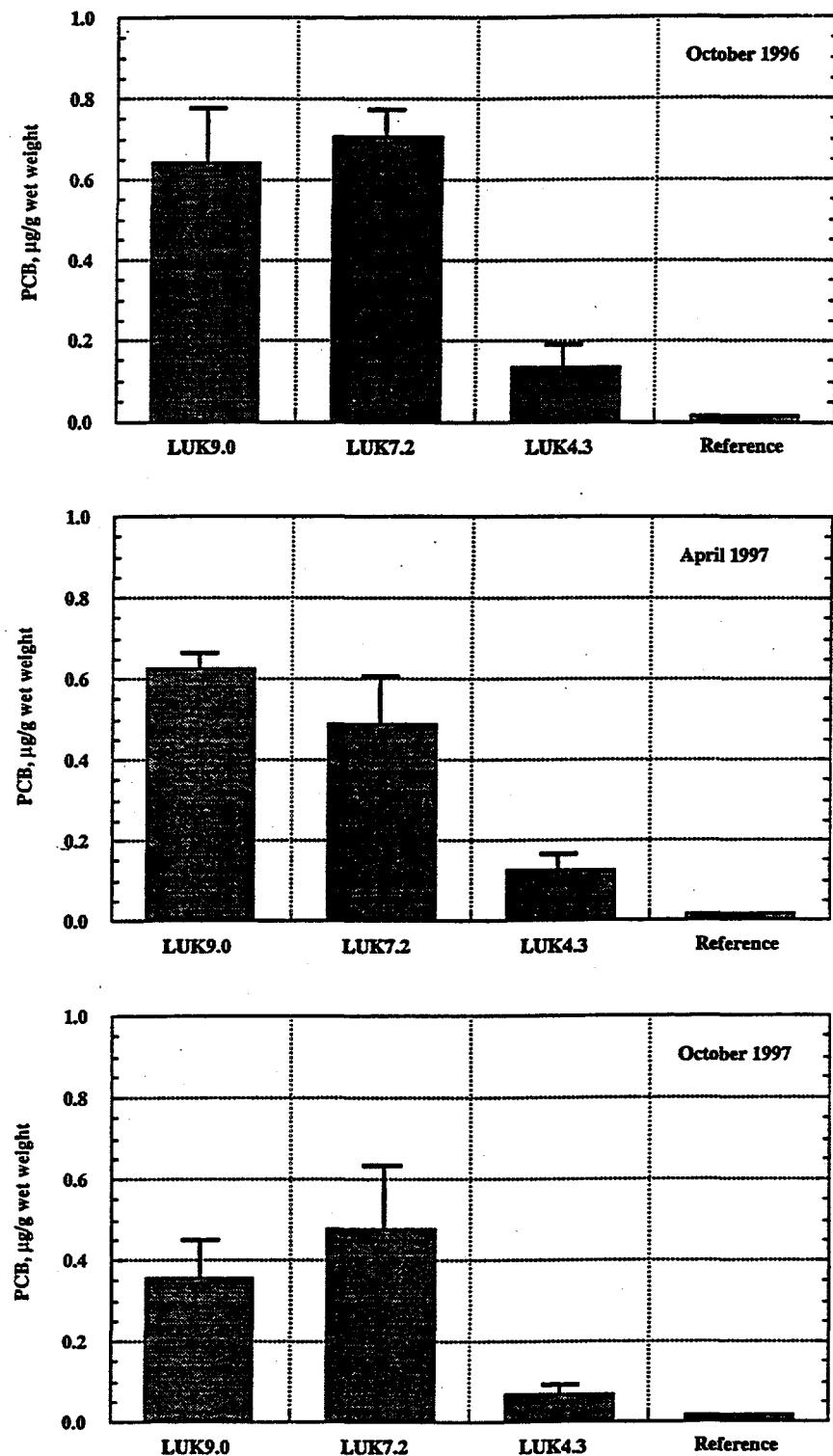


Fig. 4.1. Downstream profiles of mean PCB concentrations in longear sunfish (*Lepomis megalotis*) collected in Little Bayou Creek in October 1996, April 1997, and October 1997. Reference site locations are listed in Table 4.1.

at LUK 7.2. PCB concentrations at the two sites were similar, averaging 0.6 to 0.7  $\mu\text{g/g}$  (Table 4.1). This deviation from the typical downstream pattern would be expected if PCB inputs from the headwater site were reduced. At LUK 4.3, the mean PCB concentration in sunfish was  $0.13 \pm 0.06 \mu\text{g/g}$ . Composition of the PCB mixtures found in sunfish resembled Aroclor 1254 and 1260 at all sites.

In Big Bayou Creek, spotted bass contained an average PCB concentration of  $0.45 \pm 0.07 \mu\text{g/g}$  (range 0.30–0.58  $\mu\text{g/g}$ ). Bass from the same site averaged 0.16  $\mu\text{g/g}$  in October 1995. Although levels of PCBs in fish at this site remain well below that typical of upper Little Bayou Creek, the presence of mean concentrations of nearly 0.5  $\mu\text{g/g}$  indicates that PCB inputs to this creek are continuing. Only the highly chlorinated materials similar to Aroclor 1254/1260 were present.

**Spring 1997.** Mean PCB concentrations in sunfish from Little Bayou Creek exhibited the decreasing downstream pattern typical of all previous sampling except fall 1996 (Fig. 4.1). Highest concentrations occurred at LUK 9.0, lowest at LUK 4.3. Traces of lower chlorinated PCB congeners typical of Aroclor 1248 were found in fish from the upper two sites. The presence of these less persistent congeners in fish may be an indicator of fresh inputs of dissolved PCBs to these upper sites.

**Fall 1997.** In October 1997, the downstream pattern of mean PCB concentrations in sunfish again resembled that of fall 1996, with the concentration at LUK 7.2 exceeding that at LUK 9.0 (Fig. 4.1). Overall, concentrations were lower in fall 1997 than fall 1996. Materials typical of Aroclor 1248 were not detected, suggesting exposure to more highly weathered PCBs than was the case in the preceding spring. The pattern exhibited over the course of October 1996 through October 1997 is that expected if PCB inputs to Little Bayou Creek headwaters were reduced over the summer, and increased during the winter. If groundwater seepage into the storm drain system is an important mechanism for introducing PCBs to the surface flow, such a seasonal pattern would be expected.

PCBs in spotted bass were much lower in fall 1997 than in fall 1996, averaging only 0.07  $\mu\text{g/L}$  (Table 4.1). This may be due in part to the small size of individual fish in the fall 1997 collection, but it also indicates that PCB inputs to Big Bayou Creek are probably highly episodic.

#### 4.3.1.2 Temporal trends

The long-term pattern of mean PCB concentrations in fish at the uppermost site in Little Bayou Creek gives evidence of continued but decreased inputs of PCBs to the creek headwaters (Fig. 4.2). Considerable improvement is evident in PCB contamination in Little Bayou Creek, where average concentrations in sunfish at LUK 9.0 have decreased from nearly 2  $\mu\text{g/g}$  in spring 1992 to less than 0.4  $\mu\text{g/g}$  in fall 1997. As headwater inputs decrease, the relative importance of in-stream contamination as a source of contamination to fish increases. In the absence or reduction of continued upstream inputs, contaminated sediments should be gradually washed out and buried, and the downstream profile in which PCB concentrations at LUK 7.2 (and eventually LUK 4.3) exceed those at LUK 9.0 should become more frequent or typical. Thus, spatial patterns discussed in Section 4.3.1.1 provide additional evidence of success in reducing point source inputs over the past 5 years.

#### 4.3.2 Mercury

The bioaccumulation of mercury by fish is predominantly a food chain mediated process, thus predatory species that occupy trophic positions at or near the top of the aquatic food web would be expected to contain higher concentrations of mercury than species lower in

the food chain. Spotted bass in Big Bayou Creek occupy that role of terminal predator and are monitored by this task to evaluate the maximum mercury level likely in fish from that creek. The mean mercury concentration in spotted bass collected in October 1996 was  $0.52 \pm 0.11 \mu\text{g/g}$ , with a range of  $0.33 - 0.73 \mu\text{g/g}$ . In October 1997, the collection averaged  $0.26 \pm 0.05 \mu\text{g/g}$ , with a range of  $0.13 - 0.35 \mu\text{g/g}$ . Spotted bass appeared to be getting less abundant in this reach of Big Bayou Creek, and collection of larger individuals was difficult. As a consequence, fish collected in October 1997 were smaller than those taken previously. Mercury concentrations in predatory fish such as bass and walleye typically increase as a function of fish size, thus collections in which individuals are smaller would be expected to have lower average mercury concentrations. Mean mercury concentrations adjusted for the variation in mercury concentration with fish weight are plotted in Fig. 4.3. A slight decreasing trend is evident. The few larger bass collected from Big Bayou Creek continue to have mercury concentrations that approach common human health threshold limits.

Aqueous total mercury and methylmercury in Big Bayou Creek upstream and downstream from PGDP were measured in summer 1997 by researchers at ORNL and Frontier

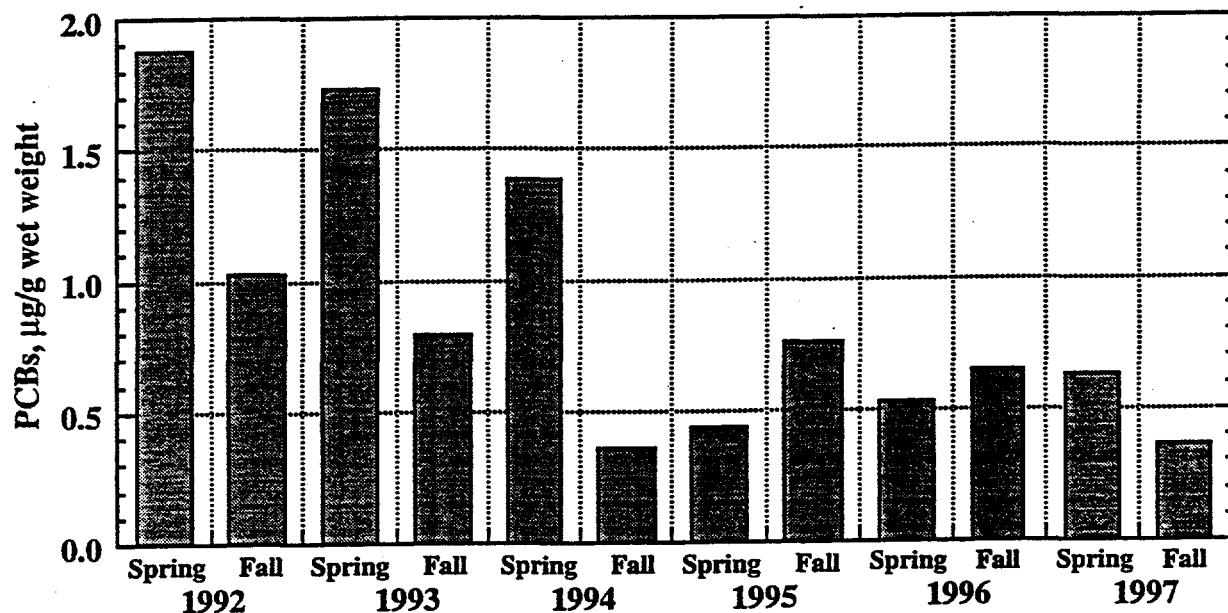


Fig. 4.2. Temporal changes in average PCB concentrations in longear sunfish (*Lepomis megalotis*) from Little Bayou Creek (LUK 9.0) near the Paducah Gaseous Diffusion Plant.

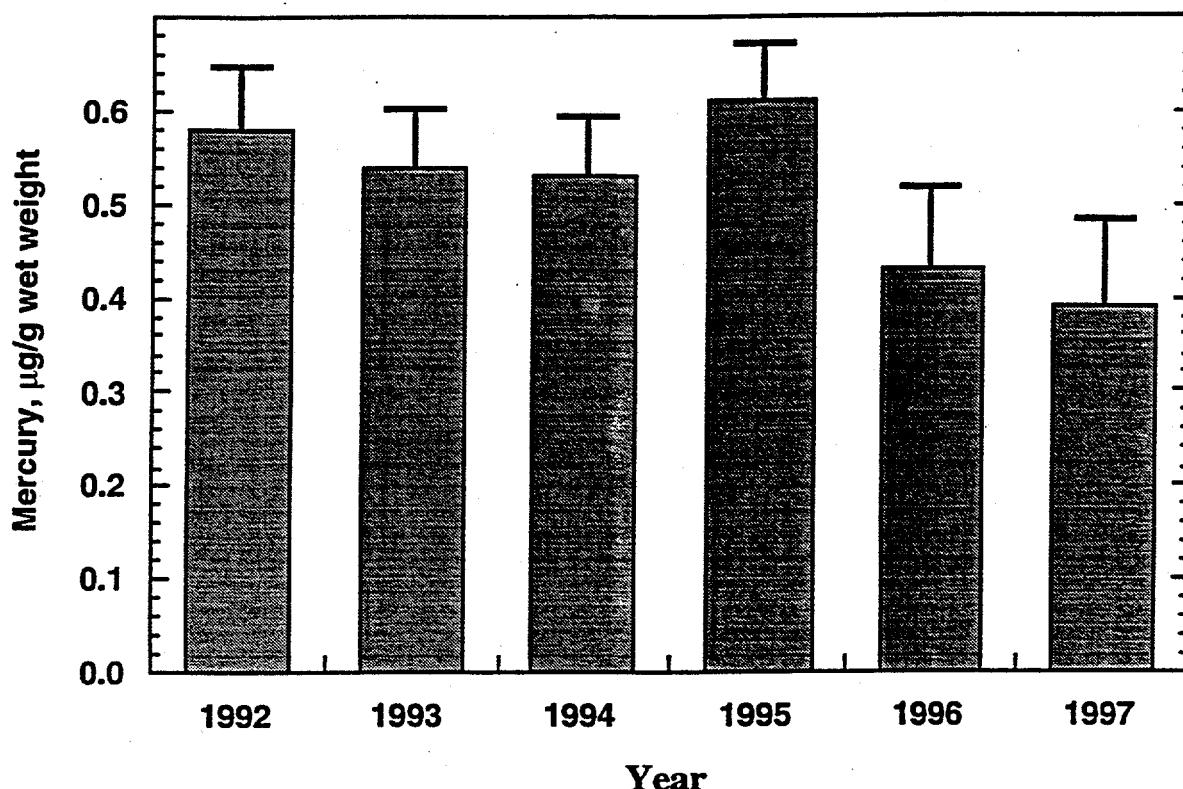


Fig. 4.3. Adjusted mean concentrations of mercury ( $\pm$  SE) in spotted bass (*Micropterus punctulatus*) from Big Bayou Creek (BBK 9.1) downstream from the Paducah Gaseous Diffusion Plant, 1992–97. Values were adjusted for the variation in mercury concentration with fish weight using analysis of covariance.

Geosciences in Seattle, Washington, as part of a study funded by the Lockheed Martin Energy Systems Y-12 Plant to investigate the relationship between waterborne mercury concentrations and mercury bioaccumulation in fish. The baseflow concentration of total mercury was 5.3 ng/L at BBK 9.1 downstream from PGDP and 1.2 ng/L at BBK 12.5, upstream from PGDP. Methylmercury concentrations were 0.12 and 0.10 ng/L at the two sites, respectively (G.R. Southworth, ORNL, unpublished data). Typical reference stream mercury concentrations in this study were 2.0–5.0 ng/L total mercury and 0.04–0.08 ng/L methylmercury for five sites in East Tennessee (Hinds Creek, Brushy Fork, Beaverdam Creek, and the Clinch River). Thus, total mercury in Big Bayou Creek falls within the range typical of uncontaminated streams in East Tennessee and elsewhere in the United States, and well below the EPA water quality criterion (12 ng/L). The accumulation of mercury in fish in this system appears to be greatly affected by the unusually high bioavailability of very low concentrations of mercury in water.

#### 4.3.3 PCBs and Metals in Forage Fish

The mean concentrations of total PCBs and a suite of metals in whole-body samples of longear sunfish and stonerollers are reported in Tables 4.2 and 4.3. PCB concentrations

**Table 4.2. Mean ( $\pm$  SE) concentrations ( $\mu\text{g/g}$ , wet weight) of various analytes in composited longear sunfish (*Lepomis auritus*) samples collected from stream sites near the PGDP and reference streams, May 1997**

| Analytes    | Little Bayou Creek sites <sup>a</sup> |                 |                              |                 | Big Bayou Creek site <sup>a</sup> |                              | Massac Creek <sup>a</sup><br>(Reference site) |
|-------------|---------------------------------------|-----------------|------------------------------|-----------------|-----------------------------------|------------------------------|---|
|             | Outfall 010                           | LUK 9.0         | LUK 7.2                      | LUK 4.3         | BBK 9.1                           | MAK 13.8                     |   |
| PCBs, total | 2.12 $\pm$ 0.13                       | 3.59 $\pm$ 0.10 | 1.74 $\pm$ 0.08              | 1.23 $\pm$ 0.06 | 0.52 $\pm$ 0.04                   |                              | < 0.003                                       |
| Antimony    | --                                    | --              | < 0.02                       | --              | < 0.02                            |                              | < 0.02  |
| Arsenic     | --                                    | --              | < 0.22                       | --              | < 0.19                            |                              | < 0.20  |
| Beryllium   | --                                    | --              | < 0.02                       | --              | < 0.02                            |                              | < 0.02  |
| Cadmium     | --                                    | --              | 0.03 $\pm$ 0.01 <sup>b</sup> | --              | 0.02 $\pm$ 0.0 <sup>c</sup>       | 0.03 $\pm$ 0.00              |   |
| Chromium    | --                                    | --              | 0.30 $\pm$ 0.01              | --              | 0.55 $\pm$ 0.09                   | 0.58 $\pm$ 0.13              |   |
| Copper      | --                                    | --              | 0.56 $\pm$ 0.03              | --              | 1.07 $\pm$ 0.32                   | 0.60 $\pm$ 0.10              |   |
| Lead        | --                                    | --              | 0.20 $\pm$ 0.02              | --              | 0.13 $\pm$ 0.02                   | 0.13 $\pm$ 0.03              |   |
| Mercury     | --                                    | --              | 0.03 $\pm$ 0.00              | --              | 0.05 $\pm$ 0.00                   | 0.04 $\pm$ 0.00              |   |
| Nickel      | --                                    | --              | 0.39 $\pm$ 0.03              | --              | 0.72 $\pm$ 0.07                   | 0.49 $\pm$ 0.02              |   |
| Selenium    | --                                    | --              | 0.96 $\pm$ 0.03              | --              | 0.70 $\pm$ 0.09                   | 0.85 $\pm$ 0.08              |   |
| Silver      | --                                    | --              | 0.02 $\pm$ 0.00 <sup>b</sup> | --              | 0.02 $\pm$ 0.00 <sup>b</sup>      | 0.03 $\pm$ 0.00 <sup>c</sup> |   |
| Thallium    | --                                    | --              | < 0.02                       | --              | 0.02 $\pm$ 0.00 <sup>b</sup>      | 0.02 $\pm$ 0.00 <sup>b</sup> |   |
| Uranium     | --                                    | --              | 0.19 $\pm$ 0.02              | --              | 0.09 $\pm$ 0.02                   | < 0.02                       |   |
| Zinc        | --                                    | --              | 17 $\pm$ 0                   | --              | 21 $\pm$ 2                        | 22 $\pm$ 1                   |   |

<sup>a</sup>Outfall 010 = lower half of Outfall 010 ditch; LUK = Little Bayou Creek kilometer; BBK = Big Bayou Creek kilometer; MAK = Massac Creek kilometer.

<sup>b</sup>2 of 3 values below the detection limit.

<sup>c</sup>1 of 3 values below the detection limit. Undetected values were used to calculate the means where at least one detected value was reported. N = 3 composite samples at each site except outfall 010 (N = 2).

**Table 4.3. Mean ( $\pm$  SE) concentrations ( $\mu\text{g/g}$ , wet weight) of various analytes in composited stonerollers (*Campostoma anomalum*) samples collected from stream sites near the PGDP and reference streams, May 1997**

| Analytes    | PGDP Sites <sup>a</sup>      |                 | Massac Creek <sup>a</sup><br>(Reference site) |
|-------------|------------------------------|-----------------|---|
|             | LUK 7.2                      | BBK 9.1         |   |
| PCBs, total | 2.32 $\pm$ 0.19              | 0.74 $\pm$ 0.03 | < 0.003                                       |
| Antimony    | < 0.02                       | < 0.02          | < 0.02  |
| Arsenic     | 0.16 $\pm$ 0.01 <sup>b</sup> | < 0.20          | 0.44 $\pm$ 0.12                               |
| Beryllium   | < 0.02                       | < 0.02          | 0.02 $\pm$ 0.00                               |
| Cadmium     | 0.04 $\pm$ 0.01              | 0.02 $\pm$ 0.00 | 0.04 $\pm$ 0.01                               |
| Chromium    | 0.68 $\pm$ 0.03              | 0.91 $\pm$ 0.40 | 1.09 $\pm$ 0.13                               |
| Copper      | 1.30 $\pm$ 0.00              | 2.7 $\pm$ 0.12  | 1.33 $\pm$ 0.09                               |
| Lead        | 0.21 $\pm$ 0.01              | 0.12 $\pm$ 0.01 | 0.60 $\pm$ 0.02                               |
| Mercury     | 0.03 $\pm$ 0.00              | 0.05 $\pm$ 0.01 | 0.03 $\pm$ 0.00                               |
| Nickel      | 0.41 $\pm$ 0.01              | 0.47 $\pm$ 0.02 | 0.74 $\pm$ 0.02                               |
| Selenium    | 0.92 $\pm$ 0.04              | 1.13 $\pm$ 0.03 | 0.57 $\pm$ 0.02                               |
| Silver      | 0.02 $\pm$ 0.00 <sup>c</sup> | 0.05 $\pm$ 0.00 | 0.02 $\pm$ 0.00                               |
| Thallium    | 0.02 $\pm$ 0.00 <sup>c</sup> | < 0.02          | < 0.02  |
| Uranium     | 0.77 $\pm$ 0.03              | 0.24 $\pm$ 0.02 | 0.05 $\pm$ 0.00                               |
| Zinc        | 24 $\pm$ 1                   | 35 $\pm$ 3      | 23 $\pm$ 1                                    |

<sup>a</sup>LUK = Little Bayou Creek kilometer; BBK = Big Bayou Creek kilometer; MAK = Massac Creek kilometer.

<sup>b</sup>2 of 3 values below the detection limit.

<sup>c</sup>1 of 3 values below the detection limit. Undetected values were used to calculate the means where at least one detected value was reported.

averaged one hundred to a thousand times higher than in fish from a local reference site, Massac Creek. As was the case for sunfish filets, the highest average PCB concentration was found in longear sunfish collected from LUK 9.0 (3.59  $\mu\text{g/g}$ ). Mean PCB concentrations were much lower downstream, averaging 1.74  $\mu\text{g/g}$  at LUK 7.2 and 1.23  $\mu\text{g/g}$  at LUK 4.3. Longear sunfish from outfall 010 were also high in PCBs (averaging 2.12  $\mu\text{g/g}$ ), strongly suggesting that this outfall, or sediments in the outfall ditch, is a major source of PCBs to downstream waters. The mean PCB concentration in Big Bayou Creek sunfish was also elevated in comparison to reference values (averaging 0.52  $\mu\text{g/g}$ ), but was much lower than any concentration reported for Little Bayou Creek fish.

PCB levels in stonerollers were approximately 30–40% higher than in longear sunfish collected from the same PGDP site. However, stonerollers were common only in the middle

reaches of each stream (LUK 7.2 and BBK 9.1) and may not be as available to terrestrial predators as longear sunfish in most stream sections.

Most average metal concentrations in fish near the PGDP were similar to, or lower than reference stream values (Tables 4.2, 4.3). Exceptions were copper, selenium, and uranium. Mean copper concentrations in both sunfish and stonerollers at BBK 9.1 exceeded the reference site mean by about a factor of two, and selenium was higher in stonerollers at both BBK 9.1 and LUK 7.2 by a similar factor. Selenium in sunfish was not elevated relative to reference site. Mean uranium concentrations in both species were 10–15 times higher at BBK 9.1, and about 5 times higher at LUK 7.2, than at the reference site. As was the case with PCBs, most metal concentrations were higher in stonerollers than in longear sunfish collected from the same site.

As expected, PCBs stand out as the most likely contaminant of potential ecological concern to fish-eating birds and mammals. The forage fish data were compared to the appropriate No Observable Effect Level (NOAEL) and Lowest Observable Effect Level (LOAEL) food benchmarks for mink and kingfisher by Sample (1997). The mean concentration of PCBs in sunfish exceeded NOAELs for kingfish at all five Paducah locations and exceeded NOAELs for mink at four locations (all in Little Bayou Creek watershed). The mean concentration in LUK 9.0 fish exceeded the LOAEL for kingfisher. With the exception of mercury, selenium, and zinc, all other analytes did not exceed NOAELs or LOAELs for each species at any location. The forage fish data suggest that fish accumulate PCBs to levels that present a risk to piscivorous wildlife, with the metal concentrations in fish being of much less concern.

## 5. FISH COMMUNITY MONITORING

*M. G. Ryon*

### 5.1 INTRODUCTION

Fish population and community studies can be used to assess the ecological effects of changes in water quality and habitat. These studies offer several advantages as indicators of environmental quality (see Karr et al. 1986, Karr 1987) and are especially relevant to assessment of the biotic integrity of Little Bayou and Big Bayou creeks. These creeks receive mixed effluents with a variety of stressors; the fish community includes species that may be sensitive to only one (e.g., temperature) or many of these stressors. Thus, analysis of the fish community may provide some indication as to which stressors are having the most impact. Monitoring of fish communities has been used by the Biological Monitoring and Abatement Program (BMAP) in ESD for receiving streams at PGDP since 1991. Changes in the fish communities in these streams have indicated impacts close to the PGDP (in Big Bayou Creek near Outfall 008; Ryon 1994b) and impacts associated with elevated temperatures (Roy et al. 1996). Fish community data have also indicated an absence of impacts at downstream locations where PGDP is less of an influence (e.g., at LUK 4.3 in Little Bayou Creek, Ryon 1996).

The objectives of the instream fish monitoring task are (1) to characterize spatial and temporal patterns in the distribution and abundance of fishes in Little Bayou and Big Bayou creeks, (2) to document the effects of PGDP operations on fish community structure and function, and (3) to document any recovery of the community associated with remedial actions conducted by PGDP.

### 5.2 STUDY SITES

Quantitative sampling of the fish community was conducted at five sites. Three sites are located on Big Bayou Creek (BBK 12.5, BBK 10.0, and BBK 9.1; Fig. 2.2); one is on Little Bayou Creek (LUK 7.2, Fig. 2.2) and one offsite reference station is located on Massac Creek (MAK 13.8, Fig. 2.1). MAK 13.8 was chosen as a reference site for BBK 9.1 and BBK 10.0. The upper site on Big Bayou Creek (BBK 12.5) was selected as a smaller reference site to be comparable to LUK 7.2.

### 5.3 MATERIALS AND METHODS

Quantitative sampling of the fish populations at the five sites in the PGDP area was conducted by electrofishing on March 17–20 and September 8–10, 1997. Data from these samples were used to estimate species richness and population size (numbers and biomass per unit area) and calculate annual production. All field sampling was conducted according to standard operating procedures (Schilling et al. 1996).

### 5.3.1 Quantitative Field Sampling Procedures

All stream sampling was conducted using two or three Smith-Root backpack electrofishers, depending on stream size. Each unit can deliver up to 1200 V of pulsed, direct current to stun fish.

After 0.64-cm-mesh seines were placed across the upper and lower boundaries of the fish sampling site to restrict fish movement, a five- to nine-person sampling team electrofished the site in an upstream direction on three consecutive passes. Stunned fish were collected and stored, by pass, in seine-net holding pens (0.64-cm-diam mesh) or buckets during further sampling.

Following the electrofishing, fish were anesthetized with MS-222 (tricaine methanesulfonate), identified, measured (total length), and weighed using Pesola spring scales. Individuals were recorded by 1-cm size classes and species. After ten individuals of a species-size class were measured and weighed, additional members of that size class were only measured. At sites with extremely high densities, specimens of some species were merely counted after a sufficient number of lengths and weights had been obtained. Length-weight regressions and length frequency distributions, based on the measured individuals, were used to estimate missing length and weight data.

After processing fish from all passes, the fish were allowed to fully recover from the anesthesia and returned to the stream. Any additional mortality that occurred as a result of processing was noted at that time. Following completion of fish sampling, the length and pool:riffle ratio and a subsample of widths and depths of the sampling reach were measured at each site.

### 5.3.2 Data Analysis

**Population Size.** Quantitative species population estimates were calculated using the method of Carle and Strub (1978). Biomass was estimated by multiplying the population estimate by the mean weight per size class. To calculate density and biomass per unit area, total numbers and biomass were divided by the surface area ( $m^2$ ) of the study reach. These data were compiled and analyzed by a comprehensive Fortran 77 program developed by ESD staff (Railsback et al. 1989).

**Annual Production.** Annual production was estimated at each site using a size-frequency method (Garman and Waters 1983) as modified by Railsback et al. (1989). Production was calculated for the period between the spring 1996 to 1997 sampling dates and the fall 1996 to 1997 sampling dates. Due to projected reductions in sampling frequency, only fall samples will be taken in the future at PGDP. Thus, the spring production estimate will be compared with the fall production estimate to see how much the two estimates vary, at least for one year, as a means to transition from a spring-based to a fall-based estimate.

## 5.4 RESULTS

The physical parameters of the sample sites showed only minor differences between the March (spring) and September (fall) samples (Table 5.1). In 1997, the sites were generally deeper and wider in spring sampling compared to fall samples. Some of the spring sampling

**Table 5.1. Length, mean width, mean depth, surface area, and pool:riffle ratio of fish sampling sites in Big Bayou Creek, Little Bayou Creek, and a reference stream, Massac Creek for 1997**

| Site <sup>a</sup>     | Length<br>(m) | Mean width<br>(m) | Mean depth<br>(cm) | Surface area<br>(m <sup>2</sup> ) | Pool:riffle<br>ratio |
|-----------------------|---------------|-------------------|--------------------|-----------------------------------|----------------------|
| <b>March 1997</b>     |               |                   |                    |                                   |                      |
| BBK 9.1               | 95            | 8.6               | 24.8               | 822                               | 1.1                  |
| BBK 10.0              | 97            | 6.1               | 17.3               | 590                               | 0.6                  |
| BBK 12.5              | 101           | 7.1               | 18.2               | 718                               | 1.7                  |
| LUK 7.2               | 102           | 3.9               | 13.2               | 395                               | 0.4                  |
| MAK 13.8              | 95            | 8.0               | 29.6               | 761                               | 3.8                  |
| <b>September 1997</b> |               |                   |                    |                                   |                      |
| BBK 9.1               | 110           | 6.5               | 25.4               | 719                               | 0.6                  |
| BBK 10.0              | 105           | 4.6               | 18.3               | 479                               | 1.1                  |
| BBK 12.5              | 97            | 5.9               | 11.6               | 573                               | 3.9                  |
| LUK 7.2               | 110           | 3.8               | 10.2               | 416                               | 0.5                  |
| MAK 13.8              | 108           | 3.8               | 17.5               | 413                               | 1.2                  |

<sup>a</sup>Site designations are Big Bayou Creek kilometer (BBK), Little Bayou Creek kilometer (LUK), and Massac Creek kilometer (MAK).

was influenced by recent rainfall. In particular, stream discharge at MAK 13.8 was much higher than normal and the sample was taken under less than optimum conditions.

#### 5.4.1 Species Richness and Composition

The species composition of 1997 samples is listed in Table 5.2. Thirty-eight species were found at the five sites, with BBK 9.1 and MAK 13.8 having the most species. The close proximity of BBK 9.1 to the Ohio River is evident in the variety of large river species (e.g., the bowfin, *Amia calva*) found in our samples. The variety at BBK 9.1 included a large number of species found in only one of the sample periods (i.e., suckers in spring and minnows in fall). One of these suckers is the black buffalo (*Ictiobus niger*), a species listed as threatened by the state (KSNPC 1996) and which has been found during qualitative sampling in other parts of the Big Bayou Creek watershed (Ryon and Carrico 1998). The other sites had a more stable fish community with most species observed in both samples.

The redspotted sunfish (*Lepomis miniatus*) was taken in the spring 1997 sample at LUK 7.2, and represents the third collection of this species from Little Bayou Creek. The presence of this species, which is classified as in need of management (KSNPC 1996), indicates the stream is capable of supporting rare species. The spring sample also included the first record of the fathead minnow (*P. promelas*) from Little Bayou Creek (Ryon and Carrico 1998). The specimens were golden in color, a morph often sold as bait and not found in wild populations, and obviously represent some type of "bait bucket" introduction. The species was not found in fall 1997 sampling at the site.

Since 1995, an expanded distribution was noted for the Mississippi silvery minnow (*Hybognathus nuchalis*). During 1991-95, the silvery minnow had only been found regularly

Table 5.2. List of species found at fish community sampling sites in Big Bayou Creek, Little Bayou Creek, and Massac Creek, 1997

| Species <sup>b</sup>  | Sites <sup>a</sup> |             |             |         |            |
|---|--------------------|-------------|-------------|---------|------------|
|   | BBK<br>9.1         | BBK<br>10.0 | BBK<br>12.5 | LU<br>K | MAK<br>7.2 |
| <b>Amidae</b>   |                    |             |             |         |            |
| Bowfin ( <i>Amia calva</i> )                                | 1 <sup>c</sup>     |             |             |         |            |
| <b>Clupeidae</b>  |                    |             |             |         |            |
| Gizzard shad ( <i>Dorosoma cepedianum</i> )                 | 1                  |             |             |         |            |
| <b>Cyprinidae</b>   |                    |             |             |         |            |
| Central stoneroller ( <i>Campostoma anomalum</i> )          | 2                  | 2           | 2           | 2       | 2          |
| Red shiner ( <i>Cyprinella lutrensis</i> )                  | 1                  |             | 1           | 2       | 1          |
| Steelcolor shiner ( <i>Cyprinella whipplei</i> )            | 1                  |             |             |         | 2          |
| Mississippi silvery minnow ( <i>Hybognathus nuchalis</i> )  | 1                  |             | 2           |         | 2          |
| Ribbon shiner ( <i>Lythrurus fumeus</i> )                   | 1                  |             |             |         |            |
| Redfin shiner ( <i>Lythrurus umbratilis</i> )               | 1                  | 1           | 2           |         | 1          |
| Golden shiner ( <i>Notemigonus crysoleucas</i> )            | 1                  |             | 2           | 1       | 1          |
| Bluntnose minnow ( <i>Pimephales notatus</i> )              | 1                  | 2           | 2           | 2       | 1          |
| Fathead minnow ( <i>Pimephales promelas</i> )               |                    |             |             | 1       |            |
| Creek chub ( <i>Semotilus atromaculatus</i> )               | 1                  | 2           | 2           | 2       | 2          |
| <b>Catostomidae</b>   |                    |             |             |         |            |
| River carpsucker ( <i>Carpoides carpio</i> )                | 1                  |             |             |         |            |
| White sucker ( <i>Catostomus commersoni</i> )               | 1                  |             |             |         |            |
| Creek chubsucker ( <i>Erimyzon oblongus</i> )               | 1                  |             | 2           |         | 2          |
| Smallmouth buffalo ( <i>Ictiobus bubalus</i> ) <sup>d</sup> | 1                  |             |             |         |            |
| Bigmouth buffalo ( <i>Ictiobus cyprinellus</i> )            | 1                  |             |             |         |            |
| Black buffalo ( <i>Ictiobus niger</i> ) <sup>d</sup>        | 1                  |             |             |         |            |
| Spotted sucker ( <i>Myoxocephalus melanops</i> )            | 2                  | 1           |             |         | 1          |
| Golden redhorse ( <i>Moxostoma erythrurum</i> )             | 1                  |             |             |         | 2          |
| <b>Ictaluridae</b>  |                    |             |             |         |            |
| Black bullhead ( <i>Ameiurus melas</i> )                    | 2                  | 2           | 1           | 1       |            |
| Yellow bullhead ( <i>Ameiurus natalis</i> )                 | 2                  | 2           | 2           | 2       | 2          |
| <b>Esocidae</b>   |                    |             |             |         |            |
| Grass pickerel ( <i>Esox americanus</i> )                   | 2                  | 2           |             | 1       | 1          |
| <b>Aphredoderidae</b>                                       |                    |             |             |         |            |
| Pirate perch ( <i>Aphredoderus sayanus</i> )                | 1                  | 1           |             | 1       | 2          |
| <b>Cyprinodontidae</b>                                      |                    |             |             |         |            |
| Blackspotted topminnow ( <i>Fundulus olivaceus</i> )        | 2                  | 2           | 2           | 2       | 2          |
| <b>Poeciliidae</b>  |                    |             |             |         |            |
| Western mosquitofish ( <i>Gambusia affinis</i> )            | 1                  | 1           | 1           | 2       | 1          |

Table 5.2 (continued)

| Species <sup>b</sup>                              | Sites <sup>a</sup> |      |      |    |      |
|---|--------------------|------|------|----|------|
|   | BBK                | BBK  | BBK  | LU | MAK  |
|   | 9.1                | 10.0 | 12.5 | K  | 13.8 |
|   |                    | 7.2  |      |    |      |
| Centrarchidae                                     |                    |      |      |    |      |
| Flier ( <i>Centrarchus macropterus</i> )          | 1                  |      |      | 1  |      |
| Green sunfish ( <i>Lepomis cyanellus</i> )        | 2                  | 2    | 2    | 2  | 2    |
| Warmouth ( <i>Lepomis gulosus</i> )               | 2                  |      | 1    | 2  |      |
| Bluegill ( <i>Lepomis macrochirus</i> )           | 2                  | 2    | 2    | 2  | 2    |
| Longear sunfish ( <i>Lepomis megalotis</i> )      | 2                  | 2    | 2    | 2  | 2    |
| Redspotted sunfish ( <i>Lepomis miniatus</i> )    |                    |      |      | 1  |      |
| Hybrid sunfish                                    | 1                  | 2    | 2    | 1  | 1    |
| Spotted bass ( <i>Micropterus punctulatus</i> )   | 2                  | 1    | 2    |    | 1    |
| Largemouth bass ( <i>Micropterus salmoides</i> )  | 1                  | 2    | 2    | 1  | 1    |
| Percidae  |                    |      |      |    |      |
| Bluntnose darter ( <i>Etheostoma chlorosoma</i> ) |                    |      |      | 1  |      |
| Slough darter ( <i>Etheostoma gracile</i> )       |                    | 1    | 1    | 2  | 1    |
| Logperch ( <i>Percina caprodes</i> )              |                    |      |      |    | 2    |
| Blackside darter ( <i>Percina maculata</i> )      |                    |      |      |    | 2    |
| TOTAL SPECIES                                     | 32                 | 17   | 20   | 20 | 24   |

<sup>a</sup>BBK = Big Bayou Creek kilometer, LUK = Little Bayou Creek kilometer, MAK = Massac Creek kilometer.

<sup>b</sup>Common and scientific names according to the American Fisheries Society (Robins et al. 1991) and Etnier and Starnes (1993).

Numbers indicate the occurrence of a species at that site, out of two total samples.

<sup>a</sup>Species identification confirmed by Dr. David A. Etnier, Department of Zoology, University of Tennessee.

in quantitative samples of Massac Creek. In September 1995, it began to appear in Big Bayou Creek and Little Bayou Creek samples (Ryon 1996). By September 1996, the silvery minnow was found at all sites and was very abundant at BBK 12.5, LUK 7.2 and MAK 13.8 (Ryon 1997). In 1997, it retreated from its widespread distribution and was collected only in Big Bayou Creek (at BBK 9.1 and BBK 12.5) and in Massac Creek. Similar patterns of expansion and retreat have been noticed for other species in previous BMAP sampling of the streams in the vicinity of the PGDP including the suckermouth minnow (*Phenacobius mirabilis*) and the pirate perch (*Aphredoderus sayanus*). These pulsed variations in density and distribution may be a function more of the natural colonization and population expansion capacities of the species than a reflection of changes in ecological health of the streams.

The mean number of species or species richness at the sites is given in Table 5.3. As was the case with the total number of species, BBK 9.1 and MAK 13.8 had the most species; for BBK 9.1, this number represented a substantial increase from means of previous (1991–96) sampling; for MAK 13.8, the 1997 species richness declined from previous levels. The mean 1997 species richness was also higher than previous values at the other two Big Bayou Creek

**Table 5.3. Fish community density (individuals/m<sup>2</sup>), biomass (g/m<sup>2</sup>), and species richness for March and September 1997 and means ( $\pm$ SD) for 1991-1996 and 1997 at sampling sites in Big Bayou Creek, Little Bayou Creek, and a reference stream, Massac Creek**

| Sampling periods      | Sites <sup>a</sup> |                  |                  |                 |                   |
|-----------------------|--------------------|------------------|------------------|-----------------|-------------------|
|                       | BBK 9.1            | BBK 10.0         | BBK 12.5         | LUK 7.2         | MAK 13.8          |
| <b>March 1997</b>     |                    |                  |                  |                 |                   |
| Density               | 0.36               | 0.88             | 1.16             | 1.26            | 0.18 <sup>b</sup> |
| Biomass               | 39.87              | 10.12            | 8.14             | 6.03            | 3.52              |
| Species richness      | 20                 | 14               | 19               | 16              | 18                |
| <b>September 1997</b> |                    |                  |                  |                 |                   |
| Density               | 1.70               | 5.50             | 4.04             | 1.74            | 6.16              |
| Biomass               | 17.60              | 23.89            | 13.11            | 6.29            | 19.10             |
| Species richness      | 23                 | 14               | 15               | 16              | 20                |
| <b>Means 1997</b>     |                    |                  |                  |                 |                   |
| Density               | 1.03               | 3.19             | 2.60             | 1.50            | 3.17              |
| Biomass               | 28.74              | 17.01            | 10.63            | 6.16            | 11.31             |
| Species richness      | 21.5               | 14.0             | 17.0             | 16.0            | 19.0              |
| <b>Means 1991-96</b>  |                    |                  |                  |                 |                   |
| Density               | 1.69 $\pm$ 0.97    | 3.91 $\pm$ 2.54  | 3.73 $\pm$ 1.21  | 3.21 $\pm$ 1.54 | 3.11 $\pm$ 1.64   |
| Biomass               | 21.56 $\pm$ 10.53  | 18.35 $\pm$ 9.78 | 14.25 $\pm$ 2.38 | 6.27 $\pm$ 2.75 | 12.37 $\pm$ 6.72  |
| Species Richness      | 15.7 $\pm$ 3.7     | 11.4 $\pm$ 2.1   | 14.5 $\pm$ 2.0   | 15.6 $\pm$ 3.9  | 20.7 $\pm$ 3.4    |

<sup>a</sup>BBK = Big Bayou Creek kilometer, LUK = Little Bayou Creek kilometer, MAK = Massac Creek kilometer.

<sup>b</sup>Sample conducted in very high abd turbid water; density and biomass values may not be representative of actual conditions.

sites, suggesting an improvement that may be related to watershed level parameters. If the improvement is a true change in water quality, the improvement would need to continue over several sampling years; otherwise, the change in species numbers may reflect normal variability of the data.

The composition of the fish community samples at the five sites is given in Table 5.4. In this context, community composition includes trophic level or feeding designation, taxonomic relationship or type of species, and tolerance/intolerance or sensitivity to stress or disruption (see Table E.1). The intolerant species are those species susceptible to habitat degradation and/or pollution (see Karr et al. 1986; Ohio EPA 1988; Mills et al. 1993). Benthic insectivores are a feeding guild that can reflect impacts on the benthic macroinvertebrate availability (Miller et al. 1988); and generalist feeders are species that are capable of switching easily between food items and, therefore, can be more successful in streams exposed to a

**Table 5.4. Fish community composition based on 1997 quantitative samples of Big Bayou Creek, Little Bayou Creek, and Massac Creek**

| Species category    | Sites <sup>a</sup>  |          |          |                     |                     |
|---------------------|---------------------|----------|----------|---------------------|---------------------|
|                     | BBK 9.1             | BBK 10.0 | BBK 12.5 | LUK 7.2             | MAK 13.8            |
| Cyprinidae          | 9 (28) <sup>b</sup> | 4 (24)   | 7 (35)   | 6 (20)              | 8 (33)              |
| Catostomidae        | 8 (25) <sup>c</sup> | 1 (6)    | 1 (5)    | 0                   | 3 (13)              |
| Centrarchidae       | 7 (22)              | 5 (29)   | 6 (30)   | 7 (35)              | 5 (21)              |
| Percidae            | 0                   | 1 (6)    | 2 (10)   | 1 (5)               | 3 (13)              |
| Tolerant species    | 7 (22)              | 4 (24)   | 5 (20)   | 6 (30) <sup>c</sup> | 4 (17) <sup>c</sup> |
| Intolerant species  | 4 (13)              | 1 (6)    | 1 (5)    | 1 (5)               | 5 (21)              |
| Piscivore           | 4 (13)              | 3 (18)   | 2 (10)   | 2 (10)              | 3 (13)              |
| Benthic Insectivore | 6 (19) <sup>c</sup> | 1 (6)    | 3 (15)   | 3 (15)              | 6 (25)              |
| Generalist feeder   | 9 (28)              | 5 (29)   | 6 (30)   | 7 (35)              | 5 (21)              |

<sup>a</sup>BBK = Big Bayou Creek kilometer; LUK = Little Bayou Creek kilometer; MAK = Massac Creek kilometer.

<sup>b</sup>Number of species at that site with percent of total species at that site in parentheses.

<sup>c</sup>Values that have changed substantially compared to the 1996 samples.

variety of stresses (Leonard and Orth 1986). Generally, a stream with better water and habitat quality will have more trophic levels, more taxonomic groups, more sensitive species, and fewer tolerant species than streams that are under atypical ecological stress.

The 1997 sample data indicate that BBK 10.0 has a more limited community than the MAK 13.8 reference. Furthermore, this site also appeared limited compared to BBK 12.5 and BBK 9.1. For many categories [darters (Percidae), suckers (Catostomidae), benthic insectivores and intolerant species], this site has fewer representatives than MAK 13.8, although BBK 10.0 did have more species in these categories in the 1997 samples than in the 1996 and earlier samples. The BBK 9.1 site has also improved in some of these categories in 1997 in comparison to 1996 and is only limited in numbers of darter species when compared to the reference samples. Whether the improvement is a result of natural variation or improved water quality should become apparent with further sampling. The LUK 7.2 site and the BBK 12.5 reference are very similar. LUK 7.2 did experience a rise in the percent of tolerant species in comparison to 1996 samples at that site.

#### 5.4.2 Density

Quantitative estimates of total density (number of individuals) for 1997 samples are given in Table 5.3, and density estimates for individual species are given in Appendix E, Tables E.2 and E.4. Mean densities for 1997 were generally lower than historic means with the exception of MAK 13.8. Density was particularly low at LUK 7.2 and BBK 9.1, being only 46 and 61%

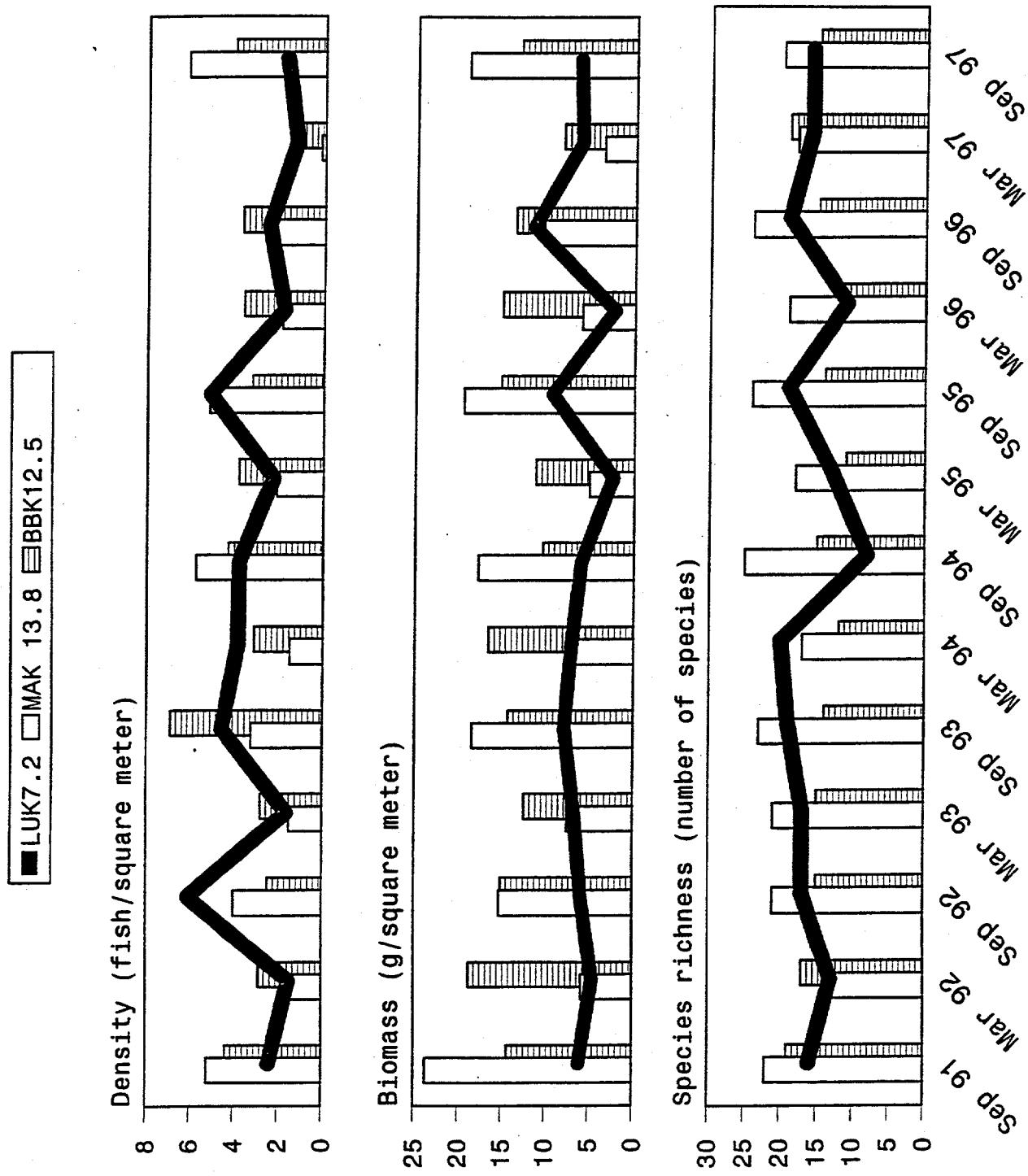
of the historic level, respectively. The low density at LUK 7.2 was outside the normal range for this site (Fig. 5.1), with the spring density being the lowest ever measured at this site. The low density at BBK 9.1 was within the previous range of values (Fig. 5.2), although again the spring value was quite low. The low densities for spring 1997 at most sites, including MAK 13.8, could be due in large part to the heavy rainfall before and during much of the spring sampling that resulted in less than ideal sampling conditions. Such conditions included high but clear water at BBK 9.1, higher turbidity than normal at LUK 7.2, and both turbid and high water at MAK 13.8. Thus, the low spring values may be explained by adverse sampling conditions. However, the fall density was also low at LUK 7.2, being the lowest fall value observed at that site, suggesting an impact not related to sampling conditions. In general, only density at BBK 10.0 exceeded the reference values, while densities at BBK 9.1 and LUK 7.2 were lower than reference values.

Although there were pronounced differences between sites in total densities, there was more agreement on which species were most abundant at the sites. The central stoneroller (*Campostoma anomalum*) and longear sunfish (*Lepomis megalotis*) were usually the most abundant species at all sites in Big Bayou Creek and at MAK 13.8 (Appendix E, Tables E.2 and E.4). Similar to 1996 findings, the presence of other species such as Mississippi silvery minnow, spotted sucker (*Mylochthys melanops*), and blackspotted topminnow (*Fundulus olivaceus*) was also noted among the most common species at these sites. At LUK 7.2, there was less consistency among the most abundant species, and several different species were found to be the most abundant, including the bluntnose minnow (*Pimephales notatus*), creek chub (*Semotilus atromaculatus*), green sunfish (*Lepomis cyanellus*), and blackspotted topminnow (Tables E.2 and E.4). The absence of large numbers of the longear sunfish and central stoneroller from this site is an established pattern, and probably reflects some difference in habitat between this site and the other sampling sites.

#### 5.4.3 Biomass

The biomass (weight of fish) estimates of the 1997 sampling are given in Tables 5.3, E.3 and E.5. The mean 1997 values were generally within the range of previous samples (Figs. 5.1 and 5.2). Mean 1997 biomass values at BBK 9.1 and BBK 10.0 were up to twice as high as the MAK 13.8 reference site, a pattern also seen in the historic data for these sites. The biomass values at LUK 7.2 did not indicate a change that would correlate with the low density in 1997, but the values were less than the mean biomass at the BBK 12.5 reference site. The lower biomass at LUK 7.2 compared to BBK 12.5 is also a pattern seen in previous samples. Unlike most years, the spring biomass at BBK 9.1 was higher than in the fall for 1997; this pattern is probably a reflection of the number of sucker species taken during that season.

As might be expected based on the density analysis, the longear sunfish and central stoneroller contributed the highest or next highest biomass at BBK 10.0 and BBK 12.5 (Tables E.3 and E.5). Other fish species that were among the larger biomass contributors at each site included the spotted sucker and golden redhorse (*Moxostoma erythrurum*) at BBK 9.1 and MAK 13.8. At LUK 7.2, the two highest biomass contributors included the creek chub, bluntnose minnow, and green sunfish, depending on sample season.



**Fig. 5.1.** Species richness, biomass, and density at Little Bayou Creek site and at two reference sites. LUK = Little Bayou Creek kilometer; MAK = Massac Creek kilometer; BBK Big Bayou Creek = kilometer.

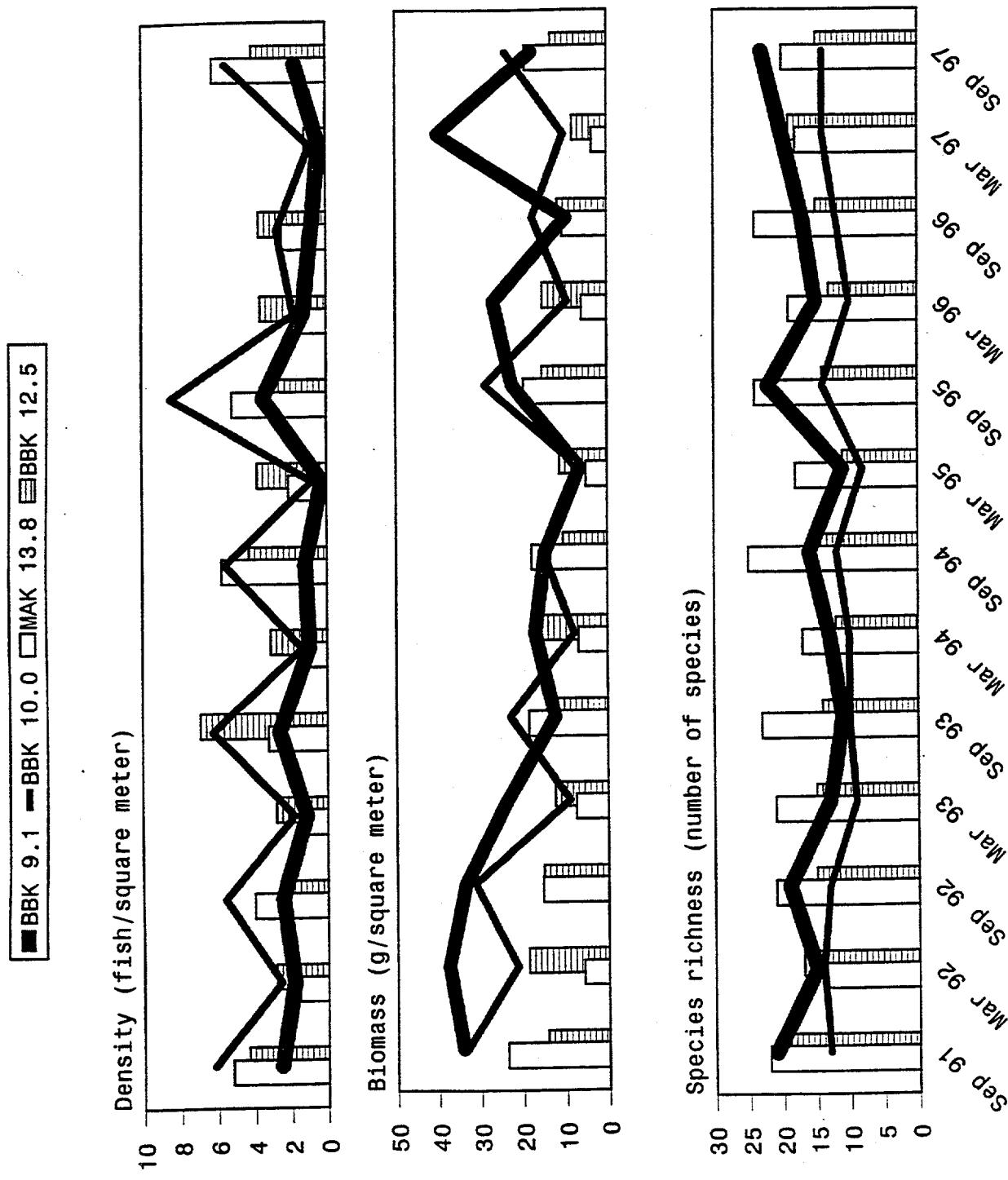


Fig. 5.2. Species richness, biomass, and density at Big Bayou Creek sites and a reference stream. BBK = Big Bayou Creek kilometer; MAK = Massac Creek kilometer.

#### 5.4.4 Production

Estimates of fish community "production" are a broader representation of overall species productivity. They represent a measurement of the accumulation of matter (or tissue) and, indirectly, a measurement of nutritional energy flow in the streams (MacFadyen 1948). The estimates track the success of reproduction in adding new individuals to a community (recruitment) and the growth of existing individuals in size and weight. This measurement goes beyond a biomass estimate, in that it compares the accumulated mass from one point in time to another, and accounts for changes between size classes within species. Thus, in this measurement, it is important not only to identify new individuals entering a size class, but growth in weight within a size class as well. Low production would suggest a failure in one or both of these components.

Because of the expected changes in sampling season from a spring and fall to a fall schedule, the production was estimated for both spring to spring and fall to fall intervals. The 1996–97 production is given for the spring period in Table 5.5 and for the fall in Table 5.6. Total spring production (in grams per square meter per year) was highest in Big Bayou Creek and increased from downstream to upstream (Table 5.5). The production at BBK 9.1 and BBK 10.0 was more than six times that at the reference site, MAK 13.8. At BBK 10.0, the production was dominated by the central stoneroller. Production at BBK 9.1 was dominated by the longear sunfish. In contrast to the past few years, spring productivity at BBK 9.1 and BBK 10.0 increased in 1997 (Fig 5.3). Productivity at the reference sites, although lower in 1997, did not show a declining trend extending more than one year. Production at LUK 7.2 was only a fifth of that at BBK 12.5 (Table 5.5). A ten-fold difference in production of central stoneroller and longear sunfish accounted for most of the disparity.

Production estimates for fall 1996 to 1997 revealed a slightly different pattern (Table 5.6 and Fig. 5.4). For this interval, production was much higher at MAK 13.8 and LUK 7.2 than in spring calculations of production. The fall levels of production at MAK 13.8 were similar to those at BBK 10.0 and more than double those at BBK 9.1. Although fall production was higher at LUK 7.2, it still remained less than half that at BBK 12.5. In general, fall production estimates were higher at all sites, except BBK 9.1. For BBK 9.1, spring production depends on a large contribution from sunfish; the fall sample did not include as much of this contribution. The overall higher production in fall samples compared to spring samples might be expected because it would include the growth of young-of-the-year fish, without a loss of individuals from winter mortality that is evident in spring sampling.

The spring production found in these streams was within the range of production values found in warmwater streams of the southeastern United States, including production estimates generated by similar methods at Oak Ridge monitoring sites (Table 5.5 in Ryon 1994c). Estimates of spring production in minimally disturbed streams in the southeastern United States (references) varied from 2.02 to 27.12  $\text{g} \cdot \text{m}^{-2} \cdot \text{year}^{-1}$  compared to 1.45 to 10.66  $\text{g} \cdot \text{m}^{-2} \cdot \text{year}^{-1}$  at PGDP area reference streams. Similarly, production at sites downstream of plant discharges that released mixed effluents ranged from 3.06 to 27.38  $\text{g} \cdot \text{m}^{-2} \cdot \text{year}^{-1}$  in the Southeast v 2.16 to 9.74  $\text{g} \cdot \text{m}^{-2} \cdot \text{year}^{-1}$  in Big Bayou Creek watershed.

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Table 5.5. Annual fish production (g/m<sup>2</sup>/yr) in Big Bayou Creek, Little Bayou Creek, and a reference stream, Massac Creek, March-April 1996 to March 1997

| Species <sup>b</sup>       | Sites <sup>a</sup> |          |          |         |          |
|----------------------------|--------------------|----------|----------|---------|----------|
|                            | BBK 9.1            | BBK 10.0 | BBK 12.5 | LUK 7.2 | MAK 13.8 |
| Bowfin                     | -0.43              | -        | -        | -       | -        |
| Gizzard shad               | -0.14              | -        | -        | -       | -        |
| Stoneroller                | 0.49               | 7.77     | 6.82     | 0.66    | 0.92     |
| Red shiner                 | -                  | -        | -0.01    | 0.20    | 0        |
| Steelcolor shiner          | -                  | -        | -        | -       | 0.06     |
| Common carp                | -0.06              | -        | -        | -       | -        |
| Mississippi silvery minnow | -                  | -        | -0.03    | -       | -0.01    |
| Ribbon shiner              | -                  | -        | -        | -       | -<0.01   |
| Redfin shiner              | -                  | -0.01    | -0.03    | -       | 0.01     |
| Golden shiner              | -                  | -        | -<0.01   | -<0.01  | -0.01    |
| Bluntnose minnow           | -                  | 0.02     | 0.11     | 0.31    | 0.02     |
| Fathead minnow             | -                  | -        | -        | 0.09    | -        |
| Creek chub                 | -0.07              | 0.59     | 1.69     | 0.46    | 0.09     |
| River carpsucker           | -0.19              | -        | -        | -       | -        |
| White sucker               | -0.02              | -        | -        | -       | -0.01    |
| Creek chubsucker           | -0.05              | -        | -0.06    | -       | 0.05     |
| Smallmouth buffalo         | -0.01              | -        | -        | -       | -        |
| Black buffalo              | -<0.01             | -        | -        | -       | -        |
| Spotted sucker             | 1.52               | -0.03    | -        | -       | -0.36    |
| Golden redhorse            | -0.18              | -        | -        | -       | -0.24    |
| Black bullhead             | -0.02              | -0.02    | -0.02    | -0.09   | -        |
| Yellow bullhead            | -0.14              | -0.02    | 0.44     | -0.01   | -0.02    |
| Grass pickerel             | <0.01              | -0.02    | -        | -       | -0.01    |
| Pirate perch               | -<0.01             | -<0.01   | -        | 0.01    | -<0.01   |
| Blackspotted topminnow     | -0.01              | 0.10     | 0.31     | 0.22    | 0.06     |
| Western mosquitofish       | 0                  | 0.04     | -        | 0.11    | -<0.01   |
| Green sunfish              | 0.20               | 0.05     | 0.58     | 0.22    | 0.16     |
| Warmouth                   | -<0.01             | -        | -<0.01   | 0.01    | -        |
| Bluegill                   | 2.98               | 0.02     | -0.06    | -0.02   | <0.01    |
| Longear sunfish            | 5.89               | 1.28     | 1.91     | 0.02    | 0.74     |

Table 5.5 (continued)

| Species <sup>b</sup> | Sites <sup>a</sup> |          |          |         |          |
|----------------------|--------------------|----------|----------|---------|----------|
|                      | BBK 9.1            | BBK 10.0 | BBK 12.5 | LUK 7.2 | MAK 13.8 |
| Redear sunfish       | -0.03              | -        | -        | -       | -        |
| Redspotted sunfish   | -                  | -        | -        | <0.01   | -        |
| Spotted bass         | -0.07              | -        | <0.01    | -       | -        |
| Largemouth bass      | -0.03              | -0.03    | 0.01     | -       | -        |
| Bluntnose darter     | -                  | -        | <0.01    | -       | -        |
| Slough darter        | 0                  | 0        | <0.01    | -0.03   | <0.01    |
| Logperch             | -                  | -        | -        | -       | <0.01    |
| Blackside darter     | -                  | -        | -        | -       | <0.01    |
| Total production     | 9.69               | 9.74     | 10.66    | 2.16    | 1.45     |

<sup>a</sup>BBK = Big Bayou Creek kilometer, LUK = Little Bayou Creek kilometer, MAK = Massac Creek kilometer.

<sup>b</sup>Common names according to the American Fisheries Society (Robins et al. 1991).

## 5.5 DISCUSSION

Data on the fish communities of Big Bayou Creek and Little Bayou Creek downstream of PGDP were compared to data from reference sites located on Big Bayou Creek above PGDP and on Massac Creek. These comparisons indicated a slight degradation in the communities downstream of PGDP.

Data indicated that the effects on the fish community were greatest just downstream from PGDP at BBK 10.0. The fish community at this site had lower mean and total species richness in comparison with MAK 13.8. However, the conditions at BBK 10.0 seemed to indicate an improvement in 1997. Unlike most sampling years, there was one sensitive species, more benthic insectivores, and more piscivores at this site. Density and biomass at BBK 10.0 were similar to or higher than those at the reference site (Fig 5.2). Although the fish community at BBK 10.0 still has demonstrated shortcomings, measures of the fish community were generally more positive in 1997. Future monitoring of the fish community will indicate whether this improvement was only a natural cycle of variation or the beginning of a trend. If water quality or stream conditions have improved, then the parameters of sensitive species, benthic insectivores, and darter species should continue to improve.

Similar to BBK 10.0, the fish community at BBK 9.1 showed signs of improvement in 1997. Mean and total species richness were higher than at the reference site, MAK 13.8. These values were also much higher than the historic means at BBK 9.1. Although there were fewer sensitive species, and at lower densities at BBK 9.1 than at MAK 13.8, more catostomids and benthic insectivores were present in 1997 at BBK 9.1. Based on sampling since 1991, density was less than or equal to that at MAK 13.8, but biomass remained high

**5-14 — Biological Monitoring Program**

**Table 5.6. Annual fish production (g/m<sup>2</sup>/yr) in Big Bayou Creek, Little Bayou Creek, and a reference stream, Massac Creek, September 1996 to September 1997**

| Species <sup>b</sup>       | Sites <sup>a</sup> |          |          |         |          |
|----------------------------|--------------------|----------|----------|---------|----------|
|                            | BBK 9.1            | BBK 10.0 | BBK 12.5 | LUK 7.2 | MAK 13.8 |
| Gizzard shad               | -0.07              | -        | -        | -       | -0.05    |
| Stoneroller                | 2.19               | 10.32    | 4.54     | -0.04   | 4.23     |
| Red shiner                 | 0                  | -        | -        | 0.01    | -<0.01   |
| Spotfin shiner             | -                  | -        | -        | -       | -<0.01   |
| Steelcolor shiner          | <0.01              | -        | -        | -0.01   | 0.08     |
| Mississippi silvery minnow | -                  | -<0.01   | 0.09     | 1.48    | 0.92     |
| Ribbon shiner              | -<0.01             | -        | -        | -       | -<0.01   |
| Redfin shiner              | <0.01              | -<0.01   | 0.02     | -<0.01  | -<0.01   |
| Golden shiner              | -<0.01             | -        | -0.03    | -<0.01  | -        |
| Bluntnose minnow           | -<0.01             | <0.01    | 0.03     | 0.82    | 0.39     |
| Fathead minnow             | -                  | -        | -        | -       | -0.01    |
| Creek chub                 | -0.03              | -0.15    | 1.91     | 1.46    | 1.78     |
| Creek chubsucker           | -                  | -        | -0.02    | -       | 0.24     |
| Bigmouth buffalo           | -0.06              | -        | -        | -       | -        |
| Spotted sucker             | 0.08               | -        | -        | -       | -        |
| Golden redhorse            | -                  | -        | -        | -       | 0.03     |
| Black bullhead             | -0.09              | 0.15     | -        | -       | -        |
| Yellow bullhead            | -0.01              | 0.01     | 0.68     | 0.16    | 0.04     |
| Grass pickerel             | -0.04              | -0.03    | -        | -0.02   | -        |
| Pirate perch               | -                  | -        | -        | -0.02   | 0.01     |
| Blackspotted topminnow     | 0.13               | 0.50     | 0.67     | 0.69    | 0.53     |
| Western mosquitofish       | 0.04               | 0.17     | 0.04     | 0.09    | <0.01    |
| Flier                      | 0.03               | -        | -        | -0.01   | -        |
| Green sunfish              | 0.34               | 0.13     | 0.87     | 0.60    | 0.41     |
| Warmouth                   | -0.01              | -        | -        | -0.01   | -<0.01   |
| Bluegill                   | 0.59               | -0.23    | 0.37     | -<0.01  | 0.13     |
| Longear sunfish            | 1.92               | 4.62     | 2.47     | 0.35    | 2.53     |
| Spotted bass               | -0.12              | 0.07     | 0        | -<0.01  | 0.01     |
| Largemouth bass            | -0.16              | -0.08    | 0.04     | -<0.01  | <0.01    |
| White crappie              | -0.02              | -        | -        | -       | -0.01    |

Table 5.6 (continued)

| Species <sup>b</sup>    | Sites <sup>a</sup> |          |          |         |          |
|-------------------------|--------------------|----------|----------|---------|----------|
|                         | BBK 9.1            | BBK 10.0 | BBK 12.5 | LUK 7.2 | MAK 13.8 |
| Slough darter           | -                  | -        | 0        | -0.01   | -<0.01   |
| Logperch                | -                  | -        | -        | -       | -0.16    |
| Blackside darter        | -                  | -        | -        | -       | -0.03    |
|                         | 4.70               | 15.46    | 11.68    | 5.54    | 11.06    |
| <u>Total production</u> |                    |          |          |         |          |

<sup>a</sup>BBK = Big Bayou Creek kilometer, LUK = Little Bayou Creek kilometer, MAK = Massac Creek kilometer.

<sup>b</sup>Common names according to the American Fisheries Society (Robins et al. 1991).

(Fig 5.2). Productivity estimates continued to increase (in spring samples) from low points in 1994-95 (Fig 5.3). The reversal of the four-fold decrease in production (from 1992-93 to 1994-95) indicates some moderation of impacts on recruitment success for the fish community at BBK 9.1. Further continuations of these improvement trends would be a stronger indication that a watershed improvement has occurred than that the changes were related to natural variation.

The fish community at LUK 7.2 was similar to the BBK 12.5 reference. The mean species richness values were similar to those of the reference site and continued to remain above the low value in fall 1994 (Fig 5.1). Biomass also remained at the mean levels of previous sampling (Table 5.3), but densities were low. Unlike conditions in Big Bayou Creek sites, productivity did not increase in 1997 (Fig 5.3).

Monitoring of the fish communities associated with PGDP streams indicated some depressed conditions, but did not specifically identify causative agents. The impacts were more evident at sites closest to the plant, which suggests that PGDP activities may be the cause. The low species richness and lack of sensitive species may be caused by poor water quality or may reflect degraded habitat. Previously, temperature extremes have been identified as a factor that could be impacting fish communities (Roy et al. 1996). The improvement in the community metrics at Big Bayou Creek sites may indicate some recovery in this section of the stream.

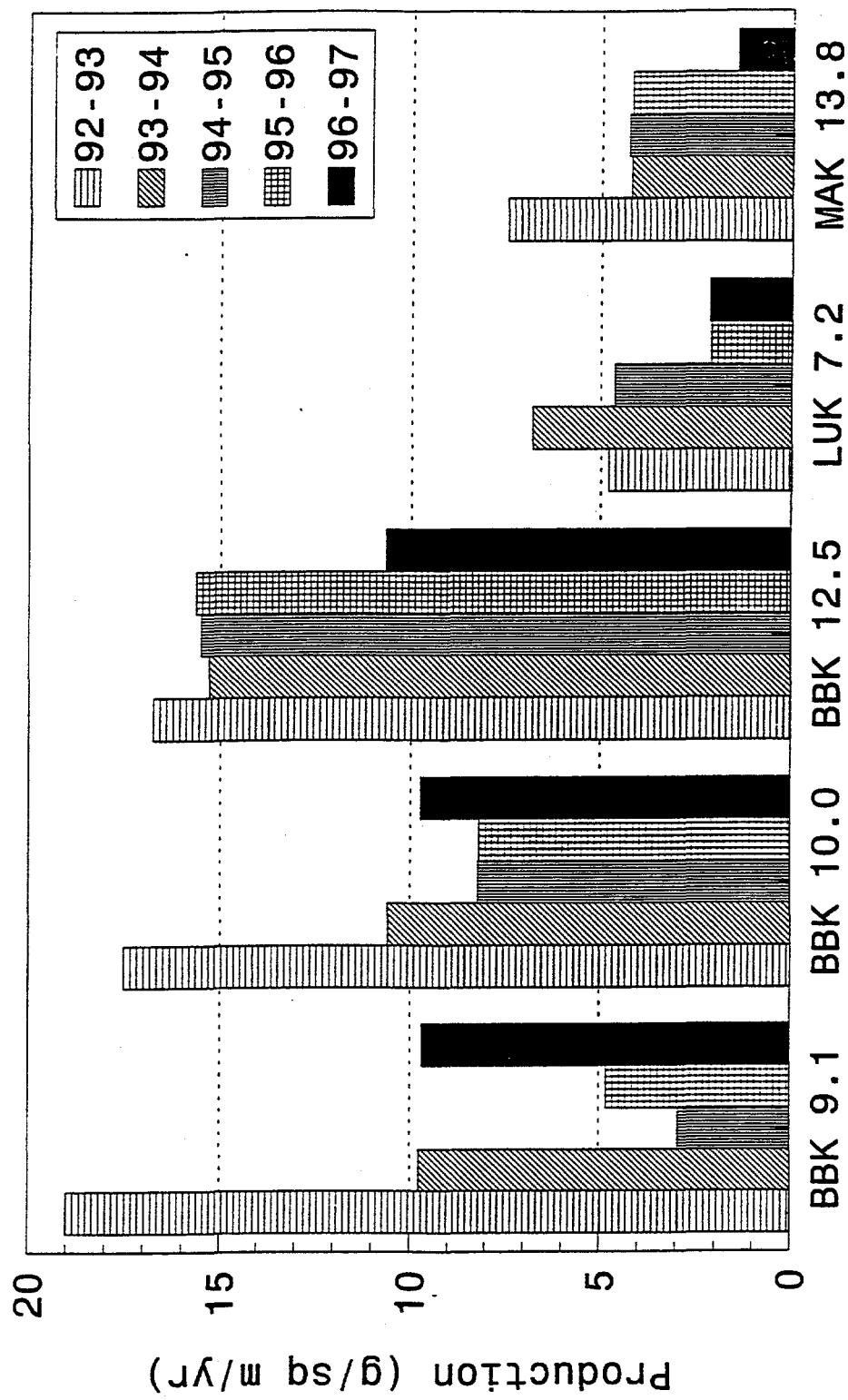


Fig. 5.3. Total annual fish community production (in grams per  $m^2$  per year) for Big Bayou Creek (BBK), Little Bayou Creek (LUK), and Massac Creek (MAK) for 1992 to 1997 based on spring-to-spring estimates.

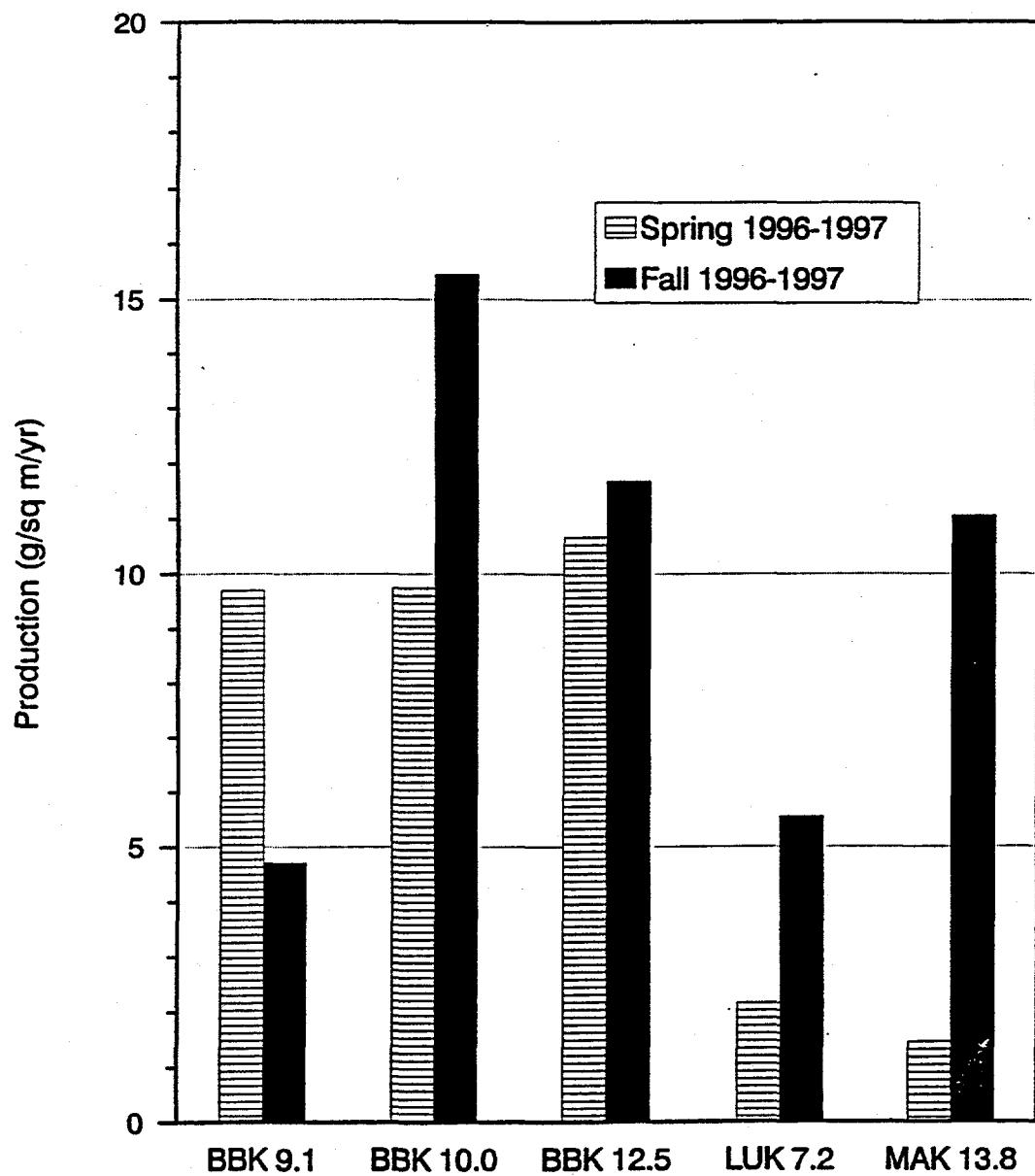


Fig. 5.4. Total annual production (in grams per  $m^2$  per year) for Big Bayou Creek, Little Bayou Creek, and Massac Creek based on a March-April 1996 to March 1997 (spring) interval and a September 1996 to September 1997 (fall) interval. BBK = Big Bayou Creek kilometer; LUK = Little Bayou Creek kilometer; MAK = Massac Creek kilometer.

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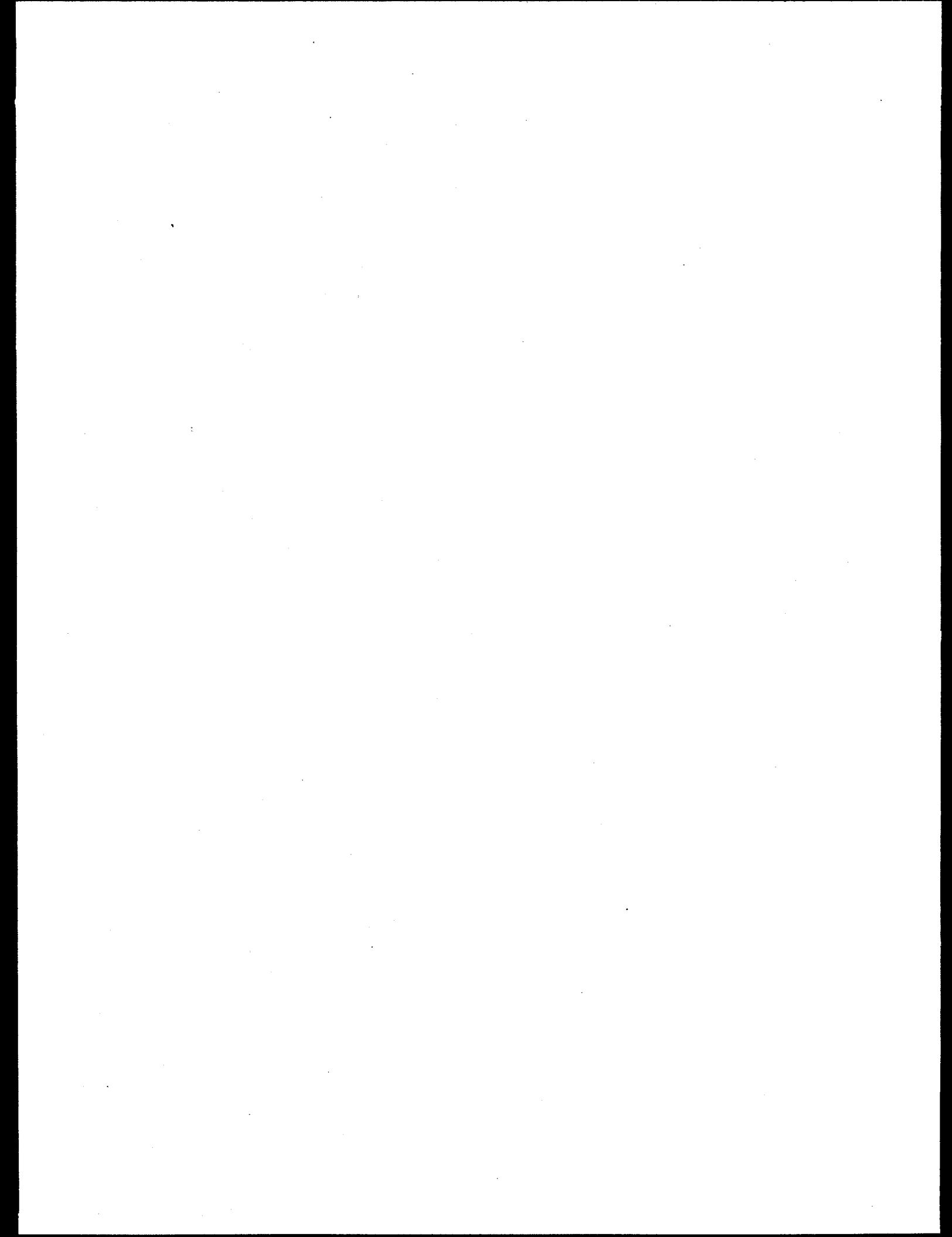
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**Appendix A**

**BIOLOGICAL MONITORING PROGRAM  
PADUCAH GASEOUS DIFFUSION PLANT  
SEMI-ANNUAL PROGRESS REPORT FOR 1997**



**Biological Monitoring Program  
Paducah Gaseous Diffusion Plant  
Semi-annual Progress Report for 1997**

|                |                  |
|----------------|------------------|
| L. A. Kszos    | M. G. Ryon       |
| B. K. Konetsky | G. R. Southworth |
| M. J. Peterson | J. G. Smith      |

Date: September 22, 1997

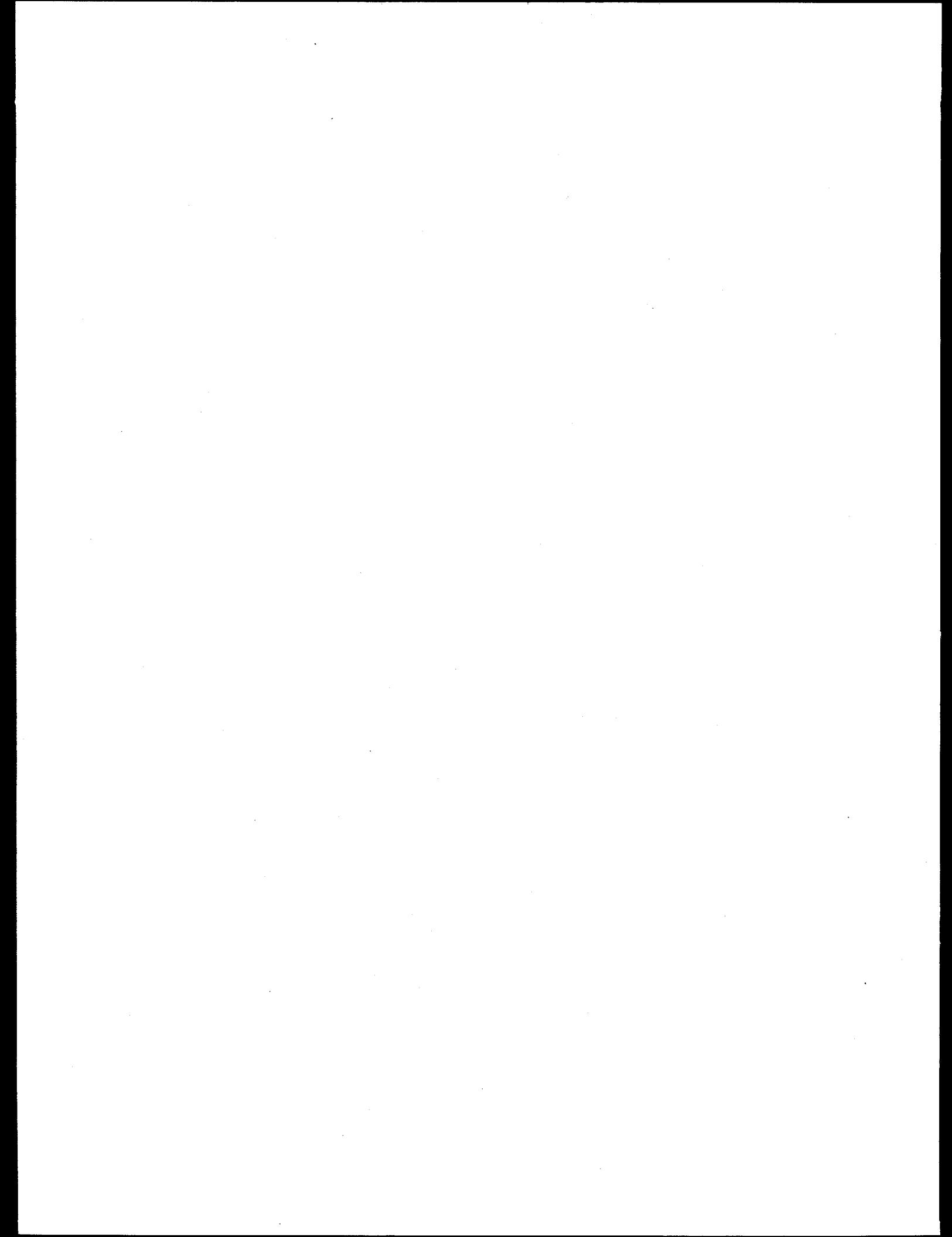
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Managed by  
LOCKHEED MARTIN ENERGY RESEARCH CORP.  
for the  
U.S. DEPARTMENT OF ENERGY  
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## A-1. INTRODUCTION

The Paducah Gaseous Diffusion Plant Biological Monitoring Program (PGDP BMP) was implemented in 1987 and consisted of ecological surveys, toxicity testing of effluents and receiving streams, identification of bioaccumulation of trace contaminants in biota, and supplemental chemical characterization of effluents. The overall goals of the BMP program are to (1) evaluate the acceptability of PGDP effluents under the Kentucky Pollutant Discharge Elimination System (KPDES) regulatory program and (2) characterize potential health and environmental impacts. Because research staff from the Environmental Sciences Division (ESD) at ORNL were experienced in biological monitoring, they served as reviewers and advisers throughout the planning and implementation of the PGDP BMP. Beginning in fall 1991, ESD/ORNL added data collection and report preparation to its responsibilities for the PGDP BMP. The BMP has continued because it has proven to be extremely valuable in (1) identifying those effluents with the potential for adversely affecting instream fauna, (2) assessing the ecological health of receiving streams, (3) guiding plans for remediation, and (4) protecting human health. For example, BMP has documented the improved health of the streams in the vicinity of PGDP; continued documentation of ecological recovery and improvement of water quality may be used to develop appropriate chemical limits and monitoring requirements.

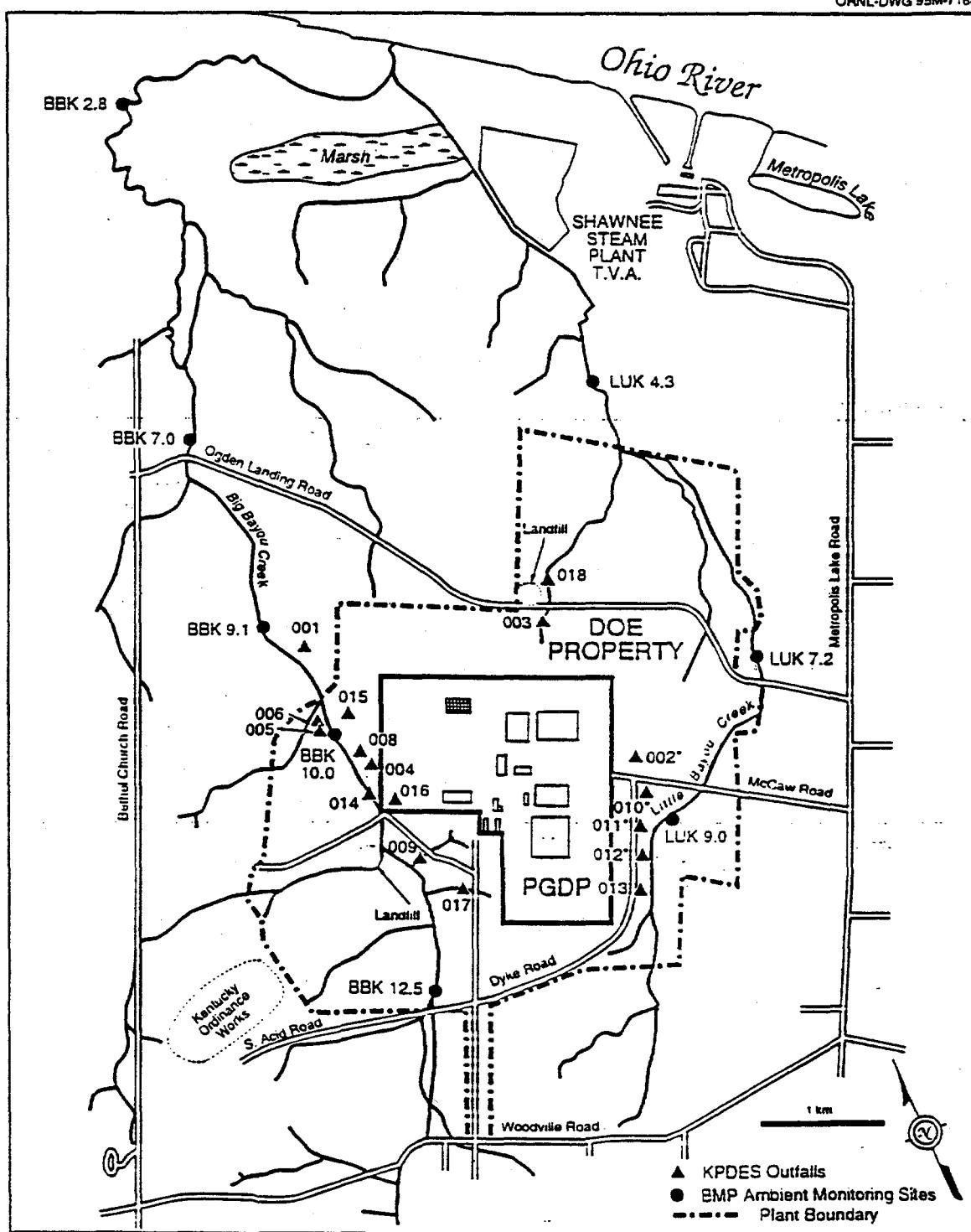
This progress report documents ESD/ORNL activities for 1996 that were not available for inclusion in the January–December, 1997 annual report (Kszos 1997) and that occurred from January to June, 1997.

## A-2. MONITORING SCHEDULE AND SAMPLING SITES

Scheduled monitoring activities for 1997 are outlined in Table 2.1. Location of sampling sites is shown in Figures 2.1, 2.2, and 2.3.

**Table A-2.1. Sampling schedule for Paducah Gaseous Diffusion Plant biological monitoring in calendar year 1997**

| Activity           | Month (1997) |      |      |      |     |      |      |      |      |      |      |      |
|--------------------|--------------|------|------|------|-----|------|------|------|------|------|------|------|
|                    | Jan.         | Feb. | Mar. | Apr. | May | Jun. | Jul. | Aug. | Sep. | Oct. | Nov. | Dec. |
| Toxicity Testing   | X            |      | X    | X    | X   |      |      |      | X    |      |      | X    |
| Benthic            |              |      |      | X    |     |      |      |      |      |      | X    |      |
| Macroinvertebrates |              |      |      |      |     |      |      |      |      |      |      |      |
| Fishes             |              |      |      | X    |     |      |      |      |      | X    |      |      |
| Bioaccumulation    |              |      |      |      |     | X    |      |      |      |      | X    |      |



\*Combined at C617 pond and discharged through 011/010

Fig. A-2.1. Location of biological monitoring sites on Big Bayou Creek and Little Bayou Creek in relation to the Paducah Gaseous Diffusion Plant (PGDP).

ORNL-DWG 95M-7163

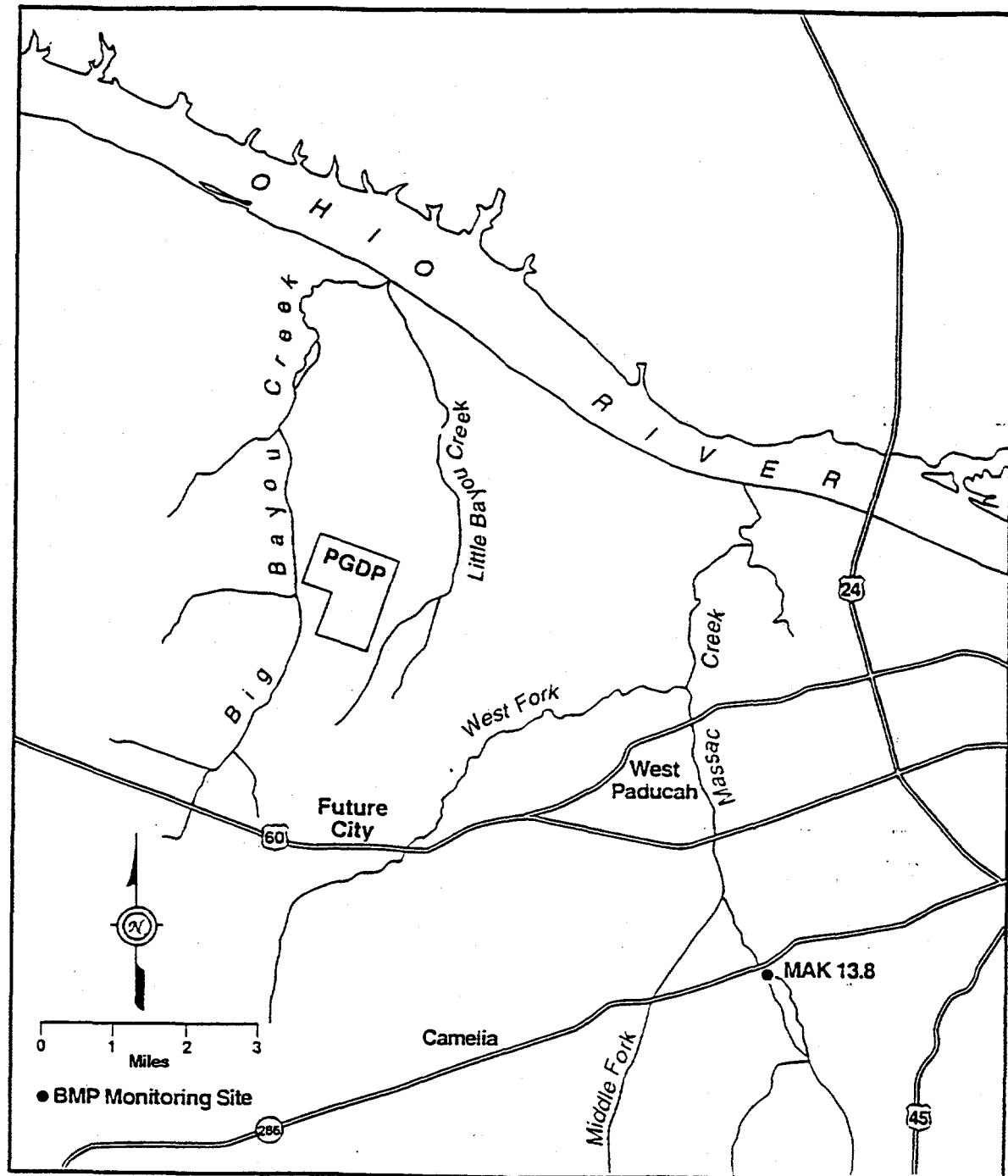
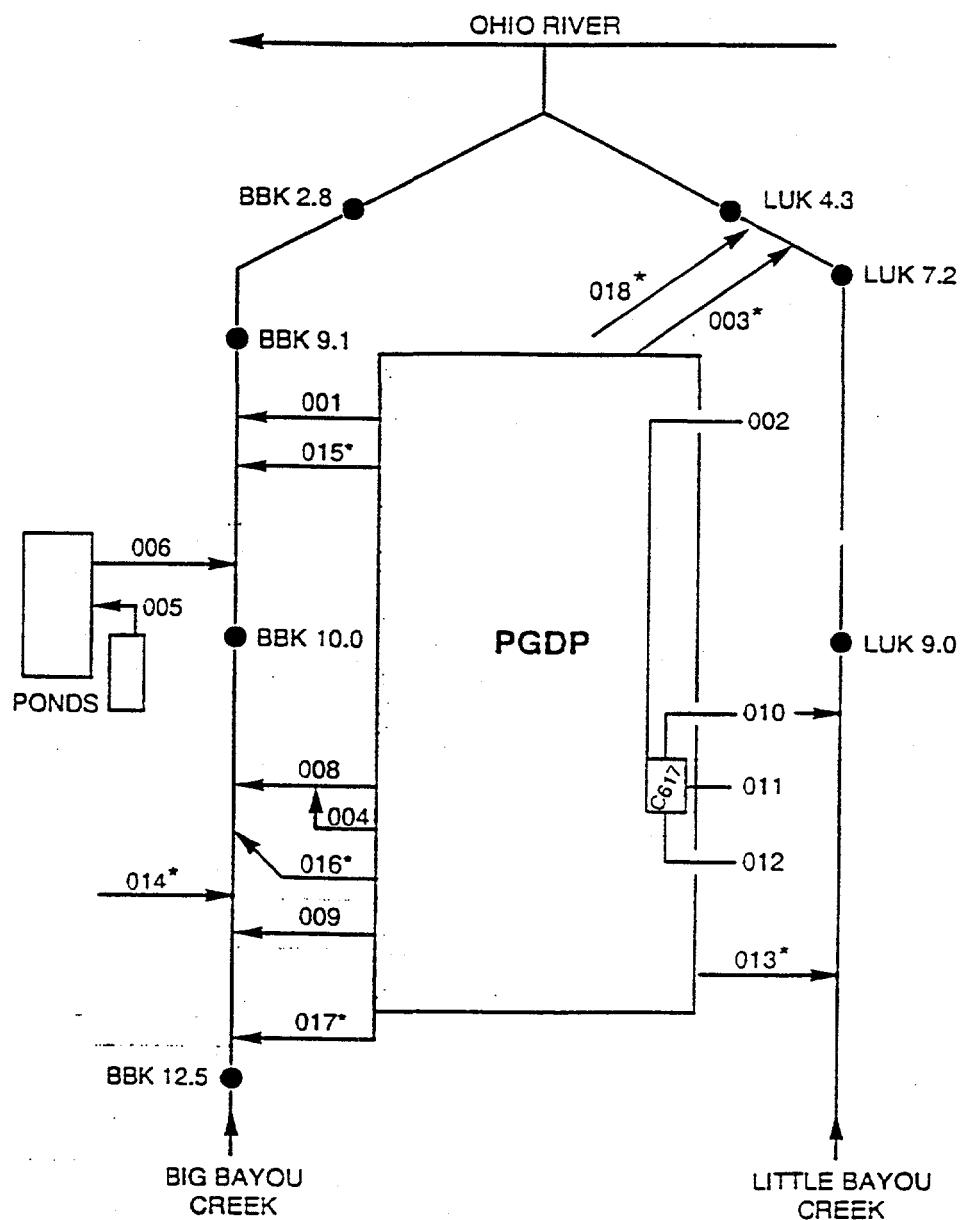


Fig. A-2.2. Location of reference site, Massac Creek kilometer (MAK) 13.8 in relation to Paducah Gaseous Diffusion Plant (PGDP).



\*INTERMITTENT FLOW

Fig. A-2.3. Schematic representation of biological monitoring sites on Big Bayou Creek and Little Bayou Creek in relation to outfalls and the Paducah Gaseous Diffusion Plant (PGDP).

### A-3. TOXICITY MONITORING (*L. A. Kszos and B. K. Konetsky*)

#### A-3.1 INTRODUCTION

The ESD Toxicology Laboratory at ORNL began evaluating the toxicity of continuous and intermittent outfalls at PGDP in October 1991. As required by a draft Agreed Order, *Ceriodaphnia* and fathead minnow tests of the continuous and intermittent outfalls were conducted quarterly. In September 1992, a renewed KPDES permit was issued to PGDP. Under the requirements of this permit, *Ceriodaphnia* and fathead minnow tests were continued on a quarterly basis. As required, the test methods used were the Cladoceran (*Ceriodaphnia dubia*) Survival and Reproduction Test (hereinafter referred to as the *Ceriodaphnia* test) and the Fathead Minnow (*Pimephales promelas*) Larval Survival and Growth Test (hereinafter referred to as the fathead minnow test; Lewis et al. 1994). After May 1995, tests of continuously flowing Outfalls 006, 008, 009, and 010 were reduced to the more sensitive species (fathead minnow larvae). Tests of continuously flowing Outfall 001 continued with *C. dubia* and fathead minnow larvae. After January 1996, tests of intermittently flowing Outfalls 013, 015, 016, 017, and 018 were reduced to the more sensitive species (fathead minnow larvae).

#### A-3.2 MATERIALS AND METHODS

Toxicity tests of effluents from the continuously flowing outfalls (001, 006, 008, 009, and 010) and the intermittently flowing outfalls (013, 015, 016, 017, and 018) were conducted according to the schedule shown in Table A-3.1. With the exception of effluent samples collected from Outfalls 008 and 010 in March 1997, samples from the continuously flowing outfalls were collected by personnel from ESD and transported to a nearby offsite laboratory at the Paducah Community College. In March 1997, samples from Outfall 008 and 010 were collected by ESD personnel and transported or shipped to ORNL for testing. Samples from the continuously flowing outfalls are time-dependent 24-h composite samples. The intermittently flowing outfalls are rainfall dependent; thus, tests were conducted using one grab sample. Samples from the intermittently flowing outfalls were collected by personnel from PGDP, refrigerated, and shipped to ESD using 24-h delivery. All samples were collected and delivered according to established chain-of-custody procedures (Kszos et al. 1989). Time of collection, water temperature, and arrival time in the laboratory were recorded.

Effluent samples from continuously flowing Outfalls 006, 008, 009, and 010 and all of the intermittently flowing outfalls were evaluated for toxicity with fathead minnows. Samples from continuously flowing Outfall 001 were also evaluated for toxicity with *C. dubia*. The *Ceriodaphnia* and fathead minnow tests are static-renewal tests, meaning that test water is replaced daily for 6 or 7 consecutive days. The fathead minnow test consists of four replicates per test concentration with ten animals per replicate. Each day before the water was replaced, the number of surviving larvae was recorded. At the end of 7 d, the larvae were dried and weighed to obtain an estimate of growth. The *Ceriodaphnia* test consists of ten replicates per test concentration with one animal per replicate. Each day the animals were transferred from a beaker containing old test solution and placed in a beaker containing fresh test solution. At this time, survival and the number of offspring produced were recorded. A control consisting of dilute mineral water augmented with trace metals was included with each test. On each fresh sample, subsamples of each effluent were routinely analyzed for pH, conductivity, alkalinity, and water hardness (Kszos et al. 1989).

Table A-3.1. Summary of toxicity test dates

| Outfall                 | Test Date          | Species                            |
|-------------------------|--------------------|------------------------------------|
| 013, 015, 016, 017, 018 | January 7-14, 1997 | fathead minnow                     |
| 001                     | March 7-14, 1997   | <i>C. dubia</i> and fathead minnow |
| 006, 009                | March 7-14, 1997   | fathead minnow                     |
| 008, 010                | March 11-18, 1997  | fathead minnow                     |
| 013, 015, 016, 017, 018 | April 8-15, 1997   | fathead minnow                     |
| 001                     | May 14-21, 1997    | <i>C. dubia</i> and fathead minnow |
| 006, 008, 009, 010      | May 14-21, 1997    | fathead minnow                     |

A linear interpolation method (Lewis et al. 1994) was used to determine the 25% inhibition concentration (IC25, that concentration causing a 25% reduction in fathead minnow growth or *Ceriodaphnia* reproduction compared to a control). A computer program [A Linear Interpolation Method for Sublethal Toxicity: Inhibition Concentration (ICp) Approach, version 2.0] distributed by the EPA (Environmental Research Laboratory, Duluth, Minnesota) was used for the calculation. The chronic toxicity unit (TUC = 100/IC25) is required as a compliance endpoint in the renewed permit (September 1992 to present). The higher the TUC, the more toxic an effluent. Because Little Bayou and Big Bayou creeks have been determined to have a low flow of zero, a TUC > 1.0 for the continuously flowing outfalls would be considered a noncompliance and an indicator of potential instream toxicity. Summary statistics (e.g., mean, standard deviation) were calculated using the Statistical Analyses System (SAS 1985a, 1985b).

### A-3.3 Results

Results of toxicity tests and chemical analyses of the continuously flowing outfalls are shown in Tables A-3.2 and A-3.3. Results of the toxicity tests and chemical analyses of the intermittently flowing outfalls are shown in Tables A-3.4 and A-3.5. Effluent samples from the continuously flowing outfalls had TUCs < 1 for all tests conducted. The water quality of the effluent samples was generally similar between test periods with the exception of outfalls 008, 009, and 010 which tended to have higher alkalinity during the March tests compared to the May test. Effluent samples from the intermittently flowing outfalls had TUCs < 1 for all tests conducted with the exception of Outfall 016 in April 1997. During April 1997, Outfall 016 had a TUC of 19.61. The cause of the toxicity is not known. It does not appear to be directly related to the concentration of total suspended solids because the concentration of total suspended solids was lower in Outfall 016 during April than in some of the other outfalls during January or April where toxicity was not observed.

**Table A-3.2. Results of toxicity tests of continuously flowing outfalls conducted during January–June, 1997**

| Outfall                  | IC <sub>25</sub> (%)       |                           | TUC                        |                           |
|--------------------------|----------------------------|---------------------------|----------------------------|---------------------------|
|                          | <i>Pimephales promelas</i> | <i>Ceriodaphnia dubia</i> | <i>Pimephales promelas</i> | <i>Ceriodaphnia dubia</i> |
| <i>March 7–14, 1997</i>  |                            |                           |                            |                           |
| 001                      | > 100                      | > 100                     | < 1                        | < 1                       |
| 006                      | > 100                      | NA                        | < 1                        | NA                        |
| 009                      | > 100                      | NA                        | < 1                        | NA                        |
| <i>March 11–18, 1997</i> |                            |                           |                            |                           |
| 008                      | > 100                      | NA                        | < 1                        | NA                        |
| 010                      | > 100                      | NA                        | < 1                        | NA                        |
| <i>May 14–21, 1997</i>   |                            |                           |                            |                           |
| 001                      | > 100                      | > 100                     | < 1                        | < 1                       |
| 006                      | > 100                      | NA                        | < 1                        | NA                        |
| 008                      | > 100                      | NA                        | < 1                        | NA                        |
| 009                      | > 100                      | NA                        | < 1                        | NA                        |
| 010                      | > 100                      | NA                        | < 1                        | NA                        |

*Note:* NA = Not applicable; IC<sub>25</sub> = the concentration causing a 25% reduction in *Pimephales promelas* (fathead minnows) growth or *Ceriodaphnia dubia* reproduction. TUC = chronic toxicity units.

**Table A-3.3. Summary (Mean  $\pm$  SD) of water chemistry analyses conducted during toxicity tests of continuously flowing outfalls at the Paducah Gaseous Diffusion Plant, January–June 1997**

| Outfall                  | pH<br>(S. U.) | Alkalinity<br>(mg/L as CaCO <sub>3</sub> ) | Hardness<br>(mg/L as CaCO <sub>3</sub> ) | Conductivity<br>( $\mu$ S/cm) |
|--------------------------|---------------|--|--|-------------------------------|
| <i>March 7–14, 1997</i>  |               |  |  |                               |
| 001                      | 7.69 (0.10)   | 42.3 (7.5)                                 | 246.7 (49.2)                             | 911.0 (181.6)                 |
| 006                      | 9.18 (0.24)   | 46.0 (2.7)                                 | 66.7 (3.1)                               | 206.7 (52.4)                  |
| 009                      | 8.07 (0.02)   | 74.3 (6.0)                                 | 106.7 (11.0)                             | 399.0 (6.9)                   |
| <i>March 11–18, 1997</i> |               |  |  |                               |
| 008                      | 7.49 (0.09)   | 51.0 (5.3)                                 | 78.0 (2.0)                               | 304.7 (24.4)                  |
| 010                      | 7.80 (0.04)   | 52.3 (1.5)                                 | 88.7 (3.1)                               | 286.0 (9.5)                   |
| <i>May 14–21, 1997</i>   |               |  |  |                               |
| 001                      | 8.95 (0.11)   | 39.3 (4.6)                                 | 228.0 (74.7)                             | 980.7 (120.8)                 |
| 006                      | 9.00 (0.03)   | 43.3 (3.1)                                 | 72.7 (10.3)                              | 214.3 (4.2)                   |
| 008                      | 7.30 (0.13)   | 29.3 (5.5)                                 | 76.0 (10.0)                              | 311.0 (7.6)                   |
| 009                      | 7.93 (0.14)   | 42.3 (4.9)                                 | 86.0 (13.1)                              | 289.7 (13.3)                  |
| 010                      | 7.69 (0.09)   | 34.0 (4.0)                                 | 92.0 (16.4)                              | 300.0 (11.4)                  |

**Table A-3.4. Results of toxicity tests of intermittently flowing outfalls conducted January–June 1997**

| Outfall                   | IC <sub>25</sub> (%) | TUc   |
|---------------------------|----------------------|-------|
| <i>January 7–14, 1997</i> |                      |       |
| 013                       | > 100                | < 1   |
| 015                       | > 100                | < 1   |
| 016                       | > 100                | < 1   |
| 017                       | > 100                | < 1   |
| 018                       | > 100                | < 1   |
| <i>April 8–15, 1997</i>   |                      |       |
| 013                       | > 100                | < 1   |
| 015                       | > 100                | < 1   |
| 016                       | 5.10                 | 19.61 |
| 017                       | > 100                | < 1   |
| 018                       | > 100                | < 1   |

*Note:* IC<sub>25</sub> = the concentration causing a 25% reduction in *Pimephales promelas* (fathead minnow) growth; TUc = chronic toxicity units.

**Table A-3.5. Summary of water chemistry analyses conducted during toxicity tests of intermittently flowing outfalls at the Paducah Gaseous Diffusion Plant, January–June 1997**

| Outfall                   | pH<br>(S. U.) | Alkalinity<br>(mg/L as CaCO <sub>3</sub> ) | Hardness<br>(mg/L as CaCO <sub>3</sub> ) | Conductivity<br>( $\mu$ S/cm) | Total<br>suspended<br>solids (mg/L) |
|---------------------------|---------------|--|--|-------------------------------|-------------------------------------|
| <i>January 7–14, 1997</i> |               |  |  |                               |                                     |
| 013                       | 7.87          | 82   | 156                                      | 448                           | 79.3                                |
| 015                       | 8.29          | 140  | 304                                      | 772                           | 50.0                                |
| 016                       | 8.21          | 150  | 210                                      | 616                           | 7.7                                 |
| 017                       | 7.92          | 182  | 210                                      | 512                           | 3.4                                 |
| 018                       | 7.66          | 71   | 90                                       | 214                           | 29.5                                |
| <i>April 8–15, 1997</i>   |               |  |  |                               |                                     |
| 013                       | 7.97          | 103  | 122                                      | 353                           | 10.6                                |
| 015                       | 7.94          | 128  | 180                                      | 514                           | 11.8                                |
| 016                       | 8.12          | 121  | 166                                      | 439                           | 33.0                                |
| 017                       | 8.05          | 142  | 154                                      | 436                           | 16.2                                |
| 018                       | 7.90          | 53   | 57                                       | 155                           | 36.6                                |

#### A-4. BIOACCUMULATION STUDIES (*M. J. Peterson and G. R. Southworth*)

##### A-4.1 INTRODUCTION

Bioaccumulation monitoring conducted to date as part of the Biological Monitoring Program at PGDP identified polychlorobiphenyl (PCB) contamination in fish in Big Bayou Creek and Little Bayou Creek as major concerns (Birge et al. 1990, 1992; Kszos et al. 1994, Kszos 1994, 1996a, 1996b, 1997). Mercury concentrations in fish from Big Bayou Creek were also found to be higher in fish collected downstream from PGDP discharges than in fish from an upstream site (Birge et al. 1990, 1992; Kszos et al. 1994, Kszos 1994, 1996a, 1996b, 1997). Concentrations of various other metals and organics in fish from Big Bayou Creek and Little Bayou Creek were well below levels of concern for human consumption.

The primary objective of the 1995-96 bioaccumulation monitoring was to evaluate spatial and temporal changes in PCB contamination in sunfish from Little Bayou Creek. PCB contamination in fish in Big Bayou Creek had declined to near background levels over the 1992-95 period, and monitoring in this stream was consequently reduced to a single site immediately downstream from the lowermost PGDP discharge to Big Bayou Creek. Similarly, mercury monitoring was conducted only at that site in Big Bayou Creek. Because Big Bayou Creek is capable of supporting a limited sport fishery for larger game fish, spotted bass were analyzed for mercury and PCBs to evaluate the maximum concentrations likely in fish near the PGDP.

##### A-4.2 STUDY SITES

In October 1996, longear sunfish (*L. megalotis*) were collected for PCB analysis at LUK 9.0, LUK 7.2 and LUK 4.3 on Little Bayou Creek (Fig. 2.1). Spotted bass (*Micropterus punctulatus*) were collected from Big Bayou Creek (BBK 9.1) near PGDP and analyzed for PCBs and mercury. Sunfish were collected at LUK 9.0, LUK 7.2, and LUK 4.3 in Little Bayou Creek for PCB analysis in May 1997. Forage fish (central stoneroller, *Campostoma anomalum*, and small longear sunfish) were also collected at these sites and the Big Bayou Creek site (BBK 9.1). These fish will be analyzed for a suite of metals and PCBs in order to provide data for evaluating ecological risks to fish-eating birds and mammals. Results of the spring 1997 sampling will be reported in the next annual report.

##### A-4.3 MATERIALS AND METHODS

Methods were the same as reported in Kszos 1997, with the following changes. The number of individual fish analyzed for PCBs at each site in Little Bayou Creek was reduced from eight to six in fiscal year 1997. In Big Bayou Creek, mercury and PCB monitoring in sunfish was dropped, and four spotted bass were collected at the BBK 9.1 site for mercury and PCB analysis. Concentrations of these substances approach local background levels in fish from Big Bayou Creek. The reduced sampling and analysis effort was deemed adequate to document whether concentrations of mercury and PCBs remain below levels of concern at this site.

At each site, forage fish were collected by electrofishing, and grouped into three subgroups each containing ten fish. Individuals in each subgroup were weighed and measured, and the sample of ten whole fish was then homogenized in a stainless steel blender and

packaged in aluminum foil for delivery to the analytical laboratory. The composite samples will be analyzed for mercury and PCBs using procedures in Kszos 1997, and for metals using inductively coupled plasma mass spectrometry (ICP/MS) by EPA procedure 200.8.

#### **A-4.4 RESULTS AND DISCUSSION**

##### **A-4.4.1 PCBs**

PCB concentrations in sunfish collected October 1996 in Little Bayou Creek continue to exhibit a pronounced decrease with distance downstream from PGDP (Fig. A-4.1). Concentrations were highest at LUK 9.0 and LUK 7.2 and decreased dramatically at LUK 4.2. Unlike the case in previous monitoring, the highest average concentration was not found at LUK 9.0, but rather at LUK 7.2. PCB concentrations at the two sites were similar, averaging ( $\pm$  SE)  $0.64 \pm 0.13 \mu\text{g/g}$  at LUK 9.0 and  $0.71 \pm 0.06 \mu\text{g/g}$  at LUK 7.2. This deviation from the typical downstream pattern is probably a consequence of the variability typical of PCB bioaccumulation and analysis. At LUK 4.2, the mean PCB concentration in sunfish was  $0.13 \pm 0.06 \mu\text{g/g}$ . Composition of the PCB mixtures found in sunfish resembled Aroclor 1254 and 1260 at all sites. Concentration ranges were  $0.35\text{--}1.2 \mu\text{g/g}$  at LUK 9.0,  $0.48\text{--}0.89 \mu\text{g/g}$  at LUK 7.2, and  $<0.01\text{--}0.32 \mu\text{g/g}$  at LUK 4.2. No fish exceeded the  $2.0 \mu\text{g/g}$  Food and Drug Administration limit.

The trend in PCB contamination in fish in Little Bayou Creek from 1992 to 1996 is depicted in Fig. A-4.2. Average concentrations have decreased to about 25% of the peak values seen in 1992, but appear to have stopped declining. For the past two years, mean PCB concentrations in sunfish at LUK 9.1 have remained around  $0.5 \mu\text{g/g}$ , suggesting that low inputs continue at a reduced level.

In Big Bayou Creek, spotted bass contained an average PCB concentration of  $0.43 \pm 0.06 \mu\text{g/g}$  (range  $0.30\text{--}0.57 \mu\text{g/g}$ ). Bass from the same site averaged  $0.16 \mu\text{g/g}$  in October 1995. Although levels of PCBs in fish at this site remain well below that typical of upper Little Bayou Creek, the presence of mean concentrations approaching  $0.5 \mu\text{g/g}$  indicates that PCB inputs to this creek are continuing. Only the highly chlorinated materials similar to Aroclor 1254/1260 were present.

##### **A-4.4.2 Mercury**

The bioaccumulation of mercury by fish is predominantly a food chain mediated process; thus, predatory species that occupy trophic positions at or near the top of the aquatic food web would be expected to contain higher concentrations of mercury than species lower in the food chain. Spotted bass in Big Bayou Creek occupy that role of terminal predator and are monitored by this task to evaluate the maximum mercury level likely in fish from that creek. The mean mercury concentration in spotted bass collected in October 1996 was  $0.52 \pm 0.11 \mu\text{g/g}$  (Fig. A-4.3), with a range of  $0.33\text{--}0.73 \mu\text{g/g}$ . No temporal trend was evident, and mercury concentrations in Big Bayou Creek bass continued to average around  $0.5 \mu\text{g/g}$ .

Aqueous total mercury and methylmercury in Big Bayou Creek upstream and downstream from PGDP were measured in summer 1997 by researchers at ORNL and Frontier Geosciences in Seattle, Washington, as part of a study funded by the Lockheed Martin Energy Systems' Y-12 Plant to investigate the relationship between waterborne mercury concentrations and mercury bioaccumulation in fish. The baseflow concentration of total mercury was  $5.3 \text{ ng/L}$  at BBK 9.1 downstream from PGDP and  $1.2 \text{ ng/L}$  at BBK 12.5 upstream from PGDP.

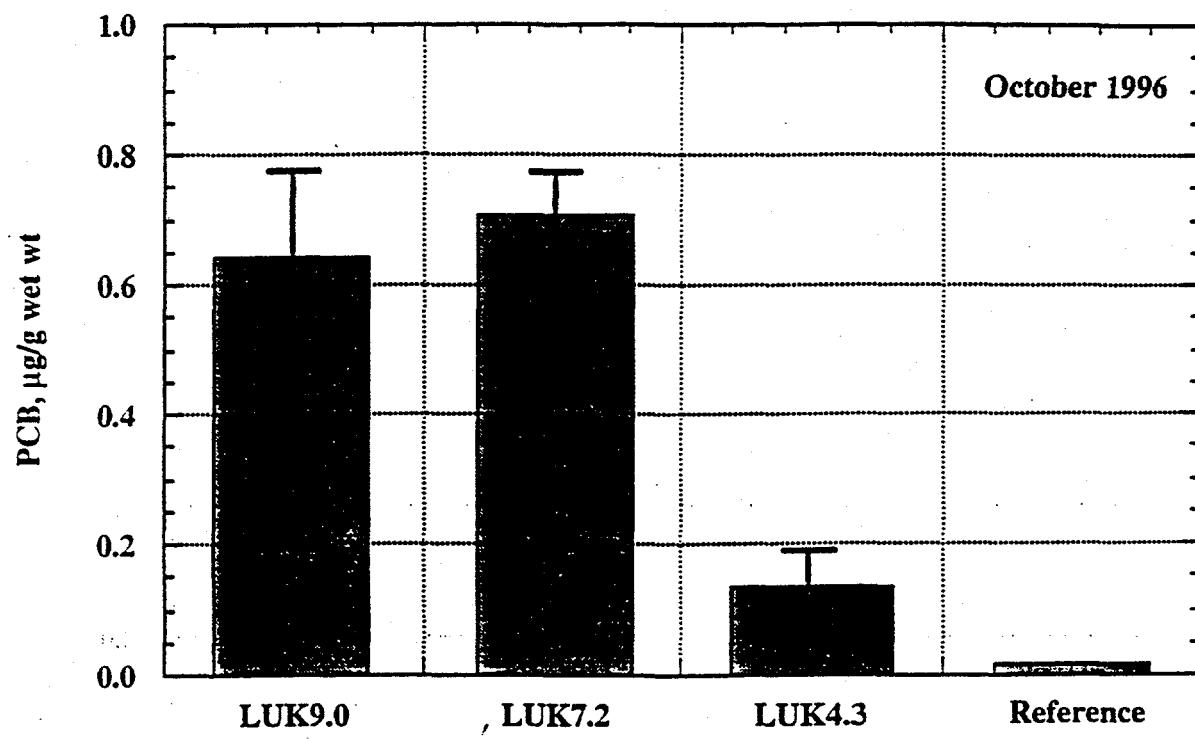


Fig. A-4.1. Average concentration of PCBs in longear sunfish filets, Little Bayou Creek. Error bars represent one standard error.

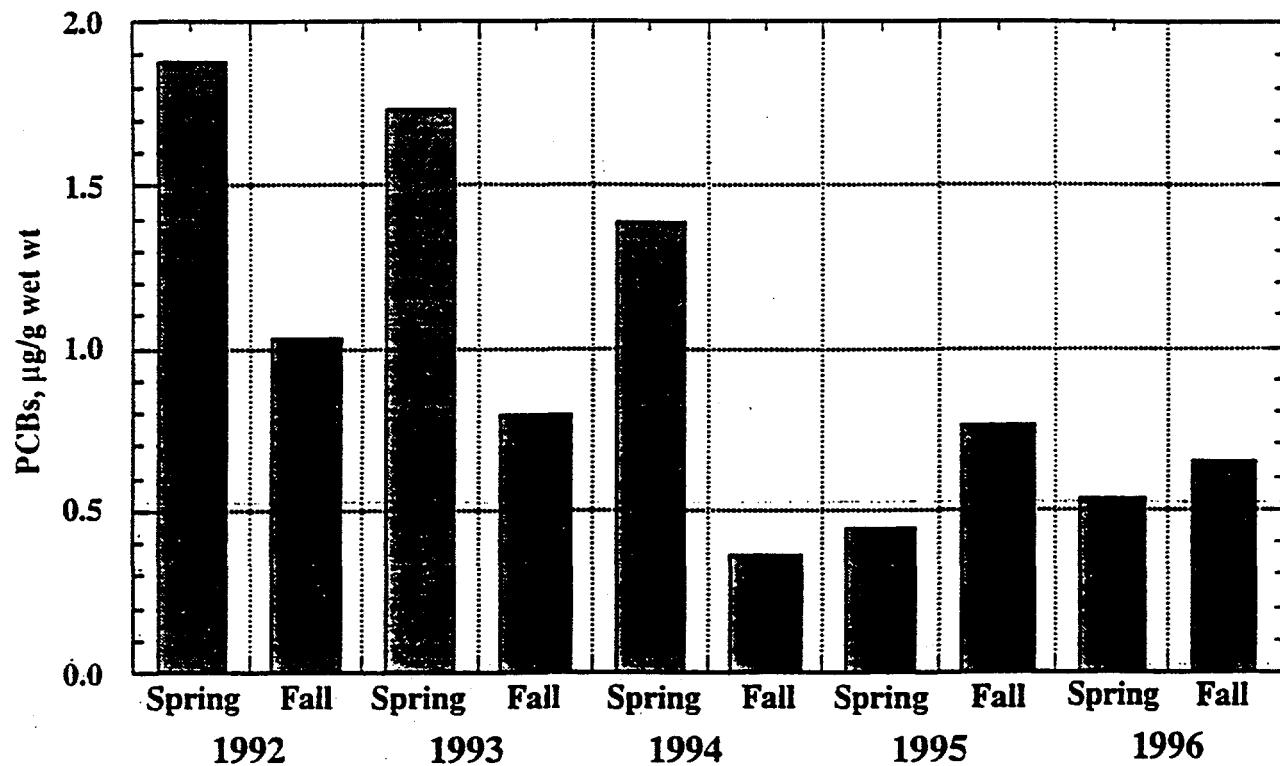


Fig. A-4.2. Average concentration of PCBs in longear sunfish in Little Bayou Creek at the site nearest PGDP (LUK 9.0), 1992-1996.

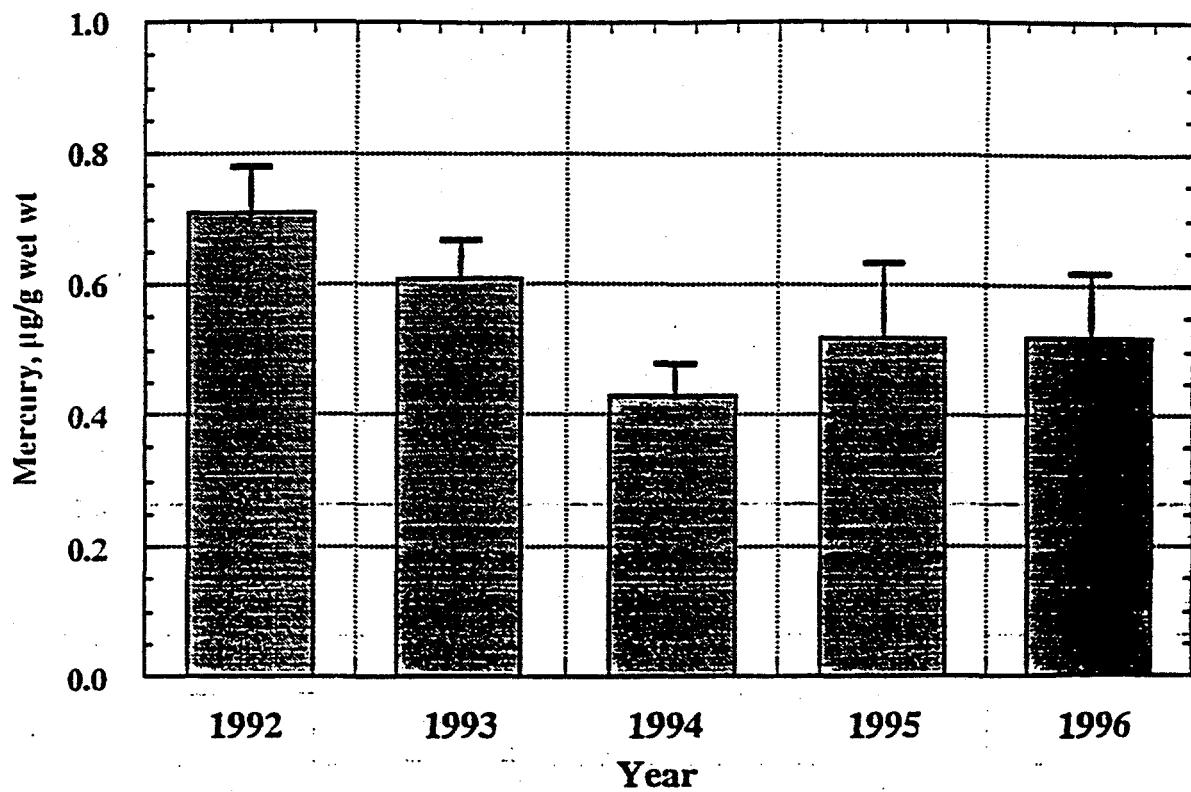


Fig. A-4.3. Mean concentration of mercury in filets of spotted bass from Big Bayou Creek near PGDP (BBK 9.1), 1992– 1996. Error bars represent one standard error.

Methylmercury concentrations were 0.12 and 0.10 ng/L at the two sites, respectively (G. R. Southworth, ORNL, unpublished data). Typical reference stream mercury concentrations in this study were 2.0–5.0 ng/L total mercury and 0.04–0.08 ng/L methylmercury. Thus, total mercury in Big Bayou Creek falls within the range typical of uncontaminated streams in East Tennessee and elsewhere in the United States, and well below the Environmental Protection Agency water quality criterion (12 ng/L). The accumulation of mercury in fish in this system appears to be greatly affected by the unusually high bioavailability of very low concentrations of mercury.

## **A-5. ECOLOGICAL MONITORING**

### **A-5.1 FISHES (*M. G. Ryon*)**

#### **A-5.1.1 Introduction**

Fish population and community studies can be used to assess the ecological effects of changes in water quality and habitat. These studies offer several advantages over other indicators of environmental quality (Karr et al. 1986, Karr 1987) and are especially relevant to assessment of the biotic integrity of Little Bayou and Big Bayou creeks. Monitoring of fish communities has been used by the Biological Monitoring and Abatement Program (BMAP) in ESD for receiving streams at ORNL (Loar et al. 1991), the East Tennessee Technology Park (Loar et al. 1992; Ryon 1993a), the Portsmouth, Ohio facility (Ryon 1994d), and the Y-12 Plant (Loar et al. 1989; Ryon 1992; Southworth et al. 1992), with some programs operational since 1984. Changes in the fish communities in these systems have indicated recovery (Ryon 1994a,c) as well as documented impacts (Ryon 1993b, 1994b).

The objectives of the instream fish monitoring task were (1) to characterize spatial and temporal patterns in the distribution and abundance of fishes in Little Bayou and Big Bayou creeks, (2) to document the effects of PGDP operations on fish community structure and function, and (3) to document any recovery of the community associated with remedial actions conducted by PGDP.

#### **A-5.1.2 Study Sites**

Quantitative sampling of the fish community was conducted at five sites. Three sites are located on Big Bayou Creek (BBK 12.5, BBK 10.0, and BBK 9.1; Fig. 2.1), one on Little Bayou Creek (LUK 7.2, Fig. 2.1), and one offsite reference station is located on Massac Creek (MAK 13.8, Fig. 2.2). MAK 13.8 was chosen as a reference site for BBK 9.1 and BBK 10.0. The upper site on Big Bayou Creek (BBK 12.5) was selected as a smaller reference site to be comparable to LUK 7.2.

#### **A-5.1.3 Materials and Methods**

Quantitative sampling of the fish populations was conducted by electrofishing on March 17–20, 1997. Data from these samples were used to estimate species richness, population size (numbers and biomass per unit area), and calculate annual production. Fish sampling sites either overlapped or were within 100 m of the sites included in the benthic macroinvertebrate monitoring task. All field sampling was conducted according to standard operating procedures (Schilling et al. 1996).

#### A-5.1.3.1 Field sampling procedures

All stream sampling was conducted using two or three Smith-Root backpack electrofishers, depending on stream size. Each unit can deliver up to 1200 V of pulsed direct current in order to stun fish. After 0.64-cm mesh seines were placed across the upper and lower boundaries of the fish sampling site to restrict fish movement, a five- to nine-person sampling team electrofished the site in an upstream direction on three consecutive passes. Stunned fish were collected and stored, by pass, in seine-net holding pens (0.64-cm diam mesh) or in buckets during further sampling.

Following the electrofishing, fish were anesthetized with MS-222 (tricaine methanesulfonate), identified, measured (total length), and weighed using Pesola spring scales. Individuals were recorded by 1-cm size classes and species. After ten individuals of a species-size class were measured and weighed, additional members of that size class were only measured. At sites with extremely high densities, specimens of some species might have only been counted after a sufficient number of lengths and weights had been obtained. Length-weight regressions based on the measured individuals were used to estimate missing length and weight data.

After processing fish from all passes, the fish were allowed to fully recover from the anesthesia and returned to the stream. Any additional mortality that occurred as a result of processing was noted at that time. Following completion of fish sampling, the length, mean width, mean depth, and pool:riffle ratio of the sampling reach were measured at each site.

#### A-5.1.3.2 Population data analysis

Quantitative species population estimates were calculated using the method of Carle and Strub (1978). Biomass was estimated by multiplying the population estimate by the mean weight per size class. To calculate density and biomass per unit area, total numbers and biomass were divided by the surface area (in square meters) of the study reach. These data were compiled and analyzed by a comprehensive Fortran 77 program developed by ESD staff (Railsback et al. 1989).

#### A-5.1.4 Results

The physical parameters of the sample sites showed the influence of the heavy rainfall and resulting increased stream flows that were prevalent during the spring of 1997. In general, stream depth and width was greater in spring 1997 than the previous spring (Table 5.1). In some cases, the higher flows resulted in a doubling of mean depth. Particularly at MAK 13.8, the greater depth and widths indicated conditions that were not favorable for obtaining a totally effective sample. However, because sampling conditions were unlikely to change in a reasonable time frame to allow useful comparisons to data collected at other sites, a spring sample was made at MAK 13.8 under these adverse conditions.

##### A-5.1.4.1 Species richness and composition

A total of 35 fish species were found at the 5 sites on Big Bayou Creek, Little Bayou Creek, and Massac Creek (Table A-5.2) for the March 1997 samples. BBK 9.1 and BBK 10.0 had 20 and 14 species for the spring sampling season, unusually high numbers for that time of the year. The number of species at BBK 9.1 is 1.3–1.8 times higher than spring species richness in 1995 and 1996 sampling (Ryon 1996, 1997) and generally higher than any other spring sample (Fig. A-5.1). A similar pattern is seen at BBK 10.0. The LUK 7.2 site had 16 species in the spring sample, while the comparable reference site, BBK 12.5 had 19 species. Like Big Bayou Creek sites, species richness at LUK 7.2 was higher than the two previous spring samples and is generally at a high level for the site (Fig. A-5.2). The core species assemblage at all sites included central

**Table A-5.1. Lengths, mean width, mean depth, surface area, and pool:riffle ratio of fish sampling sites in Big Bayou Creek, Little Bayou Creek, and a reference stream, Massac Creek, spring 1996 and 1997**

| Site <sup>a</sup>       | Length<br>(m) | Mean width<br>(m) | Mean depth<br>(cm) | Surface area<br>(m <sup>2</sup> ) | Pool:riffle<br>ratio |
|-------------------------|---------------|-------------------|--------------------|-----------------------------------|----------------------|
| <i>March 1997</i>       |               |                   |                    |                                   |                      |
| BBK 9.1                 | 95            | 8.6               | 24.8               | 822                               | 1.1                  |
| BBK 10.0                | 97            | 6.1               | 17.3               | 590                               | 0.6                  |
| BBK 12.5                | 101           | 7.1               | 18.2               | 718                               | 1.7                  |
| LUK 7.2                 | 102           | 3.9               | 13.2               | 395                               | 0.4                  |
| MAK 13.8                | 95            | 8.0               | 29.6               | 761                               | 3.8                  |
| <i>March-April 1996</i> |               |                   |                    |                                   |                      |
| BBK 9.1                 | 90            | 6.2               | 23.9               | 569                               | 1.1                  |
| BBK 10.0                | 101           | 5.1               | 12.0               | 514                               | 1.2                  |
| BBK 12.5                | 115           | 6.4               | 11.6               | 734                               | 1.7                  |
| LUK 7.2                 | 108           | 3.1               | 7.3                | 338                               | 0.4                  |
| MAK 13.8                | 99            | 5.8               | 18.7               | 572                               | 1.1                  |

<sup>a</sup>Site designations are Big Bayou Creek kilometer (BBK), Little Bayou Creek kilometer (LUK), and Massac Creek kilometer (MAK).

Table A- 5.2. Fish densities (number/m<sup>2</sup>) in Big Bayou Creek, Little Bayou Creek, and a reference stream, Massac Creek, March 1997

| Species <sup>b</sup>   | Sites <sup>a</sup>  |          |          |                   |
|------------------------|---------------------|----------|----------|-------------------|
|                        | BBK 9.1             | BBK 10.0 | BBK 12.5 | MAK 13.8          |
| Bowfin                 | < 0.01              | -        | -        | -                 |
| Gizzard shad           | < 0.01              | -        | -        | -                 |
| Stoneroller            | 0.01                | 0.55     | 0.39     | 0.01              |
| Red shiner             | -                   | -        | < 0.01   | -                 |
| Steelcolor shiner      | -                   | -        | -        | 0.03              |
| Miss. Silvery minnow   | -                   | -        | 0.01     | 0.02              |
| Redfin shiner          | -                   | -        | < 0.01   | < 0.01            |
| Golden shiner          | -                   | -        | 0.01     | < 0.01            |
| Bluntnose minnow       | -                   | 0.02     | 0.01     | -                 |
| Fathead minnow         | -                   | -        | -        | -                 |
| Creek chub             | -                   | 0.03     | 0.07     | < 0.01            |
| River carpsucker       | < 0.01              | -        | -        | -                 |
| White sucker           | < 0.01              | -        | -        | -                 |
| Creek chubsucker       | < 0.01              | -        | < 0.01   | < 0.01            |
| Smallmouth buffalo     | < 0.01 <sup>c</sup> | -        | -        | -                 |
| Black buffalo          | < 0.01 <sup>c</sup> | -        | -        | -                 |
| Spotted sucker         | 0.05                | < 0.01   | -        | < 0.01            |
| Golden redhorse        | 0.02                | -        | -        | < 0.01            |
| Black bullhead         | 0.01                | 0.01     | 0.01     | 0.01              |
| Yellow bullhead        | < 0.01              | < 0.01   | 0.03     | 0.01              |
| Grass pickerel         | 0.01                | < 0.01   | -        | < 0.01            |
| Pirate perch           | < 0.01              | < 0.01   | -        | 0.02              |
| Blackspotted topminnow | 0.01                | 0.04     | 0.03     | 0.14              |
| Western mosquitofish   | -                   | -        | -        | 0.01              |
| Green sunfish          | 0.03                | 0.05     | 0.21     | 0.22              |
| Warmouth               | < 0.01              | -        | < 0.01   | 0.05              |
| Bluegill               | 0.04                | 0.04     | 0.03     | 0.01              |
| Longear sunfish        | 0.15                | 0.13     | 0.35     | 0.10              |
| Redspotted sunfish     | -                   | -        | -        | < 0.01            |
| Hybrid sunfish         | -                   | < 0.01   | < 0.01   | 0.01              |
| Spotted bass           | 0.01                | -        | < 0.01   | -                 |
| Largemouth bass        | -                   | < 0.01   | < 0.01   | -                 |
| Bluntnose darter       | -                   | -        | < 0.01   | -                 |
| Slough darter          | -                   | < 0.01   | < 0.01   | 0.01              |
| Logperch               | -                   | -        | -        | < 0.01            |
| Blackside darter       | -                   | -        | -        | 0.01              |
| Species richness       | 20                  | 14       | 19       | 16                |
| Total density          | 0.36                | 0.88     | 1.16     | 1.26              |
|                        |                     |          |          | 0.18 <sup>d</sup> |

<sup>a</sup>BBK = Big Bayou Creek kilometer, LUK = Little Bayou Creek kilometer, MAK = Massac Creek kilometer.<sup>b</sup>Common and scientific names according to the American Fisheries Society (Robins et al. 1991).<sup>c</sup>Species identification confirmed by Dr. David A. Etnier, Department of Zoology, University of Tennessee.<sup>d</sup>Sample was affected by high water with turbid conditions. Density was much lower than normal due to these adverse collecting conditions. Species richness was appropriate for site despite higher than normal flow.

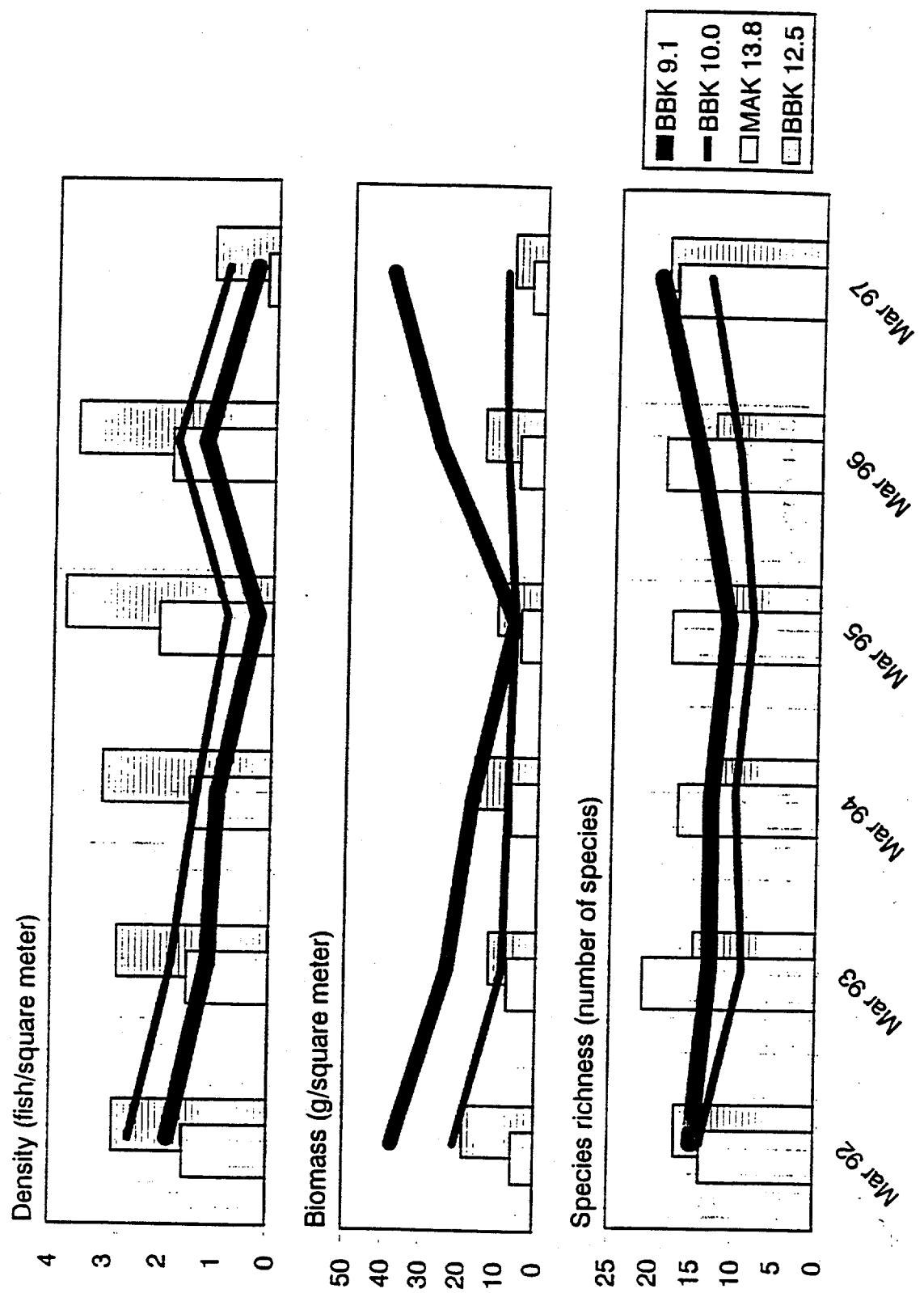


Fig. A-5.1. Species richness, biomass, and density at Big Bayou Creek and reference sites.

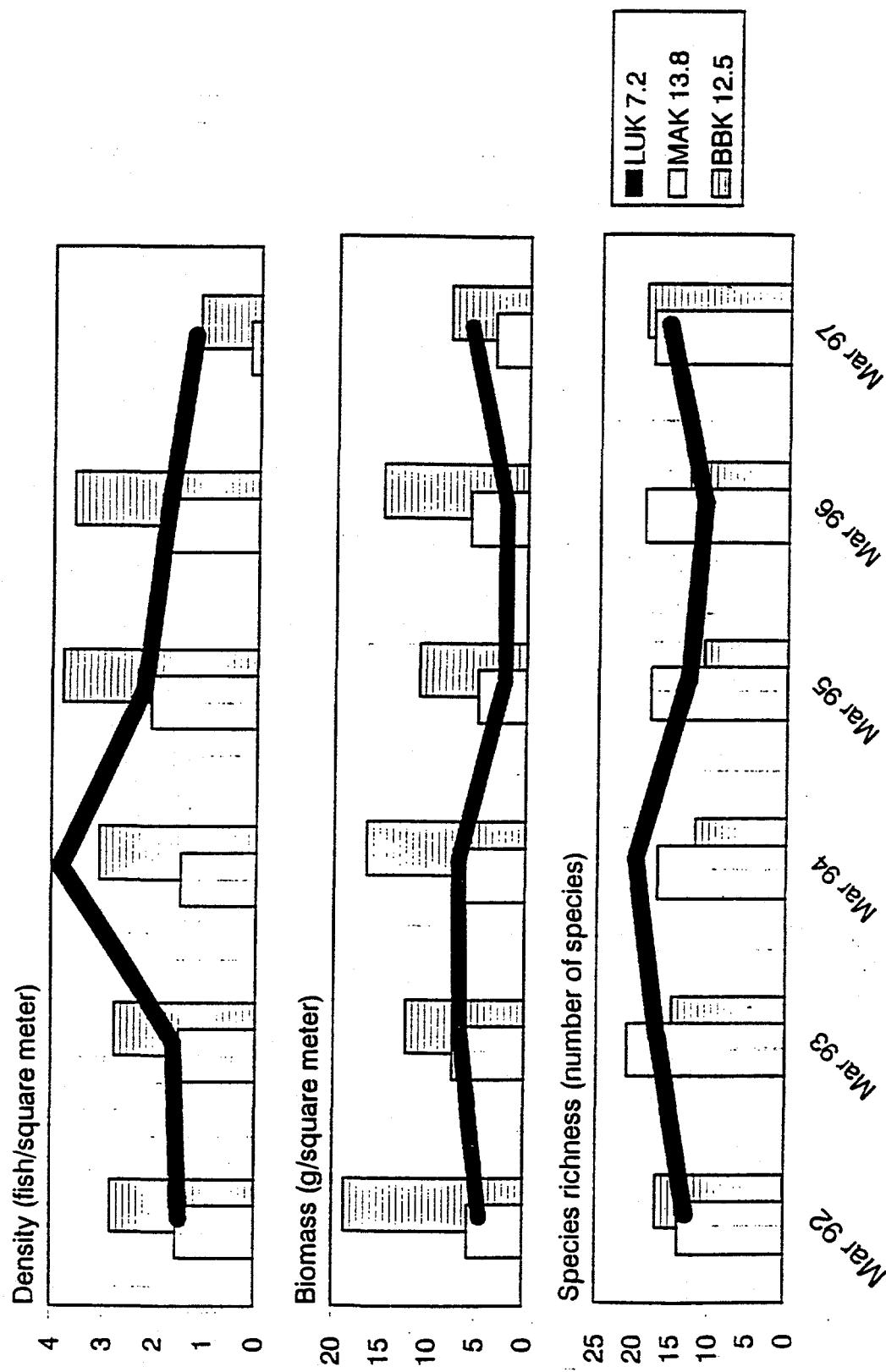


Fig. A-5.2. Species richness, biomass, density at Little Bayou Creek and reference sites.

stoneroller (*Campostoma anomalum*), creek chub (*Semotilus atromaculatus*), yellow bullhead (*Ameiurus natalis*), blackspotted topminnow (*Fundulus olivaceus*), mosquitofish (*Gambusia affinis*), green sunfish (*Lepomis cyanellus*), bluegill (*L. macrochirus*), and longear sunfish (*L. megalotis*). Four species were judged to be sensitive to water quality and/or habitat degradation (Karr et al. 1986; Ohio EPA 1987, 1988) and eight were rated as tolerant to such conditions (Appendix D, Table D.1, Kszos 1997). Noticeable in this spring sample was the considerable number of sucker species, especially at BBK 9.1, the presence of the black bullhead (*Ameiurus melas*) at all sites, and the occurrence of the fathead minnow (*Pimephales promelas*) at LUK 7.2. Two rare species, the black buffalo (*Ictiobus niger*) and the redspotted sunfish (*Lepomis miniatus*), were collected at BBK 9.1 and LUK 7.2, respectively.

At the most downstream site on Big Bayou Creek, BBK 9.1, several species were collected that probably were moving into the site from larger downstream sections of Big Bayou Creek. The river carpsucker (*Carpoides carpio*), smallmouth buffalo (*Ictiobus bubalus*), bowfin (*Amia calva*), grass pickerel (*Esox americanus*), and black buffalo were never or only rarely collected previously at this site. The heavy rainfall and associated high water levels in spring 1997 near Paducah suggests that these species were either seeking refugee from turbulent conditions or were moving upstream for spawning. This upstream displacement of species may have been responsible in part for the increased species richness at all Big Bayou watershed sites.

At BBK 10.0 the increased species richness included a sucker and a darter, species more sensitive to stress than normally found at the site. The continued presence of the grass pickerel at BBK 10.0 might indicate improving conditions, since it was not found at this site prior to 1996.

The LUK 7.2 site also had a couple of unique species occurrences not always seen previously at the site. The redspotted sunfish was found at the site, as well as a large number of fathead minnows. The fathead minnow had not been seen in Little Bayou Creek prior to this spring sample and this occurrence was a result of 'bait bucket' type of introduction. The individual fathead minnows were a light orange-gold in color, a hue seen only in stock bred for sale in bait shops. Most likely, some fisherman released a fair number of these shortly before our sample.

#### A-5.1.4.2 Density

Quantitative estimates of density were lower in this spring sample than during previous spring samples (Tables A-5.2 and A-5.3; Figs. A-5.1 and A-5.2). The low densities were probably related to the flushing that occurred as a result of the high rainfall in spring 1997. This trend was seen at the reference sites also. Particularly at MAK 13.8 where high water levels and turbid conditions made sampling difficult, the spring density values may be somewhat suspect and should be compared to past trends with some reservations.

#### A-5.1.4.3 Biomass

The biomass levels seem to be less affected by high water conditions than the densities. At BBK 9.1, BBK 10.0, and LUK 7.2, spring biomass was similar to or higher than recent spring samples (Figs. 5.1 and 5.2). The disconnect between the density and biomass patterns reflects the dependence by the density measure on numbers of small fish or individuals. These sizes are more easily influenced by high water than the larger size classes that contribute more to the biomass metric. For example, at BBK 9.1 the influx of larger sucker species was not enough to maintain the density losses relating to displacement of smaller sunfish and minnow species. However, their large size translated to an increase in overall biomass.

#### A-5.1.5 Discussion

Data on the fish communities of Big Bayou Creek and Little Bayou Creek downstream of PGDP were compared to data from reference sites located on Big Bayou Creek above PGDP and on Massac Creek. These comparisons did not indicate a noticeable degradation in the communities downstream of PGDP. Further comparisons to previous spring data at these sites indicated an improvement for many parameters, such as species richness or presence of sensitive species. However, these improvements must be put in perspective. The spring 1997 sample was made during a period when heavy rainfall and high flows disrupted the normal community balance in streams near the Paducah facilities. This disruption included lower density values and the presence of unusual species; these patterns were seen in reference as well as study sites.

The pattern of improvement is likely a reflection of this physical disruption caused by the increased flows as much as any improvement in water quality of plant discharges. If a lasting improvement is occurring, then it should be apparent in further sampling that would extend the comparisons beyond this period of unusual water conditions.

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### A-5.2 BENTHIC MACROINVERTEBRATES (*J. G. Smith*)

#### A-5.2.1 Introduction

Benthic macroinvertebrates are those organisms lacking spinal columns that are large enough to be seen without the aid of magnification and that live on or among the substrate particles of flowing and non-flowing bodies of water. The limited mobility and relatively long life spans (a few months to more than a year) of most taxa make them ideal for use in following long-term ecological trends associated with natural or unnatural changes in the environment (Platts et al. 1983). Thus, the composition and structure of the benthic macroinvertebrate community reflects the relatively recent past and can be considerably more informative than methods that rely solely on water quality analyses.

The objectives of the benthic macroinvertebrate monitoring task are to evaluate the condition of benthic macroinvertebrate communities in Big Bayou Creek and Little Bayou Creek, and identify trends that occur that may be associated with operations or remedial actions at the PGDP.

#### A-5.2.2 Study Sites

Benthic macroinvertebrate samples have been routinely collected since 1991 from three sites on Big Bayou Creek (BBK 9.1, BBK 10.0, and BBK 12.5), and one site each on Little Bayou Creek (LBK 7.2) and Massac Creek (MAK 13.8) (Figs. 2.1 and 2.2). Sites BBK 12.5 and MAK 13.8 serve as reference sites and are not known to be negatively affected by PGDP activities.

Table A-5.3. Fish biomass (g/m<sup>2</sup>) in Big Bayou Creek, Little Bayou Creek, and a reference stream, Massac Creek, March 1997.

| Species <sup>b</sup>   | Sites <sup>a</sup> |         |         |        |         |
|------------------------|--------------------|---------|---------|--------|---------|
|                        | BBK9.1             | BBK10.0 | BBK12.5 | LUK7.2 | MAK13.8 |
| Bowfin                 | 1.95               | -       | -       | -      | -       |
| Gizzard shad           | 0.36               | -       | -       | -      | -       |
| Stoneroller            | 0.09               | 5.04    | 1.86    | 0.69   | 0.06    |
| Red shiner             | -                  | -       | 0.01    | 0.08   | -       |
| Steelcolor shiner      | -                  | -       | -       | -      | 0.08    |
| Miss. Silvery minnow   | -                  | -       | 0.02    | -      | 0.03    |
| Redfin shiner          | -                  | -       | 0.01    | -      | <0.01   |
| Golden shiner          | -                  | -       | 0.03    | -      | 0.03    |
| Bluntnose minnow       | -                  | 0.10    | 0.03    | 0.78   | -       |
| Fathead minnow         | -                  | -       | -       | 0.29   | -       |
| Creek chub             | -                  | 0.98    | 1.00    | 2.12   | 0.02    |
| River carpsucker       | 1.68               | -       | -       | -      | -       |
| White sucker           | 0.26               | -       | -       | -      | -       |
| Creek chubsucker       | 0.39               | -       | 0.31    | -      | 0.06    |
| Smallmouth buffalo     | 0.13               | -       | -       | -      | -       |
| Black buffalo          | 0.10               | -       | -       | -      | -       |
| Spotted sucker         | 15.62              | 0.16    | -       | -      | 1.54    |
| Golden redhorse        | 13.22              | -       | -       | -      | 1.07    |
| Black bullhead         | 0.14               | 0.14    | 0.13    | 0.53   | -       |
| Yellow bullhead        | 0.14               | 0.05    | 1.11    | 0.10   | 0.01    |
|                        |                    |         |         |        |         |
| Grass pickerel         | 0.16               | 0.07    | -       | -      | 0.04    |
| Pirate perch           | 0.01               | 0.02    | -       | 0.16   | 0.01    |
| Blackspotted topminnow | 0.01               | 0.05    | 0.06    | 0.15   | 0.04    |
|                        |                    |         |         |        |         |
| Western mosquitofish   | -                  | -       | -       | <0.01  | -       |
| Green sunfish          | 0.10               | 0.19    | 0.63    | 0.67   | 0.23    |
| Warmouth               | 0.01               | -       | 0.01    | 0.16   | -       |
| Bluegill               | 1.10               | 0.11    | 0.27    | 0.13   | 0.02    |
| Longear sunfish        | 3.24               | 2.83    | 2.56    | 0.09   | 0.22    |
| Redspotted sunfish     | -                  | -       | -       | 0.02   | -       |
| Hybrid sunfish         | -                  | 0.03    | 0.08    | 0.05   | -       |
| Spotted bass           | 1.16               | -       | 0.01    | -      | -       |
| Largemouth bass        | -                  | 0.35    | 0.01    | -      | -       |
| Bluntnose darter       | -                  | -       | <0.01   | -      | -       |
| Slough darter          | -                  | <0.01   | <0.01   | 0.01   | -       |
| Logperch               | -                  | -       | -       | -      | 0.03    |
| Blackside darter       | -                  | -       | -       | -      | 0.03    |
|                        |                    |         |         |        |         |
| Total biomass          | 39.87              | 10.12   | 8.14    | 6.03   | 3.52    |

<sup>a</sup>BBK = Big Bayou Creek kilometer, LUK = Little Bayou Creek kilometer, MAK = Massac Creek kilometer.

<sup>b</sup>Common and scientific names according to the American Fisheries Society (Robins et al. 1991).

#### A-5.2.3 Materials and Methods

During each sampling period, three random samples were collected with a Surber sampler ( $0.09\text{ m}^2$  or  $1\text{ ft}^2$ ) equipped with a  $363\text{-}\mu\text{m}$  mesh net. Samples were collected from riffles only because this type of habitat often possesses the greatest variety of benthic organisms (e.g., Hynes 1970, Platts et al. 1983), and limiting collections to a single type of habitat reduces inter-sample variability (e.g., Plafkin et al. 1989; Resh and McElravy 1993). Samples were placed in prelabeled, polyurethane-coated, glass jars and preserved with 95% ethyl alcohol. To prevent sample decomposition due to dilution of the original preservative, the ethanol in each jar was replaced within seven days of collection.

Just before sample collection, water depth, location within the riffle (distance from permanent head-stakes on the stream bank), visual estimate of the relative current velocity (very slow, slow, moderate, or fast), and substrate types (visual estimate) based on a modified Wentworth particle size scale (Loar et al. 1985), were recorded for each sample. A detailed description of the procedures employed for site evaluation and sample collection, storage, and maintenance can be found in Smith and Smith (1995).

In the laboratory, each sample was first placed in a U.S. Standard No. 60-mesh ( $250\text{-}\mu\text{m}$  openings) sieve and rinsed with tap water. Small aliquots of a sample were then placed in a white tray partially filled with water, and the organisms were removed from the sample debris with forceps. This process was repeated with the remaining sample until it was entirely sorted. Finally, organisms were identified to the lowest practical taxon and enumerated. Details of laboratory sample processing procedures are available in Smith and Smith (1995).

Data were managed and all descriptive statistics were calculated using Statistical Analysis System software and procedures (SAS 1985a, 1985b).

#### 5.2.4 Results

Following a revision of the sampling plan in late fiscal year 1996, samples were collected in March and September 1997, but only those samples collected in March have been processed; samples collected in September will be held in controlled storage for a minimum of two years and processed only if needed.

A summary of the results for samples collected in March 1997 is presented in Fig. A-5.3; also included in Fig. A-5.3 are the results for each March sampling period since 1992. Total density, total richness (number of taxa/sample), and richness of the Ephemeroptera, Plecoptera, and Trichoptera (number of EPT taxa/sample) were all dramatically lower than in most previous years in March except 1993. Lowest values for total and EPT richness were observed at BBK 10.0 where the total number of taxa and number of EPT taxa were at least 50% less than at the other four sites. The dramatic reduction in values in 1997 was probably associated with the heavy rains that plagued the Midwest during the winter and spring months. Dramatic shifts from the previous sampling period (September 1996) were observed in the substrate at all sampling sites, indicating that a significant quantity of water had scoured the stream (J. G. Smith, Environmental Sciences Division, Oak Ridge National Laboratory, personal observation, March 17, 1997). Although major shifts appear to frequently occur in the substrate at most sampling locations, this was the first time since the project began in September 1991 that a major shift in the substrate was observed at BBK 12.5. The low values for density, total richness, and EPT richness observed in March 1993 were also attributed to

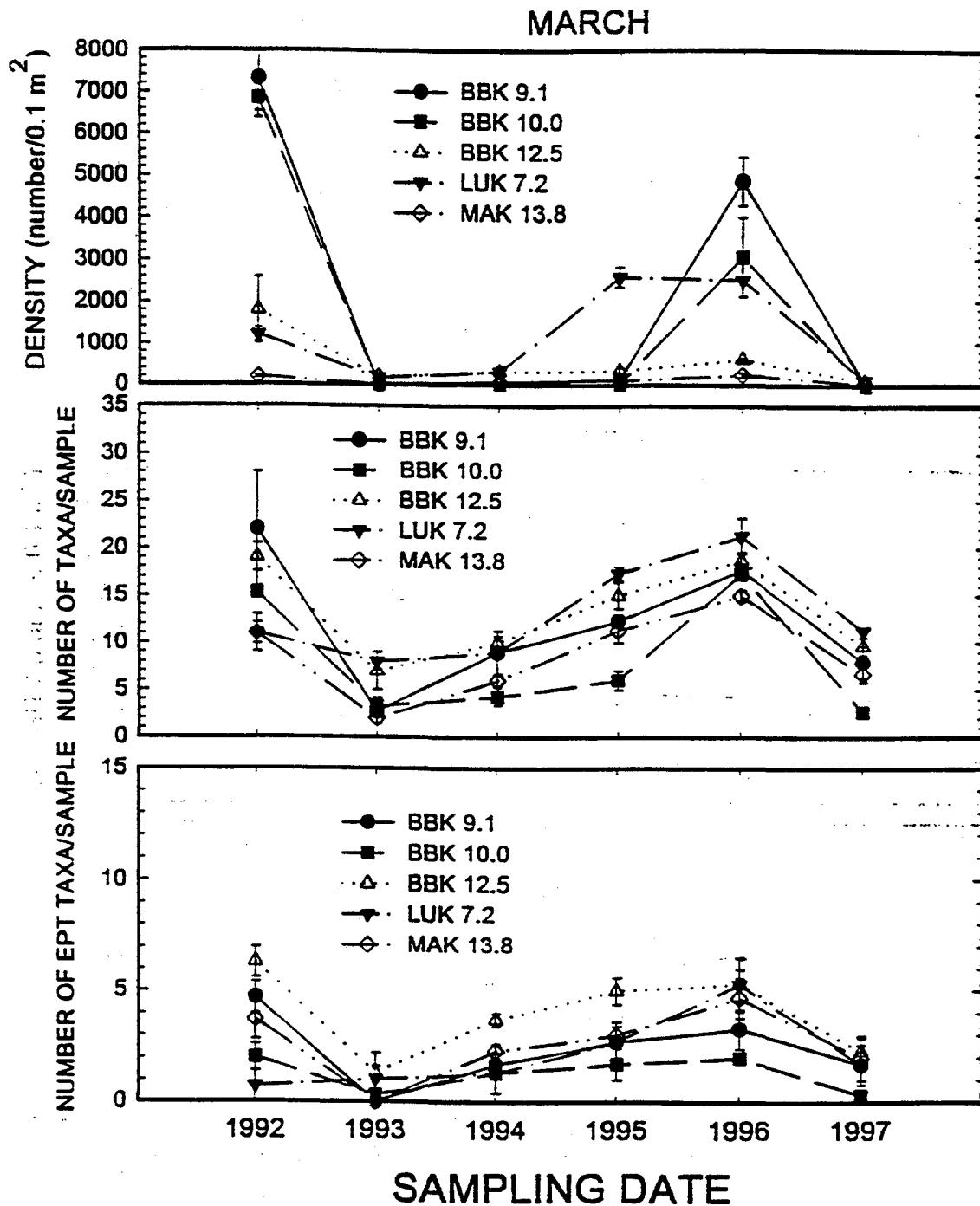


Fig. 5.3. Total density, total taxonomic richness (number of taxa/sample), and richness of the Ephemeroptera, Plecoptera, and Trichoptera taxa (number of EPT taxa/sample) in Big Bayou Creek, Little Bayou Creek, and Massac Creek. Values are the means  $\pm 1$  SE. BBK = Big Bayou Creek kilometer, LUK = Little Bayou Creek kilometer, MAK = Massac Creek kilometer.

heavy rains in the previous February (Smith 1996). Spates that cause large increases in stream discharge can devastate a macroinvertebrate community due, supposedly, to drift away from an area and mortality associated movement of the substrate (McElravy et al. 1989). The rate of recovery will depend upon the availability of potential recolonizing organisms.

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**Appendix B**

**RAINFALL**

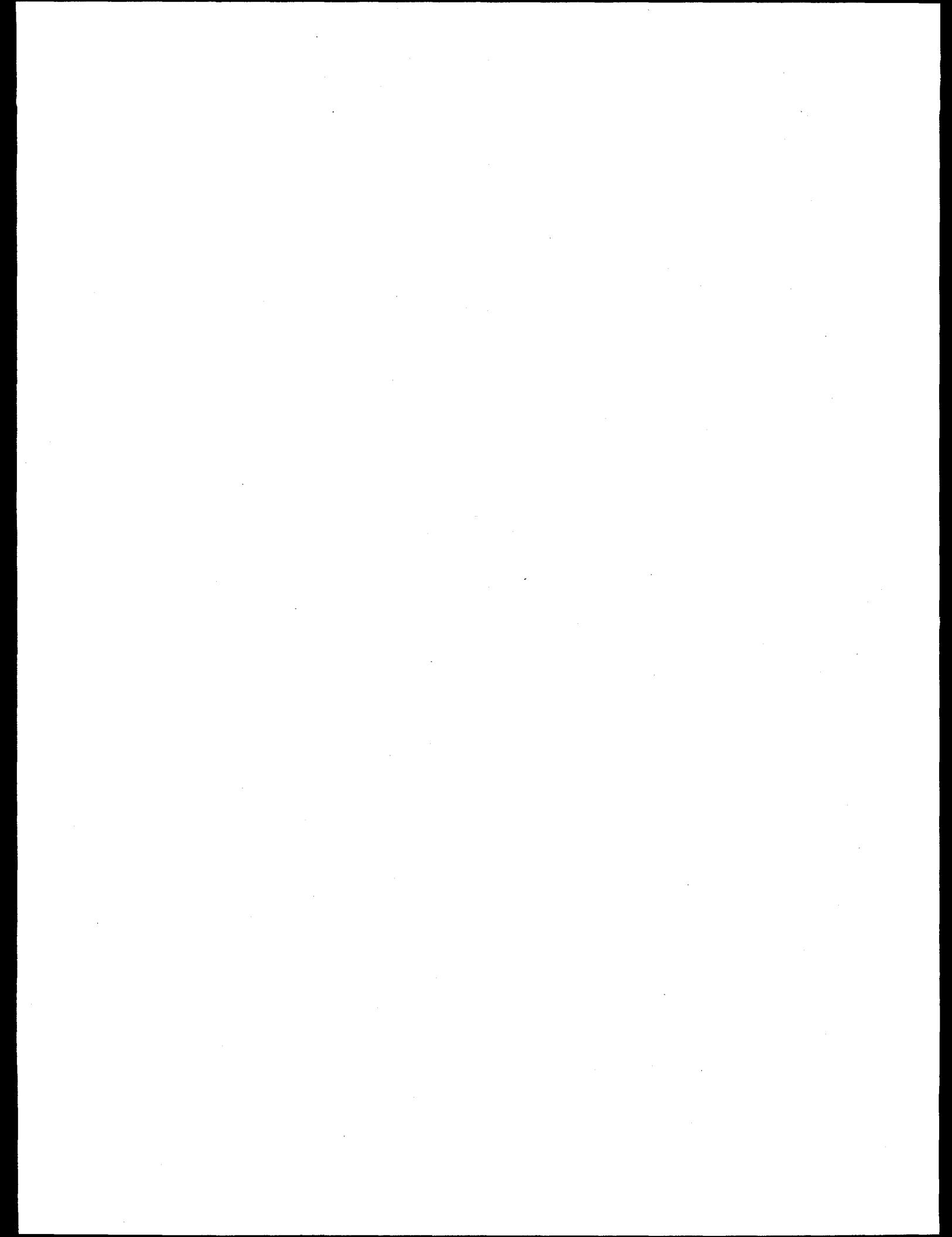


Table B.1. Daily precipitation for 1997 in Paducah Kentucky

| Month    | Day | Precipitation (cm) |
|----------|-----|--------------------|
| January  | 4   | 0.711              |
|          | 8   | 0.508              |
|          | 9   | 0.179              |
|          | 10  | 0.051              |
|          | 15  | 2.718              |
|          | 21  | 0.051              |
|          | 22  | 1.397              |
|          | 24  | 0.051              |
| February | 27  | 0.991              |
|          | 3   | 2.870              |
|          | 4   | 0.026              |
|          | 7   | 0.660              |
|          | 8   | 0.051              |
|          | 13  | 0.635              |
|          | 19  | 0.076              |
|          | 20  | 0.102              |
| March    | 21  | 1.295              |
|          | 26  | 3.277              |
|          | 28  | 4.521              |
|          | 1   | 9.070              |
|          | 2   | 0.740              |
|          | 3   | 2.840              |
|          | 9   | 1.372              |
|          | 13  | 1.245              |
| April    | 17  | 0.178              |
|          | 18  | 1.727              |
|          | 25  | 1.270              |
|          | 28  | 0.279              |
|          | 30  | 0.127              |
|          | 4   | 1.727              |
|          | 5   | 3.708              |
|          | 11  | 1.880              |
|          | 12  | 1.194              |
|          | 19  | 0.076              |
|          | 20  | 0.889              |
|          | 21  | 0.787              |

Table B.1 (continued)

| Month     | Day | Precipitation (cm) |
|-----------|-----|--------------------|
| April     | 22  | 0.102              |
|           | 26  | 0.533              |
|           | 27  | 1.600              |
|           | 30  | 0.279              |
| May       | 2   | 8.763              |
|           | 8   | 1.803              |
|           | 14  | 0.787              |
|           | 18  | 0.178              |
|           | 19  | 2.464              |
|           | 24  | 0.051              |
|           | 26  | 0.229              |
|           | 27  | 0.279              |
|           | 28  | 5.283              |
|           | 30  | 0.127              |
| June      | 31  | 0.229              |
|           | 1   | 1.880              |
|           | 5   | 0.025              |
|           | 6   | 1.346              |
|           | 7   | 0.356              |
|           | 8   | 0.432              |
|           | 13  | 3.937              |
|           | 16  | 0.076              |
|           | 17  | 0.864              |
|           | 21  | 0.762              |
|           | 22  | 0.356              |
|           | 27  | 0.305              |
| July      | 4   | 0.686              |
|           | 8   | 4.775              |
|           | 14  | 0.584              |
|           | 28  | 1.016              |
| August    | 8   | 1.219              |
|           | 9   | 0.381              |
|           | 12  | 0.229              |
|           | 14  | 0.279              |
|           | 15  | 1.930              |
|           | 19  | 1.829              |
|           | 26  | 1.219              |
| September | 30  | 0.178              |
|           | 2   | 0.102              |

Table B.1 (continued)

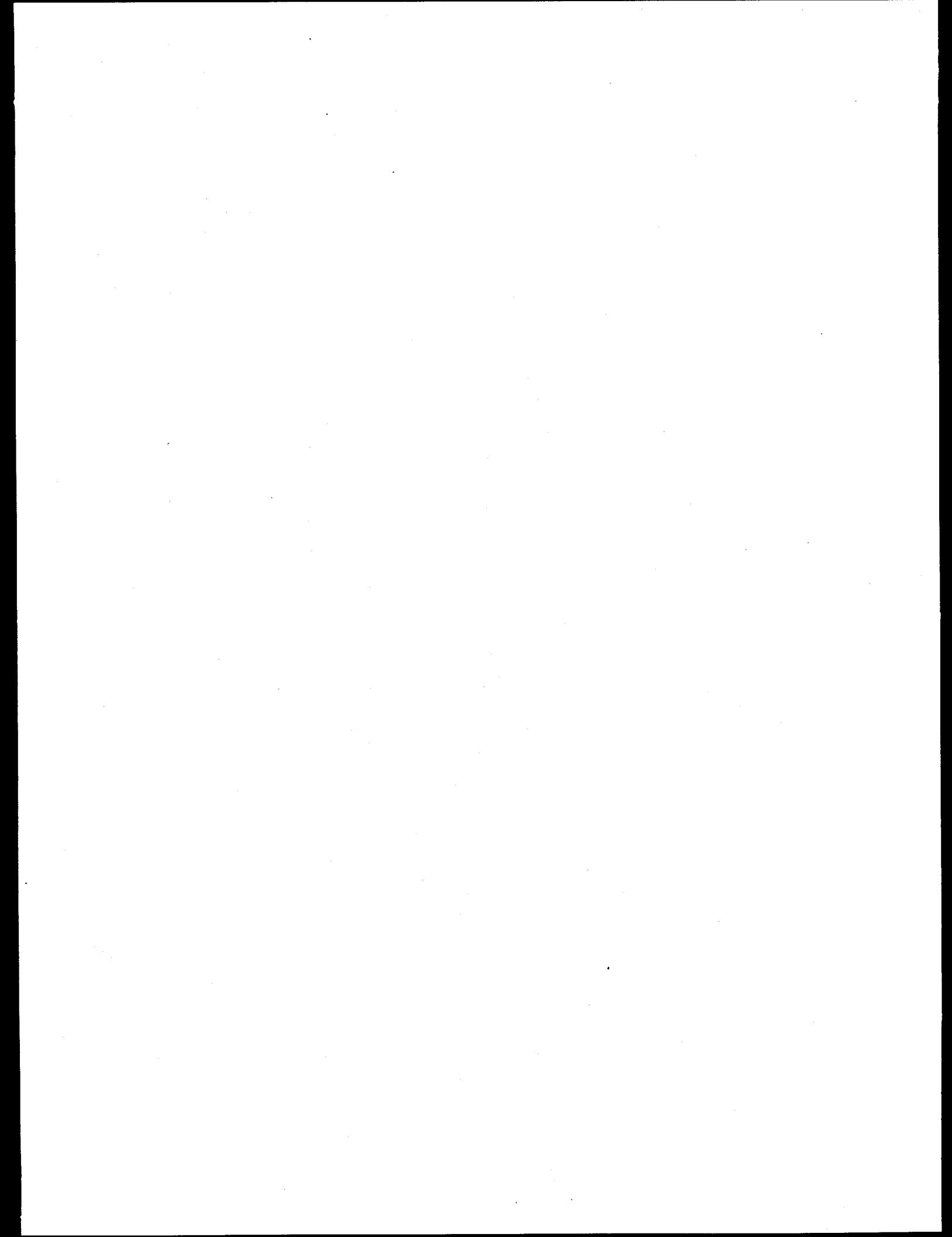
| Month     | Day | Precipitation (cm) |
|-----------|-----|--------------------|
| September | 3   | 1.245              |
|           | 8   | 0.102              |
|           | 20  | 2.311              |
|           | 23  | 1.702              |
|           | 24  | 0.584              |
|           | 28  | 0.076              |
| October   | 7   | 0.127              |
|           | 13  | 2.489              |
|           | 21  | 0.914              |
|           | 24  | 0.838              |
|           | 25  | 1.397              |
|           | 26  | 0.965              |
|           | 31  | 0.660              |
| November  | 1   | 0.178              |
|           | 2   | 0.025              |
|           | 3   | 0.279              |
|           | 5   | 0.432              |
|           | 6   | 0.965              |
|           | 10  | 0.051              |
|           | 13  | 1.829              |
|           | 21  | 0.127              |
|           | 29  | 0.178              |
|           | 30  | 1.270              |
| December  | 3   | 0.965              |
|           | 4   | 0.025              |
|           | 8   | 0.457              |
|           | 9   | 1.270              |
|           | 11  | 0.102              |
|           | 21  | 0.381              |
|           | 22  | 0.305              |
|           | 24  | 1.803              |
|           | 25  | 0.025              |
|           | 26  | 0.229              |
|           | 30  | 0.127              |

*Note:* Only days with measurable precipitation are shown

*Source:* Midwestern Climate Center, Champaign, IL, Station ID156110, Barkley Regional Airport, Paducah National Weather Service.

**Appendix C**

**TOXICITY TEST SUMMARIES PROVIDED  
TO THE  
KENTUCKY DIVISION OF WATER**



## Continuous Outfalls

Results of a *Pimephales promelas* Chronic Toxicity Test Conducted 03/07-14/97 Using Effluent from Outfall 001

| Test Solution                  | Percent Surviving (day) |      |      |      |      |      |      | Dry Weight (mg/L)  |       |
|--------------------------------|-------------------------|------|------|------|------|------|------|--|-------|
|                                | 1                       | 2    | 3    | 4    | 5    | 6    | 7    | Total  | Mean  |
| Control                        | 95.0                    | 95.0 | 92.5 | 90.0 | 90.0 | 87.5 | 87.5 | 40   | 0.475 |
| 100% Effluent                  | 100                     | 95.0 | 90.0 | 75.0 | 75.0 | 67.5 | 62.5 | 40   | 0.376 |
| 50% Effluent                   | 95.0                    | 95.0 | 92.5 | 85.0 | 72.5 | 62.5 | 59.0 | 39*  | 0.348 |
| 25% Effluent                   | 97.5                    | 92.5 | 85.0 | 77.5 | 67.5 | 57.5 | 55.0 | 40   | 0.363 |
| 12% Effluent                   | 97.5                    | 95.0 | 85.0 | 75.0 | 72.5 | 72.5 | 65.0 | 40   | 0.386 |
| 6% Effluent                    | 100                     | 97.5 | 95.0 | 92.5 | 87.5 | 85.0 | 85.0 | 40   | 0.487 |
| IC <sub>50</sub> Value: > 100% |                         |      |      |      |      |      |      | Calculated TUC Value: < 1.0  |       |
| 95% Confidence Limits: NA      |                         |      |      |      |      |      |      | Permit Limits: TUC ≥ 1.0   |       |
| UL:                            |                         |      |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |
| LL:                            |                         |      |      |      |      |      |      |  |       |
| UL = Upper Limit               |                         |      |      |      |      |      |      |  |       |
| LL = Lower Limit               |                         |      |      |      |      |      |      |  |       |

\* One test organism was missing on Day 7; therefore, percent surviving and mean growth were based on 39 test organisms.

Results of a *Pimephales promelas* Chronic Toxicity Test, 03/07-14/97 Using Effluent from Outfall 006

| Test Solution                  | Percent Surviving (day) |      |      |      |      |      |      | Dry Weight (mg/L)  |       |
|--------------------------------|-------------------------|------|------|------|------|------|------|--|-------|
|                                | 1                       | 2    | 3    | 4    | 5    | 6    | 7    | Total  | Mean  |
| Control*                       | 95.0                    | 95.0 | 92.5 | 90.0 | 90.0 | 87.5 | 87.5 | 40   | 0.475 |
| 100% Effluent                  | 100                     | 97.5 | 92.5 | 82.5 | 82.5 | 82.5 | 80.0 | 40   | 0.493 |
| 50% Effluent                   | 97.5                    | 97.5 | 92.5 | 87.5 | 87.5 | 87.5 | 80.0 | 40   | 0.439 |
| 25% Effluent                   | 100                     | 97.5 | 90.0 | 87.5 | 87.5 | 85.0 | 85.0 | 40   | 0.360 |
| 12% Effluent                   | 97.5                    | 97.5 | 90.0 | 90.0 | 90.0 | 87.5 | 85.0 | 40   | 0.486 |
| 6% Effluent                    | 100                     | 97.5 | 90.0 | 87.5 | 87.5 | 87.5 | 85.0 | 40   | 0.485 |
| IC <sub>50</sub> Value: > 100% |                         |      |      |      |      |      |      | Calculated TUC Value: < 1.0  |       |
| 95% Confidence Limits: NA      |                         |      |      |      |      |      |      | Permit Limits: TUC ≥ 1.0   |       |
| UL:                            |                         |      |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |
| LL:                            |                         |      |      |      |      |      |      |  |       |
| UL = Upper Limit               |                         |      |      |      |      |      |      |  |       |
| LL = Lower Limit               |                         |      |      |      |      |      |      |  |       |

\*The controls for this outfall were invalid due to low survival (60%); however, the controls for Outfall 001 were used to calculate the IC<sub>50</sub> since all the larvae used in the toxicity tests were obtained from the same batch.

**C-4 -- Biological Monitoring Program**

**Results of a *Pimephales promelas* Chronic Toxicity Test, 03/07-14/97 Using Effluent from Outfall 009**

| Test Solution                                      | Percent Surviving (day)     |      |      |      |      |      |      | Dry Weight (mg/L)  |       |
|--|-----------------------------|------|------|------|------|------|------|--|-------|
|  | 1                           | 2    | 3    | 4    | 5    | 6    | 7    | Total  | Mean  |
| Control  | 100                         | 95.0 | 92.5 | 92.5 | 92.5 | 90.0 | 90.0 | 40   | 0.387 |
| 100% Effluent                                      | 100                         | 95.0 | 82.5 | 82.5 | 82.5 | 82.5 | 82.5 | 40   | 0.464 |
| 50% Effluent                                       | 95.0                        | 92.5 | 87.5 | 82.5 | 82.5 | 82.5 | 77.5 | 40   | 0.456 |
| 25% Effluent                                       | 100                         | 90.0 | 85.0 | 85.0 | 85.0 | 85.0 | 85.0 | 40   | 0.460 |
| 12% Effluent                                       | 100                         | 95.0 | 87.5 | 87.5 | 85.0 | 82.5 | 80.0 | 40   | 0.461 |
| 6% Effluent  | 100                         | 97.5 | 92.5 | 92.5 | 92.5 | 90.0 | 90.0 | 40   | 0.495 |
| IC <sub>50</sub> Value: > 100%                     | Calculated TUc Value: < 1.0 |      |      |      |      |      |      |  |       |
| 95% Confidence Limits: NA                          |                             |      |      |      |      |      |      | Permit Limits: TUc ≥ 1.0   |       |
| UL:<br>LL:<br>UL = Upper Limit<br>LL = Lower Limit |                             |      |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |

**Results of a *Ceriodaphnia dubia* Chronic Toxicity Test, 03/07-14/97 Using Effluent from Outfall 001**

| Test Solution                                      | Percent Surviving (day)     |     |     |     |     |     | No. of Young |  |  |
|--|-----------------------------|-----|-----|-----|-----|-----|--------------|--|--|
|  | 1                           | 2   | 3   | 4   | 5   | 6   | Total        | Mean   |  |
| Control  | 100                         | 100 | 100 | 100 | 100 | 100 | 10           | 29.2   |  |
| 100% Effluent                                      | 100                         | 100 | 100 | 100 | 100 | 100 | 10           | 30.5   |  |
| 50% Effluent                                       | 100                         | 100 | 100 | 100 | 100 | 100 | 10           | 31.4   |  |
| 25% Effluent                                       | 100                         | 100 | 100 | 100 | 100 | 100 | 10           | 28.1   |  |
| 12% Effluent                                       | 100                         | 100 | 100 | 100 | 100 | 100 | 10           | 28.6   |  |
| 6% Effluent  | 100                         | 100 | 100 | 100 | 100 | 100 | 10           | 29.0   |  |
| IC <sub>50</sub> Value: > 100%                     | Calculated TUc Value: < 1.0 |     |     |     |     |     |              |  |  |
| 95% Confidence Limits: NA                          |                             |     |     |     |     |     |              | Permit Limits: TUc ≥ 1.0   |  |
| UL:<br>LL:<br>UL = Upper Limit<br>LL = Lower Limit |                             |     |     |     |     |     |              | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |  |

Results of a *Pimephales promelas* Chronic Toxicity Test, 5/14-21/97 Using Effluent from Outfall 001

| Test Solution                  | Percent Surviving (day) |      |      |      |      |      |      | Dry Weight (mg/L)  |       |
|--------------------------------|-------------------------|------|------|------|------|------|------|--|-------|
|                                | 1                       | 2    | 3    | 4    | 5    | 6    | 7    | Total  | Mean  |
| Control                        | 97.5                    | 95.0 | 95.0 | 95.0 | 95.0 | 95.0 | 95.0 | 40   | 0.491 |
| 100% Effluent                  | 100                     | 100  | 97.5 | 95.0 | 92.5 | 90.0 | 90.0 | 40   | 0.630 |
| 50% Effluent                   | 100                     | 100  | 100  | 100  | 100  | 100  | 100  | 40   | 0.670 |
| 25% Effluent                   | 97.5                    | 97.5 | 97.5 | 97.5 | 97.5 | 97.5 | 97.5 | 40   | 0.694 |
| 12% Effluent                   | 100                     | 100  | 100  | 100  | 100  | 100  | 100  | 40   | 0.647 |
| 6% Effluent                    | 100                     | 100  | 100  | 100  | 100  | 100  | 100  | 40   | 0.607 |
| IC <sub>50</sub> Value: > 100% |                         |      |      |      |      |      |      | Calculated TUc Value: < 1.0  |       |
| 95% Confidence Limits: NA      |                         |      |      |      |      |      |      | Permit Limits: TUc = 1.0   |       |
| UL:                            |                         |      |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |
| LL:                            |                         |      |      |      |      |      |      |  |       |
| UL = Upper Limit               |                         |      |      |      |      |      |      |  |       |
| LL = Lower Limit               |                         |      |      |      |      |      |      |  |       |

Results of a *Pimephales promelas* Chronic Toxicity Test, 05/14-21/97 Using Effluent from Outfall 006

| Test Solution                  | Percent Surviving (day) |      |      |      |      |      |      | Dry Weight (mg/L)  |       |
|--------------------------------|-------------------------|------|------|------|------|------|------|--|-------|
|                                | 1                       | 2    | 3    | 4    | 5    | 6    | 7    | Total  | Mean  |
| Control                        | 97.5                    | 97.5 | 97.5 | 97.5 | 95.0 | 95.0 | 95.0 | 40   | 0.500 |
| 100% Effluent                  | 97.5                    | 97.5 | 95.0 | 92.5 | 92.5 | 87.5 | 87.5 | 40   | 0.502 |
| 50% Effluent                   | 100                     | 100  | 100  | 100  | 100  | 100  | 100  | 40   | 0.597 |
| 25% Effluent                   | 100                     | 100  | 100  | 100  | 100  | 100  | 100  | 40   | 0.586 |
| 12% Effluent                   | 95.0                    | 95.0 | 95.0 | 95.0 | 95.0 | 95.0 | 95.0 | 40   | 0.638 |
| 6% Effluent                    | 100                     | 100  | 97.5 | 97.5 | 95.0 | 95.0 | 95.0 | 40   | 0.583 |
| IC <sub>50</sub> Value: > 100% |                         |      |      |      |      |      |      | Calculated TUc Value: < 1.0  |       |
| 95% Confidence Limits: NA      |                         |      |      |      |      |      |      | Permit Limits: TUc = 1.0   |       |
| UL:                            |                         |      |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |
| LL:                            |                         |      |      |      |      |      |      |  |       |
| UL = Upper Limit               |                         |      |      |      |      |      |      |  |       |
| LL = Lower Limit               |                         |      |      |      |      |      |      |  |       |

C-6 — Biological Monitoring Program

Results of a *Pimephales promelas* Chronic Toxicity Test, 05/14-21/97 Using Effluent from Outfall 008

| Test Solution                  | Percent Surviving (day) |      |      |      |      |      |      | Dry Weight (mg/L)  |       |
|--------------------------------|-------------------------|------|------|------|------|------|------|--|-------|
|                                | 1                       | 2    | 3    | 4    | 5    | 6    | 7    | Total  | Mean  |
| Control                        | 100                     | 100  | 100  | 97.5 | 97.5 | 97.5 | 97.5 | 40   | 0.456 |
| 100% Effluent                  | 100                     | 100  | 95.0 | 90.0 | 87.5 | 85.0 | 85.0 | 40   | 0.438 |
| 50% Effluent                   | 97.5                    | 92.5 | 92.5 | 82.5 | 72.5 | 70.0 | 70.0 | 40   | 0.353 |
| 25% Effluent                   | 100                     | 100  | 100  | 97.5 | 97.5 | 97.5 | 97.5 | 40   | 0.503 |
| 12% Effluent                   | 100                     | 100  | 100  | 95.0 | 90.0 | 90.0 | 87.5 | 40   | 0.471 |
| 6% Effluent                    | 100                     | 100  | 100  | 100  | 100  | 100  | 100  | 40   | 0.515 |
| IC <sub>50</sub> Value: > 100% |                         |      |      |      |      |      |      | Calculated TUC Value: < 1.0  |       |
| 95% Confidence Limits: NA      |                         |      |      |      |      |      |      | Permit Limits: TUC = 1.0   |       |
| UL:                            |                         |      |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |
| LL:                            |                         |      |      |      |      |      |      |  |       |
| UL = Upper Limit               |                         |      |      |      |      |      |      |  |       |
| LL = Lower Limit               |                         |      |      |      |      |      |      |  |       |

Results of a *Pimephales promelas* Chronic Toxicity Test, 05/14-21/97 Using Effluent from Outfall 009

| Test Solution                  | Percent Surviving (day) |      |      |      |      |      |      | Dry Weight (mg/L)  |       |
|--------------------------------|-------------------------|------|------|------|------|------|------|--|-------|
|                                | 1                       | 2    | 3    | 4    | 5    | 6    | 7    | Total  | Mean  |
| Control                        | 97.5                    | 97.5 | 97.5 | 95.0 | 95.0 | 95.0 | 95.0 | 40   | 0.451 |
| 100% Effluent                  | 100                     | 100  | 95.0 | 90.0 | 90.0 | 82.5 | 82.5 | 40   | 0.486 |
| 50% Effluent                   | 100                     | 100  | 97.5 | 92.5 | 85.0 | 85.0 | 85.0 | 40   | 0.469 |
| 25% Effluent                   | 100                     | 100  | 95.0 | 77.5 | 60.0 | 60.0 | 59.0 | 39 <sup>a</sup>  | 0.341 |
| 12% Effluent                   | 97.5                    | 95.0 | 95.0 | 95.0 | 95.0 | 92.5 | 90.0 | 40   | 0.566 |
| 6% Effluent                    | 100                     | 100  | 95.0 | 95.0 | 90.0 | 90.0 | 90.0 | 40   | 0.477 |
| IC <sub>50</sub> Value: > 100% |                         |      |      |      |      |      |      | Calculated TUC Value: < 1.0  |       |
| 95% Confidence Limits: NA      |                         |      |      |      |      |      |      | Permit Limits: TUC = 1.0   |       |
| UL:                            |                         |      |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |
| LL:                            |                         |      |      |      |      |      |      |  |       |
| UL = Upper Limit               |                         |      |      |      |      |      |      |  |       |
| LL = Lower Limit               |                         |      |      |      |      |      |      |  |       |

<sup>a</sup>One test organism was missing on Day 7; therefore, percent surviving and mean weight were based on 39 test organisms.

Results of a *Pimephales promelas* Chronic Toxicity Test, 05/14-21/97 Using Effluent from Outfall 010

| Test Solution                  | Percent Surviving (day) |      |      |      |      |      |      | Dry Weight (mg/L)  |       |
|--------------------------------|-------------------------|------|------|------|------|------|------|--|-------|
|                                | 1                       | 2    | 3    | 4    | 5    | 6    | 7    | Total  | Mean  |
| Control                        | 100                     | 100  | 100  | 100  | 100  | 100  | 100  | 40   | 0.557 |
| 100% Effluent                  | 97.5                    | 97.5 | 97.5 | 95.0 | 92.5 | 92.5 | 92.5 | 40   | 0.554 |
| 50% Effluent                   | 100                     | 100  | 100  | 100  | 100  | 100  | 100  | 40   | 0.639 |
| 25% Effluent                   | 100                     | 100  | 100  | 100  | 100  | 100  | 100  | 40   | 0.626 |
| 12% Effluent                   | 95.0                    | 95.0 | 95.0 | 95.0 | 92.5 | 92.5 | 92.5 | 40   | 0.583 |
| 6% Effluent                    | 100                     | 100  | 100  | 97.5 | 97.5 | 97.5 | 97.5 | 40   | 0.612 |
| IC <sub>50</sub> Value: > 100% |                         |      |      |      |      |      |      | Calculated TUC Value: < 1.0  |       |
| 95% Confidence Limits:         |                         |      |      |      |      |      |      | Permit Limits: TUC = 1.0   |       |
| UL:                            |                         |      |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |
| LL:                            |                         |      |      |      |      |      |      |  |       |
| UL = Upper Limit               |                         |      |      |      |      |      |      |  |       |
| LL = Lower Limit               |                         |      |      |      |      |      |      |  |       |

Results of a *Ceriodaphnia dubia* Chronic Toxicity Test, 05/14-20/97 Using Effluent from Outfall 001

| Test Solution                  | Percent Surviving (day) |     |     |     |     |     | No. of Young   |      |
|--------------------------------|-------------------------|-----|-----|-----|-----|-----|--|------|
|                                | 1                       | 2   | 3   | 4   | 5   | 6   | Total  | Mean |
| Control                        | 100                     | 100 | 100 | 100 | 100 | 100 | 10   | 28.0 |
| 100% Effluent                  | 100                     | 100 | 90  | 90  | 90  | 90  | 10   | 31.7 |
| 50% Effluent                   | 100                     | 100 | 100 | 100 | 100 | 100 | 10   | 32.4 |
| 25% Effluent                   | 100                     | 100 | 90  | 90  | 90  | 90  | 10   | 25.8 |
| 12% Effluent                   | 100                     | 100 | 100 | 100 | 100 | 100 | 10   | 32.0 |
| 6% Effluent                    | 100                     | 100 | 100 | 100 | 100 | 100 | 10   | 33.3 |
| IC <sub>50</sub> Value: > 100% |                         |     |     |     |     |     | Calculated TUC Value: < 1.0  |      |
| 95% Confidence Limits: NA      |                         |     |     |     |     |     | Permit Limits: TUC = 1.0   |      |
| UL:                            |                         |     |     |     |     |     | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |      |
| LL:                            |                         |     |     |     |     |     |  |      |
| UL = Upper Limit               |                         |     |     |     |     |     |  |      |
| LL = Lower Limit               |                         |     |     |     |     |     |  |      |

**C-8 – Biological Monitoring Program**

**Results of a *Pimephales promelas* Chronic Toxicity Test, 08/13–20/97 Using Effluent from Outfall 001**

| Test Solution                 | Percent Surviving (day) |      |      |      |      |      |      | Dry Weight (mg/L)  |       |
|-------------------------------|-------------------------|------|------|------|------|------|------|--|-------|
|                               | 1                       | 2    | 3    | 4    | 5    | 6    | 7    | Total  | Mean  |
| Control                       | 100                     | 100  | 100  | 100  | 100  | 100  | 100  | 40   | 0.685 |
| 100% Effluent                 | 100                     | 100  | 100  | 90.0 | 82.5 | 75.0 | 70.0 | 40   | 0.334 |
| 50% Effluent                  | 97.5                    | 95.0 | 95.0 | 95.0 | 95.0 | 95.0 | 90.0 | 40   | 0.555 |
| 25% Effluent                  | 100                     | 100  | 100  | 100  | 100  | 100  | 100  | 40   | 0.582 |
| 12% Effluent                  | 95.0                    | 92.5 | 92.5 | 90.0 | 87.5 | 77.5 | 75.0 | 40   | 0.420 |
| 6% Effluent                   | 100                     | 95.0 | 95.0 | 92.5 | 92.5 | 90.0 | 90.0 | 40   | 0.700 |
| IC <sub>50</sub> Value: 11.99 |                         |      |      |      |      |      |      | Calculated TUc Value: 8.34   |       |
| 95% Confidence Limits: NA     |                         |      |      |      |      |      |      | Permit Limits: TUc = 1.0   |       |
| UL: 72.94                     |                         |      |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |
| LL: 9.68                      |                         |      |      |      |      |      |      |  |       |
| UL = Upper Limit              |                         |      |      |      |      |      |      |  |       |
| LL = Lower Limit              |                         |      |      |      |      |      |      |  |       |

**Results of a *Pimephales promelas* Chronic Toxicity Test, 08/13–20/97 Using Effluent from Outfall 006**

| Test Solution                  | Percent Surviving (day) |      |      |      |      |      |      | Dry Weight (mg/L)  |       |
|--------------------------------|-------------------------|------|------|------|------|------|------|--|-------|
|                                | 1                       | 2    | 3    | 4    | 5    | 6    | 7    | Total  | Mean  |
| Control                        | 100                     | 100  | 90.0 | 90.0 | 87.5 | 87.5 | 87.5 | 40   | 0.549 |
| 100% Effluent                  | 100                     | 97.5 | 97.5 | 95.0 | 95.0 | 92.5 | 92.5 | 40   | 0.698 |
| 50% Effluent                   | 100                     | 97.5 | 97.5 | 97.5 | 92.5 | 92.5 | 90.0 | 40   | 0.733 |
| 25% Effluent                   | 100                     | 100  | 100  | 97.5 | 97.5 | 97.5 | 97.5 | 40   | 0.766 |
| 12% Effluent                   | 95.0                    | 92.5 | 92.5 | 92.5 | 90.0 | 87.5 | 85.0 | 40   | 0.636 |
| 6% Effluent                    | 100                     | 100  | 97.5 | 97.5 | 97.5 | 97.5 | 95.0 | 40   | 0.732 |
| IC <sub>50</sub> Value: > 100% |                         |      |      |      |      |      |      | Calculated TUc Value: < 1.0  |       |
| 95% Confidence Limits: NA      |                         |      |      |      |      |      |      | Permit Limits: TUc = 1.0   |       |
| UL:                            |                         |      |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |
| LL:                            |                         |      |      |      |      |      |      |  |       |
| UL = Upper Limit               |                         |      |      |      |      |      |      |  |       |
| LL = Lower Limit               |                         |      |      |      |      |      |      |  |       |

Results of a *Pimephales promelas* Chronic Toxicity Test, 08/13–20/97 Using Effluent from Outfall 008

| Test Solution                  | Percent Surviving (day) |      |      |      |      |      |      | Dry Weight (mg/L)  |       |
|--------------------------------|-------------------------|------|------|------|------|------|------|--|-------|
|                                | 1                       | 2    | 3    | 4    | 5    | 6    | 7    | Total  | Mean  |
| Control                        | 95.0                    | 92.5 | 90.0 | 87.5 | 87.5 | 85.0 | 85.0 | 40   | 0.448 |
| 100% Effluent                  | 100                     | 100  | 97.5 | 97.5 | 95.0 | 95.0 | 92.5 | 40   | 0.538 |
| 50% Effluent                   | 95.0                    | 92.5 | 92.5 | 90.0 | 87.5 | 87.5 | 87.5 | 40   | 0.545 |
| 25% Effluent                   | 97.5                    | 97.5 | 97.5 | 97.5 | 97.5 | 95.0 | 95.0 | 40   | 0.634 |
| 12% Effluent                   | 100                     | 97.5 | 97.5 | 97.5 | 97.5 | 97.5 | 97.5 | 40   | 0.633 |
| 6% Effluent                    | 100                     | 100  | 100  | 100  | 100  | 100  | 100  | 40   | 0.574 |
| IC <sub>50</sub> Value: > 100% |                         |      |      |      |      |      |      | Calculated TUc Value: < 1.0  |       |
| 95% Confidence Limits: NA      |                         |      |      |      |      |      |      | Permit Limits: TUc = 1.0   |       |
| UL:                            |                         |      |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |
| LL:                            |                         |      |      |      |      |      |      |  |       |
| UL = Upper Limit               |                         |      |      |      |      |      |      |  |       |
| LL = Lower Limit               |                         |      |      |      |      |      |      |  |       |

Results of a *Pimephales promelas* Chronic Toxicity Test, 08/13–20/97 Using Effluent from Outfall 009

| Test Solution                  | Percent Surviving (day) |      |      |      |      |      |      | Dry Weight (mg/L)  |       |
|--------------------------------|-------------------------|------|------|------|------|------|------|--|-------|
|                                | 1                       | 2    | 3    | 4    | 5    | 6    | 7    | Total  | Mean  |
| Control                        | 100                     | 95.0 | 95.0 | 95.0 | 95.0 | 95.0 | 95.0 | 40   | 0.590 |
| 100% Effluent                  | 100                     | 100  | 100  | 95.0 | 90.0 | 90.0 | 87.5 | 40   | 0.575 |
| 50% Effluent                   | 100                     | 97.5 | 97.5 | 95.0 | 90.0 | 90.0 | 90.0 | 40   | 0.600 |
| 25% Effluent                   | 95.0                    | 95.0 | 95.0 | 92.5 | 92.5 | 87.5 | 87.5 | 40   | 0.557 |
| 12% Effluent                   | 100                     | 100  | 97.5 | 97.5 | 97.5 | 95.0 | 95.0 | 40   | 0.650 |
| 6% Effluent                    | 100                     | 100  | 97.5 | 97.5 | 97.5 | 97.5 | 90.0 | 40   | 0.578 |
| IC <sub>50</sub> Value: > 100% |                         |      |      |      |      |      |      | Calculated TUc Value: < 1.0  |       |
| 95% Confidence Limits: NA      |                         |      |      |      |      |      |      | Permit Limits: TUc = 1.0   |       |
| UL:                            |                         |      |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |
| LL:                            |                         |      |      |      |      |      |      |  |       |
| UL = Upper Limit               |                         |      |      |      |      |      |      |  |       |
| LL = Lower Limit               |                         |      |      |      |      |      |      |  |       |

**C-10 — Biological Monitoring Program**

**Results of a *Pimephales promelas* Chronic Toxicity Test, 08/13-20/97 Using Effluent from Outfall 010**

| Test Solution                  | Percent Surviving (day)  |      |      |      |      |      |      | Dry Weight (mg/L)        |       |
|--------------------------------|--|------|------|------|------|------|------|--------------------------|-------|
|                                | 1  | 2    | 3    | 4    | 5    | 6    | 7    | Total                    | Mean  |
| Control                        | 100  | 97.5 | 97.5 | 97.5 | 97.5 | 97.5 | 92.5 | 40                       | 0.587 |
| 100% Effluent                  | 100  | 100  | 100  | 100  | 100  | 100  | 100  | 40                       | 0.658 |
| 50% Effluent                   | 100  | 97.5 | 97.5 | 97.5 | 97.5 | 97.5 | 97.5 | 40                       | 0.664 |
| 25% Effluent                   | 100  | 100  | 100  | 100  | 100  | 100  | 100  | 40                       | 0.758 |
| 12% Effluent                   | 100  | 100  | 100  | 100  | 100  | 100  | 100  | 40                       | 0.740 |
| 6% Effluent                    | 100  | 100  | 97.5 | 97.5 | 95.0 | 95.0 | 95.0 | 40                       | 0.680 |
| IC <sub>50</sub> Value: > 100% | Calculated TUc Value: < 1.0  |      |      |      |      |      |      |                          |       |
| 95% Confidence Limits:         |  |      |      |      |      |      |      | Permit Limits: TUc = 1.0 |       |
| UL:                            | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |      |      |      |      |      |      |                          |       |
| LL:                            |  |      |      |      |      |      |      |                          |       |
| UL = Upper Limit               |  |      |      |      |      |      |      |                          |       |
| LL = Lower Limit               |  |      |      |      |      |      |      |                          |       |

**Results of a *Ceriodaphnia dubia* Chronic Toxicity Test, 08/13-19/97 Using Effluent from Outfall 001**

| Test Solution                  | Percent Surviving (day)  |     |     |     |     |     | No. of Young |                          |  |
|--------------------------------|--|-----|-----|-----|-----|-----|--------------|--------------------------|--|
|                                | 1  | 2   | 3   | 4   | 5   | 6   | Total        | Mean                     |  |
| Control                        | 100  | 100 | 100 | 100 | 100 | 100 | 10           | 33.1                     |  |
| 100% Effluent                  | 100  | 100 | 100 | 100 | 100 | 100 | 10           | 35.7                     |  |
| 50% Effluent                   | 100  | 100 | 100 | 100 | 100 | 100 | 10           | 27.1                     |  |
| 25% Effluent                   | 100  | 100 | 100 | 100 | 100 | 100 | 10           | 32.7                     |  |
| 12% Effluent                   | 100  | 100 | 100 | 100 | 100 | 100 | 10           | 32.9                     |  |
| 6% Effluent                    | 100  | 100 | 100 | 100 | 100 | 100 | 10           | 32.0                     |  |
| IC <sub>50</sub> Value: > 100% | Calculated TUc Value: < 1.0  |     |     |     |     |     |              |                          |  |
| 95% Confidence Limits: NA      |  |     |     |     |     |     |              | Permit Limits: TUc = 1.0 |  |
| UL:                            | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |     |     |     |     |     |              |                          |  |
| LL:                            |  |     |     |     |     |     |              |                          |  |
| UL = Upper Limit               |  |     |     |     |     |     |              |                          |  |
| LL = Lower Limit               |  |     |     |     |     |     |              |                          |  |

Results of a *Pimephales promelas* Chronic Toxicity Test, 09/04-11/97 Using Effluent from Outfall 001

| Test Solution                  | Percent Surviving (day) |     |      |      |      |      |      | Dry Weight (mg/L)  |       |
|--------------------------------|-------------------------|-----|------|------|------|------|------|--|-------|
|                                | 1                       | 2   | 3    | 4    | 5    | 6    | 7    | Total  | Mean  |
| Control                        | 100                     | 100 | 100  | 97.5 | 97.5 | 97.5 | 97.5 | 40   | 0.457 |
| 100% Effluent                  | 100                     | 100 | 95.0 | 92.5 | 90.0 | 90.0 | 87.5 | 40   | 0.444 |
| 50% Effluent                   | 100                     | 100 | 97.5 | 97.5 | 92.5 | 90.0 | 82.5 | 40   | 0.473 |
| 25% Effluent                   | 100                     | 100 | 100  | 100  | 100  | 100  | 100  | 40   | 0.559 |
| 12% Effluent                   | 100                     | 100 | 97.5 | 97.5 | 97.5 | 97.5 | 92.5 | 40   | 0.476 |
| 6% Effluent                    | 100                     | 100 | 100  | 100  | 100  | 100  | 97.5 | 40   | 0.560 |
| IC <sub>50</sub> Value: > 100% |                         |     |      |      |      |      |      | Calculated TUC Value: < 1.0  |       |
| 95% Confidence Limits: NA      |                         |     |      |      |      |      |      | Permit Limits: TUC = 1.0   |       |
| UL:                            |                         |     |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |
| LL:                            |                         |     |      |      |      |      |      |  |       |
| UL = Upper Limit               |                         |     |      |      |      |      |      |  |       |
| LL = Lower Limit               |                         |     |      |      |      |      |      |  |       |

Results of a *Ceriodaphnia dubia* Chronic Toxicity Test, 11/06-12/97 Using Effluent from Outfall 001

| Test Solution                  | Percent Surviving (day) |     |     |     |     |     | No. of Young   |      |
|--------------------------------|-------------------------|-----|-----|-----|-----|-----|--|------|
|                                | 1                       | 2   | 3   | 4   | 5   | 6   | Total  | Mean |
| Control                        | 100                     | 100 | 100 | 100 | 100 | 100 | 10   | 28.6 |
| 100% Effluent                  | 100                     | 100 | 100 | 100 | 100 | 90  | 10   | 29.3 |
| 50% Effluent                   | 100                     | 100 | 100 | 100 | 100 | 100 | 10   | 31.3 |
| 25% Effluent                   | 100                     | 100 | 100 | 100 | 100 | 100 | 10   | 32.0 |
| 12% Effluent                   | 100                     | 100 | 100 | 100 | 100 | 100 | 10   | 29.4 |
| 6% Effluent                    | 100                     | 100 | 100 | 100 | 100 | 100 | 10   | 28.4 |
| IC <sub>50</sub> Value: > 100% |                         |     |     |     |     |     | Calculated TUC Value: < 1.0  |      |
| 95% Confidence Limits: NA      |                         |     |     |     |     |     | Permit Limits: TUC = 1.0   |      |
| UL:                            |                         |     |     |     |     |     | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |      |
| LL:                            |                         |     |     |     |     |     |  |      |
| UL = Upper Limit               |                         |     |     |     |     |     |  |      |
| LL = Lower Limit               |                         |     |     |     |     |     |  |      |

C-12 – Biological Monitoring Program

Results of a *Pimephales promelas* Chronic Toxicity Test, 12/03–10/97 Using Effluent from Outfall 001

| Test Solution                  | Percent Surviving (day)  |     |      |      |      |      |      | Dry Weight (mg/L)        |       |
|--------------------------------|--|-----|------|------|------|------|------|--------------------------|-------|
|                                | 1  | 2   | 3    | 4    | 5    | 6    | 7    | Total                    | Mean  |
| Control                        | 100  | 100 | 95.0 | 95.0 | 95.0 | 95.0 | 92.5 | 40                       | 0.407 |
| 100% Effluent                  | 100  | 100 | 100  | 100  | 97.5 | 97.5 | 95.0 | 40                       | 0.492 |
| 50% Effluent                   | 100  | 100 | 97.5 | 97.5 | 95.0 | 95.0 | 95.0 | 40                       | 0.474 |
| 25% Effluent                   | 100  | 100 | 100  | 100  | 100  | 100  | 100  | 39 <sup>a</sup>          | 0.477 |
| 12% Effluent                   | 100  | 100 | 100  | 97.5 | 97.5 | 97.5 | 97.5 | 40                       | 0.497 |
| 6% Effluent                    | 100  | 100 | 97.5 | 97.5 | 95.0 | 95.0 | 95.0 | 40                       | 0.447 |
| IC <sub>50</sub> Value: > 100% | Calculated TUC Value: < 1.0  |     |      |      |      |      |      |                          |       |
| 95% Confidence Limits: NA      |  |     |      |      |      |      |      | Permit Limits: TUC = 1.0 |       |
| UL:                            | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |     |      |      |      |      |      |                          |       |
| LL:                            |  |     |      |      |      |      |      |                          |       |
| UL = Upper Limit               |  |     |      |      |      |      |      |                          |       |
| LL = Lower Limit               |  |     |      |      |      |      |      |                          |       |

<sup>a</sup> One test organism was inadvertently killed on Day 6; therefore, mean weight and percent surviving are based on 39 test organisms.

Results of a *Pimephales promelas* Chronic Toxicity Test, 12/03–10/97 Using Effluent from Outfall 006

| Test Solution                  | Percent Surviving (day)  |      |      |      |      |      |      | Dry Weight (mg/L)        |       |
|--------------------------------|--|------|------|------|------|------|------|--------------------------|-------|
|                                | 1  | 2    | 3    | 4    | 5    | 6    | 7    | Total                    | Mean  |
| Control                        | 100  | 100  | 100  | 100  | 100  | 100  | 100  | 40                       | 0.414 |
| 100% Effluent                  | 100  | 97.5 | 97.5 | 97.5 | 95.0 | 95.0 | 95.0 | 40                       | 0.469 |
| 50% Effluent                   | 97.5   | 97.5 | 94.9 | 87.2 | 82.1 | 64.1 | 61.5 | 39 <sup>a</sup>          | 0.324 |
| 25% Effluent                   | 100  | 100  | 100  | 97.5 | 97.5 | 97.5 | 97.5 | 40                       | 0.474 |
| 12% Effluent                   | 100  | 100  | 100  | 100  | 100  | 100  | 100  | 40                       | 0.485 |
| 6% Effluent                    | 100  | 100  | 100  | 100  | 97.5 | 97.5 | 97.5 | 40                       | 0.460 |
| IC <sub>50</sub> Value: > 100% | Calculated TUC Value: < 1.0  |      |      |      |      |      |      |                          |       |
| 95% Confidence Limits: NA      |  |      |      |      |      |      |      | Permit Limits: TUC = 1.0 |       |
| UL:                            | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |      |      |      |      |      |      |                          |       |
| LL:                            |  |      |      |      |      |      |      |                          |       |
| UL = Upper Limit               |  |      |      |      |      |      |      |                          |       |
| LL = Lower Limit               |  |      |      |      |      |      |      |                          |       |

<sup>a</sup> One test organism was missing on Day 3; therefore, mean weight and percent surviving are based on 39 test organisms.

Results of a *Pimephales promelas* Chronic Toxicity Test, 12/03-10/97 Using Effluent from Outfall 008

| Test Solution                                      | Percent Surviving (day)                                 |     |      |      |      |      |      | Dry Weight (mg/L)  |       |
|--|---|-----|------|------|------|------|------|--|-------|
|  | 1   | 2   | 3    | 4    | 5    | 6    | 7    | Total  | Mean  |
| Control  | 100   | 100 | 100  | 100  | 100  | 100  | 100  | 39 <sup>a</sup>  | 0.408 |
| 100% Effluent                                      | 100   | 100 | 100  | 100  | 100  | 100  | 100  | 40   | 0.432 |
| 50% Effluent                                       | 100   | 100 | 100  | 100  | 97.5 | 95.0 | 95.0 | 40   | 0.436 |
| 25% Effluent                                       | 100   | 100 | 97.5 | 97.5 | 97.5 | 95.0 | 95.0 | 40   | 0.452 |
| 12% Effluent                                       | 100   | 100 | 100  | 100  | 100  | 100  | 100  | 40   | 0.456 |
| 6% Effluent  | 100   | 100 | 100  | 100  | 100  | 97.5 | 97.5 | 40   | 0.464 |
| IC <sub>50</sub> Value: > 100%                     | Calculated TUC Value: < 1.0<br>Permit Limits: TUC = 1.0 |     |      |      |      |      |      |  |       |
| 95% Confidence Limits: NA                          |   |     |      |      |      |      |      |  |       |
| UL:<br>LL:<br>UL = Upper Limit<br>LL = Lower Limit |   |     |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |

<sup>a</sup> One test organism was missing on Day 1; therefore, mean weight and percent surviving are based on 39 test organisms.

Results of a *Pimephales promelas* Chronic Toxicity Test, 12/03-10/97 Using Effluent from Outfall 009

| Test Solution                                      | Percent Surviving (day)                                 |     |      |      |      |      |      | Dry Weight (mg/L)  |       |
|--|---|-----|------|------|------|------|------|--|-------|
|  | 1   | 2   | 3    | 4    | 5    | 6    | 7    | Total  | Mean  |
| Control  | 100   | 100 | 100  | 100  | 100  | 100  | 100  | 40   | 0.411 |
| 100% Effluent                                      | 100   | 100 | 95.0 | 92.5 | 82.5 | 82.5 | 80.0 | 40   | 0.373 |
| 50% Effluent                                       | 100   | 100 | 95.0 | 92.5 | 90.0 | 82.5 | 82.5 | 40   | 0.371 |
| 25% Effluent                                       | 100   | 100 | 97.5 | 97.5 | 97.5 | 97.5 | 97.5 | 40   | 0.452 |
| 12% Effluent                                       | 100   | 100 | 100  | 100  | 97.5 | 90.0 | 90.0 | 40   | 0.394 |
| 6% Effluent  | 100   | 100 | 97.5 | 97.5 | 95.0 | 85.0 | 85.0 | 40   | 0.395 |
| IC <sub>50</sub> Value: > 100%                     | Calculated TUC Value: < 1.0<br>Permit Limits: TUC = 1.0 |     |      |      |      |      |      |  |       |
| 95% Confidence Limits: NA                          |   |     |      |      |      |      |      |  |       |
| UL:<br>LL:<br>UL = Upper Limit<br>LL = Lower Limit |   |     |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |

C-14 — Biological Monitoring Program

Results of a *Pimephales promelas* Chronic Toxicity Test, 12/03-10/97 Using Effluent from Outfall 010

| Test Solution                                   | Percent Surviving (day) |     |      |      |      |      |      | Dry Weight (mg/L)  |       |
|---|-------------------------|-----|------|------|------|------|------|--|-------|
|   | 1                       | 2   | 3    | 4    | 5    | 6    | 7    | Total  | Mean  |
| Control   | 100                     | 100 | 100  | 100  | 100  | 100  | 100  | 40   | 0.405 |
| 100% Effluent                                   | 100                     | 100 | 100  | 97.5 | 97.5 | 92.5 | 92.5 | 40   | 0.332 |
| 50% Effluent                                    | 100                     | 100 | 95.0 | 87.5 | 82.5 | 77.5 | 52.5 | 40   | 0.240 |
| 25% Effluent                                    | 100                     | 100 | 100  | 100  | 100  | 97.5 | 97.5 | 40   | 0.433 |
| 12% Effluent                                    | 100                     | 100 | 100  | 97.5 | 97.5 | 95.0 | 95.0 | 40   | 0.375 |
| 6% Effluent                                     | 100                     | 100 | 100  | 100  | 100  | 100  | 100  | 40   | 0.438 |
| IC <sub>25</sub> Value: > 100% (See Note below) |                         |     |      |      |      |      |      | Calculated TUC Value: < 1.0  |       |
| 95% Confidence Limits: NA                       |                         |     |      |      |      |      |      | Permit Limits: TUC = 1.0   |       |
| UL:   |                         |     |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |
| LL:   |                         |     |      |      |      |      |      |  |       |
| UL = Upper Limit                                |                         |     |      |      |      |      |      |  |       |
| LL = Lower Limit                                |                         |     |      |      |      |      |      |  |       |

NOTE: The mean dry weight of the fish in the 50% concentration (0.240 mg/larvae) is an outlier. Charles Roth, at the Kentucky Division of Water, approved excluding the 50% concentration from the IC<sub>25</sub> calculation. Therefore, the IC<sub>25</sub> value for this outfall is > 100%.

## Intermittent Outfalls

**Results of a *Pimephales promelas* Chronic Toxicity Test, 01/07-14/97 Using Effluent from Outfall 013**

| Test Solution                  | Percent Surviving (day) |      |      |      |      |      |      | Dry Weight (mg/L)  |       |
|--------------------------------|-------------------------|------|------|------|------|------|------|--|-------|
|                                | 1                       | 2    | 3    | 4    | 5    | 6    | 7    | Total  | Mean  |
| Control                        | 100                     | 100  | 100  | 100  | 100  | 97.5 | 97.5 | 40   | 0.398 |
| 100% Effluent                  | 100                     | 97.5 | 97.5 | 92.5 | 92.5 | 92.5 | 87.5 | 40   | 0.538 |
| 50% Effluent                   | 100                     | 95.0 | 92.5 | 90.0 | 87.5 | 85.0 | 85.0 | 40   | 0.492 |
| 25% Effluent                   | 97.5                    | 97.5 | 97.5 | 97.5 | 95.0 | 90.0 | 90.0 | 40   | 0.524 |
| 12% Effluent                   | 100                     | 97.5 | 97.5 | 97.5 | 97.5 | 97.5 | 97.5 | 40   | 0.497 |
| 6% Effluent                    | 100                     | 95.0 | 95.0 | 95.0 | 92.5 | 90.0 | 90.0 | 40   | 0.464 |
| IC <sub>50</sub> Value: > 100% |                         |      |      |      |      |      |      | Calculated TUc Value: < 1.0  |       |
| 95% Confidence Limits: NA      |                         |      |      |      |      |      |      | Permit Limits: TUc ≥ 1.0   |       |
| UL:                            |                         |      |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |
| LL:                            |                         |      |      |      |      |      |      |  |       |
| UL = Upper Limit               |                         |      |      |      |      |      |      |  |       |
| LL = Lower Limit               |                         |      |      |      |      |      |      |  |       |

**Results of a *Pimephales promelas* Chronic Toxicity Test, 01/07-14/97 Using Effluent from Outfall 015**

| Test Solution                  | Percent Surviving (day) |      |      |      |      |      |      | Dry Weight (mg/L)  |       |
|--------------------------------|-------------------------|------|------|------|------|------|------|--|-------|
|                                | 1                       | 2    | 3    | 4    | 5    | 6    | 7    | Total  | Mean  |
| Control                        | 100                     | 100  | 100  | 100  | 100  | 100  | 100  | 40   | 0.365 |
| 100% Effluent                  | 100                     | 100  | 100  | 97.5 | 97.5 | 95.0 | 95.0 | 40   | 0.632 |
| 50% Effluent                   | 100                     | 100  | 100  | 100  | 100  | 100  | 97.5 | 40   | 0.567 |
| 25% Effluent                   | 100                     | 97.5 | 97.5 | 95.0 | 92.5 | 92.5 | 92.5 | 40   | 0.613 |
| 12% Effluent                   | 100                     | 100  | 100  | 100  | 100  | 100  | 100  | 40   | 0.521 |
| 6% Effluent                    | 100                     | 100  | 92.5 | 92.5 | 92.5 | 90.0 | 90.0 | 40   | 0.450 |
| IC <sub>50</sub> Value: > 100% |                         |      |      |      |      |      |      | Calculated TUc Value: < 1.0  |       |
| 95% Confidence Limits: NA      |                         |      |      |      |      |      |      | Permit Limits: TUc ≥ 1.0   |       |
| UL:                            |                         |      |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |
| LL:                            |                         |      |      |      |      |      |      |  |       |
| UL = Upper Limit               |                         |      |      |      |      |      |      |  |       |
| LL = Lower Limit               |                         |      |      |      |      |      |      |  |       |

**C-16 — Biological Monitoring Program**

**Results of a *Pimephales promelas* Chronic Toxicity Test, 01/07-14/97 Using Effluent from Outfall 016**

| Test Solution                  | Percent Surviving (day) |     |      |      |      |      |      | Dry Weight (mg/L)  |       |
|--------------------------------|-------------------------|-----|------|------|------|------|------|--|-------|
|                                | 1                       | 2   | 3    | 4    | 5    | 6    | 7    | Total  | Mean  |
| Control                        | 100                     | 100 | 100  | 100  | 97.5 | 97.5 | 97.5 | 40   | 0.420 |
| 100% Effluent                  | 100                     | 100 | 100  | 100  | 100  | 100  | 100  | 40   | 0.708 |
| 50% Effluent                   | 100                     | 100 | 100  | 97.4 | 97.4 | 97.4 | 97.4 | 39 <sup>a</sup>  | 0.628 |
| 25% Effluent                   | 100                     | 100 | 100  | 100  | 100  | 100  | 100  | 40   | 0.617 |
| 12% Effluent                   | 100                     | 100 | 97.5 | 97.5 | 97.5 | 95.0 | 92.5 | 40   | 0.573 |
| 6% Effluent                    | 100                     | 100 | 100  | 100  | 100  | 100  | 100  | 40   | 0.519 |
| IC <sub>50</sub> Value: > 100% |                         |     |      |      |      |      |      | Calculated TUC Value: < 1.0  |       |
| 95% Confidence Limits: NA      |                         |     |      |      |      |      |      | Permit Limits: TUC ≥ 1.0   |       |
| UL:                            |                         |     |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |
| LL:                            |                         |     |      |      |      |      |      |  |       |
| UL = Upper Limit               |                         |     |      |      |      |      |      |  |       |
| LL = Lower Limit               |                         |     |      |      |      |      |      |  |       |

<sup>a</sup>One test organism was missing on Day 3; therefore, percent surviving and mean weight were based on 39 test organisms.

**Results of a *Pimephales promelas* Chronic Toxicity Test, 01/07-14/97 Using Effluent from Outfall 017**

| Test Solution                  | Percent Surviving (day) |      |      |      |      |      |      | Dry Weight (mg/L)  |       |
|--------------------------------|-------------------------|------|------|------|------|------|------|--|-------|
|                                | 1                       | 2    | 3    | 4    | 5    | 6    | 7    | Total  | Mean  |
| Control                        | 97.5                    | 97.5 | 95.0 | 95.0 | 95.0 | 92.5 | 92.5 | 40   | 0.385 |
| 100% Effluent                  | 100                     | 97.5 | 97.5 | 97.5 | 97.5 | 97.5 | 87.5 | 40   | 0.515 |
| 50% Effluent                   | 95.0                    | 95.0 | 95.0 | 95.0 | 92.5 | 89.7 | 89.7 | 39 <sup>a</sup>  | 0.528 |
| 25% Effluent                   | 100                     | 97.5 | 95.0 | 95.0 | 95.0 | 95.0 | 95.0 | 40   | 0.514 |
| 12% Effluent                   | 100                     | 100  | 97.5 | 97.5 | 97.5 | 97.5 | 97.5 | 40   | 0.484 |
| 6% Effluent                    | 100                     | 100  | 100  | 100  | 95.0 | 95.0 | 95.0 | 40   | 0.494 |
| IC <sub>50</sub> Value: > 100% |                         |      |      |      |      |      |      | Calculated TUC Value: < 1.0  |       |
| 95% Confidence Limits: NA      |                         |      |      |      |      |      |      | Permit Limits: TUC ≥ 1.0   |       |
| UL:                            |                         |      |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |
| LL:                            |                         |      |      |      |      |      |      |  |       |
| UL = Upper Limit               |                         |      |      |      |      |      |      |  |       |
| LL = Lower Limit               |                         |      |      |      |      |      |      |  |       |

<sup>a</sup>One test organism was killed on Day 6; therefore, percent surviving and mean weight were based on 39 test organisms.

Results of a *Pimephales promelas* Chronic Toxicity Test, 01/07-14/97 Using Effluent from Outfall 001

| Test Solution                  | Percent Surviving (day)   |      |      |      |      |      |      | Dry Weight (mg/L) |       |
|--------------------------------|---|------|------|------|------|------|------|-------------------|-------|
|                                | 1   | 2    | 3    | 4    | 5    | 6    | 7    | Total             | Mean  |
| Control                        | 100   | 100  | 100  | 100  | 100  | 97.5 | 95.0 | 40                | 0.380 |
| 100% Effluent                  | 100   | 97.5 | 97.5 | 95.0 | 95.0 | 95.0 | 92.5 | 40                | 0.576 |
| 50% Effluent                   | 100   | 100  | 100  | 100  | 100  | 100  | 97.5 | 40                | 0.624 |
| 25% Effluent                   | 100   | 100  | 100  | 100  | 100  | 97.5 | 97.5 | 40                | 0.607 |
| 12% Effluent                   | 100   | 100  | 100  | 100  | 100  | 95.0 | 95.0 | 40                | 0.560 |
| 6% Effluent                    | 97.5  | 97.5 | 97.5 | 95.0 | 92.5 | 90.0 | 90.0 | 40                | 0.503 |
| IC <sub>50</sub> Value: > 100% | Calculated TUC Value: < 1.0<br>Permit Limits: TUC ≥ 1.0<br><br>If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |      |      |      |      |      |      |                   |       |
| 95% Confidence Limits:         |   |      |      |      |      |      |      |                   |       |
| UL: 52.49                      |   |      |      |      |      |      |      |                   |       |
| LL: 5.87                       |   |      |      |      |      |      |      |                   |       |
| UL = Upper Limit               |   |      |      |      |      |      |      |                   |       |
| LL = Lower Limit               |   |      |      |      |      |      |      |                   |       |

Results of a *Pimephales promelas* Chronic Toxicity Test, 04/08-15/97 Using Effluent from Outfall 013

| Test Solution                  | Percent Surviving (day)  |     |      |      |      |      |      | Dry Weight (mg/L) |                    |
|--------------------------------|--|-----|------|------|------|------|------|-------------------|--------------------|
|                                | 1  | 2   | 3    | 4    | 5    | 6    | 7    | Total             | Mean               |
| Control 1                      | 100  | 100 | 100  | 100  | 100  | 100  | 100  | 80                | 0.376 <sup>a</sup> |
| Control 2                      | 100  | 100 | 100  | 100  | 100  | 100  | 100  |                   |                    |
| 100% Effluent                  | 100  | 100 | 100  | 100  | 97.5 | 97.5 | 97.5 | 40                | 0.479              |
| 50% Effluent                   | 100  | 100 | 100  | 100  | 97.5 | 97.5 | 97.5 | 40                | 0.463              |
| 25% Effluent                   | 100  | 100 | 97.5 | 97.5 | 97.5 | 97.5 | 97.5 | 40                | 0.491              |
| 12% Effluent                   | 100  | 100 | 97.5 | 95.0 | 95.0 | 95.0 | 92.5 | 40                | 0.445              |
| 6% Effluent                    | 100  | 100 | 100  | 100  | 90.0 | 90.0 | 90.0 | 40                | 0.418              |
| IC <sub>50</sub> Value: > 100% | Calculated TUC Value: < 1.0<br>Permit Limits: NA<br><br>If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |     |      |      |      |      |      |                   |                    |
| 95% Confidence Limits: NA      |  |     |      |      |      |      |      |                   |                    |
| UL:                            |  |     |      |      |      |      |      |                   |                    |
| LL:                            |  |     |      |      |      |      |      |                   |                    |
| UL = Upper Limit               |  |     |      |      |      |      |      |                   |                    |
| LL = Lower Limit               |  |     |      |      |      |      |      |                   |                    |

<sup>a</sup>Normally, a control is included with each outfall tested; however, only two controls were included during the test because of a lack of sufficient larvae. Mean control weight was calculated by using eight control replicates.

**C-18 — Biological Monitoring Program**

**Results of a *Pimephales promelas* Chronic Toxicity Test, 04/08-15/97 Using Effluent from Outfall 015**

| Test Solution                  | Percent Surviving (day)  |      |      |      |      |      |      | Dry Weight (mg/L) |                    |
|--------------------------------|--|------|------|------|------|------|------|-------------------|--------------------|
|                                | 1  | 2    | 3    | 4    | 5    | 6    | 7    | Total             | Mean               |
| Control 1                      | 100  | 100  | 100  | 100  | 100  | 100  | 100  | 80                | 0.376 <sup>a</sup> |
| Control 2                      | 100  | 100  | 100  | 100  | 100  | 100  | 100  |                   |                    |
| 100% Effluent                  | 100  | 100  | 95.0 | 89.7 | 89.7 | 89.7 | 89.7 | 39 <sup>b</sup>   | 0.459              |
| 50% Effluent                   | 100  | 97.5 | 97.5 | 97.5 | 97.5 | 95.0 | 95.0 | 40                | 0.484              |
| 25% Effluent                   | 100  | 100  | 100  | 100  | 100  | 100  | 100  | 40                | 0.479              |
| 12% Effluent                   | 100  | 100  | 100  | 100  | 97.5 | 95.0 | 95.0 | 40                | 0.490              |
| 6% Effluent                    | 97.5   | 97.5 | 95.0 | 95.0 | 95.0 | 92.5 | 90.0 | 40                | 0.405              |
| IC <sub>50</sub> Value: > 100% | Calculated TUC Value: < 1.0  |      |      |      |      |      |      | Permit Limits: NA |                    |
| 95% Confidence Limits: NA      |  |      |      |      |      |      |      |                   |                    |
| UL:                            | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |      |      |      |      |      |      | Limit Values: NA  |                    |
| LL:                            |  |      |      |      |      |      |      |                   |                    |
| UL = Upper Limit               |  |      |      |      |      |      |      |                   |                    |
| LL = Lower Limit               |  |      |      |      |      |      |      |                   |                    |

<sup>a</sup>Normally, a control is included with each outfall tested; however, only two controls were included during the test because of a lack of sufficient larvae. Mean control weight was calculated by using eight control replicates.

<sup>b</sup>One test organism was inadvertently killed on Day 4; therefore, percent survival and mean growth were based on 39 test organisms.

**Results of a *Pimephales promelas* Chronic Toxicity Test, 04/08-15/97 Using Effluent from Outfall 016**

| Test Solution                | Percent Surviving (day)  |      |      |      |      |      |      | Dry Weight (mg/L) |                    |
|------------------------------|--|------|------|------|------|------|------|-------------------|--------------------|
|                              | 1  | 2    | 3    | 4    | 5    | 6    | 7    | Total             | Mean               |
| Control 1                    | 100  | 100  | 100  | 100  | 100  | 100  | 100  | 80                | 0.376 <sup>a</sup> |
| Control 2                    | 100  | 100  | 100  | 100  | 100  | 100  | 100  |                   |                    |
| 100% Effluent                | 100  | 100  | 62.5 | 35.0 | 20.0 | 20.0 | 20.0 | 40                | 0.108              |
| 50% Effluent                 | 100  | 97.5 | 70.0 | 55.0 | 40.0 | 40.0 | 40.0 | 40                | 0.158              |
| 25% Effluent                 | 97.5   | 97.5 | 95.0 | 80.0 | 50.0 | 47.5 | 42.5 | 40                | 0.155              |
| 12% Effluent                 | 100  | 100  | 90.0 | 65.0 | 55.0 | 55.0 | 50.0 | 40                | 0.219              |
| 6% Effluent                  | 100  | 100  | 100  | 85.0 | 65.0 | 60.0 | 57.5 | 40                | 0.265              |
| IC <sub>50</sub> Value: 5.10 | Calculated TUC Value: 19.61  |      |      |      |      |      |      | Permit Limits: NA |                    |
| 95% Confidence Limits: NA    |  |      |      |      |      |      |      |                   |                    |
| UL: 11.12                    | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |      |      |      |      |      |      | Limit Values: NA  |                    |
| LL: 3.17                     |  |      |      |      |      |      |      |                   |                    |
| UL = Upper Limit             |  |      |      |      |      |      |      |                   |                    |
| LL = Lower Limit             |  |      |      |      |      |      |      |                   |                    |

<sup>a</sup>Normally, a control is included with each outfall tested; however, only two controls were included during the test because of a lack of sufficient larvae. Mean control weight was calculated by using eight control replicates.

**Results of a *Pimephales promelas* Chronic Toxicity Test, 04/08-15/97 Using Effluent from Outfall 017**

| Test Solution                  | Percent Surviving (day)  |      |      |      |      |      |      | Dry Weight (mg/L)  |                    |
|--------------------------------|--|------|------|------|------|------|------|--|--------------------|
|                                | 1  | 2    | 3    | 4    | 5    | 6    | 7    | Total  | Mean               |
| Control 1                      | 100  | 100  | 100  | 100  | 100  | 100  | 100  | 80   | 0.376 <sup>a</sup> |
| Control 2                      | 100  | 100  | 100  | 100  | 100  | 100  | 100  |  |                    |
| 100% Effluent                  | 100  | 100  | 87.5 | 77.5 | 67.5 | 67.5 | 67.5 | 40   | 0.354              |
| 50% Effluent                   | 97.5   | 97.5 | 97.5 | 90.0 | 77.5 | 75.0 | 62.5 | 40   | 0.340              |
| 25% Effluent                   | 100  | 100  | 100  | 90.0 | 82.5 | 77.5 | 77.5 | 40   | 0.353              |
| 12% Effluent                   | 100  | 100  | 97.5 | 75.0 | 45.0 | 37.5 | 32.5 | 40   | 0.172              |
| 6% Effluent                    | 97.5   | 97.5 | 95.0 | 95.0 | 95.0 | 95.0 | 95.0 | 40   | 0.442              |
| IC <sub>50</sub> Value: > 100% | Calculated TUC Value: < 1.0  |      |      |      |      |      |      | Permit Limits: NA  |                    |
| 95% Confidence Limits: NA      |  |      |      |      |      |      |      |  |                    |
| UL:                            | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |      |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |                    |
| LL:                            |  |      |      |      |      |      |      |  |                    |
| UL = Upper Limit               |  |      |      |      |      |      |      |  |                    |
| LL = Lower Limit               |  |      |      |      |      |      |      |  |                    |

<sup>a</sup>Normally, a control is included with each outfall tested; however, only two controls were included during the test because of a lack of sufficient larvae. Mean control weight was calculated by using eight control replicates.

**Results of a *Pimephales promelas* Chronic Toxicity Test, 04/08-15/97 Using Effluent from Outfall 018**

| Test Solution                  | Percent Surviving (day)  |      |      |      |      |      |      | Dry Weight (mg/L)  |                    |
|--------------------------------|--|------|------|------|------|------|------|--|--------------------|
|                                | 1  | 2    | 3    | 4    | 5    | 6    | 7    | Total  | Mean               |
| Control 1                      | 100  | 100  | 100  | 100  | 100  | 100  | 100  | 80   | 0.376 <sup>a</sup> |
| Control 2                      | 100  | 100  | 100  | 100  | 100  | 100  | 100  |  |                    |
| 100% Effluent                  | 97.5   | 97.5 | 92.5 | 65.0 | 60.0 | 60.0 | 60.0 | 40   | 0.371              |
| 50% Effluent                   | 97.5   | 97.5 | 95.0 | 72.5 | 62.5 | 60.0 | 60.0 | 40   | 0.287              |
| 25% Effluent                   | 97.5   | 97.5 | 97.5 | 90.0 | 90.0 | 87.5 | 82.5 | 40   | 0.364              |
| 12% Effluent                   | 100  | 100  | 100  | 87.5 | 85.0 | 85.0 | 82.5 | 40   | 0.367              |
| 6% Effluent                    | 100  | 100  | 97.5 | 97.5 | 97.5 | 97.5 | 97.5 | 40   | 0.359              |
| IC <sub>50</sub> Value: > 100% | Calculated TUC Value: < 1.0  |      |      |      |      |      |      | Permit Limits: NA  |                    |
| 95% Confidence Limits:         |  |      |      |      |      |      |      |  |                    |
| UL:                            | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |      |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |                    |
| LL:                            |  |      |      |      |      |      |      |  |                    |
| UL = Upper Limit               |  |      |      |      |      |      |      |  |                    |
| LL = Lower Limit               |  |      |      |      |      |      |      |  |                    |

<sup>a</sup>Normally, a control is included with each outfall tested; however, only two controls were included during the test because of a lack of sufficient larvae. Mean control weight was calculated by using eight control replicates.

**C-20 — Biological Monitoring Program**

**Results of a *Pimephales promelas* Chronic Toxicity Test, 07/10-17/97 Using Effluent from Outfall 013**

| Test Solution                  | Percent Surviving (day)  |      |      |      |      |      |      | Dry Weight (mg/L) |       |
|--------------------------------|--|------|------|------|------|------|------|-------------------|-------|
|                                | 1  | 2    | 3    | 4    | 5    | 6    | 7    | Total             | Mean  |
| Control                        | 97.5   | 97.5 | 97.5 | 97.5 | 97.5 | 95.0 | 95.0 | 40                | 0.484 |
| 100% Effluent                  | 100  | 100  | 100  | 100  | 100  | 100  | 97.4 | 39 <sup>a</sup>   | 0.643 |
| 50% Effluent                   | 90.0   | 87.5 | 87.5 | 87.5 | 87.5 | 82.5 | 82.5 | 40                | 0.520 |
| 25% Effluent                   | 100  | 100  | 100  | 97.5 | 97.5 | 97.5 | 97.5 | 40                | 0.621 |
| 12% Effluent                   | 100  | 100  | 97.5 | 97.5 | 95.0 | 95.0 | 95.0 | 40                | 0.565 |
| 6% Effluent                    | 97.5   | 97.5 | 97.5 | 97.5 | 97.5 | 97.5 | 97.5 | 40                | 0.495 |
| IC <sub>50</sub> Value: > 100% | Calculated TUc Value: < 1.0  |      |      |      |      |      |      |                   |       |
| 95% Confidence Limits: NA      |  |      |      |      |      |      |      | Permit Limits: NA |       |
| UL:                            | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |      |      |      |      |      |      |                   |       |
| LL:                            |  |      |      |      |      |      |      |                   |       |
| UL = Upper Limit               |  |      |      |      |      |      |      |                   |       |
| LL = Lower Limit               |  |      |      |      |      |      |      |                   |       |

<sup>a</sup>One test organism was missing on Day 4; therefore, percent surviving and mean growth were based on 39 test organisms.

**Results of a *Pimephales promelas* Chronic Toxicity Test, 07/10-17/97 Using Effluent from Outfall 015**

| Test Solution                 | Percent Surviving (day)  |      |      |      |      |      |      | Dry Weight (mg/L) |       |
|-------------------------------|--|------|------|------|------|------|------|-------------------|-------|
|                               | 1  | 2    | 3    | 4    | 5    | 6    | 7    | Total             | Mean  |
| Control                       | 97.5   | 97.5 | 97.5 | 87.5 | 87.5 | 85.0 | 82.5 | 40                | 0.379 |
| 100% Effluent                 | 97.5   | 95.0 | 90.0 | 90.0 | 87.5 | 75.0 | 47.5 | 40                | 0.287 |
| 50% Effluent                  | 92.5   | 92.5 | 92.5 | 85.0 | 85.0 | 82.5 | 47.5 | 40                | 0.208 |
| 25% Effluent                  | 100  | 100  | 97.5 | 95.0 | 92.5 | 92.5 | 90.0 | 40                | 0.656 |
| 12% Effluent                  | 95.0   | 92.5 | 92.5 | 90.0 | 87.5 | 87.5 | 85.0 | 40                | 0.566 |
| 6% Effluent                   | 100  | 100  | 97.5 | 95.0 | 92.5 | 92.5 | 92.5 | 40                | 0.576 |
| IC <sub>50</sub> Value: 36.46 | Calculated TUc Value: 2.74   |      |      |      |      |      |      |                   |       |
| 95% Confidence Limits:        |  |      |      |      |      |      |      | Permit Limits: NA |       |
| UL: 38.46                     | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |      |      |      |      |      |      |                   |       |
| LL: 34.86                     |  |      |      |      |      |      |      |                   |       |
| UL = Upper Limit              |  |      |      |      |      |      |      |                   |       |
| LL = Lower Limit              |  |      |      |      |      |      |      |                   |       |

Results of a *Pimephales promelas* Chronic Toxicity Test, 07/10-17/97 Using Effluent from Outfall 016

| Test Solution                  | Percent Surviving (day)  |      |      |      |      |      |      | Dry Weight (mg/L) |       |
|--------------------------------|--|------|------|------|------|------|------|-------------------|-------|
|                                | 1  | 2    | 3    | 4    | 5    | 6    | 7    | Total             | Mean  |
| Control                        | 95.0   | 95.0 | 95.0 | 90.0 | 82.5 | 80.0 | 80.0 | 40                | 0.388 |
| 100% Effluent                  | 97.5   | 97.5 | 97.5 | 95.0 | 95.0 | 95.0 | 95.0 | 40                | 0.590 |
| 50% Effluent                   | 95.0   | 95.0 | 87.5 | 80.0 | 77.5 | 70.0 | 70.0 | 40                | 0.467 |
| 25% Effluent                   | 95.0   | 92.5 | 90.0 | 90.0 | 87.5 | 87.5 | 87.5 | 40                | 0.523 |
| 12% Effluent                   | 100  | 100  | 100  | 100  | 100  | 100  | 97.5 | 40                | 0.668 |
| 6% Effluent                    | 100  | 100  | 95.0 | 95.0 | 92.5 | 92.5 | 92.5 | 40                | 0.457 |
| IC <sub>50</sub> Value: > 100% | Calculated TUC Value: < 1.0  |      |      |      |      |      |      | Permit Limits: NA |       |
| 95% Confidence Limits: NA      |  |      |      |      |      |      |      |                   |       |
| UL:                            | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |      |      |      |      |      |      | Permit Limits: NA |       |
| LL:                            |  |      |      |      |      |      |      |                   |       |
| UL = Upper Limit               |  |      |      |      |      |      |      |                   |       |
| LL = Lower Limit               |  |      |      |      |      |      |      |                   |       |

Results of a *Pimephales promelas* Chronic Toxicity Test, 07/10-17/97 Using Effluent from Outfall 017

| Test Solution                  | Percent Surviving (day)  |      |      |      |      |      |      | Dry Weight (mg/L) |       |
|--------------------------------|--|------|------|------|------|------|------|-------------------|-------|
|                                | 1  | 2    | 3    | 4    | 5    | 6    | 7    | Total             | Mean  |
| Control                        | 95.5   | 95.0 | 95.0 | 92.5 | 92.5 | 87.5 | 87.5 | 40                | 0.338 |
| 100% Effluent                  | 100  | 100  | 100  | 92.5 | 90.0 | 87.5 | 87.5 | 40                | 0.539 |
| 50% Effluent                   | 100  | 100  | 100  | 97.5 | 97.5 | 87.5 | 82.5 | 40                | 0.469 |
| 25% Effluent                   | 100  | 100  | 97.5 | 97.5 | 97.5 | 97.5 | 95.0 | 40                | 0.474 |
| 12% Effluent                   | 97.5   | 97.5 | 97.5 | 97.5 | 95.0 | 95.0 | 90.0 | 40                | 0.448 |
| 6% Effluent                    | 90.0   | 90.0 | 87.5 | 85.0 | 85.0 | 85.0 | 85.0 | 40                | 0.492 |
| IC <sub>50</sub> Value: > 100% | Calculated TUC Value: < 1.0  |      |      |      |      |      |      | Permit Limits: NA |       |
| 95% Confidence Limits: NA      |  |      |      |      |      |      |      |                   |       |
| UL:                            | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |      |      |      |      |      |      | Permit Limits: NA |       |
| LL:                            |  |      |      |      |      |      |      |                   |       |
| UL = Upper Limit               |  |      |      |      |      |      |      |                   |       |
| LL = Lower Limit               |  |      |      |      |      |      |      |                   |       |

**C-22 — Biological Monitoring Program**

**Results of a *Pimephales promelas* Chronic Toxicity Test, 07/10-17/97 Using Effluent from Outfall 018**

| Test Solution                  | Percent Surviving (day)  |      |      |      |      |      |      | Dry Weight (mg/L) |       |  |  |
|--------------------------------|--|------|------|------|------|------|------|-------------------|-------|--|--|
|                                | 1  | 2    | 3    | 4    | 5    | 6    | 7    | Total             | Mean  |  |  |
| Control                        | 97.5   | 97.5 | 97.5 | 97.5 | 97.5 | 97.5 | 97.5 | 40                | 0.414 |  |  |
| 100% Effluent                  | 92.5   | 92.5 | 92.5 | 90.0 | 90.0 | 87.5 | 85.0 | 40                | 0.531 |  |  |
| 50% Effluent                   | 100  | 100  | 100  | 97.5 | 97.5 | 97.5 | 97.5 | 40                | 0.625 |  |  |
| 25% Effluent                   | 100  | 100  | 100  | 95.0 | 94.9 | 94.9 | 94.9 | 39 <sup>a</sup>   | 0.652 |  |  |
| 12% Effluent                   | 100  | 100  | 100  | 100  | 100  | 97.5 | 97.5 | 40                | 0.628 |  |  |
| 6% Effluent                    | 100  | 95.0 | 95.0 | 90.0 | 90.0 | 90.0 | 90.0 | 40                | 0.536 |  |  |
| IC <sub>50</sub> Value: > 100% | Calculated TUC Value: < 1.0  |      |      |      |      |      |      | Permit Limits: NA |       |  |  |
| 95% Confidence Limits:         |  |      |      |      |      |      |      |                   |       |  |  |
| UL:                            | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |      |      |      |      |      |      |                   |       |  |  |
| LL:                            |  |      |      |      |      |      |      |                   |       |  |  |
| UL = Upper Limit               |  |      |      |      |      |      |      |                   |       |  |  |
| LL = Lower Limit               |  |      |      |      |      |      |      |                   |       |  |  |

<sup>a</sup>One test organism was inadvertently killed on Day 5; therefore, percent surviving and mean growth were based on 39 test organisms.

**Results of a *Pimephales promelas* Chronic Toxicity Test, 12/02-09/97 Using Effluent from Outfall 013**

| Test Solution                  | Percent Surviving (day)  |     |      |      |      |      |      | Dry Weight (mg/L) |       |  |  |
|--------------------------------|--|-----|------|------|------|------|------|-------------------|-------|--|--|
|                                | 1  | 2   | 3    | 4    | 5    | 6    | 7    | Total             | Mean  |  |  |
| Control                        | 100  | 100 | 100  | 100  | 100  | 100  | 100  | 40                | 0.400 |  |  |
| 100% Effluent                  | 100  | 100 | 87.5 | 77.5 | 72.5 | 72.5 | 72.5 | 40                | 0.355 |  |  |
| 50% Effluent                   | 100  | 100 | 87.5 | 72.5 | 70.0 | 70.0 | 67.5 | 40                | 0.356 |  |  |
| 25% Effluent                   | 100  | 100 | 97.5 | 85.0 | 85.0 | 85.0 | 85.0 | 40                | 0.380 |  |  |
| 12% Effluent                   | 100  | 100 | 97.5 | 97.5 | 92.5 | 90.0 | 87.5 | 40                | 0.366 |  |  |
| 6% Effluent                    | 100  | 100 | 100  | 90.0 | 87.5 | 87.5 | 87.5 | 40                | 0.370 |  |  |
| IC <sub>50</sub> Value: > 100% | Calculated TUC Value: < 1.0  |     |      |      |      |      |      | Permit Limits: NA |       |  |  |
| 95% Confidence Limits: NA      |  |     |      |      |      |      |      |                   |       |  |  |
| UL:                            | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |     |      |      |      |      |      |                   |       |  |  |
| LL:                            |  |     |      |      |      |      |      |                   |       |  |  |
| UL = Upper Limit               |  |     |      |      |      |      |      |                   |       |  |  |
| LL = Lower Limit               |  |     |      |      |      |      |      |                   |       |  |  |

Results of a *Pimephales promelas* Chronic Toxicity Test, 12/02-09/97 Using Effluent from Outfall 015

| Test Solution                  | Percent Surviving (day) |      |      |      |      |      |      | Dry Weight (mg/L)  |       |
|--------------------------------|-------------------------|------|------|------|------|------|------|--|-------|
|                                | 1                       | 2    | 3    | 4    | 5    | 6    | 7    | Total  | Mean  |
| Control                        | 100                     | 100  | 100  | 100  | 100  | 97.5 | 97.5 | 40   | 0.430 |
| 100% Effluent                  | 97.5                    | 97.5 | 90.0 | 80.0 | 67.5 | 67.5 | 67.5 | 40   | 0.400 |
| 50% Effluent                   | 97.5                    | 97.5 | 97.5 | 90.0 | 80.0 | 67.5 | 67.5 | 40   | 0.430 |
| 25% Effluent                   | 97.5                    | 97.5 | 92.5 | 90.0 | 80.0 | 77.5 | 77.5 | 40   | 0.417 |
| 12% Effluent                   | 97.5                    | 97.5 | 97.5 | 97.5 | 92.5 | 92.5 | 92.5 | 40   | 0.552 |
| 6% Effluent                    | 97.5                    | 97.5 | 97.5 | 97.5 | 92.5 | 92.5 | 92.5 | 40   | 0.485 |
| IC <sub>25</sub> Value: > 100% |                         |      |      |      |      |      |      | Calculated TUc Value: < 1.0  |       |
| 95% Confidence Limits: NA      |                         |      |      |      |      |      |      | Permit Limits: NA  |       |
| UL:                            |                         |      |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |
| LL:                            |                         |      |      |      |      |      |      |  |       |
| UL = Upper Limit               |                         |      |      |      |      |      |      |  |       |
| LL = Lower Limit               |                         |      |      |      |      |      |      |  |       |

Results of a *Pimephales promelas* Chronic Toxicity Test, 12/02-09/97 Using Effluent from Outfall 016

| Test Solution                                   | Percent Surviving (day) |      |      |      |      |      |      | Dry Weight (mg/L)  |       |
|---|-------------------------|------|------|------|------|------|------|--|-------|
|   | 1                       | 2    | 3    | 4    | 5    | 6    | 7    | Total  | Mean  |
| Control   | 100                     | 100  | 100  | 100  | 100  | 100  | 100  | 40   | 0.462 |
| 100% Effluent                                   | 100                     | 100  | 97.5 | 90.0 | 75.0 | 70.0 | 70.0 | 40   | 0.376 |
| 50% Effluent                                    | 97.5                    | 97.4 | 92.3 | 87.2 | 66.7 | 53.8 | 53.8 | 39*  | 0.312 |
| 25% Effluent                                    | 97.5                    | 97.5 | 92.5 | 85.0 | 62.5 | 62.5 | 60.0 | 40   | 0.361 |
| 12% Effluent                                    | 100                     | 100  | 97.5 | 92.5 | 82.5 | 72.5 | 65.0 | 40   | 0.422 |
| 6% Effluent                                     | 97.5                    | 97.5 | 97.5 | 97.5 | 95.0 | 92.5 | 90.0 | 40   | 0.502 |
| IC <sub>25</sub> Value: > 100% (See Note below) |                         |      |      |      |      |      |      | Calculated TUc Value: < 1.0  |       |
| 95% Confidence Limits: See Note below           |                         |      |      |      |      |      |      | Permit Limits: NA  |       |
| UL:   |                         |      |      |      |      |      |      | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |
| LL:   |                         |      |      |      |      |      |      |  |       |
| UL = Upper Limit                                |                         |      |      |      |      |      |      |  |       |
| LL = Lower Limit                                |                         |      |      |      |      |      |      |  |       |

\*One test organism was inadvertently killed on Day 2; therefore, percent surviving and mean growth were based on 39 test organisms.

NOTE: The mean dry weight of the fish in the 50% concentration (0.312 mg/larvae) is an outlier. Charles Roth, at the Kentucky Division of Water, approved excluding the 50% concentration from the IC<sub>25</sub> calculation. Therefore, the IC<sub>25</sub> value for this outfall is > 100%.

**C-24 — Biological Monitoring Program**

**Results of a *Pimephales promelas* Chronic Toxicity Test, 12/02-09/97 Using Effluent from Outfall 017**

| Test Solution                  | Percent Surviving (day) |      |      |      |      |      |                             | Dry Weight (mg/L)  |       |
|--------------------------------|-------------------------|------|------|------|------|------|-----------------------------|--|-------|
|                                | 1                       | 2    | 3    | 4    | 5    | 6    | 7                           | Total  | Mean  |
| Control                        | 100                     | 100  | 100  | 100  | 100  | 100  | 100                         | 40   | 0.472 |
| 100% Effluent                  | 97.5                    | 97.5 | 97.5 | 95.0 | 82.5 | 82.5 | 80.0                        | 40   | 0.589 |
| 50% Effluent                   | 97.5                    | 95.0 | 90.0 | 90.0 | 72.5 | 70.0 | 70.0                        | 40   | 0.438 |
| 25% Effluent                   | 100                     | 97.5 | 92.5 | 92.5 | 87.5 | 85.0 | 82.5                        | 40   | 0.532 |
| 12% Effluent                   | 100                     | 100  | 97.5 | 97.5 | 97.5 | 97.5 | 97.5                        | 40   | 0.587 |
| 6% Effluent                    | 100                     | 100  | 100  | 100  | 100  | 100  | 100                         | 40   | 0.590 |
| IC <sub>50</sub> Value: > 100% |                         |      |      |      |      |      | Calculated TUc Value: < 1.0 |  |       |
| 95% Confidence Limits: NA      |                         |      |      |      |      |      | Permit Limits: NA           |  |       |
| UL:                            |                         |      |      |      |      |      |                             | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |
| LL:                            |                         |      |      |      |      |      |                             |  |       |
| UL = Upper Limit               |                         |      |      |      |      |      |                             |  |       |
| LL = Lower Limit               |                         |      |      |      |      |      |                             |  |       |

**Results of a *Pimephales promelas* Chronic Toxicity Test, 12/02-09/97 Using Effluent from Outfall 018**

| Test Solution                  | Percent Surviving (day) |      |      |      |      |      |                             | Dry Weight (mg/L)  |       |
|--------------------------------|-------------------------|------|------|------|------|------|-----------------------------|--|-------|
|                                | 1                       | 2    | 3    | 4    | 5    | 6    | 7                           | Total  | Mean  |
| Control                        | 100                     | 100  | 100  | 100  | 100  | 100  | 100                         | 40   | 0.490 |
| 100% Effluent                  | 100                     | 100  | 97.5 | 97.5 | 92.5 | 90.0 | 90.0                        | 40   | 0.582 |
| 50% Effluent                   | 100                     | 97.5 | 95.0 | 95.0 | 90.0 | 90.0 | 90.0                        | 40   | 0.635 |
| 25% Effluent                   | 100                     | 100  | 100  | 97.5 | 82.5 | 80.0 | 80.0                        | 40   | 0.516 |
| 12% Effluent                   | 100                     | 100  | 100  | 97.5 | 97.5 | 95.0 | 95.0                        | 40   | 0.624 |
| 6% Effluent                    | 100                     | 100  | 100  | 100  | 100  | 100  | 100                         | 40   | 0.606 |
| IC <sub>50</sub> Value: > 100% |                         |      |      |      |      |      | Calculated TUc Value: < 1.0 |  |       |
| 95% Confidence Limits:         |                         |      |      |      |      |      | Permit Limits: NA           |  |       |
| UL:                            |                         |      |      |      |      |      |                             | If acute test, method used to determine LC <sub>50</sub> and Confidence Limit Values: NA |       |
| LL:                            |                         |      |      |      |      |      |                             |  |       |
| UL = Upper Limit               |                         |      |      |      |      |      |                             |  |       |
| LL = Lower Limit               |                         |      |      |      |      |      |                             |  |       |

**Appendix D**

**RESULTS OF ANALYSES OF INDIVIDUAL FISH SAMPLES**

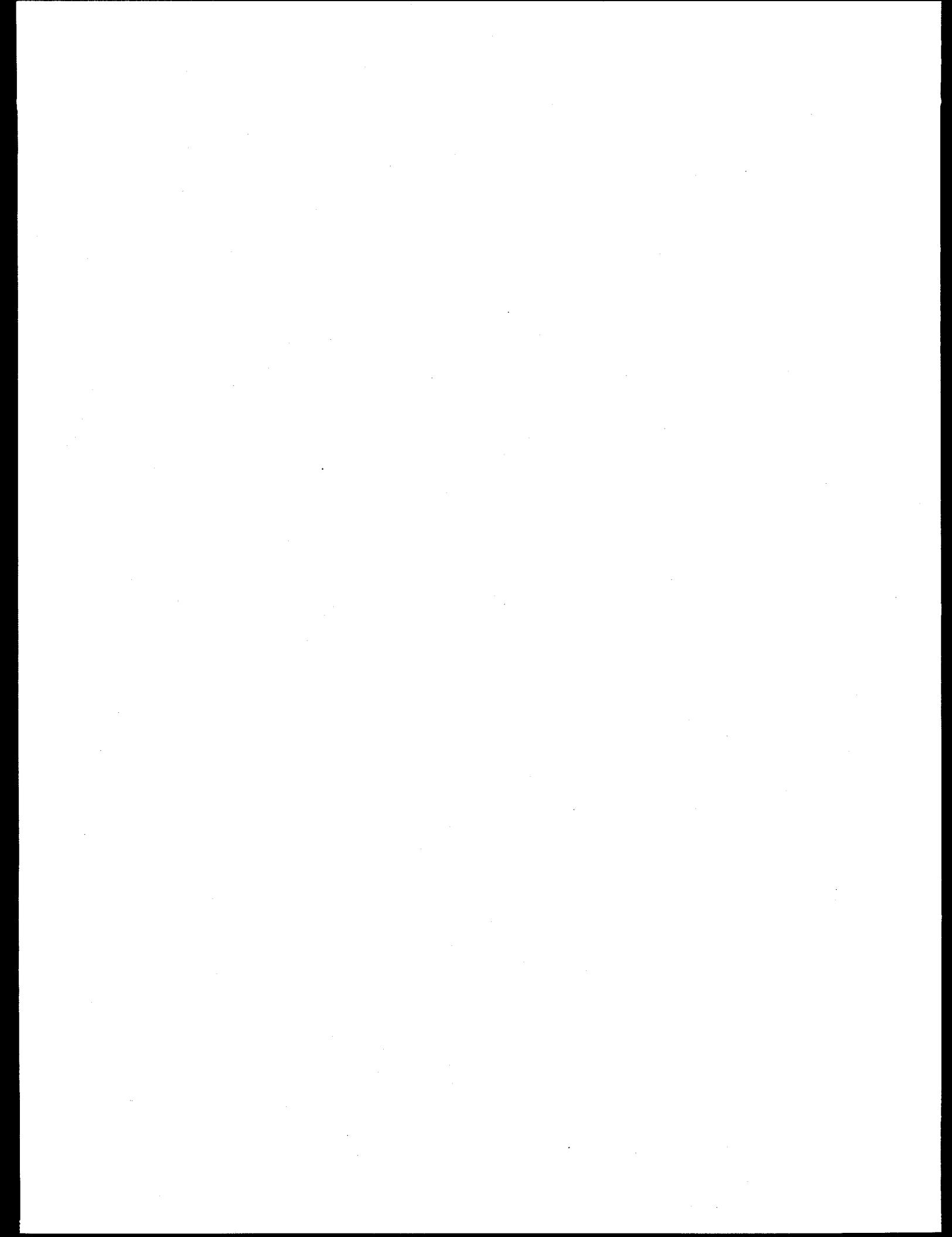


Table D.1. Concentrations of mercury and PCBs in individual fish collected from Little Bayou Creek and Big Bayou Creek

| Site <sup>a</sup> | Date <sup>b</sup> | Spp. <sup>c</sup> | Sex | Sample | Type <sup>d</sup> | Wt. <sup>e</sup> | Lgt. <sup>f</sup> | Hg <sup>g</sup> | 1248 <sup>h</sup> | Qual <sup>i</sup> | 1254 <sup>j</sup> | Qual | 1260 <sup>k</sup> | Qual | Lipids <sup>l</sup> |
|-------------------|-------------------|-------------------|-----|--------|-------------------|------------------|-------------------|-----------------|-------------------|-------------------|-------------------|------|-------------------|------|---------------------|
| BBK9.1            | 10/24/96          | SPOBASS           | F   | 10776  | R                 | 681              | 35.7              | 0.69            | 0.058             | U                 | 0.16              | .    | 0.13              | .    | 0.44                |
| BBK9.1            | 10/24/96          | SPOBASS           | M   | 10777  | R                 | 332              | 29.5              | 0.34            | 0.095             | U                 | 0.18              | .    | 0.17              | .    | 0.27                |
| BBK9.1            | 10/24/96          | SPOBASS           | F   | 10778  | R                 | 390              | 29.0              | 0.33            | 0.099             | U                 | 0.38              | .    | 0.18              | .    | 0.87                |
| BBK9.1            | 10/24/96          | SPOBASS           | M   | 10779  | R                 | 470              | 33.0              | 0.73            | 0.093             | U                 | 0.09              | .    | 0.48              | .    | 1.31                |
| BBK9.1            | 10/24/96          | SPOBASS           | .   | 10776  | D                 | .                | .                 | 0.51            | 0.1               | U                 | 0.11              | .    | 0.12              | .    | 0.19                |
| BBK9.1            | 10/24/96          | SPOBASS           | .   | 10777  | D                 | .                | .                 | .               | 0.1               | U                 | 0.10              | .    | 0.10              | .    | 0.13                |
| LUK9.0            | 10/24/96          | LONEAR            | .   | 10770  | R                 | 27.6             | 11.8              | .               | 0.14              | U                 | 0.14              | U    | 0.45              | .    | 0.41                |
| LUK9.0            | 10/24/96          | LONEAR            | .   | 10771  | R                 | 28.3             | 12.0              | .               | 0.22              | U                 | 0.22              | U    | 1.19              | .    | 0.09                |
| LUK9.0            | 10/24/96          | LONEAR            | .   | 10772  | R                 | 25.3             | 11.2              | .               | 0.12              | U                 | 0.24              | .    | 0.18              | .    | 0.34                |
| LUK9.0            | 10/24/96          | LONEAR            | .   | 10773  | R                 | 31.5             | 12.2              | .               | 0.11              | U                 | 0.57              | .    | 0.24              | .    | 0.25                |
| LUK9.0            | 10/24/96          | LONEAR            | .   | 10774  | R                 | 31.5             | 12.1              | .               | 0.098             | U                 | 0.25              | .    | 0.10              | .    | .                   |
| LUK9.0            | 10/24/96          | LONEAR            | .   | 10775  | R                 | 27.6             | 11.3              | .               | 0.1               | U                 | 0.42              | .    | 0.20              | .    | 0.19                |
| LUK7.2            | 10/24/96          | LONEAR            | M   | 10780  | R                 | 37.2             | 12                | .               | 0.094             | U                 | 0.67              | .    | 0.22              | .    | 0.84                |
| LUK7.2            | 10/24/96          | LONEAR            | M   | 10781  | R                 | 42.9             | 13.6              | .               | 0.07              | U                 | 0.42              | .    | 0.32              | .    | 0.07                |
| LUK7.2            | 10/24/96          | LONEAR            | M   | 10782  | R                 | 34.0             | 12.5              | .               | 0.089             | J                 | 0.56              | .    | 0.28              | .    | 0.36                |
| LUK7.2            | 10/24/96          | LONEAR            | M   | 10783  | R                 | 34.3             | 12.4              | .               | 0.08              | U                 | 0.34              | .    | 0.14              | .    | 0.21                |
| LUK7.2            | 10/24/96          | LONEAR            | M   | 10784  | R                 | 41.7             | 12.8              | .               | 0.075             | U                 | 0.60              | .    | 0.13              | .    | 1.68                |
| LUK7.2            | 10/24/96          | LONEAR            | M   | 10785  | R                 | 36.7             | 12.2              | .               | 0.095             | U                 | 0.45              | .    | 0.12              | .    | 2.59                |
| LUK4.3            | 10/24/96          | LONEAR            | M   | 10790  | R                 | 37.8             | 12.7              | .               | 0.099             | U                 | 0.09              | J    | 0.10              | U    | 0.12                |
| LUK4.3            | 10/24/96          | LONEAR            | M   | 10791  | R                 | 43.7             | 31.1              | .               | 0.079             | U                 | 0.08              | U    | 0.08              | U    | 0.4                 |
| LUK4.3            | 10/24/96          | LONEAR            | M   | 10792  | R                 | 47.8             | 13.3              | .               | .                 | .                 | .                 | .    | .                 | .    | .                   |
| LUK4.3            | 10/24/96          | LONEAR            | M   | 10793  | R                 | 50.9             | 13.3              | .               | 0.065             | U                 | 0.14              | .    | 0.08              | .    | 0.56                |
| LUK4.3            | 10/24/96          | LONEAR            | F   | 10794  | R                 | 43.4             | 12.8              | .               | 0.088             | U                 | 0.09              | U    | 0.09              | U    | 0.85                |
| LUK4.3            | 10/24/96          | LONEAR            | M   | 10795  | R                 | 41.6             | 13.2              | .               | 0.086             | U                 | 0.21              | .    | 0.11              | .    | 0.29                |
| HCK20.6           | 1/15/97           | REDBRE            | .   | 10826  | R                 | 53.8             | 14.6              | .               | 0.11              | U                 | 0.11              | U    | 0.11              | U    | 0.36                |
| HCK20.6           | 1/15/97           | REDBRE            | .   | 10827  | R                 | 78.6             | 16.4              | .               | 0.11              | U                 | 0.11              | U    | 0.01              | U    | 1.63                |
| HCK20.6           | 6/5/96            | REDBRE            | F   | 2731   | R                 | 87.0             | 16.4              | .               | 0.06              | U                 | 0.06              | U    | 0.06              | U    | 0.82                |
| HCK20.6           | 6/5/96            | REDBRE            | M   | 2735   | R                 | 93.5             | 16.6              | .               | 0.064             | U                 | 0.06              | U    | 0.06              | U    | 1.77                |
| LUK9.0            | 5/7/97            | LONEAR            | M   | 10960  | R                 | 36.2             | 12.3              | .               | 0.12              | .                 | 0.37              | .    | 0.29              | .    | 0.72                |
| LUK9.0            | 5/7/97            | LONEAR            | M   | 10961  | R                 | 42.1             | 12.9              | .               | 0.09              | .                 | 0.37              | .    | 0.23              | .    | 0.67                |
| LUK9.0            | 5/7/97            | LONEAR            | M   | 10962  | R                 | 41.8             | 13.0              | .               | 0.11              | P                 | 0.25              | .    | 0.24              | .    | 0.94                |
| LUK9.0            | 5/7/97            | LONEAR            | M   | 10963  | R                 | 64.6             | 14.0              | .               | 0.09              | U                 | 0.38              | P    | 0.09              | .    | 1.68                |
| LUK9.0            | 5/7/97            | LONEAR            | M   | 10964  | R                 | 54.5             | 14.0              | .               | 0.08              | P                 | 0.09              | P    | 0.42              | .    | 0.71                |
| LUK9.0            | 5/7/97            | LONEAR            | M   | 10965  | R                 | 47.9             | 13.4              | .               | 0.08              | .                 | 0.18              | P    | 0.31              | .    | 1.06                |
| LUK7.2            | 5/7/97            | LONEAR            | M   | 10970  | R                 | 34.2             | 11.4              | .               | 0.08              | U                 | 0.13              | P    | 0.09              | .    | 1.50                |
| LUK7.2            | 5/7/97            | LONEAR            | F   | 10971  | R                 | 49.6             | 12.6              | .               | 0.07              | U                 | 0.14              | .    | 0.09              | .    | 1.66                |
| LUK7.2            | 5/7/97            | LONEAR            | M   | 10972  | R                 | 52.3             | 12.4              | .               | 0.14              | P                 | 0.27              | .    | 0.17              | .    | 0.99                |
| LUK7.2            | 5/7/97            | LONEAR            | M   | 10973  | R                 | 54.8             | 12.3              | .               | 0.16              | .                 | 0.54              | P    | 0.15              | .    | 1.71                |
| LUK7.2            | 5/7/97            | LONEAR            | M   | 10974  | R                 | 43.8             | 12.0              | .               | 0.07              | U                 | 0.13              | .    | 0.12              | .    | 1.15                |
| LUK7.2            | 5/7/97            | LONEAR            | M   | 10975  | R                 | 58.3             | 13.0              | .               | 0.21              | .                 | 0.37              | .    | 0.18              | .    | 1.88                |

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Table D.1 (continued)

| Site <sup>a</sup> | Date <sup>b</sup> | Spp. <sup>c</sup> | Sex | Sample | Type <sup>d</sup> | Wt. <sup>e</sup> | Lgt. <sup>f</sup> | Hg <sup>g</sup> | 1248 <sup>h</sup> | Qual <sup>i</sup> | 1254 <sup>j</sup> | Qual <sup>k</sup> | 1260 <sup>k</sup> | Qual <sup>l</sup> | Lipids <sup>l</sup> |
|-------------------|-------------------|-------------------|-----|--------|-------------------|------------------|-------------------|-----------------|-------------------|-------------------|-------------------|-------------------|-------------------|-------------------|---------------------|
| LUK4.3            | 5/7/97            | LONEAR            | M   | 10990  | R                 | 42.6             | 11.7              | .               | 0.10              | U                 | 0.08              | J                 | 0.05              | J                 | 0.93                |
| LUK4.3            | 5/7/97            | LONEAR            | F   | 10991  | R                 | 61.5             | 13.0              | .               | 0.06              | U                 | 0.07              | .                 | 0.04              | J                 | 5.13                |
| LUK4.3            | 5/7/97            | LONEAR            | M   | 10992  | R                 | 49.5             | 12.4              | .               | 0.06              | U                 | 0.04              | J                 | 0.053             | J                 | 0.45                |
| LUK4.3            | 5/7/97            | LONEAR            | M   | 10993  | R                 | 48.8             | 12.7              | .               | 0.14              | U                 | 0.14              | U                 | 0.14              | U                 | 1.42                |
| LUK4.3            | 5/7/97            | LONEAR            | M   | 10994  | R                 | 37.0             | 11.1              | .               | 0.08              | U                 | 0.20              | P                 | 0.07              | J                 | 1.85                |
| MAK13.8           | 5/8/97            | LONEAR            | M   | 10976  | R                 | 46.1             | 12.1              | .               | 0.06              | U                 | 0.06              | U                 | 0.06              | U                 | 2.35                |
| MAK13.8           | 5/8/97            | LONEAR            | M   | 10977  | R                 | 44.4             | 12.0              | .               | 0.14              | U                 | 0.14              | U                 | 0.14              | U                 | 3.45                |
| MAK13.8           | 5/8/97            | LONEAR            | M   | 10978  | R                 | 35.8             | 11.4              | .               | 0.09              | U                 | 0.09              | U                 | 0.09              | U                 | 2.28                |
| MAK13.8           | 5/8/97            | LONEAR            | F   | 10979  | R                 | 38.8             | 11.7              | .               | 0.09              | U                 | 0.09              | U                 | 0.09              | U                 | 2.25                |
| LUK9.0            | 10/28/97          | LONEAR            | M   | 2800   | R                 | 33.0             | 13.2              | .               | 0.17              | U                 | 0.17              | U                 | 0.66              | .                 | 0.60                |
| LUK9.0            | 10/28/97          | LONEAR            | M   | 2801   | R                 | 28.4             | 11.7              | .               | 0.10              | U                 | 0.10              | U                 | 0.20              | .                 | 1.02                |
| LUK9.0            | 10/28/97          | LONEAR            | M   | 2802   | R                 | 27.9             | 11.4              | .               | 0.10              | U                 | 0.10              | U                 | 0.13              | .                 | 1.72                |
| LUK9.0            | 10/28/97          | LONEAR            | M   | 2803   | R                 | 26.1             | 11.6              | .               | 0.12              | U                 | 0.12              | U                 | 0.18              | .                 | 0.50                |
| LUK9.0            | 10/28/97          | LONEAR            | M   | 2804   | R                 | 31.1             | 12.4              | .               | 0.11              | U                 | 0.11              | U                 | 0.63              | .                 | 0.51                |
| LUK9.0            | 10/28/97          | LONEAR            | M   | 2805   | R                 | 25.8             | 10.8              | .               | 0.11              | U                 | 0.11              | U                 | 0.42              | .                 | 0.77                |
| LUK9.0            | 10/28/97          | LONEAR            | M   | 2800   | D                 | .                | .                 | .               | 0.14              | U                 | 0.14              | U                 | 0.51              | .                 | 0.26                |
| LUK7.2            | 10/28/97          | LONEAR            | M   | 2810   | R                 | 36.9             | 12.8              | .               | 0.10              | U                 | 0.10              | U                 | 0.57              | .                 | 0.76                |
| LUK7.2            | 10/28/97          | LONEAR            | M   | 2811   | R                 | 43.0             | 13.2              | .               | 0.11              | U                 | 0.22              | .                 | 0.42              | .                 | 0.55                |
| LUK7.2            | 10/28/97          | LONEAR            | M   | 2812   | R                 | 36.0             | 12.1              | .               | 0.08              | U                 | 0.08              | U                 | 0.12              | .                 | 0.84                |
| LUK7.2            | 10/28/97          | LONEAR            | M   | 2813   | R                 | 38.2             | 13.2              | .               | 0.10              | U                 | 0.10              | U                 | 0.27              | .                 | 0.64                |
| LUK7.2            | 10/28/97          | LONEAR            | M   | 2814   | R                 | 38.6             | 12.8              | .               | 0.09              | U                 | 0.09              | U                 | 0.14              | .                 | 1.54                |
| LUK7.2            | 10/28/97          | LONEAR            | M   | 2815   | R                 | 34.6             | 12.7              | .               | 0.11              | U                 | 0.35              | .                 | 0.76              | .                 | 0.67                |
| LUK7.2            | 10/28/97          | LONEAR            | M   | 2811   | D                 | .                | .                 | .               | 0.12              | U                 | 0.31              | .                 | 0.59              | .                 | 0.73                |
| LUK4.3            | 10/28/97          | LONEAR            | M   | 2820   | R                 | 43.9             | 12.3              | .               | 0.07              | U                 | 0.07              | U                 | 0.07              | U                 | 0.88                |
| LUK4.3            | 10/28/97          | LONEAR            | M   | 2821   | R                 | 39.5             | 12.6              | .               | 0.09              | U                 | 0.09              | U                 | 0.06              | J                 | 1.21                |
| LUK4.3            | 10/28/97          | LONEAR            | M   | 2822   | R                 | 41.7             | 12.9              | .               | 0.08              | U                 | 0.08              | U                 | 0.12              | .                 | 0.93                |
| LUK4.3            | 10/28/97          | LONEAR            | M   | 2823   | R                 | 37.6             | 12.4              | .               | 0.08              | U                 | 0.08              | U                 | 0.06              | J                 | 0.91                |
| LUK4.3            | 10/28/97          | LONEAR            | M   | 2824   | R                 | 49.5             | 12.7              | .               | 0.07              | U                 | 0.07              | U                 | 0.07              | J                 | 0.93                |
| LUK4.3            | 10/28/97          | LONEAR            | M   | 2825   | R                 | 57.9             | 14.5              | .               | 0.12              | U                 | 0.12              | U                 | 0.07              | J                 | 0.84                |
| LUK4.3            | 10/28/97          | LONEAR            | M   | 2825   | D                 | .                | .                 | .               | 0.11              | U                 | 0.11              | U                 | 0.09              | J                 | 0.49                |
| BBK9.1            | 10/28/97          | SPOBAS            | M   | 2830   | R                 | 296.3            | 28.7              | 0.35            | 0.02              | U                 | 0.02              | U                 | 0.06              | .                 | 0.53                |
| BBK9.1            | 10/28/97          | SPOBAS            | F   | 2831   | R                 | 270.9            | 26.9              | 0.33            | 0.03              | U                 | 0.03              | U                 | 0.07              | .                 | 0.90                |
| BBK9.1            | 10/28/97          | SPOBAS            | M   | 2832   | R                 | 269.0            | 27.5              | 0.22            | 0.03              | U                 | 0.03              | U                 | 0.06              | .                 | 0.67                |
| BBK9.1            | 10/28/97          | SPOBAS            | F   | 2833   | R                 | 143.1            | 22.4              | 0.13            | 0.04              | U                 | 0.04              | U                 | 0.07              | .                 | 0.58                |
| BBK9.1            | 10/28/97          | SPOBAS            | .   | 2830   | D                 | .                | .                 | 0.37            |                   |                   |                   |                   |                   |                   |                     |
| MAK13.8           | 10/28/97          | LONEAR            | F   | 2807   | R                 | 26.9             | 11.8              | .               | 0.14              | U                 | 0.14              | U                 | 0.14              | U                 | 0.68                |
| MAK13.8           | 10/28/97          | LONEAR            | M   | 2808   | R                 | 33.4             | 12.4              | .               | 0.10              | U                 | 0.10              | U                 | 0.10              | U                 | 0.77                |
| MAK13.8           | 10/28/97          | LONEAR            | M   | 2809   | R                 | 26.8             | 11.3              | .               | 0.12              | U                 | 0.12              | U                 | 0.12              | U                 | 0.99                |
| MAK13.8           | 10/28/97          | LONEAR            | M   | 2817   | R                 | 37.7             | 12.5              | .               | 0.10              | U                 | 0.10              | U                 | 0.10              | U                 | 0.72                |
| MAK13.8           | 10/28/97          | LONEAR            | M   | 2818   | R                 | 32.6             | 12.7              | .               | 0.11              | U                 | 0.11              | U                 | 0.11              | U                 | 0.69                |
| MAK13.8           | 10/28/97          | LONEAR            | M   | 2819   | R                 | 34.9             | 13.3              | .               | 0.13              | U                 | 0.13              | U                 | 0.13              | U                 | 0.76                |

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<sup>a</sup>Site designations are as follows: BBK = Big Bayou Creek kilometer; LUK = Little Bayou Creek kilometer; MAK = Massac Creek kilometer.

<sup>b</sup>Collection date.

<sup>c</sup>Species designations are as follows: LONEAR - Longear sunfish; SPOBASS - Spotted bass; REDBRE = redbreast sunfish.

<sup>d</sup>Type designations are as follows: R - regular sample; D - duplicate sample.

<sup>e</sup>Weight of fish measured in grams.

<sup>f</sup>Total length of fish measured in centimeters.

<sup>g</sup>Concentrations of Hg reported as  $\mu\text{g/g}$  wet weight.

<sup>h</sup>Concentrations in fish filets of Aroclor 1248 in  $\mu\text{g/g}$  wet weight.

<sup>i</sup>Data qualifiers for the three Aroclors. "U" indicates compound was analyzed for but not detected. The sample **quantitation limit** is listed. (*detection limits are estimated by using one tenth the quantitation limit*). "J" indicates an estimated value that is below the quantitation limit. "P" indicates greater than a 25% difference between the primary and secondary column results.

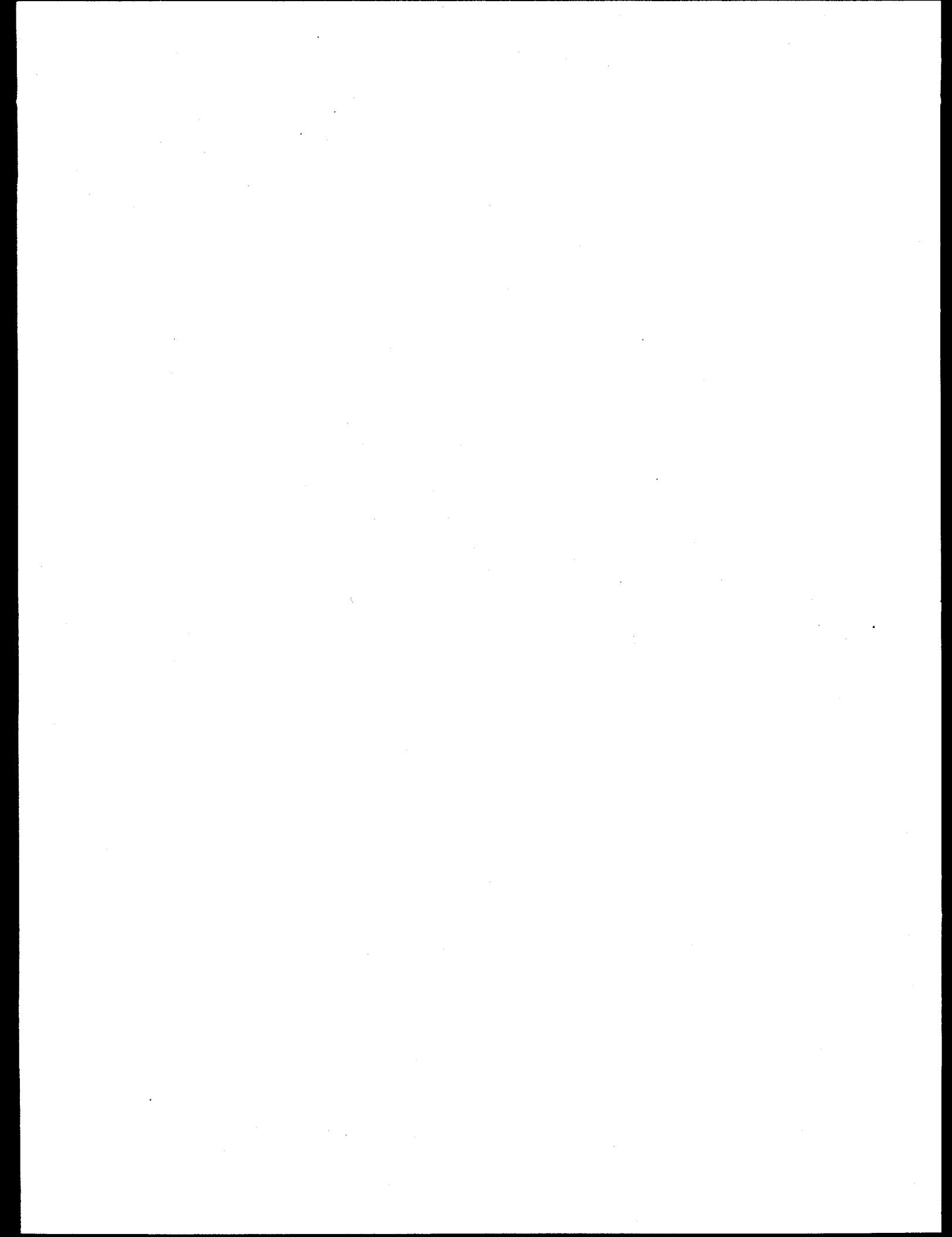
<sup>j</sup>Concentrations in fish filets of Aroclor 1254 in  $\mu\text{g/g}$  wet weight.

<sup>k</sup>Concentrations in fish filets of Aroclor 1260 in  $\mu\text{g/g}$  wet weight.

<sup>l</sup>Percent lipids reported for that sample.

**Appendix E**

**SPECIES CHARACTERISTICS, DENSITY, AND BIOMASS FOR  
FISH COMMUNITY DATA COLLECTED FROM BIG BAYOU  
CREEK, LITTLE BAYOU CREEK, AND MASSAC  
CREEK DURING MARCH AND  
SEPTEMBER 1997**



**Table E.1. Tolerance, feeding guilds, and lithophilic spawners for species found in and near the drainages of Big Bayou Creek, Little Bayou Creek, and Massac Creek**

| Species  | Tolerance <sup>a</sup> | Feeding guild <sup>b</sup> | Lithophilic spawner <sup>c</sup> |
|--|------------------------|----------------------------|----------------------------------|
| Spotted gar ( <i>Lepisosteus oculatus</i> )            |                        | PIS                        |                                  |
| Longnose gar ( <i>Lepisosteus osseus</i> )             |                        | PIS                        |                                  |
| Shortnose gar ( <i>Lepisosteus platostomus</i> )       |                        | PIS                        |                                  |
| Bowfin ( <i>Amia calva</i> )                           |                        | PIS                        |                                  |
| Gizzard shad ( <i>Dorosoma cepedianum</i> )            | TOL                    | GEN                        |                                  |
| Goldfish ( <i>Carassius auratus</i> )                  | TOL                    | GEN                        |                                  |
| Red shiner ( <i>Cyprinella lutrensis</i> )             | TOL                    |                            |                                  |
| Spotfin shiner ( <i>Cyprinella spiloptera</i> )        | TOL                    |                            |                                  |
| Steelcolor shiner ( <i>Cyprinella whipplei</i> )       | INTOL                  |                            |                                  |
| Common carp ( <i>Cyprinus carpio</i> )                 | TOL                    | GEN                        |                                  |
| Ribbon shiner ( <i>Lythrurus fumeus</i> )              | INTOL                  |                            |                                  |
| Silver chub ( <i>Macrhybopsis storeiana</i> )          |                        | BIN                        |                                  |
| Emerald shiner ( <i>Notropis atherinoides</i> )        |                        |                            | LITH                             |
| River shiner ( <i>Notropis blennius</i> )              |                        |                            | LITH                             |
| Sand shiner ( <i>Notropis stramineus</i> )             | INTOL                  |                            |                                  |
| Mimic shiner ( <i>Notropis volucellus</i> )            | INTOL                  |                            |                                  |
| Suckermouth minnow ( <i>Phenacobius mirabilis</i> )    |                        | BIN                        | LITH                             |
| Fathead minnow ( <i>Pimephales promelas</i> )          | TOL                    | GEN                        |                                  |
| Creek chub ( <i>Semotilus atromaculatus</i> )          | TOL                    | GEN                        |                                  |
| White sucker ( <i>Catostomus commersoni</i> )          | TOL                    | GEN                        | LITH                             |
| Creek chubsucker ( <i>Erimyzon oblongus</i> )          |                        | BIN                        |                                  |
| Smallmouth buffalo ( <i>Ictiobus bubalus</i> )         |                        | BIN                        |                                  |
| Bigmouth buffalo ( <i>Ictiobus cyprinellus</i> )       |                        | BIN                        |                                  |
| Black buffalo ( <i>Ictiobus niger</i> )                |                        | BIN                        |                                  |
| Spotted sucker ( <i>Minytrema melanops</i> )           | INTOL                  | GEN                        | LITH                             |
| Black redhorse ( <i>Moxostoma duquesnei</i> )          | INTOL                  | BIN                        | LITH                             |
| Golden redhorse ( <i>Moxostoma erythrurum</i> )        | INTOL                  | BIN                        | LITH                             |
| Black bullhead ( <i>Ameiurus melas</i> )               | TOL                    | GEN                        |                                  |
| Yellow bullhead ( <i>Ameiurus natalis</i> )            | TOL                    | GEN                        |                                  |
| Brown bullhead ( <i>Ameiurus nebulosus</i> )           | TOL                    | GEN                        |                                  |
| Tadpole madtom ( <i>Noturus gyrinus</i> )              | INTOL                  | BIN                        |                                  |
| Freckled madtom ( <i>Noturus nocturnus</i> )           | INTOL                  | BIN                        |                                  |
| Grass pickerel ( <i>Esox americanus vermiculatus</i> ) |                        | PIS                        |                                  |

Table E.1 (continued)

| Species   | Tolerance <sup>a</sup> | Feeding guild <sup>b</sup> | Lithophilic spawner <sup>c</sup> |
|---|------------------------|----------------------------|----------------------------------|
| Pirate perch ( <i>Aphredoderus sayanus</i> )      | BIN                    |                            |                                  |
| Brook silversides ( <i>Labidesthes sicculus</i> ) | INTOL                  |                            |                                  |
| Green sunfish ( <i>Lepomis cyanellus</i> )        | TOL                    |                            |                                  |
| Warmouth ( <i>Lepomis gulosus</i> )               |                        | GEN                        |                                  |
| Bluegill ( <i>Lepomis macrochirus</i> )           |                        | GEN                        |                                  |
| Longear sunfish ( <i>Lepomis megalotis</i> )      |                        | GEN                        |                                  |
| Redspotted sunfish ( <i>Lepomis miniatus</i> )    | BIN                    |                            |                                  |
| Spotted bass ( <i>Micropterus punctulatus</i> )   |                        | PIS                        |                                  |
| Largemouth bass ( <i>Micropterus salmoides</i> )  |                        | PIS                        |                                  |
| Mud darter ( <i>Etheostoma asprigene</i> )        | BIN                    |                            | LITH                             |
| Bluntnose darter ( <i>Etheostoma chlorosoma</i> ) | INTOL                  | BIN                        |                                  |
| Slough darter ( <i>Etheostoma gracile</i> )       |                        | BIN                        |                                  |
| Logperch ( <i>Percina caprodes</i> )              | INTOL                  | BIN                        | LITH                             |
| Blackside darter ( <i>Percina maculata</i> )      | INTOL                  | BIN                        | LITH                             |

<sup>a</sup>Tolerant (TOL) and sensitive (INTOL) species were tentatively identified for the Paducah area using collection records (Ryon and Carrico 1998) and text discussions in Becker 1983, Burr and Warren 1986, Cross and Collins 1975, Etnier and Starnes 1993, Karr et al. 1986, Lee et al. 1980, Ohio EPA 1988, Pfieger 1975, Robison and Buchanan 1988, Smith 1979, and Trautman 1981. Complete citations for references listed in this table are in Section 6 of this report.

<sup>b</sup>Feeding guilds are assigned to categories of interest in assessing impacts. Guilds include species that are primarily *generalists* (GEN), fish that feed on many types of food items and from many areas of the stream; *benthic insectivores* (BIN), those that eat macroinvertebrates associated with bottom substrates; and *piscivores* (PIS), fish that eat other fish.

<sup>c</sup>Lithophilic spawners (LITH) are species that release eggs randomly or without parental care in or onto gravel substrates. These species are especially vulnerable to siltation or low dissolved oxygen.

Table E.2. Fish densities (number/m<sup>2</sup>) in Big Bayou Creek, Little Bayou Creek, and a reference stream, Massac Creek, March 1997

| Species <sup>b</sup>   | Sites <sup>a</sup> |         |         |        |                      |
|------------------------|--------------------|---------|---------|--------|----------------------|
|                        | BBK9.1             | BBK10.0 | BBK12.5 | LUK7.2 | MAK13.8 <sup>a</sup> |
| Bowfin                 | <0.01              | -       | -       | -      | -                    |
| Gizzard shad           | <0.01              | -       | -       | -      | -                    |
| Central stoneroller    | 0.01               | 0.55    | 0.39    | 0.07   | 0.01                 |
| Red shiner             | -                  | -       | <0.01   | 0.10   | -                    |
| Steelcolor shiner      | -                  | -       | -       | -      | 0.03                 |
| Miss. silvery minnow   | -                  | -       | 0.01    | -      | 0.02                 |
| Redfin shiner          | -                  | -       | <0.01   | -      | <0.01                |
| Golden shiner          | -                  | -       | 0.01    | -      | <0.01                |
| Bluntnose minnow       | -                  | 0.02    | 0.01    | 0.27   | -                    |
| Fathead minnow         | -                  | -       | -       | 0.17   | -                    |
| Creek chub             | -                  | 0.03    | 0.07    | 0.06   | <0.01                |
| River carpsucker       | <0.01              | -       | -       | -      | -                    |
| White sucker           | <0.01              | -       | -       | -      | -                    |
| Creek chubsucker       | <0.01              | -       | <0.01   | -      | <0.01                |
| Smallmouth buffalo     | <0.01 <sup>c</sup> | -       | -       | -      | -                    |
| Black buffalo          | <0.01 <sup>c</sup> | -       | -       | -      | -                    |
| Spotted sucker         | 0.05               | <0.01   | -       | -      | <0.01                |
| Golden redhorse        | 0.02               | -       | -       | -      | <0.01                |
| Black bullhead         | 0.01               | 0.01    | 0.01    | 0.01   | -                    |
| Yellow bullhead        | <0.01              | <0.01   | 0.03    | 0.01   | <0.01                |
| Grass pickerel         | 0.01               | <0.01   | -       | -      | <0.01                |
| Pirate perch           | <0.01              | <0.01   | -       | 0.02   | <0.01                |
| Blackspotted topminnow | 0.01               | 0.04    | 0.03    | 0.14   | 0.03                 |
| Western mosquitofish   | -                  | -       | -       | 0.01   | -                    |
| Green sunfish          | 0.03               | 0.05    | 0.21    | 0.22   | 0.02                 |
| Warmouth               | <0.01              | -       | <0.01   | 0.05   | -                    |
| Bluegill               | 0.04               | 0.04    | 0.03    | 0.01   | 0.01                 |
| Longear sunfish        | 0.15               | 0.13    | 0.35    | 0.10   | 0.03                 |
| Redspotted sunfish     | -                  | -       | -       | <0.01  | -                    |
| Hybrid sunfish         | -                  | <0.01   | <0.01   | 0.01   | -                    |
| Spotted bass           | 0.01               | -       | <0.01   | -      | -                    |
| Largemouth bass        | -                  | <0.01   | <0.01   | -      | -                    |

Table E.2 (continued)

| Species <sup>b</sup> | Sites <sup>a</sup> |         |         |        |         |
|----------------------|--------------------|---------|---------|--------|---------|
|                      | BBK9.1             | BBK10.0 | BBK12.5 | LUK7.2 | MAK13.8 |
| Bluntnose darter     | -                  | -       | <0.01   | -      | -       |
| Slough darter        | -                  | <0.01   | <0.01   | 0.01   | -       |
| Logperch             | -                  | -       | -       | -      | <0.01   |
| Blackside darter     | -                  | -       | -       | -      | 0.01    |
| Species richness     | 20                 | 14      | 19      | 16     | 18      |
| Total density        | 0.36               | 0.88    | 1.16    | 1.26   | 0.18    |

<sup>a</sup>BBK = Big Bayou Creek kilometer, LUK = Little Bayou Creek kilometer, MAK = Massac Creek kilometer.

<sup>b</sup>Common and scientific names according to the American Fisheries Society (Robins et al. 1991).

Species identification confirmed by Dr. David A. Etnier, Department of Zoology, University of Tennessee.

<sup>a</sup>Sample was affected by high water with turbid conditions. Density was much lower than normal due to these adverse collecting conditions. Species richness was appropriate for site despite higher than normal flow.

Table E.3. Fish biomass (g/m<sup>2</sup>) in Big Bayou Creek, Little Bayou Creek, and a reference stream, Massac Creek, March 1997

| Species <sup>b</sup>   | Sites <sup>a</sup> |         |         |        |         |
|------------------------|--------------------|---------|---------|--------|---------|
|                        | BBK9.1             | BBK10.0 | BBK12.5 | LUK7.2 | MAK13.8 |
| Bowfin                 | 1.95               | -       | -       | -      | -       |
| Gizzard shad           | 0.36               | -       | -       | -      | -       |
| Central stoneroller    | 0.09               | 5.04    | 1.86    | 0.69   | 0.06    |
| Red shiner             | -                  | -       | 0.01    | 0.08   | -       |
| Steelcolor shiner      | -                  | -       | -       | -      | 0.08    |
| Miss. silvery minnow   | -                  | -       | 0.02    | -      | 0.03    |
| Redfin shiner          | -                  | -       | 0.01    | -      | <0.01   |
| Golden shiner          | -                  | -       | 0.03    | -      | 0.03    |
| Bluntnose minnow       | -                  | 0.10    | 0.03    | 0.78   | -       |
| Fathead minnow         | -                  | -       | -       | 0.29   | -       |
| Creek chub             | -                  | 0.98    | 1.00    | 2.12   | 0.02    |
| River carpsucker       | 1.68               | -       | -       | -      | -       |
| White sucker           | 0.26               | -       | -       | -      | -       |
| Creek chubsucker       | 0.39               | -       | 0.31    | -      | 0.06    |
| Smallmouth buffalo     | 0.13               | -       | -       | -      | -       |
| Black buffalo          | 0.10               | -       | -       | -      | -       |
| Spotted sucker         | 15.62              | 0.16    | -       | -      | 1.54    |
| Golden redhorse        | 13.22              | -       | -       | -      | 1.07    |
| Black bullhead         | 0.14               | 0.14    | 0.13    | 0.53   | -       |
| Yellow bullhead        | 0.14               | 0.05    | 1.11    | 0.10   | 0.01    |
| Grass pickerel         | 0.16               | 0.07    | -       | -      | 0.04    |
| Pirate perch           | 0.01               | 0.02    | -       | 0.16   | 0.01    |
| Blackspotted topminnow | 0.01               | 0.05    | 0.06    | 0.15   | 0.04    |
| Western mosquitofish   | -                  | -       | -       | <0.01  | -       |
| Green sunfish          | 0.10               | 0.19    | 0.63    | 0.67   | 0.23    |
| Warmouth               | 0.01               | -       | 0.01    | 0.16   | -       |
| Bluegill               | 1.10               | 0.11    | 0.27    | 0.13   | 0.02    |
| Longear sunfish        | 3.24               | 2.83    | 2.56    | 0.09   | 0.22    |
| Redspotted sunfish     | -                  | -       | -       | 0.02   | -       |
| Hybrid sunfish         | -                  | 0.03    | 0.08    | 0.05   | -       |
| Spotted bass           | 1.16               | -       | 0.01    | -      | -       |
| Largemouth bass        | -                  | 0.35    | 0.01    | -      | -       |
| Bluntnose darter       | -                  | -       | <0.01   | -      | -       |
| Slough darter          | -                  | <0.01   | <0.01   | 0.01   | -       |
| Logperch               | -                  | -       | -       | -      | 0.03    |
| Blackside darter       | -                  | -       | -       | -      | 0.03    |
| Total biomass          | 39.87              | 10.12   | 8.14    | 6.03   | 3.52    |

<sup>a</sup>BBK = Big Bayou Creek kilometer, LUK = Little Bayou Creek kilometer, MAK = Massac Creek kilometer.

<sup>b</sup>Common and scientific names according to the American Fisheries Society (Robins et al. 1991).

Table E.4. Fish densities (number/m<sup>2</sup>) in Big Bayou Creek, Little Bayou Creek, and a reference stream, Massac Creek, September 1997

| Species <sup>b</sup>   | Sites <sup>a</sup> |         |         |        |         |
|------------------------|--------------------|---------|---------|--------|---------|
|                        | BBK9.1             | BBK10.0 | BBK12.5 | LUK7.2 | MAK13.8 |
| Central stoneroller    | 1.02               | 3.89    | 1.84    | 0.05   | 2.75    |
| Red shiner             | <0.01              | -       | -       | 0.06   | <0.01   |
| Steelcolor shiner      | 0.01               | -       | -       | -      | 0.13    |
| Miss. silvery minnow   | 0.01               | -       | <0.01   | -      | 0.80    |
| Ribbon shiner          | 0.01               | -       | -       | -      | -       |
| Redfin shiner          | 0.03               | 0.01    | 0.01    | -      | -       |
| Golden shiner          | <0.01              | -       | 0.03    | <0.01  | -       |
| Bluntnose minnow       | <0.01              | 0.01    | <0.01   | 0.08   | 0.48    |
| Creek chub             | 0.05               | 0.03    | 0.38    | 0.42   | 0.58    |
| Creek chubsucker       | -                  | -       | 0.02    | -      | 0.09    |
| Bigmouth Buffalo       | <0.01              | -       | -       | -      | -       |
| Spotted sucker         | 0.01               | -       | -       | -      | -       |
| Golden redhorse        | -                  | -       | -       | -      | 0.07    |
| Black bullhead         | 0.01               | 0.02    | -       | -      | -       |
| Yellow bullhead        | 0.02               | 0.04    | 0.15    | 0.17   | 0.03    |
| Grass pickerel         | <0.01              | <0.01   | -       | <0.01  | -       |
| Pirate perch           | -                  | -       | -       | -      | 0.02    |
| Blackspotted topminnow | 0.12               | 0.31    | 0.52    | 0.39   | 0.41    |
| Western mosquitofish   | 0.05               | 0.21    | 0.16    | 0.22   | 0.01    |
| Flier                  | 0.01               | -       | -       | <0.01  | -       |
| Green sunfish          | 0.03               | 0.05    | 0.20    | 0.21   | 0.11    |
| Warmouth               | <0.01              | -       | -       | 0.02   | -       |
| Bluegill               | 0.05               | 0.26    | 0.06    | 0.01   | 0.11    |
| Longear sunfish        | 0.24               | 0.62    | 0.64    | 0.09   | 0.46    |
| Hybrid sunfish         | <0.01              | <0.01   | <0.01   | -      | <0.01   |
| Spotted bass           | 0.01               | 0.01    | 0.01    | -      | 0.02    |
| Largemouth bass        | 0.01               | 0.04    | 0.02    | <0.01  | 0.01    |
| Slough darter          | -                  | -       | -       | 0.02   | 0.03    |
| Logperch               | -                  | -       | -       | -      | 0.03    |
| Blackside darter       | -                  | -       | -       | -      | 0.02    |
| Species richness       | 23                 | 14      | 15      | 16     | 20      |
| Total density          | 1.70               | 5.50    | 4.04    | 1.74   | 6.16    |

<sup>a</sup>BBK = Big Bayou Creek kilometer, LUK = Little Bayou Creek kilometer, MAK = Massac Creek kilometer.

<sup>b</sup>Common and scientific names according to the American Fisheries Society (Robins et al. 1991).

**Table E.5. Fish biomass (g/m<sup>2</sup>) in Big Bayou Creek, Little Bayou Creek, and a reference stream, Massac Creek, September 1997**

| Species <sup>b</sup>   | Sites <sup>a</sup> |         |         |        |         |
|------------------------|--------------------|---------|---------|--------|---------|
|                        | BBK9.1             | BBK10.0 | BBK12.5 | LUK7.2 | MAK13.8 |
| Central stoneroller    | 2.32               | 10.85   | 4.69    | 0.14   | 3.39    |
| Red shiner             | <0.01              | -       | -       | 0.06   | <0.01   |
| Steelcolor shiner      | 0.02               | -       | -       | -      | 0.49    |
| Miss. silvery minnow   | 0.09               | -       | 0.03    | -      | 4.15    |
| Ribbon shiner          | 0.01               | -       | -       | -      | -       |
| Redfin shiner          | 0.02               | 0.01    | 0.03    | -      | -       |
| Golden shiner          | 0.01               | -       | 0.24    | 0.01   | -       |
| Bluntnose minnow       | <0.01              | 0.01    | 0.01    | 0.13   | 0.36    |
| Creek chub             | 0.15               | 0.38    | 2.04    | 1.94   | 2.54    |
| Creek chubsucker       | -                  | -       | 0.07    | -      | 1.03    |
| Bigmouth buffalo       | 0.95               | -       | -       | -      | -       |
| Spotted sucker         | 3.81               | -       | -       | -      | -       |
| Golden redhorse        | -                  | -       | -       | -      | 0.28    |
| Black bullhead         | 0.69               | 1.21    | -       | -      | -       |
| Yellow bullhead        | 0.29               | 0.46    | 1.10    | 0.67   | 0.42    |
| Grass pickerel         | 0.18               | 0.12    | -       | 0.10   | -       |
| Pirate perch           | -                  | -       | -       | -      | 0.11    |
| Blackspotted topminnow | 0.14               | 0.34    | 0.60    | 0.49   | 0.60    |
| Western mosquitofish   | 0.02               | 0.07    | 0.05    | 0.07   | <0.01   |
| Flier                  | 0.26               | -       | -       | 0.05   | -       |
| Green sunfish          | 0.52               | 0.77    | 1.04    | 1.49   | 0.76    |
| Warmouth               | 0.13               | -       | -       | 0.36   | -       |
| Bluegill               | 0.81               | 3.67    | 0.60    | 0.08   | 0.50    |
| Longear sunfish        | 5.52               | 4.93    | 2.37    | 0.67   | 3.98    |
| Hybrid sunfish         | 0.05               | 0.03    | 0.03    | -      | 0.02    |
| Spotted bass           | 0.96               | 0.20    | 0.06    | -      | 0.10    |
| Largemouth bass        | 0.65               | 0.84    | 0.18    | 0.02   | 0.08    |
| Slough darter          | -                  | -       | -       | 0.01   | 0.02    |
| Logperch               | -                  | -       | -       | -      | 0.19    |
| Blackside darter       | -                  | -       | -       | -      | 0.08    |
| Total biomass          | 17.60              | 23.89   | 13.11   | 6.29   | 19.10   |

<sup>a</sup>BBK = Big Bayou Creek kilometer, LUK = Little Bayou Creek kilometer, MAK = Massac Creek kilometer.

<sup>b</sup>Common and scientific names according to the American Fisheries Society (Robins et al. 1991).

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