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OAK RIDGE NATIONAL LABORATORY

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THIRD REPORT ON THE OAK RIDGE K-25 SITE BIOLOGICAL MONITORING AND ABATEMENT PROGRAM FOR MITCHELL BRANCH

R. L. Hinzman, Editor

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Environmental Sciences Division
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FOR THE UNITED STATES
DEPARTMENT OF ENERGY

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ENVIRONMENTAL SCIENCES DIVISION

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ACRONYMS AND INITIALISMS

ALT	alanine aminotransferase
ANOVA	analysis of variance
ASO	Analytical Services Organization
BCK	Bear Creek kilometer
BMAP	Biological Monitoring and Abatement Program
BMP	best management plan
BUN	blood urea nitrogen
CDFC	condition factor
CNF	Central Neutralization Facility
CRK	Clinch River kilometer
CRRI	Clinch River Remedial Investigation
DDT	Dichlorodiphenyl-trichloroethone-a pesticide
DO	dissolved oxygen
DOE	U.S. Department of Energy
EDTA	ethylenediaminetetracetic acid
EFK	East Fork Poplar Creek kilometer
EFPC	East Fork Poplar Creek
EPA	U.S. Environmental Protection Agency
EPT	Ephemeroptera, Plecoptera, and Tricoptera
EROD	7-ethoxyresorufin 0-deethylase
ESD	Environmental Sciences Division
FDA	U.S. Department of Agriculture Food and Drug Administration
GCK	Grassy Creek kilometer
GC/ECD	gas chromatography/electron capture detector
GC/MS	gas chromatography/mass spectrometry
GLM	general linear model
GSI	gonadal-somatic index
LC_{50}	concentration level (of toxin) that kills 50% of test organisms
LFPMA	functional parenchyma in liver
LMACA	macrophage aggregates in liver
LPARS	liver parasites
LSI	liver-somatic index
MAF	mean annual flow
MEK	Melton Branch kilometer

MGD	million gallons per day
MHR	Melton Hill Reservoir
MIK	Mitchell Branch kilometer
NAD(H)	nicotinamide-adenine dinucleotide (reduced form)
NADP(H)	nicotinamide-adenine dinucleotide phosphate (reduced form)
NOEL	no observed-effect level
NPDES	National Pollutant Discharge Elimination System
NTK	Northwest Tributary kilometer
ORGDP	Oak Ridge Gaseous Diffusion Plant
ORNL	Oak Ridge National Laboratory
ORR	Oak Ridge Reservation
PAH	polycyclic aromatic hydrocarbons
PCB	polychlorinated biphenyl
PCK	Poplar Creek kilometer
PHK	Pinhook Branch kilometer
PGV	preliminary guidance values
PPM	parts per million
QA	quality assurance
RIA	radioimmunoassay
SAS	Statistical Analysis System
SCK	Scarboro Creek kilometer
SD	storm drain
SE	standard error
STP	sewage treatment plant
TDEC	Tennessee Department of Environment and Conservation
TDS	total dissolved solids
TL	total length
TRC	total residual chlorine
TRK	Tennessee River kilometer
TSCA	Toxic Substances Control Act
TSS	total suspended solids
TVA	Tennessee Valley Authority
USGS	U.S. Geological Survey
VSI	visceral-somatic index
WCK	White Oak Creek kilometer
WOC	White Oak Creek
WOL	White Oak Lake

PREFACE

On September 11, 1986, a modified National Pollutant Discharge Elimination System (NPDES) permit was issued for the Oak Ridge Gaseous Diffusion Plant (now referred to as the Oak Ridge K-25 Site), a former uranium-enrichment production facility operated by Martin Marietta Energy Systems, Inc., for the U.S. Department of Energy. As required in Part III (L) of the permit, a plan for the biological monitoring of Mitchell Branch (K-1700 stream) was prepared and submitted for approval to the U.S. Environmental Protection Agency and the Tennessee Department of Environment and Conservation (Loar et al. 1992a). The plan, referred to as the Oak Ridge K-25 Site Biological Monitoring and Abatement Program (BMAP), described biomonitoring activities that would be conducted over the duration of the permit.

On October 1, 1992, a renewed NPDES permit was issued for the K-25 Site. A biological monitoring plan was submitted and approved for Mitchell Branch, Poplar Creek, Poplar Creek embayment of the Clinch River and any unnamed tributaries of these streams (Kszos et al. 1993).

This document is the third in a series of reports presenting the results of the studies that were conducted from June 1990 through December 1993 for the Oak Ridge K-25 Site BMAP although studies conducted outside this time period are included as appropriate. This report also addresses any significant modifications in the scope of work outlined in Kszos et al. 1993, but does not address changes in the scope of the BMAP which were implemented in March of 1995. Thus, the "future studies" sections in this report, written in early 1994, do not accurately reflect work conducted in 1995. The most recent changes to BMAP were directed by the K-25 Site office and approved by the Tennessee Department of Conservation and will be outlined in a new biological monitoring plan for the K-25 Site.

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EXECUTIVE SUMMARY

As a condition of the modified National Pollutant Discharge Elimination System (NPDES) permit issued to the Oak Ridge Gaseous Diffusion Plant (ORGDP; now referred to as the Oak Ridge K-25 Site) on September 11, 1986, a Biological Monitoring and Abatement Program (BMAP) was developed for the receiving stream (Mitchell Branch or K-1700 stream).

On October 1, 1992, a renewed NPDES permit was issued for the K-25 Site. A biological monitoring plan was submitted for Mitchell Branch, Poplar Creek, Poplar Creek Embayment of the Clinch River and any unnamed tributaries of these streams. The objectives of BMAP are to (1) demonstrate that the effluent limitations established for the Oak Ridge K-25 Site protect and maintain the use of Mitchell Branch for growth and propagation of fish and other aquatic life and (2) document the effects on stream biota resulting from operation of major new pollution abatement facilities, including the Central Neutralization Facility (CNF) and the Toxic Substances Control Act (TSCA) incinerator. The BMAP consists of four tasks: (1) toxicity monitoring; (2) bioaccumulation monitoring; (3) assessment of fish health; and (4) instream monitoring of biological communities, including benthic macroinvertebrates and fish. This document, the third in a series, reports on the results of the Oak Ridge K-25 Site BMAP; it describes studies that were conducted over various periods of time between June 1990 and December 1993, although monitoring conducted outside this time period is included, as appropriate.

Background

Mitchell Branch is a small stream that originates near the northeast boundary of the Oak Ridge K-25 Site; it flows only 1.5 km from its headwaters to its mouth on Poplar Creek at kilometer 7.0. The water quality of Mitchell Branch is influenced by the geology of the drainage basin, effluents entering the stream via storm drains, leachate from waste disposal sites, and remediation projects. The water quality of lower Mitchell Branch was characterized by (1) moderate levels of dissolved solids and occasionally high total suspended solids; (2) relatively low levels of nutrients; (3) elevated levels of some metals and organics; and (4)

elevated temperatures. The geology of Mitchell Branch watershed contributes to periods of no flow in its upper reaches, whereas flow in its lower reaches is augmented by as much as 10% by discharges from the Oak Ridge K-25 Site. Samples for BMAP were routinely collected from eight primary sites in Mitchell Branch. The site farthest upstream served as an undisturbed reference; the remaining sites were selected to coincide with the ambient NPDES monitoring station or to bracket known areas or sources of ecological disturbance. Additional reference sites on nearby streams were used for some tasks, depending on the specific objectives of the task.

Toxicity Testing

Tests were conducted bimonthly over the 1990 to 1994 sampling period with *Ceriodaphnia* dubia and fathead minnow larvae to assess the acute and chronic toxicity of (1) effluent from the CNF (K-1407-J) and the K-1203 Treatment Facility, (2) effluent from storm drains (SDs) 170, 180 and 190, and (3) ambient water from six sites on Mitchell Branch and five sites on Poplar Creek.

In February 1990 and April 1991 effluent from CNF would have exceeded the 2.11% no observed effect level (NOEL) in the new permit. However, the K-25 Site was operating under the old permit at that time, therefore, no violation occurred. CNF has not exceeded permit limits since the new permit became effective.

In July 1993 and March 1994, effluent from K-1203 exceeded the 4.2% NOEL limit. Confirmatory tests were conducted within 2 weeks of each exceedance and no toxicity was observed.

Full strength effluent from SDs 170 and 190 has consistently reduced survival and/or reproduction of *Ceriodaphnia*. Effluent from SD 180 reduced *Ceriodaphnia* survival or reproduction in less than half of the tests conducted. Samples of water from Mitchell Branch collected downstream of the storm drains only occasionally reduced survival or reproduction of *Ceriodaphnia*. Following dechlorination of the storm drains in 1991, survival of the *Ceriodaphnia* in laboratory tests was high during the first 48 hours of the test, thus indicating an elimination of acute toxicity in the storm drain samples.

Although the majority of tests from ambient sites show no chronic toxicity, *Ceriodaphnia* reproduction has been reduced 15 times at Mitchell Branch kilometers (MIKs) 0.71, 0.54,

0.45, and 0.12 in 1992–93. *Ceriodaphnia* have been shown to been more sensitive than fathead minnows to ambient water from Mitchell Branch. In each case where fathead minnow toxicity was observed, toxicity to *Ceriodaphnia* was also observed. As in past years, fathead minnow survival at MIK 1.43 remains low, indicating that this site is an inappropriate reference site for toxicity tests. It remains our hypothesis that the low survival is due to a naturally occurring pathogen, rather than to anthropogenic effects.

Bioaccumulation Studies

Mercury and polychlorinated biphenyls (PCBs) in fish and clams at sites near the K-25 Site were higher than levels found in fish and clams at two reference streams. Mercury bioaccumulation was observed in fish in Mitchell Branch and nearby sites on Poplar Creek. Mercury concentrations did not exceed the U.S. Department of Agriculture Food and Drug Administration (FDA) limit (1 μg/g), but some fish did exceed more conservative screening thresholds (e.g., preliminary guidance value, 0.42 μg/g). Experiments using caged and resident minnows suggests that the mercury contamination in Mitchell Branch fish arises from a source within that watershed, rather than being a consequence of contaminated fish immigrating from nearby Poplar Creek.

PCB concentrations were high in fish from two localized aquatic habitats at the K-25 Site (Mitchell Branch, K-1007-P1), but were much lower in fish from Poplar Creek in the vicinity of discharges from these sites. PCB screening of largemouth bass in the ponded tributaries at the K-25 site revealed significant contamination in fish from the K-1007-P1 pond, where the average concentration was 16 μg/g. PCB concentrations in caged clams that sampled waterborne PCBs for a fixed time interval were used to infer relative fluxes of PCBs from specific sources. Channel catfish from Poplar Creek continue to contain PCB concentrations similar to other sites in the Clinch River/Watts Bar system, which has resulted in the present fish consumption advisory posted by the Tennessee Department of Environment and Conservation (TDEC).

Metals screening of fish from lower Poplar Creek, Mitchell Branch, and the pond sites revealed little evidence of this form of contamination. Concentrations were either below detection limits or not substantially higher than reference values. Screening for organics other than PCBs in caged clams detected only traces of other organic compounds at very low concentrations, in most cases below the quantification limit of the analytical method.

Future studies will focus on documenting changes in mercury and PCB contamination in fish over time, improving estimates of the importance of K-25 PCB inputs to Poplar Creek relative to other sources, and assisting in locating the specific sources of PCB contamination at the K-25 Site.

Waterfowl were censused weekly at 13 locations on the K-25 Site. Because very low levels of PCBs and other contaminants occur in geese at the K-25 Site, and sufficient data have been collected to begin modeling of the Canada goose population, the census will be reduced to four sites: Oak Ridge National Laboratory (ORNL), the K-25 Site, the Y-12 Plant, and Melton Hill Dam. Contaminant levels will continue to be monitored by collecting geese and other waterfowl annually for radiological and nonradiological analyses. Where contaminants are known to exist (e.g., K-1007-B and K-901A ponds) additional samples will be taken and analyzed.

Twelve raccoons were trapped near Poplar Creek downstream of the K-25 Site in 1993. Fat and hair samples have been submitted for contaminant analyses.

Ten gizzard shad were collected from the K-1007-P1 pond and from Poplar Creek (near Mitchell Branch) for whole body contaminant analysis. Aliquots from homogenized samples were submitted for analysis of metals and PCBs.

Fish Health Studies

In all years (1988–1992) the integrated health responses (detoxification enzyme induction, organ dysfunction histopathology, overall health or condition, and nutritional or feeding status) for redbreast sunfish from Mitchell Branch were statistically distinct from those of sunfish collected from three reference sites, thus indicating that sunfish in Mitchell Branch ar in an unhealthy state. Although the difference in health response between sunfish from Mitchell Branch and the reference sites increased from 1989 to 1991 (suggesting a decline in health), it decreased in 1992 (suggesting an *improvement* in health). Dechlorination of the storm drain effluents may have accounted for part of this improvement. It was evident from the fish health response analysis that there was no consistent pattern among years in the importance of variables relative to intra-year site discrimination. This pattern suggests that fish health is influenced primarily by episodic events that occur in the system and that the primary stressors for these events may not be the same across time.

Length-frequency distributions and growth analyses of redbreast sunfish have improved over time. The 1992 distribution of sizes was considered balanced and was similar to the size distribution of sunfish found in the reference stream, Paint Rock Creek. By 1992 there was also no significant difference in total length at capture (growth) between Mitchell Branch and the reference stream for any age group.

Redbreast sunfish in Mitchell Branch are reproductively competent, but are spending an abnormally high amount of resources on gamete production compared with fish from the reference stream. This enhanced gamete production may be a stress response to xenobiotics in the diet or water, a result of nutrient enrichment, the consequence of elevated temperatures, the result of lessened competition, or some combination of these or other factors.

Medaka-embryo tests were used to evaluate the effect of water from several sites along Poplar Creek and two sites in Mitchell Branch on fish developmental processes. In three of the four tests conducted during 1993"94, medaka survival was significantly reduced in wateer from lower Mitchell Branch (MIK 0.12) compared with water from upper Mitchell Branch (MIK 1.43). The results of tests conducted on water from Poplar Creek were variable and suggest additional contaminant sources upstream of the K-25 Site and the ORR. Medaka studies in Mitchell Branch will be expanded to include additional reaches of stream closer to potential contaminant sources in Mitchell Branch (e.g., storm drains).

Population and Community Studies

The benthic macroinvertebrate community of Mitchell Branch within the main boundary of the K-25 Site continued to indicate the presence of poor water quality. Data collected through April 1993 showed a depression in total and EPT* richness from MIK 0.86 downstream to MIK 0.45 when compared with the reference site. Sites below MIK 1.43 were characterized by substantial fluctuations in density, total richness, and EPT richness between sampling periods. In contrast, the reference site was characterized by a relatively stable and diverse macroinvertebrate community that exhibited small fluctuations in sample parameters. Changes in EPT richness and total richness after 1989 indicate some recovery at all four

*Ephemeroptera, Plecoptera, and Tricoptera.

downstream sites. The appearance of pollution-intolerant groups, such as mayflies and stoneflies, in 1990 was also indicative of improved conditions. The period of greatest ecological change coincided closely with an improvement in water quality that was documented by ambient toxicity tests after 1988. Because recovery began prior to any significant operational changes at the K-25 Site, and soon after the end of a drought, the improvements may have been caused by increased dilution resulting from increased stream flows.

Fish population data from Mitchell Branch indicate that improvements in water quality have occurred. There has been an increase in the number of fish present in the fall 1990 through fall 1993 period. Length-frequency distributions indicate more stable populations of fish in Mitchell Branch in 1993 compared with earlier years. However, the species richness is still depressed compared with the reference stream. This lack of species may be due to past extermination and the prevention of recolonization from Poplar Creek and possibly by continued disturbances to the stream.

Future Studies

Routine sampling will continue for all tasks will the following changes. Because toxicity is rarely present at SD 180, monitoring of this storm drain will be eliminated. Due to the higher sensitivity of Ceriodaphnia and low minnow survival at MIK 1.43, subsequent ambient toxicity tests will be conducted with Ceriodaphnia only. These changes will allow us to concentrate on (1) investigating the source(s) of toxicity to SDs 170 and 190, (2) increasing the frequency of testing at Mitchell Branch sites from four times per year to six, (3) conducting tests of Mitchell Branch and the storm drains concurrently and (4) continuing to investigate potential sources of toxicity from other storm drains. Because no toxicity was observed in water from any site on Poplar Creek and the Clinch River, toxicity tests of these sites will be discontinued. Medaka embryo-larval tests will be expanded to include additional reaches of the stream closer to potential sources of toxic effects. A key objective of the biomonitoring efforts will be to identify and characterize PCB sources in Mitchell Branch and the K-1007 ponds. In addition to fish sampling, caged clams will be used to help evaluate these sources.

1. INTRODUCTION

R. L. Hinzman and L. A. Kszos

On September 11, 1986, a modified National Pollutant Discharge Elimination System (NPDES) permit was issued for the Oak Ridge Gaseous Diffusion Plant (ORGDP; now referred to as the Oak Ridge K-25 Site), a former enriched-uranium production facility currently operated by Martin Marietta Energy Systems, Inc., for the U.S. Department of Energy (DOE). As specified in Part III (L) of the permit, a plan for the biological monitoring of the receiving stream (K-1700 stream or Mitchell Branch) was submitted for approval to the U.S. Environmental Protection Agency (EPA) and the Tennessee Department of Environment and Conservation (TDEC) in December 1986. Because it was anticipated that the chemical composition of several effluents would be altered shortly after the permit modifications were issued, the Oak Ridge K-25 Site Biological Monitoring and Abatement Program (BMAP) was implemented in August 1986, before the plan had been formally approved by the regulatory agencies. On October 1, 1992, a renewed NPDES permit was issued for the K-25 Site. A biological monitoring plan was submitted for Mitchell Branch, Poplar Creek, Poplar Creek embayment of the Clinch River, and any unnamed tributaries of these streams as specified in Part III (F) (Kszos et al. 1993).

The initial BMAP for Mitchell Branch was developed to meet two major objectives. First, studies were designed to provide data sufficient to determine whether the interim effluent limits established for the K-25 Site protect and maintain the use of Mitchell Branch for growth and propagation of fish and other aquatic life. A second major objective was to document the effects on stream biota resulting from construction and operation of major new pollution abatement facilities, including the Central Neutralization Facility (CNF) and the Toxic Substances Control Act (TSCA) incinerator. The ecological effects of remedial actions (e.g., closure of the K-1407-B and K-1407-C holding ponds) can also be evaluated by this monitoring program.

The initial BMAP was modified based on results of the biological monitoring conducted since 1986 and discussion with the TDEC on November 12, 1992. The current objectives of BMAP place more emphasis on temporal than spatial trends, while continuing to identify and investigate contaminant sources associated with observed adverse effects. This change places

more emphasis on monitoring recovery through time in order to assess the effects (and effectiveness) of remedial actions.

The effluents discharged to Mitchell Branch are chemically complex and contain various trace metals, organic chemicals, neutral salts, and radionuclides (Sect. 2.2). Moreover, the composition of these effluents will change as various pollution abatement measures are implemented over the next several years. Although many contaminant inputs to the stream originate primarily as point sources from existing plant operations, area or non-point sources such as the classified burial grounds cannot be eliminated as potential sources of contaminants. An integrated approach to biological monitoring was developed to address this complexity. The Mitchell Branch BMAP consists of four major tasks: (1) toxicity monitoring, (2) bioaccumulation monitoring, (3) assessment of fish health and (4) instream monitoring of biological communities (e.g., benthic macroinvertebrates and fish).

2. DESCRIPTION OF STUDY AREA

R. L. Hinzman and W. K. Roy

Mitchell Branch is a small stream located near the northeast boundary of the Oak Ridge K-25 Site (Figs. 2.1 and 2.2). The stream has a drainage area of 1.78 km^2. Mitchell Branch flows only 1.5 km from its headwaters to its mouth, where it drains into Poplar Creek about 150 m downstream of the Blair Road bridge. The confluence of the two streams is about 1.5 km below the mouth of East Fork Poplar Creek (EFPC) and 7 km above the confluence of Poplar Creek with the Clinch River (Fig. 2.1).

Poplar Creek, which has a drainage area of 352 km^2, originates northeast of Oliver Springs, Tennessee, on the Cumberland Plateau (Figs. 2.1 and 2.3). The general direction of streamflow of Poplar Creek is southwesterly, entering the DOE Oak Ridge Reservation (ORR) upstream of the K-25 Site, and then flowing for a distance of 10 km through the plant area before entering the Clinch River near Clinch River kilometer (CRK) 19.3. EFPC, a major tributary of Poplar Creek, originates from springs on the northwest slope of Chestnut Ridge within the boundaries of the Oak Ridge Y-12 Plant. Streamflow is controlled by Lake Reality, a 1.0-ha settling pond located ~1.5 km below the spring. The creek flows for ~24 km before joining the West Fork at Poplar Creek kilometer (PCK) 8.8.

2.1 GEOHYDROLOGY *(R. L. Hinzman)*

2.1.1 Mitchell Branch

Mitchell Branch originates near the base of a small knoll southwest of McKinney Ridge. The knoll is underlain by the Conasauga Group, which consists of a calcareous shale interbedded with thinner layers of limestone and siltstone (DOE 1979). Streams such as Mitchell Branch, which are underlain by shale and sandstone, have a smaller low-flow discharge and greater range in flow than streams underlain by carbonate rocks such as Knox Dolomite (McMaster 1967).

Periods of zero discharge sometimes occur in portions of Mitchell Branch just upstream of the BMAP sampling site at Mitchell Branch kilometer (MIK) 0.86 [J. G. Smith, Environmental Sciences Division (ESD), Oak Ridge National Laboratory (ORNL), personal observation]. Periods of zero discharge are also characteristic of the upper reaches of Melton

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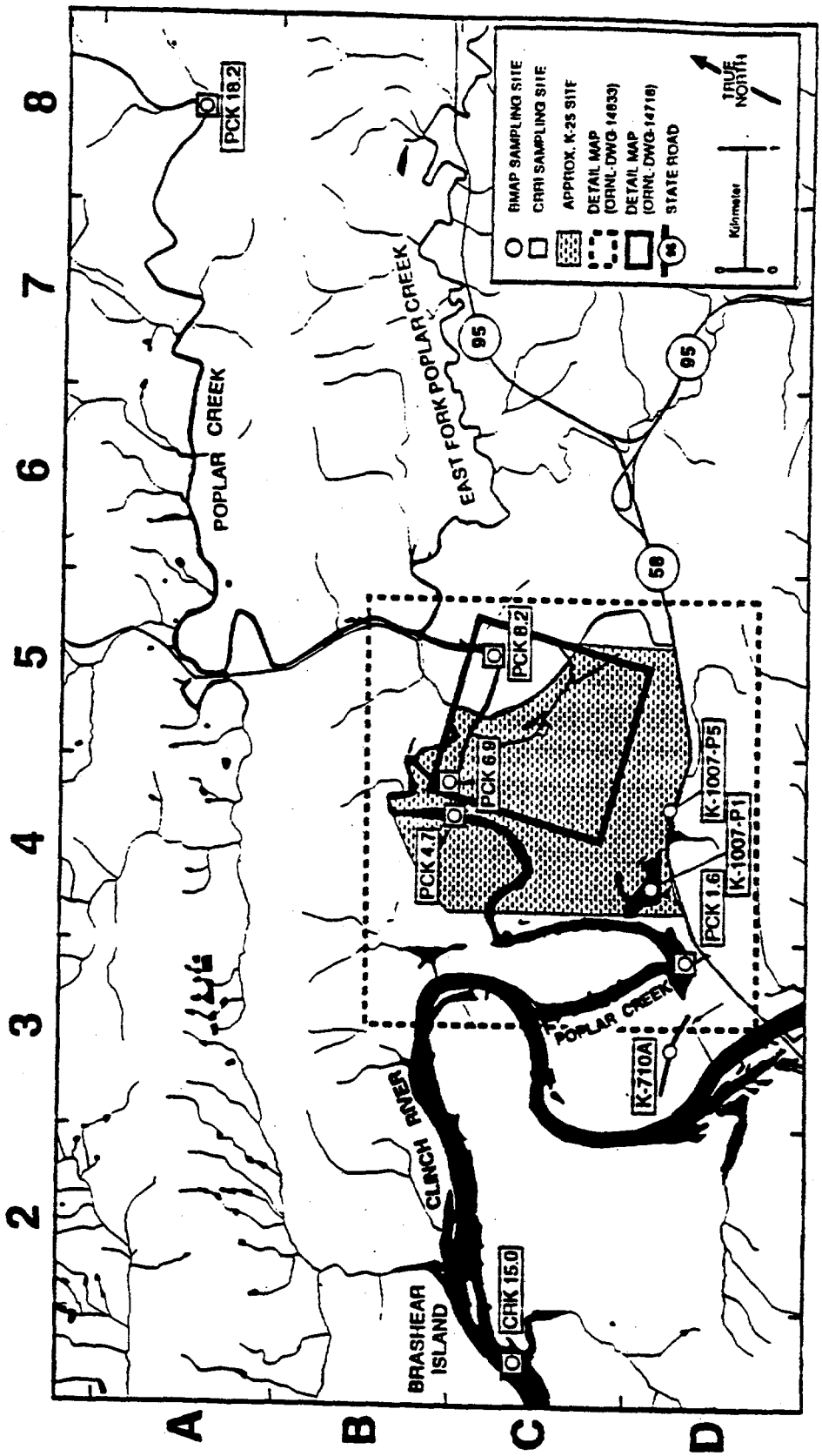

Fig. 2.1. Map of the Oak Ridge K-25 Site area showing the location of sampling sites on Poplar Creek. The drawing is based on a Geographic Information System data base of the 1969 S-15A map of the Oak Ridge, Tennessee, area. The area delineated as "detail map" is shown in Fig. 2.2. BMAP = Biological Monitoring and Abatement Program; CRRI = Clinch River Remedial Investigation; PCK = Poplar Creek kilometer; and CRK = Clinch River kilometer.

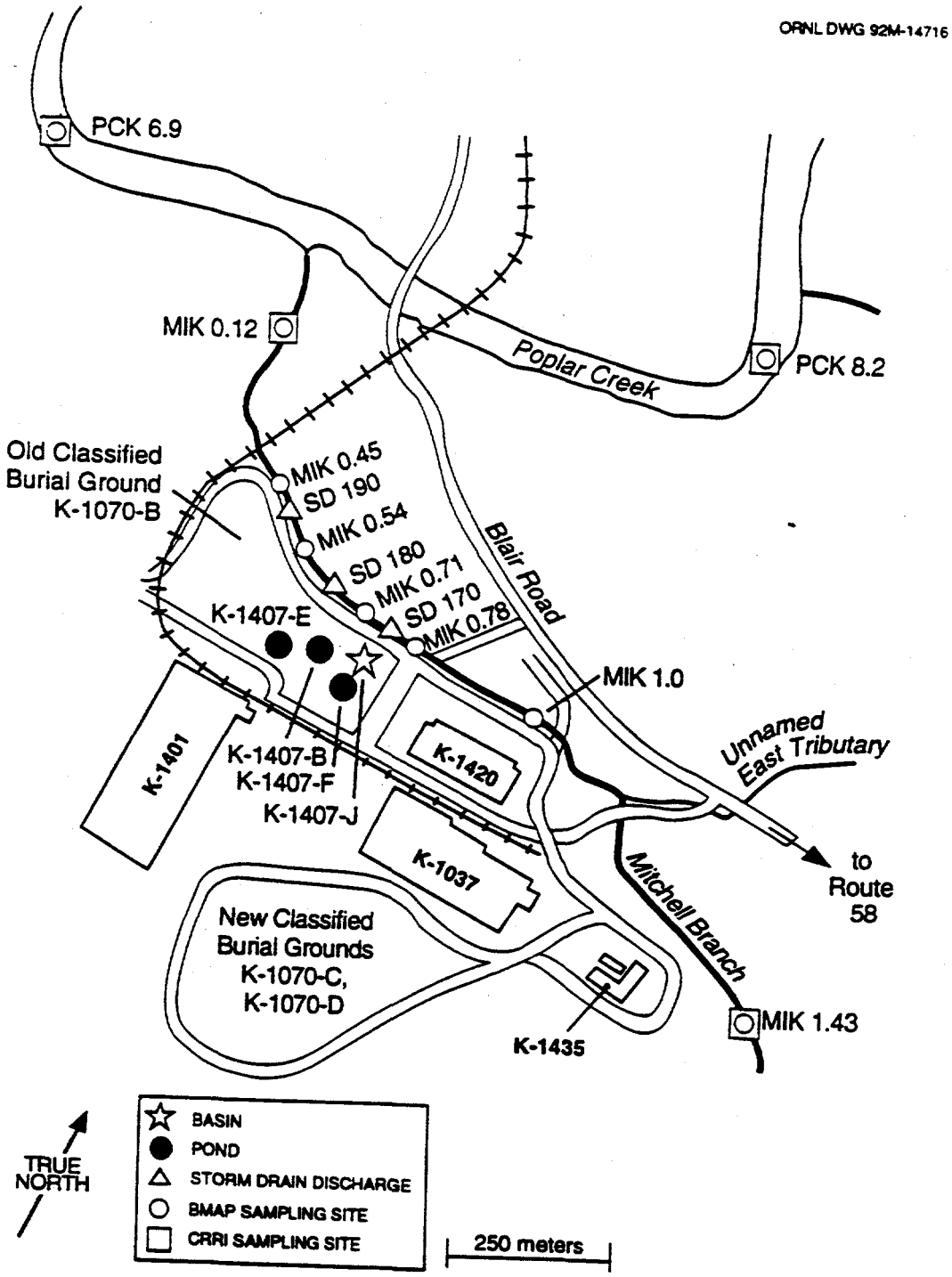

Fig. 2.2. Map of Mitchell Branch and a portion of the Oak Ridge K-25 Site showing the locations of biological monitoring sites (○) in relation to selected storm drains (△) and the K-1407-E/F ponds and the K-1407-J basin, which replaced the K-1407-B pond on November 1, 1988. BMAP = Biological Monitoring and Abatement Program; CRRI = Clinch River Remedial Investigation; MIK = Mitchell Branch kilometer; PCK = Poplar Creek kilometer and SD = storm drain.

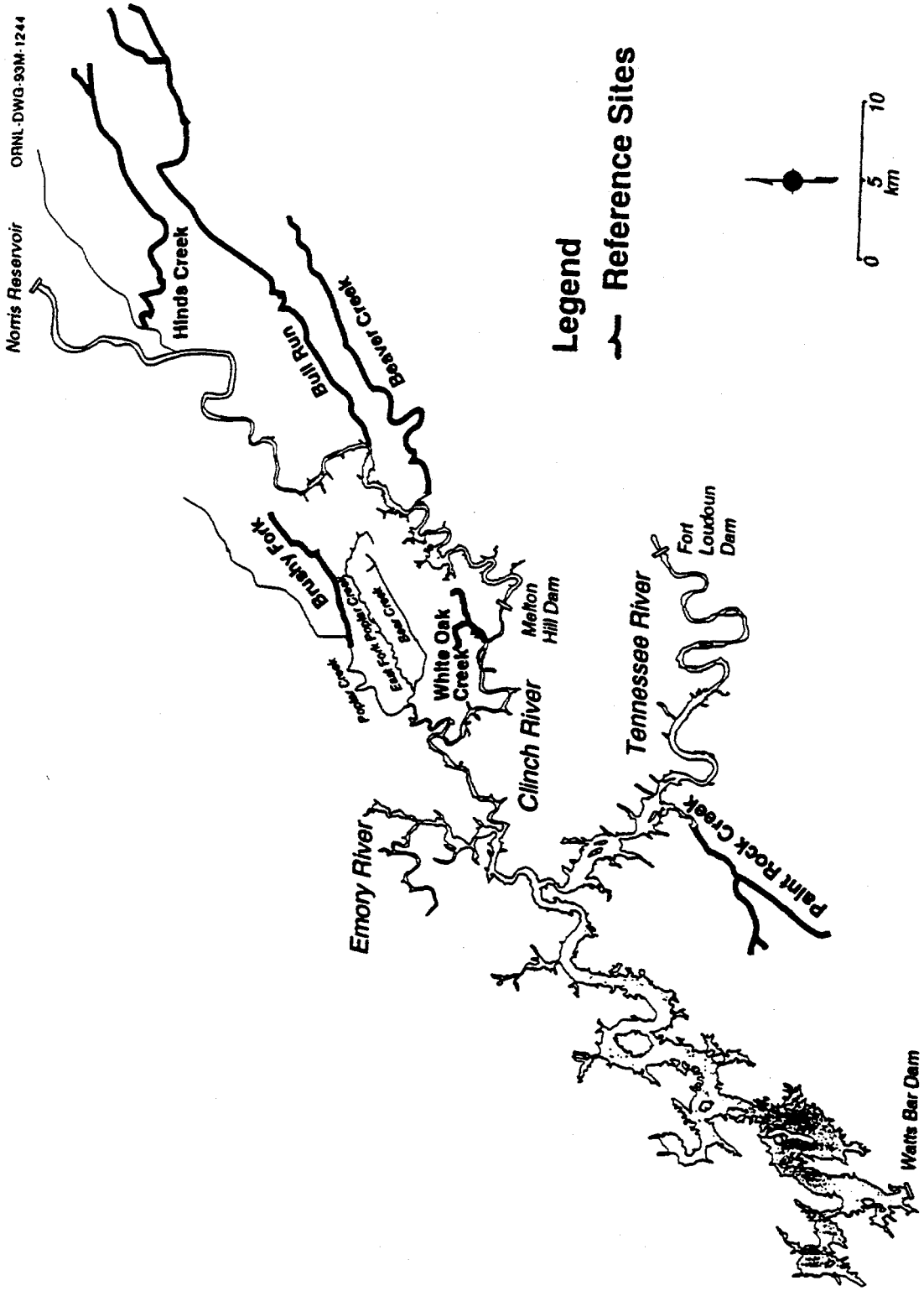

Fig. 2.3. Map of the Oak Ridge area showing locations of the reference (control) sites.

Branch near ORNL (Loar 1990, Table 2.1 in Smith et al. 1994), a small stream that is also underlain predominantly by shale and sandstone (Table 10 in McMaster 1967).

Discharges from the Oak Ridge K-25 Site augment the flow of Mitchell Branch downstream of MIK 0.78. Once-through cooling water and process water account for < 10% of the base streamflow at monitoring station K-1700 on lower Mitchell Branch at MIK 0.12. Surface run-off and groundwater probably account for most of the remaining flow. Based on these estimates, about 10% of the flow in lower Mitchell Branch can be attributed to discharges from the Oak Ridge K-25 Site (J. A. Kovacs, Environmental Management Division, personnel communication, 1994). However, in years of below-normal precipitation and minimal runoff, such as 1985–87, nearly 100% of the flow in the stream could be plant effluent.

Mitchell Branch has a relatively low temporal variability in discharge volume compared with streams without flow augmentation (Table 2.1 in Smith et al. 1994). The variability in discharge in Mitchell Branch was higher than in EFPC at East Fork Poplar Creek kilometer (EFK) 5.3, where flow is augmented by ~50% in dry years. The effect of flow augmentation may be beneficial to some biota by reducing dewatering and thus minimizing the loss of habitat. For example, Horowitz (1978) found that greater numbers of fish species occurred in streams where flow was more constant. However, increased flow stability can reduce environmental heterogeneity (i.e., habitat diversity) which can adversely affect species richness and/or diversity.

Annual precipitation was 151.8 cm in 1990, 152.8 cm in 1991, 111.2 cm in 1992, and 124.5 cm in 1993 (109%, 111%, 81%, and 91% of normal respectively). There were four major storms (> 5 cm in 24 h) in 1990, six in 1991, zero in 1992, and one in 1993. Mean annual flow (MAF) in Mitchell Branch was 35.5 L/s in 1990, 31.5 L/s in 1991, 25.8 L/s in 1992, and 19.1 L/s (values for April through December, 1993, only) in 1993.

The number of days of zero discharge in upper Melton Branch also provides a relative measure of ecologically meaningful differences in the hydrographs of the past 4 years. For example, no flow was recorded for 39 d in 1990, 51 d in 1991, 42 days in 1992, and 137 d in 1993 (Table 2.1). The number of days with zero discharge in 1993 (137 d) approached those seen during the drought period of 1986–1988. These data indicate that any adverse ecological effects resulting from reduced streamflow may have been greater in 1993 because of below-normal precipitation in 1992 and 1993.

Table 2.1. Number of days of zero discharge (number of consecutive days in parentheses) in upper Melton Branch at kilometer 1.93 (USGS gaging station 03537100)

	May	June	July	August	September	October	November	December	Total
1985	0	4(2)	8(6)	0	2(2)	0	0	0	14
1986	0	10(10)	15(15)	31(31)	27(24)	20(12)	0	0	103
1987	0	11(8)	21(19)	21(31)	30(30)	31(31)	30(30)	18(14)	172
1988	2(2)	30(30)	21(12)	30(26)	25(11)	31(31)	3(3)		142
1989	0	0	1(1)	21(17)	7(5)	0	0	0	29
1990	0	5(5)	5(3)	3(3)	22(11)	4(3)	0	0	39
1991	0	0	3(2)	16(8)	26(16)	6(4)	0	0	51
1992	0	4(4)	1(1)	7(3)	27(16)	3(3)	0	0	42
1993	0	13(7)	28(13)	28(18)	29(26)	28(21)	11(7)	0	137

Source: Lowery et al. (1986, 1987, 1988, 1989, 1990) and USGS provisional data for 1990, 1991, 1992, and 1993 (L. D. Voorhees, ORNL, unpublished data). Full citations available in Chapter 8, this document.

2.1.2 Poplar Creek

Poplar Creek is the largest tributary of the Clinch River between Melton Hill Dam and the northwest boundary of ORR. It has an average annual discharge that is approximately ten times greater than the combined discharges of other tributaries in this 21-km reach of the river (Loar et al. 1981). It flows from Poplar Creek Valley, which is underlain by the Conasauga shale, through a gap in Black Oak Ridge, which is underlain by the Knox Formation, and enters the Clinch River southwest of the Oak Ridge K-25 Site. With the exception of the silty shale and siltstone-sandstone members of the Rome Foundation adjacent to Route 58, southeast of the plant, most of the K-25 Site is underlain by Chickamauga Limestone (DOE 1979).

Poplar Creek exhibits seasonal fluctuations in discharge that, in general, reflect precipitation and runoff patterns typical of this region of East Tennessee. Maximum precipitation is in the winter (December–February), when ~31% of the annual precipitation occurs; the wettest months being February and March (NOAA 1990). Maximum runoff is also likely to occur in January, February, or March, when rainfall is normally high and soil moisture and groundwater storage are highest (McMaster 1967). Discharge in Poplar Creek has historically been highest during the winter months and lowest in late summer, when rainfall is normally low and runoff is minimal (Fig. 2.4 in Smith et al. 1994). Water levels in lower Poplar Creek are also influenced by the operation of two Tennessee Valley Authority (TVA) dams: Melton Hill Dam at CRK 37.2 (completed in 1963) upstream of the confluence and Watts Bar Dam located at Tennessee River kilometer (TRK) 852 (completed in 1942)

downstream of the confluence. Dam operations affect the magnitude and frequency of water level fluctuations in the Poplar Creek embayment as they influence the water level at the Clinch River, but they also influence stream velocities and flow direction in the lower reaches of Poplar Creek.

2.2 WATER QUALITY

Water quality of Mitchell Branch is influenced by the geology of the drainage basin (Sect. 2.1), point source discharges of effluents that enter the stream via storm drains, and leachate from waste disposal sites. The following characterizations of water quality are preceded by general descriptions of the sources of effluents discharged by the Oak Ridge K-25 Site. The characterizations of water quality are based on data from monitoring station K-1700 (Sect. 2.2.2.1), effluent data from the K-1407-J basin (Sect. 2.2.2.2), and BMAP-related measurements of water temperatures (Sect. 2.2.3). Water quality characterizations of Mitchell Branch associated with toxicity testing are given in Sect. 3.2.3.1.

2.2.1 Description of the Oak Ridge K-25 Site Discharges

2.2.1.1 Mitchell Branch

Point-source discharges to Mitchell Branch from current Oak Ridge K-25 Site operations are generally from storm drain effluents. Prior to October 1988, wastes from the uranium recovery facility, the metals cleaning facility, the chemical process development facility, the steam plant, and the coal yard were neutralized in a 113,500-L vat prior to discharge into the K-1407-B pond. Metal hydroxides were allowed to settle to the bottom of the pond, and the supernatant was conveyed, via a 1.6-km long ditch, to Mitchell Branch (Fig. 2.1).

Closure of the K-1407-B pond began on October 31, 1988, under the Resource Conservation and Recovery Act. The pond now receives rainwater and surface runoff, which is pumped to the CNF for treatment before being discharged to Poplar Creek. K-1407 B Pond is now in the process of being closed. The pond is being backfilled with an engineered rock fill and overlain with an engineered soil cover.

Many storm drains (SD) enter Mitchell Branch. Although some of these drains contribute only runoff water from roofs and parking lots to the stream during rainfall events, others may also convey groundwater. In the new NPDES permit, storm drains are classified according to their source and potential for contamination. Six storm drains are classified as Category IV

outfalls (those outfalls that pose the risk of conveying pollutants that could be released to the waters of the state). Of these six, three discharge to Mitchell Branch through SDs 170, 180, or 190. The remaining Category IV outfalls discharge to Poplar Creek through the K-1007 ponds. There are 34 Category III outfalls that enter Mitchell Branch or Poplar Creek, 29 Category II and 66 Category I outfalls that enter Mitchell Branch, Poplar Creek, or Watts Bar Reservoir.

In March 1991, following verbal approval from TDEC, discharge from the K-1407-E/F ponds was ceased as a routine operation. Instead, the coal pile and steam plant wastewaters were routed through the CNF for additional treatment and discharged through the K-1407-J basins to Poplar Creek (Kornegay et al. 1993).

Several abatement activities over the sampling period may have affected water quality in Mitchell Branch (Table 2.2). Chlorinated discharges to SD 190 from K-1401 were removed in 1992. In 1992, the K-25 Site began treating runoff from the Steam Plant coal pile at CNF. Prior to that time, acidic runoff was discharged to Mitchell Branch via SD 170. The coal pile was removed in winter 1993 and the area was reseeded. The Powerhouse coal pile was limed in summer 1993 to reduce acidic runoff through SD 992. This coal pile is scheduled to be reclaimed (capped) by September 1994. Chlorinated discharges to storm drains were eliminated by October 1993. The process primarily involved discharges to SDs 100, 124, 170, 180, and 190. Discharges were dechlorinated slowly, one by one, during 1993. A best management plan (BMP) was developed for the fire training facility in early 1993 that reduced the amount of petroleum products and water being discharged through SD 190. In May 1993, a procedure was initiated to drain a dike containing 600,000 gallons of diesel fuel at the Steam Plant, resulting in the elimination of discharges of small amounts of fiesel fuel to SD 170. The K-25 Site began collecting and treating effluent from the SW-31 Seep in January 1994, reducing discharges of organic compounds (especially trichloroethylene) through SDs 170 and 180. A BMP was developed for the K-1414 garage in March 1994 that eliminated almost all oil spills and discharges from that facility to SD 180. Erosion control measures were initiated in May 1994 at the soil disposal area north of Mitchell Branch across from SD 190.

2.2.1.2 Poplar Creek

The CNF (Building K-1407-H) went on-line in October 1987. The CNF can treat waste streams that require only neutralization and chemical precipitation. Filtration is also used at

Table 2.2. Partial list of events that may have affected water quality in Mitchell Branch, 1987 through 1994

Event	Result
1987	
CNF constructed	
K-1407-B pond closed	
1988	
Coal pile & steam plant effluents discharged through K-1407-E/F (11/88)	
End of drought	Increased flow
CNF pipeline operational	Discharges to Mitchell Branch
TSCA incinerator performance test burns	
1989	
K-1407-J effluent re-routed to Poplar Creek (9/89)	CNF discharges to Mitchell Branch cease
1990	
TSCA incinerator start-up tests (not fully operational)	
1991	
TSCA incinerator fully operational	
K-1407-E/F discharges to CNF	Discharges of K-1407-E/F to Mitchell Branch cease
Dechlorinators added to SDs 170, 180 and 190 (6/91)	Reduction of chlorinated discharges to Mitchell Branch
Repackaging at drum yard begins	
1992	
Engineering project to eliminate chlorinated discharges to Mitchell Branch complete, dechlorinators at SDs removed	
Chlorinated sources removed SDs 100, 124, 170, 180, 190	
Treatment of Steam Plant coal pile discharges	Reduce acidic discharges via SD 170
Coal pile removed (11-12/92)	Removed acid discharges to SD 170
1993	
Steam Plant dike drained (5/93)	Remove diesel source to SD 170
Powerhouse coal pile limed (summer 1993)	Reduce acidic runoff through SD 992
All chlorinated discharges to Mitchell Branch ceased (10/93)	
Steam Plant coal pile removed (winter 1993)	
BMP developed for fire training facility	Remove release of firewater & petroleum to SD 190
1994	
SW-31 seep removed from SDs 170, 180 (1/94)	Reduction of organic compounds through SDs 170 and 180
BMP developed for K-1414 garage (3/94)	Eliminated oil discharge to SD 180
Rerouted effluent from Bldg. 1001 - SD 130 (6/94)	
Erosion control measures initiated at the soil disposal area north of Mitchell Branch across from SD 190	

Note: CNF = Central Neutralization Facility. TSCA = Toxic Substances Control Act. SD = storm drain. BMP = Best Management Plan.

the CNF for additional solids removal. At the CNF, both hazardous and nonhazardous waste streams are treated. The primary hazardous waste stream is wastewater from the TSCA incinerator. The principal nonhazardous waste stream is wastewater generated at the K-1501 steam plant. The waste streams from the TSCA incinerator and the steam plant contribute > 95% of the influent to the CNF. In addition to these primary waste streams, the CNF treats miscellaneous waste streams which meet the waste acceptance criteria for the CNF. The effluent from the CNF is currently discharged into Poplar Creek at kilometer 5.1.

2.2.2 NPDES Monitoring

2.2.2.1 Mitchell Branch

The following characterization of water quality in Mitchell Branch is based on routine monitoring of parameters at station K-1700. This site is located on lower Mitchell Branch (MIK 0.12) downstream of all point-source and most area-source discharges (Figs. 2.1 and 2.2).

From 1990 through 1993, water quality at MIK 0.12 was characterized by (1) moderate levels of dissolved solids and occasionally high levels of suspended solids, (2) relatively low levels of nutrients, (3) elevated levels of some metals (e.g., aluminum and iron) and some organics, and (4) high temperatures (discussed in Sect. 2.2.3). Mean concentrations for substances in water at K-1700 above detection limits are summarized in Table A.1. The effects of elevated concentrations of these substances on the aquatic community cannot be adequately addressed because of the uncertainty of magnitude and duration of the substances in the stream. Water samples were often one-time grab samples and therefore may not have been collected during the peak of a potential release.

Mean dissolved solids concentrations at K-1700 have decreased in the 1990–1993 sampling period. However, maximum recorded values exceeded the maximum drinking water contaminant level in 1990, 1991, and 1992 (Table A.1).

Concentrations of nitrogen in Mitchell Branch are low, whereas concentrations of phosphorus are high. Even though concentrations of nitrate nitrogen were higher than background (~0.1 mg/L; Boyle et al. 1982, Table 3.16), they averaged less than 1 mg/L and never exceeded 6 mg/L at MIK 0.12 (Appendix A, Tables A.1–A.4). Phosphorus is not monitored in Mitchell Branch, but water from the K-1407-J basin had a mean phosphorus

concentration of 1.5 mg/L in 1990 and <3.1 mg/L in 1991, with a maximum recorded value of 3.7 mg/L in 1989 (Appendix A, Table A.3).

Many of the metals (Appendix A, Tables A.1–A.5) in Mitchell Branch exceeded concentrations that are typical of small, relatively undisturbed streams on the ORR (Boyle et al. 1982, Tables 3.16 and 4.16). Only manganese had a mean concentration above the reference value at K-1700 (Table A.1). Aluminum, iron, and manganese had maximum values that exceeded the reference limits at K-1700 (Tables A.1 and A.5). Of the organics measured in Mitchell Branch, all were above detection limits at least some of the time (Table A.1). Except for the decrease in dissolved solids that has occurred in since 1990, the water quality of Mitchell Branch has remained relatively constant over the last 4 years (1990–93).

2.2.2.2 Discharges to Poplar Creek

Effluent from the K-1407-J basin was characterized by high concentrations of dissolved solids with mean values exceeding reference values from 1990–1992. (Table A.3).

Concentrations of nitrate, phosphorous, and sulfate were all elevated in K-1407-J. Although the mean value for nitrate in 1991 was lower than the reference, the maximum value exceeded the reference value (Table A.3). The mean concentration of phosphorous in 1990 was 1.5, and the maximum concentration was 15.0 mg/L. The mean concentration of sulfate was higher than 250 mg/L in 1991 and 1992; maximum values in these 2 years were 1500 and 1000 mg/L respectively.

Concentrations of all metals that were monitored from 1990–93 were elevated at some time during this period, and most had mean values above the detection limits (Table A.3). However, only manganese had mean concentrations above the reference values (1991, Table A.3).

Although most of the organics that were monitored had mean values at or below detection limits, many were detectable at some time during the sampling period (Table A.3).

2.2.3 Temperature *(W. K. Roy)*

Ryan Tempmentor digital temperature recorders were deployed on April 9, 1987, in Mitchell Branch at MIK 0.50 immediately below the outfall of SD 190 and in Grassy Creek, a nearby drainage with similar geology (Sect. 2.1; McMaster 1967, Table 10). Temperature values were obtained every 20 min. until July 1, 1987, and every 60 min. thereafter. The

record is incomplete due to periodic equipment problems or recorders being out of water (either inadvertently or for data retrieval).

Grassy Creek kilometer (GCK) 2.4 site was dropped as a reference site in April 1991, in part due to its intermittent nature. It was also becoming increasingly difficult to access this site because it was situated within the boundaries of the newly created Surface Danger Zone for the Central Training Facility firing range on Bear Creek Road. Furthermore, the associated construction activity upstream of GCK 2.4 was causing increasing siltation at this site (M. G. Ryon, pers. comm., May 26, 1994). The site was replaced with one on Scarboro Creek, a reference stream located approximately 16 km northeast of Grassy Creek, in Anderson County. Temperature monitoring was initiated at Scarboro Creek kilometer (SCK) 2.2 on October 28, 1991.

Through the period of record, Mitchell Branch temperatures were high relative to unimpacted ORR streams. Mean weekly temperatures in Mitchell Branch sometimes exceeded 25°C but were generally below 20°C in Grassy Creek and always below 20°C in Scarboro Creek (Fig 2.4). Temperatures in Mitchell Branch averaged ~4 to 8°C higher than in Grassy Creek, and up to 7°C higher than in Scarboro Creek. Winter water temperatures in Scarboro Creek more closely approximate those found in Mitchell Branch than do those in Grassy Creek. In fact, mean monthly temperatures in Scarboro Creek were slightly higher than in Mitchell Branch during January, February, and March of 1994. Summer water temperatures, however, are often much higher in Mitchell Branch than in the reference streams. The *maximum* temperature at MIK 0.50 in June 1987 was 19°C higher than that at GCK 2.4, and in August 1992 it surpassed the SCK 2.2 maximum temperature by more than 25°C (Table A.6).

The temperature of Mitchell Branch is influenced strongly by warm cooling-water discharges (via storm drains) and the discharge of effluent from open retention ponds (K-1407-B prior to October 1988 and K-1407-E/F after this period) prior to discharge just 130 m above the monitoring station at MIK 0.50. Furthermore, except for the headwaters, there is little canopy cover above MIK 0.50 to help moderate temperatures.

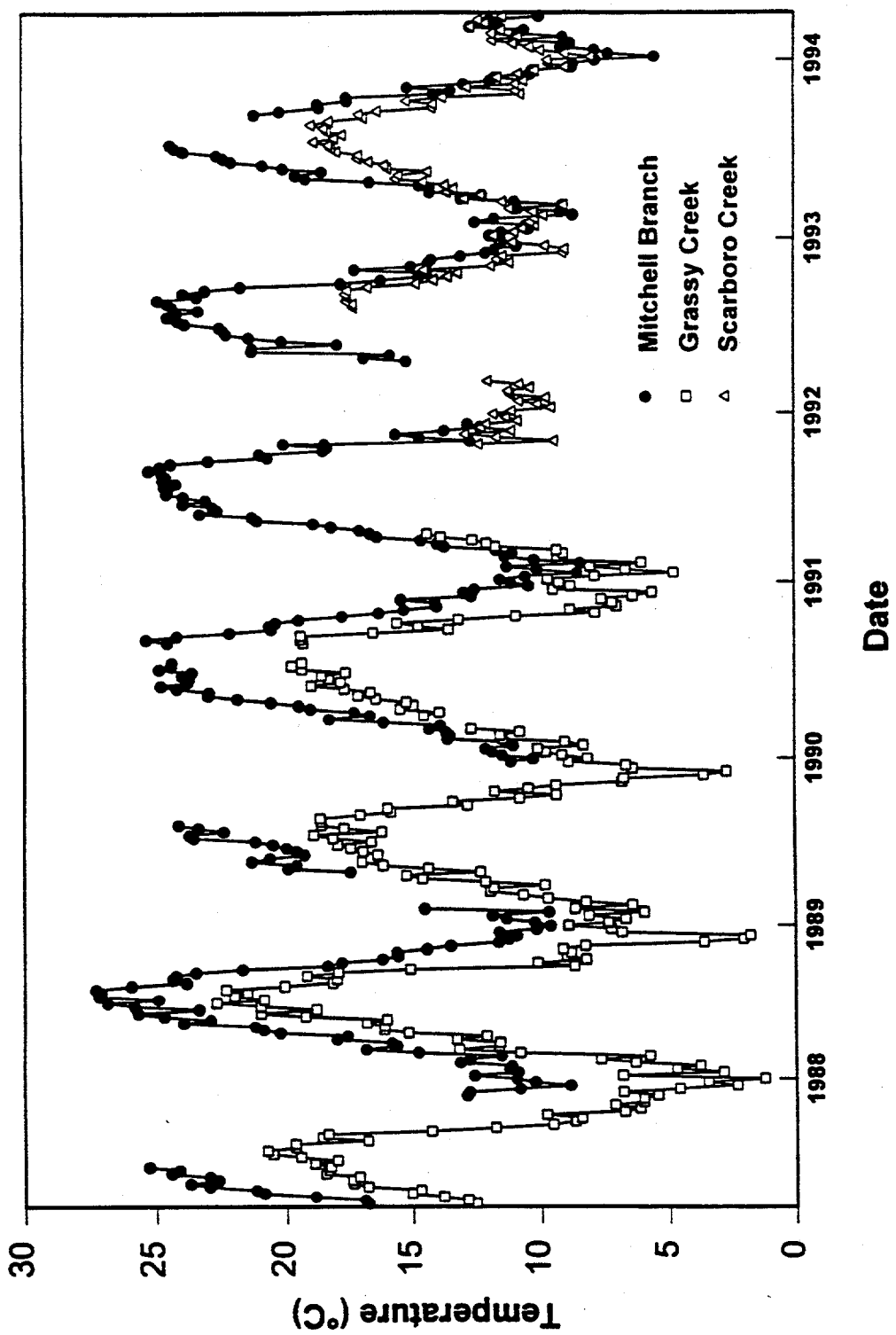

Fig. 2.4. Weekly mean temperatures for Mitchell Branch kilometer 0.50, 1987–1994, Grassy Creek kilometer 2.4, 1987–1991, and Scarboro Creek kilometer 2.2, 1991–1994. Temperature record is incomplete as a result of equipment failure.

2.3 BIOLOGICAL MONITORING SITES *(R. L. Hinzman)*

During the initial BMAP study, eight sites on Mitchell Branch (Figs. 2.1 and 2.2 and Table 2.3) were routinely sampled to assess ecological conditions in the stream. Although the upper site (MIK 1.43) was the primary reference site, sites on other area streams were also used as reference stations (Fig. 2.3). The lowermost site at MIK 0.12 coincided with the location of the (then NPDES) K-1700 monitoring station. A sampling site for the bioaccumulation task was located at PCK 6.9, downstream of the mouth of Mitchell Branch. Four of the remaining six sites (MIKs 0.78, 0.71, 0.54, and 0.45) were selected based on the location of the three most significant discharges to Mitchell Branch (SDs 170, 180, and 190); these monitoring sites were located above and below each of the outfalls respectively. The remaining two sites (MIK 0.86 and MIK 1.10) were selected to assess the potential for adverse impacts associated with (1) construction of a storage yard located immediately northeast of Mitchell Branch and (2) minor inputs from several storm drains located further upstream.

Sampling locations for the current BMAP are shown in Figs. 2.1 and 2.2 and are discussed in Table 2.3 and the description for each task. Many of the tasks share Poplar Creek and Clinch River sites with the Clinch River Remedial Investigation (CRRI). Task 1 (Toxicity Monitoring) maintained six sites in Mitchell Branch and added new sites in Poplar Creek and the Clinch River. Task 2a (Contaminant Accumulation in Aquatic Biota) maintained five sites (one each in Mitchell Branch and Clinch River and three in Poplar Creek) and added three new ones: Poplar Creek (one site) and two on unnamed tributaries. Task 2b (Contaminant Accumulation in Terrestrial Biota) will consist of site-wide monitoring, including sites both within and adjacent to the K-25 Site. Task 3a (Assessment of Fish health) maintained a site in Mitchell Branch and added sites in Poplar Creek, Clinch River, and an unnamed tributary. Task 3b (Reproductive Effects) is a new subtask and includes sites in Mitchell Branch, Poplar Creek, and the Clinch River. Task 4a (Instream Monitoring of Benthic Invertebrates) maintained four sites in Mitchell Branch and unnamed tributaries. Task 4b (Instream Monitoring of Fish) maintained two sites in Mitchell Branch.

Table 2.3. Location of the biological monitoring sites on Mitchell Branch, Poplar Creek, and unnamed tributaries

Site	Toxicity Monitoring	Bioaccumulation monitoring			Assessment of fish health		Instream monitoring	
		Aquatic biota	Food-chain effects[a]	Waterfowl[b]	Bioindicators	Reproductive effects	Benthic invertebrates	Fish
Mitchell Branch								
MIK 0.12	X	X			X	X[b]		
MIK 0.45	X						X	X
MIK 0.54	X							
MIK 0.71	X						X	X
MIK 0.78	X						X	
MIK 1.0	X[e]							
MIK 1.43 (ref.)	X					X	X	
Others (ref.)	X[d]	X			X[c,d]	X[d]		X
Poplar Creek								
PCK 1.6	X[d]	X[c]			X[d]	X[d]		
PCK 4.7	X[d]					X[d]		
PCK 6.9	X[d]	X[c]			X[d]	X[d]		
PCK 8.2	X[d]	X[c]			X[d]	X[d]		
PCK 18.2 (ref.)	X[d]	X			X	X		
Clinch River								
CRK 15.0	X[d]	X[c]			X[d]	X[d]		
Other sites								
K-1007 (P1 & P5)		X			X			
Tributary to K-1007-P5							X	
Unnamed East Tributary							X	
K-710A		X						

Note: Reference sites include designation "ref." following kilometer reading. MIK = Mitchell Branch kilometer, PCK = Poplar Creek kilometer, and CRK = Clinch River kilometer.

[a]Task consists of site-wide monitoring (see Sect. 2.2.3).

[b]Other sites on Mitchell Branch to be determined after initial tests are completed.

[c]Site not included in original Biological Monitoring and Abatement Program plan, but added later.

[d]Sites included in Sampling and Analysis Plan (Phase 2) of Clinch River Remedial Investigation (R. B. Cook, et al., unpublished data).

[e]This sampling site was moved to MIK 0.78 in April 1993 to coincide with benthic macroinvertebrate sampling.

3. TOXICITY MONITORING (TASK 1)

L. A. Kszos

The toxicity monitoring task for the K-25 Site consists of three subtasks. The first subtask measures the toxicity of effluents as required by the NPDES permit. The second subtask monitors ambient water toxicity of six sites in Mitchell Branch, five sites in Poplar Creek, and one site in the Clinch River. The third subtask measures the toxicity of the three major storm drains that enter Mitchell Branch. The effluent and storm drain toxicity data are presented in Sect. 3.1; the ambient toxicity data are presented in Sect. 3.2.

3.1 EFFLUENT AND STORM DRAIN TOXICITY

3.1.1 Introduction

The EPA supports the use of aquatic test organisms to determine the chronic toxicity of a test water (Weber et al. 1989). Toxicity monitoring at the K-25 Site uses two tests concurrently to characterize the toxicity of effluents from two outfalls that discharge to Poplar Creek. They are 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, Weber et al. 1989). These two tests are EPA-approved for use in the NPDES program to estimate (1) the chronic toxicity of effluents collected at the end of the discharge pipe and tested with a standard dilution water, (2) the toxicity of receiving water downstream from or within the influence of the outfall and (3) the effects of multiple discharges on the quality of the receiving water (Weber et al. 1989). These tests are also part of the BMAPs at ORNL, the Oak Ridge Y-12 Plant, and the Biological Monitoring Program at the Paducah Gaseous Diffusion Plant.

The Toxicology Laboratory of ESD at ORNL began evaluating the toxicity of effluent from CNF (K-1407-J, Outfall 011) in February 1990. Tests of the K-1203 Treatment Facility (Outfall 005; primarily a sewage treatment facility) began in June 1991, but bimonthly testing was not implemented until after the renewed NPDES permit became final (October 1992). Under the renewed permit, toxicity tests of CNF and K-1203 were conducted bimonthly beginning in November 1992. According to the NPDES permit, the testing frequency can be

reduced to twice per year if no toxicity is observed for six consecutive tests. Because this was the case at CNF, testing frequency was reduced to twice per year in November 1993.

The three major storm drains that discharge to Mitchell Branch were monitored under the BMAP because (1) they discharge into a reach of Mitchell Branch that is known to be stressed (Smith et al. 1993, 1994); (2) toxicity tests conducted during 1987–89 indicated that these effluents were toxic, and (3) when the pipeline to Poplar Creek was constructed for CNF, the storm drains became the major source of anthropogenic input to Mitchell Branch. During 1990–92, effluent from SDs 170, 180, and 190 was evaluated for toxicity to fathead minnows two or three times each year. As part of the revised BMAP (Kszos et al., 1993) *Ceriodaphnia* tests of the storm drains have been conducted every other month since April 1993.

3.1.2 Materials and Methods

This report includes tests conducted from January 1990 to April 1994. Effluent from K-1407-J (Outfall 011) was evaluated five to six times per year and effluent from K-1203 (Outfall 005) was evaluated twice in 1991, once in 1992, eight times in 1993, and three times in 1994. Effluent from SDs 170, 180, and 190 was evaluated eight times in 1990–92 and six times in 1993–94. Prior to November 1992, tests of CNF were conducted using seven consecutive daily grab samples collected at the NPDES discharge point. Subsequent tests used seven 24-h composite samples as required by the renewed NPDES permit. All tests of K-1203 were conducted using composite samples. All tests of storm drains were conducted using daily grab samples. Samples from CNF and K-1203 were collected by the K-25 Site personnel and delivered according to established chain-of-custody procedures (Kszos et al. 1989). Samples from storm drains were collected by ESD personnel.

Tests with *Ceriodaphnia* and fathead minnows were typically conducted concurrently following procedures outlined in Weber et al. (1989) and Kszos et al. (1989). These tests are static, renewal tests, meaning that test water is replaced daily for six or seven consecutive days. Animals are fed daily. 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 laboratory control consisting of dilute mineral water augmented with trace metals was included with each test. On each day of a test, subsamples of each effluent were routinely analyzed for pH, conductivity, alkalinity, water hardness, and total residual and free chlorine (Kszos et al. 1989). In addition, pH and dissolved oxygen were measured in 24-h old samples.

For each toxicity test, the no-observed-effect level (NOEL) and the 96-h LC_{50} were determined using methods described in Weber et al. (1989). The NOEL is the highest concentration of effluent tested that does not reduce fathead minnow survival or growth or *Ceriodaphnia* survival or reproduction. The 96-h LC_{50} is the concentration that kills 50% of the test organisms in 96 hours. In the renewed permit, a NOEL for CNF of ≤ 2.11% effluent or a 96-h LC_{50} of ≤ 7.05% effluent constitutes a violation of levels required by the permit. At K-1203, a NOEL of ≤ 4.2% effluent or a 96-h LC_{50} of ≤ 14.6% effluent constitutes a violation of permit levels. The compliance points were determined by the TDEC and were based upon the dilution of the effluents once they enter Poplar Creek. The results of each test are submitted to the K-25 Site and subsequently submitted to the TDEC. These reports contain daily survival and reproduction information, statistical results, water chemistry for at least three concentrations of effluent, quality assurance information, etc. In the following section, only the NOEL, 96-h LC_{50}, and water chemistry for full-strength effluent are reported. Toxicity tests of the storm drains are not required in the NPDES permit and are conducted only as part of the BMAP.

3.1.3 Results

3.1.3.1 CNF (K-1407-J, Outfall 011)

A summary of the NOELs for all toxicity tests conducted from January 1990 to November 1993 are provided in Table 3.1. Results of tests conducted during 1988–1989 are in Kszos (1994). In February 1990 and April 1991, effluent from CNF would have exceeded the NOEL limit in the renewed permit (2.11%) if it had been in effect. However, under the previous permit, which was in effect at that time, the exceedences were not violations of any toxicity limit. Since the renewed permit became effective, effluent from the CNF has not exceeded the permit limits. Thus, in November 1993, the test frequency was reduced to twice per year.

Table 3.1. Toxicity test results for the Central Neutralization Facility (CNF; K-1407-J) and K-1203

K-25 Site Outfall	Test Date	Species	NOEL[a] (%)	LC_{50}[b] (%)
CNF (Outfall 011)	February 1990	Fathead minnows	100	>100
		Ceriodaphnia	1	17.3
	April 1990	Fathead minnows	50	>100
		Ceriodaphnia	100	>100
	June 1990	Fathead minnows	100	>100
		Ceriodpahnia	100	>100
	August 1990	Fathead minnows	100	>100
		Ceriodaphnia	100	>100
	October 1990	Fathead minnows	100	>100
		Ceriodaphnia	100	>100
	December 1990	Fathead minnows	100	>100
		Ceriodaphnia	100	>100
	February 1991	Fathead minnows	100	>100
		Ceriodaphnia	50	>100
	April 1991	Fathead minnows	100	>100
		Ceriodaphnia	<1	35.4
	May 1991[c]	*Ceriodaphnia*	50	>100
	June 1991	Fathead minnows	100	>100
		Ceriodaphnia	50	>100
	August 1991	Fathead minnows	100	>100
		Ceriodaphnia	100	>100
	October 1991	Fathead minnows	100	>100
		Ceriodaphnia	50	>100
	December 1991	Fathead minnows	100	>100
		Ceriodaphnia	100	>100
	February 1992	Fathead minnows	100	>100
		Ceriodaphnia	50	>100
	April 1992	Fathead minnows	100	>100
		Ceriodaphnia	25	73.5
	June 1992	Fathead minnows	100	>100
		Ceriodaphnia	100	>100
	August 1992	Fathead minnows	100	>100
		Ceriodaphnia	100	>100
	November 1992	Fathead minnows	75	>75
		Ceriodaphnia	75	>75
	January 1993	Fathead minnows	75	>75
		Ceriodaphnia	25	>75
	March 1993[d]	Fathead minnows	25	>75
	April 1993	*Ceriodaphnia*	75	>75

Table 3.1 (continued)

K-25 Site Outfall	Test Date	Species	NOEL[a] (%)	LC_{50}[b] (%)
	May 1993	Fathead minnows	75	>75
		Ceriodaphnia	75	>75
	July 1993	Fathead minnows	7.05	>7.05
		Ceriodaphnia	25	>25
	September 1993	Fathead minnows	75	>75
		Ceriodaphnia	75	>75
	November 1993	Fathead minnows	75	>75
		Ceriodaphnia	25	>75
K-1203 (Outfall 005)	June 1991	Fathead minnows	100	>100
		Ceriodaphnia	100	>100
	August 1991	Fathead minnows	100	>100
		Ceriodaphnia	100	>100
	November 1992	Fathead minnows	100	>100
		Ceriodaphnia	100	>100
	January 1993	Fathead minnows	100	>100
		Ceriodaphnia	100	>100
	March 1993[d]	Fathead minnows	100	>100
	April 1993	*Ceriodaphnia*	100	>100
	May 1993	Fathead minnows	100	>100
		Ceriodaphnia	100	>100
	July 1993	Fathead minnows	14.6	>14.6
		Ceriodaphnia	<4.2	>30
	August 1993[d]	*Ceriodaphnia*	30	>100
	September 1993	Fathead minnows	100	>100
		Ceriodaphnia	30	>100
	November 1993	Fathead minnows	100	>100
		Ceriodaphnia	14.6	>100
	January 1994	Fathead minnows	100	>100
		Ceriodaphnia	100	>100
	March 1994	Fathead minnows	<4.2	>54.8
		Ceriodaphnia	100	>100
	March 1994[c]	Fathead minnows	30	>100

[a]No-observed-effect level (the highest concentration of effluent tested that does not significantly reduce fathead minnow survival or growth or *Ceriodaphnia* survival or reproduction).

[b]96-h lethal concentration for 50% of the test organisms.

[c]Confirmatory test.

[d]Invalid *Ceriodaphnia* test (unacceptable control survival); a retest was conducted in April 1993.

A summary of water quality parameters for CNF is provided in Table 3.2. The chemical characteristics of the effluent have remained similar from 1990 to 1993 with the exception of one high mean conductivity value in April 1991 (8663 µS/cm) and lower mean alkalinity during 1993. Mean conductivity ranged from 800 to 8663 µS/cm with typical values around 1500 to 3000 µS/cm. Mean alkalinity typically ranged from about 40 to 200 mg/L with one mean value equal to 462 mg/L. Mean hardness ranged from 221 to 717 mg/L.

3.1.3.2 K-1203 Treatment Facility

A summary of the NOELs for all toxicity tests conducted during January 1990 to March 1994 are provided in Table 3.1. In July 1993 and March 1994, effluent from K-1203 exceeded the NOEL limit in the renewed permit (4.2%). Both of these exceedences were violations of the NPDES permit. Confirmatory tests were conducted within 2 weeks of the violation and no toxicity was observed (NOEL = 30%).

A summary of water quality parameters for K-1203 is provided in Table 3.2. The chemical characteristics of the effluent have remained similar for 1991 to 1994. Mean conductivity ranged from 326 to 435 µS/cm, and mean alkalinity ranged from 81 to 109 mg/L. Mean pH ranged from 7.61 to 8.09.

3.1.3.3 Storm drains 170, 180, and 190

A summary of the NOELs and survival at 96 hours for toxicity tests conducted during January 1990 to April 1994, are provided in Table 3.3. Results of tests conducted during 1988–1989 are in Kszos (1994). During 1990–92, the NOEL for effluent from SDs 170, 180, and 190 was frequently, and for SD 170 and SD 190, *Ceriodaphnia* survival at 96 h was 0% for 3 to 5 tests. Because a complete dilution series was not used, the level of toxicity below full-strength could not be determined. Beginning in April 1993, test results were much more definitive. For SD 170, the NOEL for *Ceriodaphnia* was ≤ 50% in six of the seven tests conducted in 1993–94, and *Ceriodaphnia* survival at 96 h was ≥ 70%. Full-strength effluent was non-toxic in only one test. After 1992, effluent from SD 180 was typically non-toxic to *Ceriodaphnia*: the NOEL was 100% in five of the seven tests conducted in 1993–94. Test results for SD 190 were similar to those for SD 170: full strength effluent was non-toxic in only one test in 1993–94. The NOEL was ≤ 50% effluent in six of the seven tests conducted

Table 3.2. Average water quality parameters measured during toxicity tests of the Central Neutralization Facility (CNF; K-1407-J) and K-1203

Values are averages of full-strength wastewater for each test (n = 7)

Facility (Outfall)	Test Date	pH (standard units)	Conductivity (µS/cm)	Alkalinity (mg/L $CaCO_3$)	Hardness (mg/L $CaCO_3$)
CNF (011)	February 1990	7.82	1286	94	418
	April 1990	8.13	676	177	263
	June 1990	8.65	1927	462	197
	August 1990	8.54	952	151	244
	October 1990	8.31	1441	113	280
	December 1990	8.06	800	54	221
	February 1991	7.75	2979	61	596
	April 1991	7.84	8663	122	359
	May 1991	8.06	1886	120	431
	June 1991	7.87	1601	163	443
	August 1991	8.12	2187	224	456
	October 1991	7.90	3114	143	555
	December 1991	7.72	2249	126	611
	February 1992	8.10	2231	71	717
	April 1992	8.11	3206	113	677
	June 1992	7.97	1875	72	520
	August 1992	8.01	1454	85	424
	November 1992	7.81	1497	46	698
	January 1993	7.80	2831	52	594
	March 1993	7.69	1831	74	363
	April 1993	7.73	1484	41	568
	May 1993	7.76	1899	41	511
	July 1993[a]	7.92	3007	80	438
	July 1993[b]	7.57	4044	41	455
	September 1993	7.65	1808	54	639
	November 1993	7.63	1615	64	680
K-1203 (005)	June 1991	7.64	326	87	131
	August 1991	7.61	350	85	142
	November 1992	8.03	384	107	161
	January 1993	7.97	390	101	149
	March 1993	8.09	380	101	172

Table 3.2 (continued)

Facility (Outfall)	Test Date	pH (standard units)	Conductivity (µS/cm)	Alkalinity (mg/L $CaCO_3$)	Hardness (mg/L $CaCO_3$)
	April 1993	8.02	390	96	158
	May 1993	7.93	402	85	147
	July 1993[a]	7.71	400	86	155
	July 1993[b]	7.88	414	90	154
	August 1993	7.83	389	81	143
	September 1993	7.90	391	92	147
	November 1993	7.89	435	107	151
	January 1994	8.05	383	109	154
	March 1994[c]	8.01	373	108	151
	March 1994[d]	7.95	331	99	132

[a]Data are for test conducted July 8-15, 1993.
[b]Data are for test conducted July 29-August 5, 1993.
[c]Data are for test conducted March 3-10, 1994.
[d]Data are for test conducted March 24-31, 1994.

Table 3.3. Toxicity test results for storm drains 170, 180, and 190, 1990–1994

Storm Drain	Test Period	Fathead Minnow NOEL[a]	Fathead Minnow Survival[b] at 96 h (%)	*Ceriodaphnia dubia* NOEL[a]	*Ceriodaphnia dubia* Survival[b] at 96 h (%)
170	Jan. 1990	<100	0	<100	0
	dechlor.	100	100	<100	100
	May 1990	<100	100	100	100
	dechlor.	100	97.5	100	100
	Nov. 1990	<100	0	<100	0
	dechlor.	100	95	<50	0
	May 1991	100	85	<100	0
	dechlor.	100	90	<100	0
	Nov. 1991	NT	NT	<75	100 (0)[c]
	Mar. 1992	I	I	75	100
	May 1992	100	97.5	<75	80 (40)[d]
	July 1992	100	95	50	90
	Sept. 1992	100	NT	<50	20
	Nov. 1992	NT	NT	<50	90
	Apr. 1993	NT	NT	<12	100
	June 1993	NT	NT	100	100
	Aug. 1993	NT	NT	25	100
	Oct. 1993	NT	NT	25	100
	Dec. 1993	NT	NT	25	70
	Feb. 1994	NT	NT	<50	100
	Apr. 1994	NT	NT	50	100
180	Jan. 1990	100	95	<100	80 (30)[e]
	May 1990	100	100	100	100
	Nov. 1990	<100	0	<100	100
	May 1991	<100	62.5	100	100
	dechlor.	<100	57.5	100	100
	Nov. 1991	NT	NT	100	100
	Mar. 1992	I	I	100	100
	May 1992	<100	72.5	<75	100
	July 1992	100	97.5	50	80
	Apr. 1993	NT	NT	50	60
	June 1993	NT	NT	100	100

Table 3.3 (continued)

Storm Drain	Test Period	Fathead Minnow NOEL[a]	Fathead Minnow Survival[b] at 96 h (%)	*Ceriodaphnia dubia* NOEL[a]	*Ceriodaphnia dubia* Survival[b] at 96 h (%)
	Aug. 1993	NT	NT	100	100
	Oct. 1993	NT	NT	100	100
	Dec. 1993	NT	NT	50	100
	Feb. 1994	NT	NT	100	100
	Apr. 1994	NT	NT	100	100
SD190	Jan. 1990	100	95	<100	0
	dechlor.	100	90	100	90
	May 1990	100	95	75	0
	Nov. 1990	<100	0	<100	0
	dechlor.	100	100	100	100
	May 1991	100	92.5	<100	0
	dechlor.	100	92.5	100	100
	Nov. 1991	NT	NT	<100[f]	0[f]
	dechlor.	NT	NT	<75	90
	Mar. 1992	I	I	100	100
	May 1992	100	92.5	<75	100
	July 1992	100	97.5	50	90
	Apr. 1993	NT	NT	<12	0
	June 1993	NT	NT	<12	0
	Aug. 1993	NT	NT	100	100
	Oct. 1993	NT	NT	50	90
	Dec. 1993	NT	NT	12	80
	Feb. 1994	NT	NT	<50	0
	Apr. 1994	NT	NT	<50	0

Note: I = invalid test (unacceptable control survival); NT = not tested; dechlor. = dechlorinated with sodium thiosulfate.

[a]No-observed-effect level (the highest concentration of effluent tested that does not significantly reduce fathead minnow survival or growth or *Ceriodaphnia* survival or reproduction).
[b]Survival in full-strength effluent.
[c]Survival was 0% in 5 d.
[d]Survival was 40% in 7 d.
[e]Survival was 30% in 7 d.
[f]Based on total residual chlorine concentration known to kill *Ceriodaphnia*; actual survival was not measured.

in 1993–94. Survival of *Ceriodaphnia* at 96 h remained low in 1993–94: survival was 0% in 4 of the 7 tests.

A summary of water quality parameters for the storm drains is provided in Table 3.4, and mean and maximum concentrations of total residual chlorine (TRC) are given in Table 3.5. The chemical characteristics of each effluent have remained similar from 1991 to 1994 and are similar to each other. Mean conductivity at SDs 170, 180, and 190 ranged from 301–501 µS/cm, 358–572 µS/cm, and 261–560 µS/cm respectively. Mean hardness at SDs 170, 180, and 190 ranged from 135–286 mg/L, 175–207 mg/L, and 135–250 mg/L respectively. Mean alkalinity at SDs 170, 180, and 190 ranged from 94–140 mg/L, 129–162 mg/L, and 101–195 mg/L respectively. During 1990, TRC was always present in effluent from SDs 170 and 190; the maximum concentration measured in SDs 170 and 190 was 0.91 mg/L and 1.03 mg/L respectively. In 1991, the maximum TRC concentrations measured in SDs 170 and 190 were 0.02 and 0.21 mg/L respectively. Measurements taken after November 1991 are not shown because TRC was never greater than 0.03 mg/L.

3.1.4 Discussion

Effluent from CNF has not exceeded the permit limits since the effective date of the renewed permit (October 1992). In February 1990 and April 1991, the effluent was highly toxic to *Ceriodaphnia* (NOEL ≤ 1). Thus, effluent from CNF was toxic (as defined by the permit) in 2 of the past 47 tests. In terms of toxicity, it is very unlikely that there is any continuous impact on the aquatic biota of Poplar Creek. However, the two cases of toxicity at low concentrations of effluent indicate that an episodic event could have impacted the biota of Poplar Creek. The toxicity observed in April 1991 coincided with the highest mean conductivity (8663 µS/cm) measured during 1990-93. Conductivity (from the presence of dissolved solids) does not really measure the toxicant, rather it is an indicator of the presence of a toxicant; a 1% concentration of effluent would contain very low dissolved solids, yet a 1% concentration of effluent was toxic to *Ceriodaphnia*. Water quality data collected in February 1990 did not provide any information concerning the source of the toxicant. The cause or the source of toxicity in the CNF effluent was not identified. Because there was no biotic sampling ongoing during either of the toxicity tests, the level of impact is unknown; however, no fish kills in Poplar Creek were reported to ESD during either toxicity test.

Table 3.4. Average water quality parameters measured during toxicity tests of storm drains 170, 180 and 190

Values are averages of full-strength wastewater for each test (n = 7)

Storm Drain	Test Date	pH (standard units)	Conductivity (µS/cm)	Alkalinity (mg/L $CaCO_3$)	Hardness (mg/L $CaCO_3$)
170	January 1990	7.98	501	94	135
	May 1990	8.08	490	108	286
	November 1990	8.12	403	122	175
	May 1991	8.02	405	140	184
	November 1991	7.90	339	126	175
	March 1992	8.17	321	111	160
	May 1992	8.20	329	118	166
	July 1992	8.24	339	115	162
	September 1992	8.21	346	120	164
	November 1992	8.26	333	119	162
	April 1993	8.20	324	99	190
	June 1993	8.10	352	123	179
	August 1993	8.20	358	119	189
	October 1993	8.10	346	115	167
	December 1993	8.10	358	106	153
	February 1994	8.10	346	105	169
	April 1994	8.20	301	99	151
180	January 1990	8.05	572	109	203
	May 1990	8.19	358	131	175
	November 1990	8.15	407	153	195
	May 1991	8.05	456	162	207
	November 1991	7.96	367	137	192
	March 1992	8.12	415	153	203
	May 1992	8.20	364	142	189
	July 1992	8.31	365	141	180
	April 1993	8.30	377	146	207
	June 1993	8.10	380	141	204
	August 1993	8.20	379	139	199
	October 1993	8.10	389	137	186

Table 3.4 (continued)

Storm Drain	Test Date	pH (standard units)	Conductivity (µS/cm)	Alkalinity (mg/L $CaCO_3$)	Hardness (mg/L $CaCO_3$)
	December 1993	8.20	410	129	183
	February 1994	8.20	409	144	187
	April 1994	8.20	388	145	201
190	January 1990	8.02	315	110	157
	May 1990	8.04	318	118	159
	November 1990	8.12	295	115	145
	May 1991	7.96	308	113	148
	November 1991	7.90	261	111	148
	March 1992	8.07	311	117	153
	May 1992	8.12	284	101	149
	July 1992	8.18	308	107	135
	April 1993	8.30	560	195	266
	June 1993	8.10	434	146	205
	August 1993	8.00	442	143	206
	October 1993	7.90	432	153	209
	December 1993	7.80	482	159	209
	February 1994	7.90	462	160	239
	April 1994	8.00	482	173	250

Table 3.5. Summary (mean and range) of total residual chlorine measured during toxicity tests of storm drains and ambient waters from Mitchell Branch

Sample	Date	Total Residual Chlorine (mg/L)	Range
MIK 0.71	Jan. 1990	0.03	0.00–0.06
MIK 0.54		0.03	0.00–0.04
MIK 0.45		0.03	0.00–0.05
MIK 0.12		0.01	0.00–0.03
SD 170		0.37	0.00–0.64
SD 180		0.00	0.00–0.00
SD 190		0.17	0.00–0.26
MIK 0.71	March 1990	0.07	0.01–0.17
MIK 0.54		0.04	0.01–0.11
MIK 0.45		0.04	0.02–0.08
MIK 0.12		0.02	0.00–0.06
MIK 0.71	May 1990	0.00	0.00–0.00
MIK 0.54		0.00	0.00–0.00
MIK 0.45		0.01	0.00–0.08
MIK 0.12		0.00	0.00–0.00
SD 170		0.11	0.00–0.69
SD 180		0.02	0.00–0.05
SD 190		0.03	0.00–0.06
MIK 0.71	July 1990	0.31	0.14–0.48
MIK 0.54		0.12	0.06–0.16
MIK 0.45		0.16	0.09–0.22
MIK 0.12		0.04	0.02–0.05
MIK 1.00	Sep. 1990	0.01	0.00–0.04
MIK 0.71		0.60	0.13–0.84
MIK 0.54		0.27	0.12–0.40
MIK 0.45	Sep. 1990	0.33	0.25–0.42
MIK 0.12		0.08	0.04–0.13
MIK 0.71	Nov. 1990	0.44	0.01–0.78
MIK 0.54		0.25	0.00–0.44
MIK 0.45		0.28	0.01–0.47
MIK 0.12		0.09	0.00–0.12

Table 3.5 (continued)

Sample	Date	Total Residual Chlorine (mg/L)	Range
SD 170		0.56	0.05–0.91
SD 180		0.01	0.00–0.01
SD 190		0.75	0.17–1.03
MIK 0.71	Jan. 1991	0.04	0.02–0.05
MIK 0.54		0.03	0.02–0.04
MIK 0.45		0.03	0.02–0.06
MIK 0.12		0.01	0.00–0.01
MIK 0.71	March 1991	0.00	0.00–0.00
MIK 0.54		0.00	0.00–0.00
MIK 0.45		0.01	0.00–0.04
MIK 0.12		0.00	0.00–0.00
MIK 0.71	May 1991	0.00	0.00–0.00
MIK 0.54		0.00	0.00–0.00
MIK 0.45		0.02	0.00–0.05
MIK 0.12		0.00	0.00–0.00
SD 170		0.00	0.00–0.02
SD 180		0.00	0.00–0.00
SD 190		0.13	0.11–0.21
MIK 0.71	July 1991	0.00	0.00–0.00
MIK 0.54		0.00	0.00–0.01
MIK 0.45		0.00	0.00–0.01
MIK 0.12		0.00	0.00–0.00
MIK 0.71	Sep. 1991	0.01	0.00–0.02
MIK 0.54		0.02	0.01–0.03
MIK 0.45		0.07	0.00–0.11
MIK 0.12		0.01	0.00–0.01
MIK 0.71	Nov. 1991	0.00	0.00–0.00
MIK 0.54		0.00	0.00–0.00
MIK 0.45		0.00	0.00–0.00
MIK 0.12		0.00	0.00–0.00
SD 170		0.00	0.00–0.00
SD 180		0.00	0.00–0.00

Table 3.5 (continued)

Sample	Date	Total Residual Chlorine (mg/L)	Range
SD 190		0.66	0.18–0.98
MIK 0.71	Jan. 1992	0.00	0.00–0.00
MIK 0.54		0.00	0.00–0.00
MIK 0.45		0.00	0.00–0.00
MIK 0.12		0.00	0.00–0.00
MIK 0.71	March 1992	0.00	0.00–0.00
MIK 0.54		0.00	0.00–0.00
MIK 0.45		0.00	0.00–0.00
MIK 0.12		0.00	0.00–0.00
SD 170		0.00	0.00–0.01
SD 180		0.00	0.00–0.00
SD 190		0.00	0.00–0.00
MIK 0.71	May 1992	0.00	0.00–0.00
MIK 0.54		0.00	0.00–0.00
MIK 0.45		0.00	0.00–0.00
MIK 0.12		0.00	0.00–0.00
SD 170	May 1992	0.00	0.00–0.00
SD 180		0.00	0.00–0.01
SD 190		0.00	0.00–0.00
MIK 0.71	July 1992	0.00	0.00–0.01
MIK 0.54		0.00	0.00–0.00
MIK 0.45		0.00	0.00–0.00
MIK 0.12		0.00	0.00–0.00
SD 170		0.00	0.00–0.00
SD 180		0.00	0.00–0.00
SD 190		0.00	0.00–0.00
MIK 0.71	Sep. 1992	0.00	0.00–0.02
MIK 0.54		0.00	0.00–0.00
MIK 0.45		0.00	0.00–0.00
MIK 0.12		0.00	0.00–0.01
SD 170		0.01	0.01–0.02
MIK 0.71	Nov 1992	0.00	0.00–0.03

Table 3.5 (continued)

Sample	Date	Total Residual Chlorine (mg/L)	Range
MIK 0.54		0.00	0.00–0.00
MIK 0.45		0.00	0.00–0.00
SD 170		0.02	0.01–0.04
MIK 0.71	Feb. 1993	0.00	0.00–0.01
MIK 0.54		0.00	0.00–0.02
MIK 0.45		0.00	0.00–0.01
MIK 0.12		0.00	0.00–0.02
MIK 0.71	Apr. 1993	0.00	0.00–0.01
MIK 0.54		0.00	0.00–0.01
MIK 0.45		0.00	0.00–0.02
MIK 0.12		0.00	0.00–0.01
SD 170	Apr. 1993	0.01	0.00–0.03
SD 180		0.02	0.01–0.03
SD 190		0.00	0.00–0.01
SD 170	June 1993	0.00	0.00–0.00
SD 180		0.00	0.00–0.01
SD 190		0.00	0.00–0.00
MIK 0.71	July 1993	0.00	0.00–0.00
MIK 0.54		0.00	0.00–0.00
MIK 0.45		0.00	0.00–0.00
MIK 0.12		0.00	0.00–0.00
SD 170	Aug. 1993	0.00	0.00–0.00
SD 180		0.00	0.00–0.00
SD 190		0.00	0.00–0.00
MIK 0.71	Oct. 1993	0.00	0.00–0.00
MIK 0.54		0.00	0.00–0.00
MIK 0.45		0.00	0.00–0.00
MIK 0.12		0.00	0.00–0.00
SD 170		0.00	0.00–0.00
SD 180		0.00	0.00–0.00
SD 190		0.00	0.00–0.00
SD 170	Dec. 1993	0.00	0.00–0.00

Table 3.5 (continued)

Sample	Date	Total Residual Chlorine (mg/L)	Range
SD 180		0.00	0.00–0.00
SD 190		0.00	0.00–0.00
MIK 0.71	Jan. 1994	0.00	0.00–0.00
MIK 0.54		0.00	0.00–0.00
MIK 0.45		0.00	0.00–0.00
MIK 0.12		0.00	0.00–0.00

Note: SD = storm drain. MIK = Mitchell Branch kilometer.

Effluent from K-1203 has exceeded the permit limits twice since testing began. In July 1993, *Ceriodaphnia* reproduction was significantly reduced at 4.2%, and in March 1994, fathead minnow survival was significantly reduced at 4.2%. In both cases, a test was conducted within 2 weeks and no effect on reproduction or survival was observed at 30% (the highest concentration tested). This lack of toxicity indicates that the toxicity observed in July 1993 and March 1994 was transient. At this time, the cause or source of the toxicity has not been identified. Although the effluent exceeded the permit limits twice, there have been 22 tests in which no toxicity (as defined by the permit) has been observed; thus, it is unlikely that there is any continuous impact on the biota. However, as was the case for CNF, the two cases of toxicity at low concentrations of effluent indicate that an episodic event could have impacted the biota of Poplar Creek. At this time, the cause or source of the toxicity has not been identified. No fish kills in Poplar Creek were reported to ESD during either toxicity test.

Effluent from SD 180 is typically non-toxic to *Ceriodaphnia*. Effluent from SDs 170 and 190 has consistently produced NOELs of ≤ 50%. Previous to 1991, much of the toxicity (but not all) was attributed to TRC (Kszos 1994). Dechlorination of the storm drains at the discharge point began in 1991 and removed some of the acute toxicity, as evidenced by the increase in survival at 96 h (Table 3.3). Dechlorination did not remove all the chronic or acute toxicity to *Ceriodaphnia*. In particular, effluent from SD 190 decreased *Ceriodaphnia* 96-h survival to 0% in 4 of the 7 tests conducted in 1993–94. The low survival is not due to total residual chlorine because concentrations have not been measured above 0.03 mg/L since 1991. During 1990-91, concentrations of TRC in effluent from SDs 170 and 190 frequently exceeded the EPA water quality criteria for chronic exposure (0.011 mg/L) (EPA 1986b). Since November 1991, TRC concentrations have typically been 0.0 mg/L. See section 3.2.4 for further discussion of the impact of storm drain effluents on Mitchell Branch.

3.2 AMBIENT TOXICITY

3.2.1 Introduction

Ambient toxicity monitoring at the K-25 Site employed the *Ceriodaphnia* and fathead minnow tests described in Sect. 3.1. Toxicity monitoring was incorporated into BMAP in order to (1) evaluate area source contributions to stream toxicity, (2) characterize patterns of toxicity in Mitchell Branch and Poplar Creek, (3) document changes in water quality attributable to changes in operations at the K-25 Site and (4) provide data demonstrating that the effluent

limitations established for the K-25 Site protect and maintain the use of Mitchell Branch, Poplar Creek, and Clinch River for growth and propagation of fish and aquatic life. Sites in Poplar Creek and Clinch River were added in April 1993 when the revised BMAP (Kszos et al. 1993) was published. The sites chosen for testing on Mitchell Branch, Poplar Creek, and the Clinch River were selected to bracket area and point source discharges into the stream and to correspond closely to those selected as instream monitoring study sites. The Poplar Creek and Clinch River sites are shared with the CRRI (R. B. Cook et al., 1993).

3.2.2 Materials and Methods

Ambient toxicity was evaluated using the fathead minnow and *Ceriodaphnia* tests as described in Sect. 3.1 for effluents with the following exceptions: (1) no dilutions were tested, (2) each test of Mitchell Branch used seven consecutive, daily grab samples of stream water, and (3) each test of Poplar Creek and Clinch River used three grab samples collected over 6 days. Six ambient sites on Mitchell Branch (MIK 1.43, MIK 1.0 or MIK 0.78, MIK 0.71, MIK 0.54, MIK 0.45, and MIK 0.12; Fig. 2.2), five sites on Poplar Creek (PCK 18.2, PCK 8.2, PCK 6.9, PCK 4.7, and PCK 1.6, Fig. 2.1), and one site on the Clinch River (CRK 15.0; Fig. 2.1) were evaluated for toxicity. These Mitchell Branch sites are similar to those selected for the ecological monitoring component of BMAP (Sect. 5). The site at MIK 1.0 was moved to MIK 0.78 in April 1993 to coincide with the benthic macroinvertebrate sampling site. Testing of the Poplar Creek and Clinch River sites began in April 1993. Toxicity tests with *Ceriodaphnia* and fathead minnows were conducted approximately every other month from January 1990 to February 1993. Quarterly testing began with implementation of the new BMAP plan (Kszos et al. 1993) in April 1993. Water sampling and water chemistry analyses were conducted as described for the effluents in Sect. 3.1.2. Results of toxicity tests conducted in 1987–89 are presented in Kszos (1993 and 1994).

SAS statistical software (Statistical Analysis System for personal computers, release 6.03) was used to analyze all data. Survival percentages for fathead minnow larvae and for *Ceriodaphnia* were transformed (arcsine square root; Steel and Torrie 1960) before being analyzed. All fecundity values are for females that survived all 6 or 7 d of the test. Differences in hardness, alkalinity, conductivity, fathead minnow survival and growth, and *Ceriodaphnia* reproduction between sites were evaluated with an analysis of variance (ANOVA) by using the SAS general linear models (GLM) procedure (SAS 1985a, 1985b);

significant differences were then separated with Tukey's studentized range test. The Tukey test was selected because it accommodates unequal sample sizes. Because significant differences existed from test to test in *Ceriodaphnia* reproduction and fathead minnow survival and growth, the GLM procedure was inappropriate for separating differences among all sites and tests. Thus, separate GLM analyses were conducted for each test period. For each test period, significant reductions in *Ceriodaphnia* survival were determined by using Fisher's Exact Test (Weber et al. 1989). Significant reductions in *Ceriodaphnia* survival and reproduction in Mitchell Branch, Poplar Creek, and Clinch River sites are based on comparisons with the reference sites (MIK 1.43 for Mitchell Branch and PCK 18.2 for Poplar Creek and Clinch River). Fathead minnow survival at the reference site MIK 1.43 is typically low and is hypothesized to be due to a natural pathogen (Kszos 1993, 1994). Thus, significant reductions in minnow survival or growth at the sites were based on a comparison with survival and growth in a laboratory control water. Unless otherwise noted, statements of significance (probability) are based on $p = 0.05$.

3.2.3 Results

3.2.3.1 Water chemistry

Mitchell Branch

Mean pH, alkalinity, hardness, and conductivity for each site and test from 1990 through 94 are provided in Appendix B (Table B.1). Annual mean conductivity, alkalinity, and hardness are presented in Fig. 3.1. Chemistry measurements for 1990–94 are presented in Fig. 3.2.

Each year (Fig. 3.1), conductivity, alkalinity, and hardness were significantly lower at MIK 1.43 than at sites further downstream. Conductivity increased with distance downstream from approximately 180 µS/cm at MIK 1.43 to 350 µS/cm at MIK 0.12 (Figs. 3.1 and 3.2). In 1990, 1991, and 1992, conductivity at MIK 1.0 was significantly higher than at MIK 1.43 and significantly lower than at sites further downstream. In 1993, conductivity was not different from MIK 0.78 (which replaced MIK 1.0) downstream to MIK 0.12. Each year, conductivity at MIK 0.71 was significantly lower than at MIK 0.12 but not from MIK 0.71 downstream to MIK 0.45. The comparison of conductivity for all test dates (Table 3.6) showed that MIK 0.45 could be distinguished from MIK 0.71 and that adjacent sites were not different from each other.

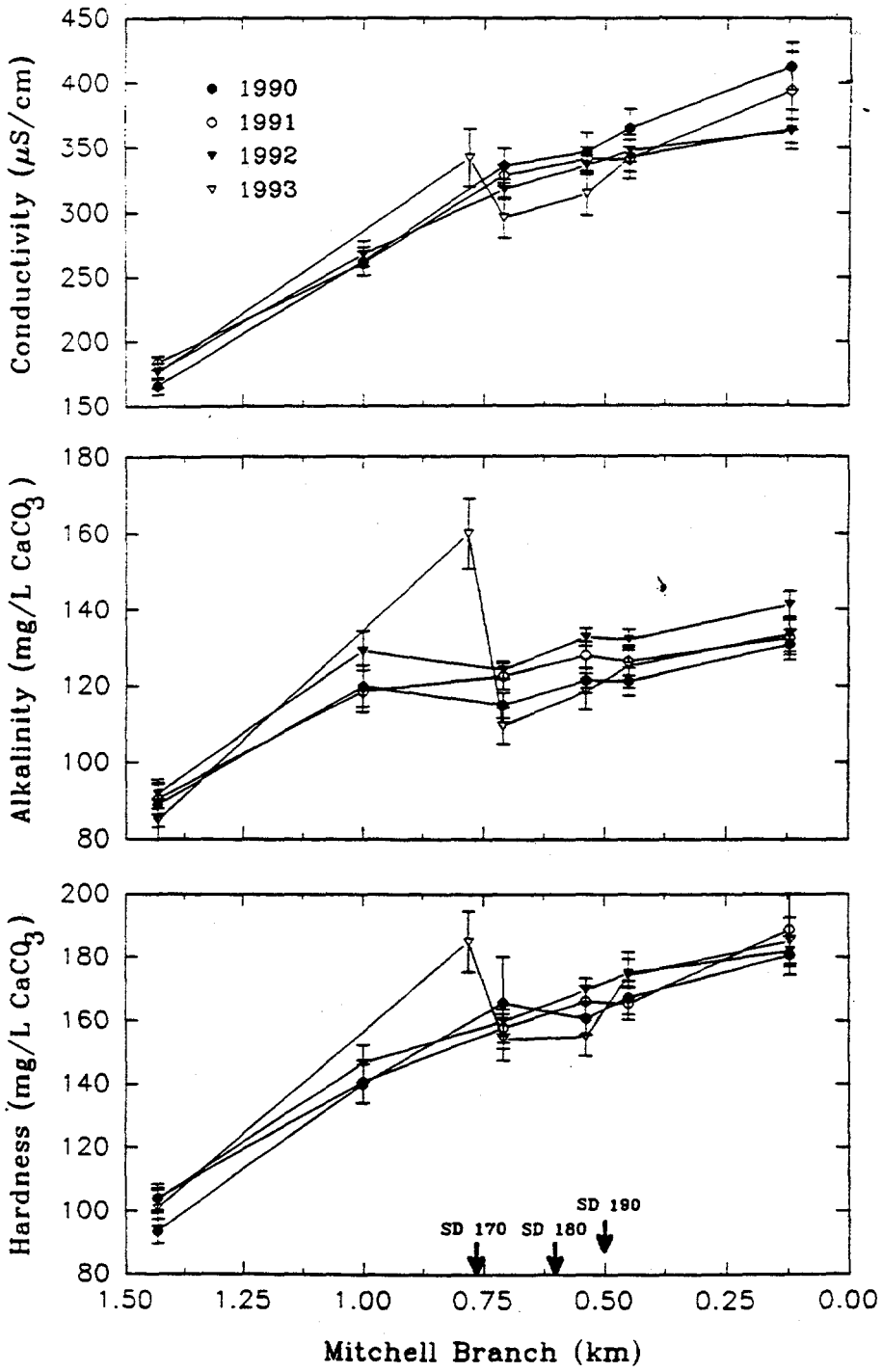

Fig. 3.1. Annual mean conductivity, alkalinity and hardness for each site in Mitchell Branch, 1990–1993. Vertical bars represent ± 1 SE. SD = storm drain.

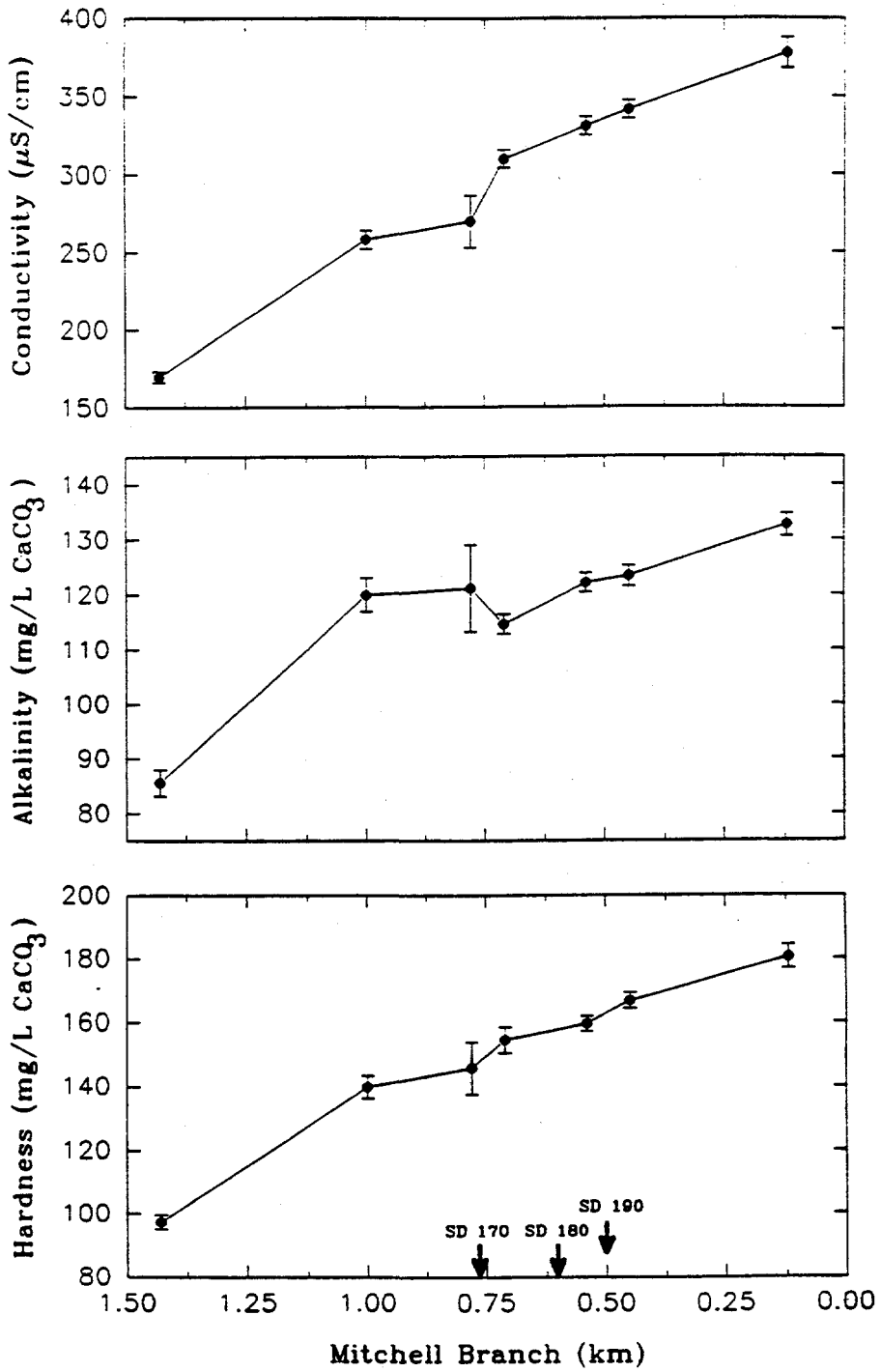

Fig. 3.2. Multi-year mean conductivity, alkalinity and hardness for each site in Mitchell Branch. Values are for tests conducted January 1990 to January 1994. Vertical bars represent ± 1 SE. SD = storm drain.

Alkalinity increased from approximately 90 mg/L at MIK 1.43 to 120 mg/L at MIK 0.12 (Fig. 3.1 and 3.2). Alkalinity (each year and for all test dates combined; Table 3.6) was not different from MIK 1.0 downstream to MIK 0.12 with the exception of MIK 0.78 in 1993 (Figs. 3.1 and 3.2). Alkalinity at MIK 0.78 was significantly higher than at all sites except MIK 0.12.

Table 3.6. Tukey groupings for conductivity and hardness for tests completed during 1990-1994

Site (MIK)	Conductivity						Hardness				
1.43	A						A				
1.00		B						B			
0.78		B	C					B	C		
0.71			C	D					C	D	
0.54				D	E				C	D	
0.45					E					D	
0.12						F					E

Note: Sides with the same letter are not significantly different. MIK = Mitchell Branch kilometer

Hardness increased from approximately 100 mg/L at MIK 1.43 to 170 mg/L at MIK 0.12 (Figs 3.1 and 3.2). During 1990 and 1991, hardness at MIK 1.0 was significantly lower than at MIK 0.12 but not different from the remaining sites downstream (MIK 0.71 to MIK 0.45), and hardness at MIK 0.12 was not different from the three sites upstream (MIKs 0.45, 0.54, and 0.71). The comparison of hardness for all test dates (Table 3.6) showed that MIK 0.12 could be distinguished from all sites; however, differences in hardness were not significantly different from MIK 0.78 downstream to MIK 0.54 and from MIK 0.71 downstream to MIK 0.45. Hardness was not different between MIK 1.0 and MIK 0.78.

TRC measured at each site is shown in Table 3.5. From January to November 1990, TRC was present at all sites from MIK 0.71 downstream to MIK 0.12. For example, maximum TRC at MIK 0.71 ranged from 0.00 to 0.78 mg/L. Maximum TRC at MIK 0.45 ranged from 0.03 to 0.47 mg/L. From January 1991 to November 1991, maximum TRC was

above 0.06 mg/L only once at MIK 0.45. Measurements taken after November 1991 are not shown in Table 3.5 because TRC was never greater than 0.03 mg/L.

Poplar Creek and Clinch River

Mean pH, alkalinity, hardness, and conductivity for each test and site from 1990 to 94 are provided in Appendix B (Table B.1). Mean water quality changed very little with distance downstream in Poplar Creek with the exception of the test conducted in April 1993. Mean alkalinity, hardness, and conductivity were lower at all sites in April 1993 and January 1994 as compared with July and October 1993. In April 1993 and January 1994, alkalinity ranged from about 40 to 70 mg/L for the Poplar Creek sites and averaged about 100 mg/L for the Clinch River site; hardness ranged from about 70 to 130 mg/L for the Poplar Creek sites and 125 to 150 mg/L for the Clinch River site; conductivity ranged from about 160 to 225 μS/cm for the Poplar Creek sites and about 255 to 260 μS/cm for the Clinch River site. In July and October 1993, alkalinity ranged from about 112 to 140 mg/L for the Poplar Creek and Clinch River sites; hardness ranged from about 150 to 190 mg/L for the Poplar Creek and Clinch River sites; conductivity ranged from about 250 to 375 μS/cm for the Poplar Creek and Clinch River sites.

3.2.3.2 Fathead minnow tests

Mitchell Branch

Survival and growth of fathead minnows in water from each site and each test are presented in Appendix B (Table B.2). Mean survival and growth in minnows in water from each site in each year are plotted in Fig. 3.3. Results for 1987–89 are presented in Kszos (1993, 1994). As in previous years, minnow survival in water from the reference site (MIK 1.43) was low (about 60%) during each year. At MIK 1.0, mean survival was also low each year (about 60%), compared with other sites and compared with 1987–89, when mean survival was about 80 to 90%. Mean survival at MIKs 0.78, 0.71, 0.54, 0.45, and 0.12 was about 90% in each year with the exception of low mean survival (60%) at MIK 0.45 in 1990. This low mean survival was because of two tests in which none of the minnows survived. Mean survival at MIK 0.71 and MIK 0.45 is artificially elevated because one test at each site was not conducted when TRC was high enough to have killed all the test organisms on the first day of the test. Thus, had the tests been conducted, mean survival at these sites would be

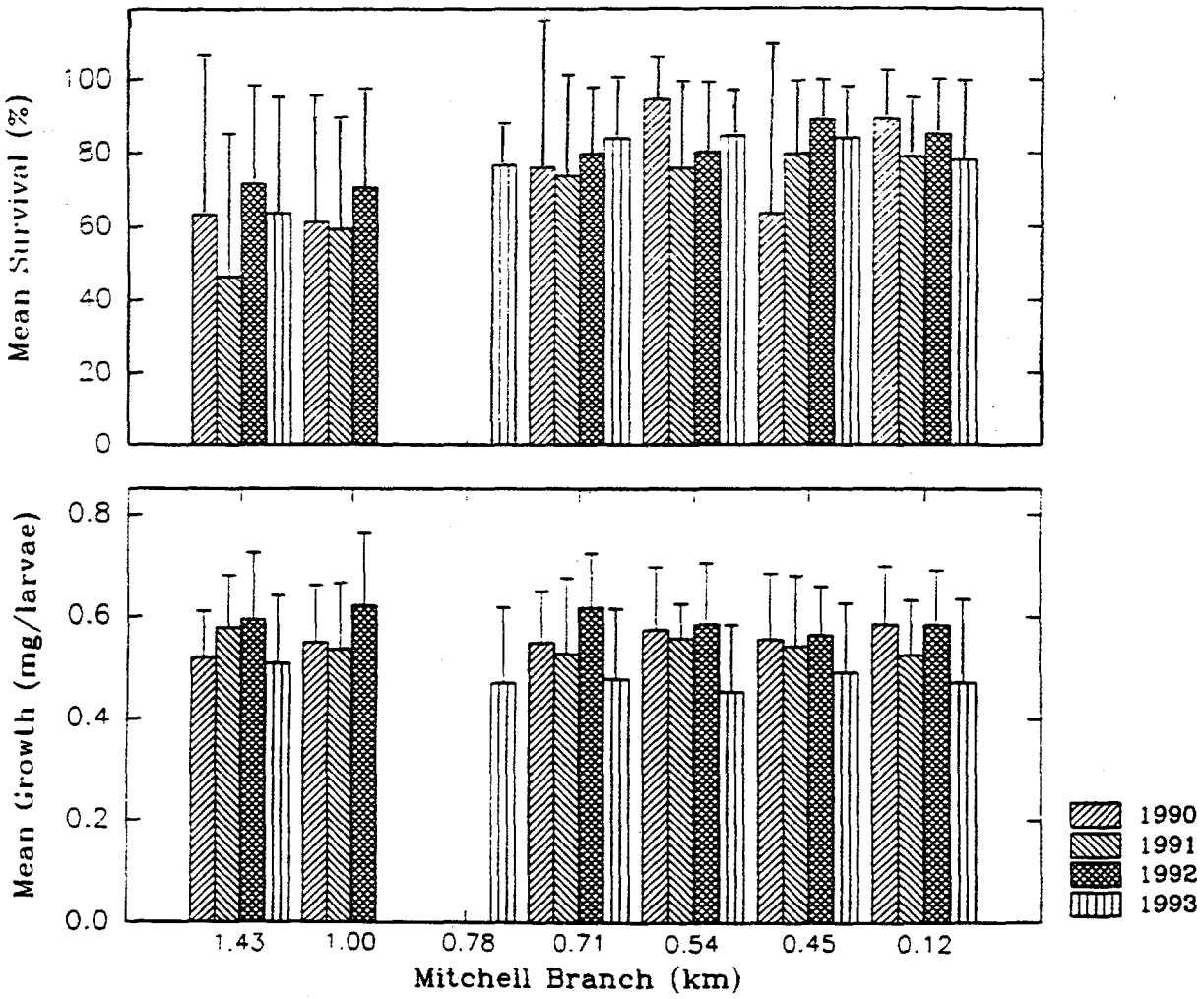

Fig. 3.3. Mean percentage survival (a) and growth (b) of fathead minnows in Mitchell Branch, 1990–1993. Vertical bars represent ± SD.

closer to 70%. Mean growth between sites in each year was similar and was lower at all sites in 1993. Significant differences in survival and growth are presented in Table 3.7. No toxicity to fathead minnows occurred in 95 of 106 test/site combinations. Acute toxicity occurred once at MIK 0.71 and twice at MIK 0.45 in 1990. Chronic toxicity was found in 5 of 106 test/site combinations: 3 times at MIK 1.00 in 1990, 1 time at MIK 0.71 in 1993, and 1 time each at MIK 0.54 and MIK 0.45 in 1992.

Poplar Creek and Clinch River

Survival and growth of fathead minnows in water from each site and for each test are presented in Appendix B (Table B.2). Minnow survival and growth in water from each site were never significantly lower than that in the reference site (PCK 18.2).

3.2.3.3 *Ceriodaphnia* tests

Mitchell Branch

Survival and reproduction of *Ceriodaphnia* in water from each site and for each test are presented in Appendix B (Table B.2). Mean survival and reproduction in water from each site in each year are plotted in Fig. 3.4. Results for 1987–89 are presented in Kszos (1993, 1994). Mean survival and reproduction at MIKs 1.43 and 1.00 were consistently high. Mean survival at MIKs 0.71, 0.54, and 0.45 was the lowest (about 40%) during 1990. From 1990 to 1993, survival at all sites was ≥ 70%. Mean reproduction at all sites from MIK 0.71 downstream to MIK 0.12 was lower in 1990 and 1992 than in 1991 or 1993. Significant differences in survival and reproduction are presented in Table 3.7. No toxicity to *Ceriodaphnia* occurred in 78 of 108 test/site combinations. Acute toxicity occurred in 14 of 108 test/site combinations with 11 of the 18 occurring during 1990 (three times each at MIK 0.71 and MIK 0.54, four times at MIK 0.45, and once at MIK 0.12). Chronic toxicity was found in 15 of 108 test/site combinations with 10 of the 15 occurring at MIKs 0.71, 0.54, and 0.45 during 1992. The only incidence of chronic toxicity in 1993 occurred at MIK 0.71.

Poplar Creek and Clinch River

Survival and reproduction of *Ceriodaphnia* in water from each site and for each test are presented in Appendix B (Table B.2). *Ceriodaphnia* survival and reproduction in water from each site were never significantly lower than the reference site (PCK 18.2).

Table 3.7. Number of fathead minnow and *Ceriodaphnia* tests where no toxicity, acute toxicity or chronic toxicity was observed for Mitchell Branch sites

			No Toxicity[a]		Acute Toxicity[b]		Chronic Toxicity[c]	
Site (MIK)	Year	n[d]	FHM	CD	FHM	CD	FHM	CD
1.00	1990	6	3	6	0	0	3	0
	1991	4/5	3	5	0	0	1	0
	1992	6	5	6	0	0	1	0
	1993	1	1	1	0	0	0	0
	All	17/18	12	18	0	0	4	0
0.78	1993	3	3	3	0	0	0	0
0.71	1990	5[e]	4	2	1	3	0	0
	1991	6	6	5	0	0	0	1
	1992	6	6	1	0	1	0	4
	1993	4	4	2	0	1	0	1
	All	21	20	10	1	5	0	6
0.54	1990	5[e]/6	5	3	0	3	0	1
	1991	6	6	6	0	0	0	0
	1992	6	5	1	0	1	1	4
	1993	4	4	4	0	0	0	0
	All	21/22	20	14	0	4	1	5
0.45	1990	6	4	2	2	4	0	0
	1991	6	6	6	0	0	0	0
	1992	6	6	4	0	0	0	2
	1993	4	4	4	0	0	0	0
	All	22	20	16	2	4	0	2
0.12	1990	6	6	5	0	1	0	0
	1991	6	6	6	0	0	0	0
	1992	6	6	4	0	0	0	2
	1993	4	4	4	0	0	0	0
	All	22	22	18	0	1	0	2

Note: MIK = Mitchell Branch kilometer; FHM = fathead minnow; CD = *Ceriodaphnia dubia*.

[a]There was no significant reduction in minnow survival or growth or *Ceriodaphnia* survival or reproduction.

[b]Survival ≤20% in 48 h.

[c]Minnow survival or growth significantly reduced compared to laboratory control or *Ceriodaphnia* survival or reproduction significantly reduced compared to reference site.

[d]n = number of tests given for both species or as fathead minnow/*Ceriodaphnia*.

[e]Test was not completed because total residual chlorine would have killed all organisms on the first day of the test

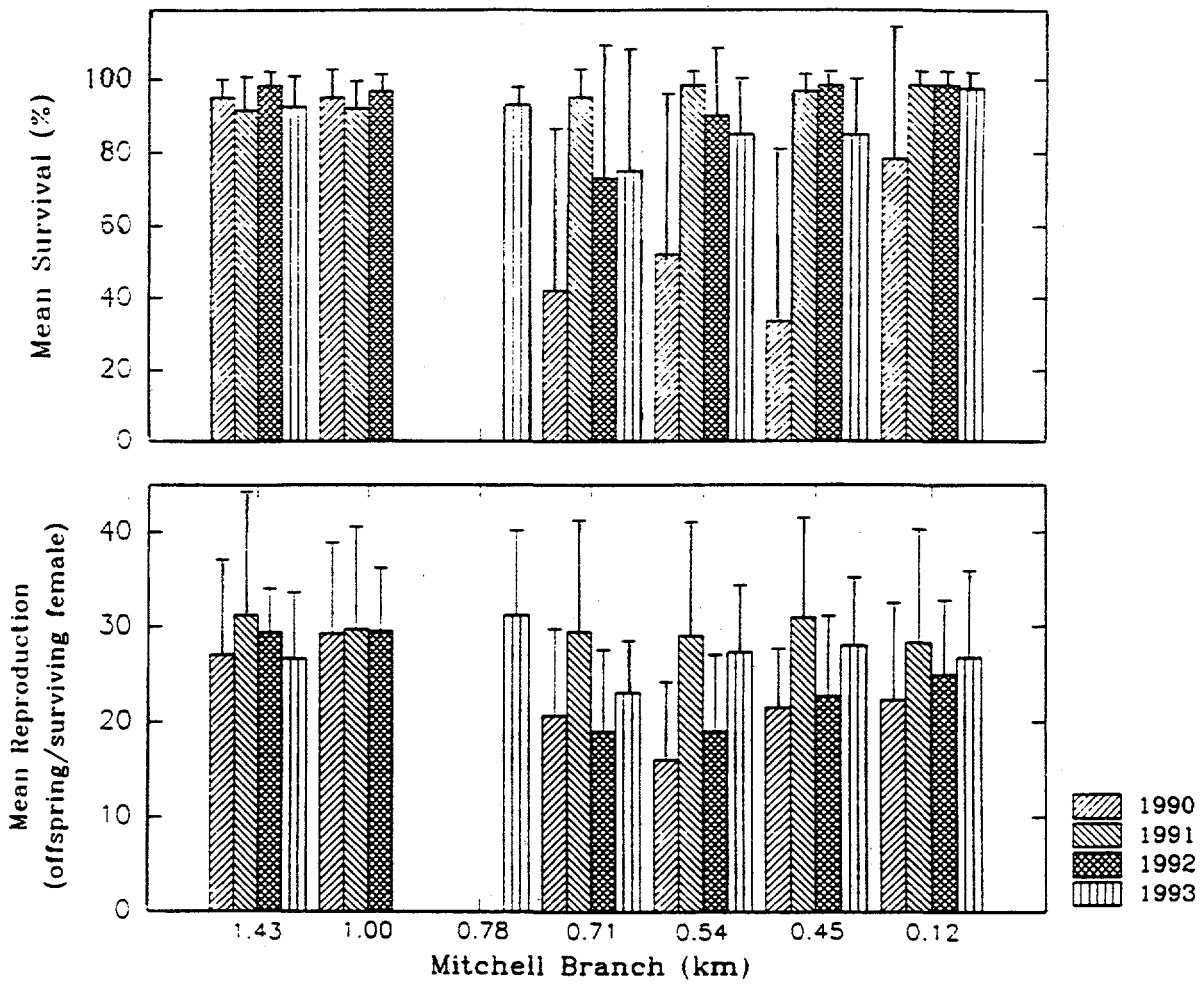

Fig. 3.4. Mean percentage survival (a) and reproduction (b) of *Ceriodaphnia* for each site in Mitchell Branch, 1990–1993. Vertical bars represent ± SD.

3.2.4 Discussion

The K-25 Site successfully eliminated the acute toxicity of water from Mitchell Branch to fathead minnows and *Ceriodaphnia* with the dechlorination of the storm drain effluents in 1991 at the NPDES discharge points. This is evidenced by the dramatic reduction in concentrations of TRC coincident with a near elimination of acute toxicity. However, the ambient toxicity test data and results of the storm drain toxicity tests indicate that the effluent from SDs 170 and 190 continues to adversely impact Mitchell Branch. Although the majority of tests for Mitchell Branch sites show no chronic toxicity, *Ceriodaphnia* reproduction has been reduced at MIKs 0.71 (below SD 170), 0.54 (below SD 180), 0.45 (below SD 190) and MIK 0.12 fifteen times in 1992–93. In addition, chronic toxicity at SDs 170 and 190 is the norm for tests conducted in 1993–94.

Ceriodaphnia have been shown to be more sensitive than fathead minnows to water from all of the ambient sites except MIK 1.0. In each case where toxicity to fathead minnows was observed, toxicity to *Ceriodaphnia* was also observed. In other cases, no toxicity to fathead minnows was observed but *Ceriodaphnia* survival and/or reproduction was reduced. As in previous years, minnow survival at MIK 1.43 remains low, indicating that this site is an inappropriate reference site for ambient toxicity tests with fathead minnows. It remains our hypothesis that the toxicity at MIK 1.43 is due to a natural pathogen rather than an anthropogenic input (cf. Kszos 1993, 1994; Stewart 1994). Evidence which supports this hypothesis includes (1) lack of toxicity in samples treated with ultraviolet light or filtration, (2) high beaker to beaker variability in minnow survival and (3) low mean survival and beaker to beaker variability in water from non-contaminated reference sites (Kszos and Stewart 1992). Due to the higher sensitivity of the *Ceriodaphnia* and the low minnow survival at MIK 1.43, subsequent ambient toxicity tests will be conducted with only *Ceriodaphnia.* Because toxicity is rarely present at SD 180, testing at this storm drain will be reduced to several times per year. These changes will allow us to concentrate on (1) investigating the source(s) of toxicity in SDs 170 and 190, (2) increase the frequency of testing at Mitchell Branch sites from four times per year to six, (3) conduct tests of Mitchell Branch and the storm drains concurrently, and (4) continue to investigate potential sources of toxicity from other storm drains.

The magnitude of the increases in conductivity, alkalinity, and hardness with distance downstream in Mitchell Branch have remained the same during 1990–93. Analyses of water

chemistry data indicates that the increase in conductivity and hardness is due to cumulative inputs from the K-25 Site rather than substantial input from one particular storm drain. Chemically, MIKs 1.43 and 0.12 could be distinguished from all other sites but adjacent sites could not be distinguished from each other (e.g., MIK 0.45 and MIK 0.54). During 1990–91, concentrations of TRC in water from MIKs 0.71, 0.54, and 0.45 frequently exceeded the EPA water quality criteria for chronic exposure of 0.011 mg/L (EPA 1986b). Since November 1991, TRC concentrations have typically been 0.0 mg/L.

Toxicity tests of water from Poplar Creek and Clinch River were conducted for the first time during 1993. Water quality data indicate that at least two different flow conditions were evaluated for Poplar Creek. In April 1993 and January 1994, mean alkalinity, hardness, and conductivity were lower at all sites than in July and October 1993. For all tests conducted, no toxicity was evident in water from any site. The water chemistry and toxicity data provide no evidence that effluents entering via Mitchell Branch (enters Poplar Creek above PCK 6.9) or CNF (enters Poplar Creek above PCK 4.7) impact Poplar Creek. In addition, the toxicity tests do not detect any impact from the Power House area (above PCK 1.6). Toxicity tests of water from Poplar Creek and Clinch River will therefore be discontinued.

4. BIOACCUMULATION STUDIES (TASK 2)

M. J. Peterson, G. R. Southworth, B. G. Blaylock,
B. R. O'Neal, D. K. Cox, and T. L. Ashwood

During the period 1987–92, bioaccumulation monitoring for the K-25 Site BMAP focused on the extent of contamination in aquatic biota in Mitchell Branch and nearby waters of Poplar Creek. In 1993, to address modifications cited in the new (1992) NPDES permit, the bioaccumulation monitoring task increased in scope to evaluate potential biological contamination in other aquatic habitats associated with the K-25 Site and to assess contamination in the terrestrial environment.

The bioaccumulation studies task has four primary objectives: (1) identify contaminants that accumulate in biota, (2) determine the degree of contamination, (3) identify and evaluate contaminant sources and (4) evaluate spatial and temporal changes in biotic contamination as a measure of the success of remedial actions. This task has two components: contaminant accumulation in aquatic biota (subtask 2a), and contaminant accumulation in terrestrial biota (subtask 2b).

4.1 CONTAMINANT ACCUMULATION IN AQUATIC BIOTA (SUBTASK 2a)

(M. J. Peterson and G. R. Southworth)

4.1.1 Introduction

The results presented in previous BMAP reports (Smith et al. 1993, 1994) encompassed monitoring activities at the K-25 site from 1987 to 1990. Mercury and polychlorinated biphenyls (PCBs) in fish and clams at sites near the K-25 Site were higher than levels found in fish and clams at two reference streams. Although Mitchell Branch was a likely source of PCBs to biota in Poplar Creek, the PCB contribution from Mitchell Branch appeared to be small relative to other possible sources (i.e., East Fork Poplar Creek and Bear Creek inputs and Poplar Creek sediments).

Mercury was clearly elevated in Mitchell Branch fish; however, levels were below the FDA action level of 1.0 μg/g (FDA 1984b) and the EPA's interim recommended screening value of 0.6 μg/g (EPA 1993). The close proximity of the Mitchell Branch site to Poplar Creek, coupled with the similar mercury concentrations (after size adjustment) in fish from the two systems, suggested that fish collected from Mitchell Branch may have migrated from

mercury-contaminated Poplar Creek. The significance of Mitchell Branch as a source of mercury contamination to its own biota, or to biota in downstream waters, was unresolved.

In 1991 and 1992, bioaccumulation monitoring efforts continued to focus on Mitchell Branch and nearby waters of Poplar Creek with little change from past years. The exception was a special study using caged dace in 1992 that attempted to evaluate the significance of Mitchell Branch as source of mercury contamination to its resident biota. In 1993, to address modifications cited in the new (1992) NPDES permit, monitoring increased in scope to evaluate potential biological contamination in other aquatic habitats associated with the K-25 Site. A contaminant screening study was implemented to assess the bioaccumulation of a wide range of contaminants at various locations, including at some ponds near the K-25 Site that had not been previously sampled. Resident fish and transplanted clams were the primary monitoring tools used to assess biotic contamination. In spring 1993, to monitor spatial and temporal trends in mercury and PCB contamination, sunfish were sampled at an expanded number of sites on Poplar Creek and the Clinch River.

4.1.2 Methods

Sunfish were collected by electrofishing from Mitchell Branch (MIK 0.2) and two nearby sites on Poplar Creek (PCKs 6.9 and 8.2) once yearly from 1987 to 1992. For comparison, sunfish were also collected from an uncontaminated reference stream, Hinds Creek. Sunfish from additional sites on Poplar Creek (PCKs 10.4 and 1.6) and the Clinch River (CRK 15.0) were sampled during this time, but less frequently. For each monitoring period, fish from Mitchell Branch were also analyzed for a suite of other metals, and asiatic caged clams were placed in Mitchell Branch to monitor organic contaminants. Clams are used because they provide a time-averaged measure of current exposure, and because they have been shown to be effective monitors of organic compounds that are more readily metabolized and excreted (and therefore accumulated) by fish.

In 1993, monitoring of mercury and PCB contamination in sunfish continued at sites monitored in past years (CRK 15.0, MIK 0.2, PCK 1.6, PCK 6.9, and PCK 8.2), with the addition of a site upstream of any K-25 Site impacts (PCK 18.2). Fish were collected from PCK 1.6, PCK 18.2, MIK 0.2, and three ponded tributaries of Poplar Creek (K-1007-P1, K-1007-P5, and K-710A ponds) to be analyzed for a variety of metals in addition to mercury and PCBs. To assess the presence of organic contaminants, caged asiatic clams were placed in

Mitchell Branch (MIK 0.2), Poplar Creek (PCKs 18.2 and 1.6), and the K-1007-P1 and K-710A ponds.

Bluegill (*Lepomis macrochirus*) was the primary fish species utilized to track mercury and PCB contamination in Poplar Creek and the Clinch River. In Mitchell Branch, where bluegill were not abundant, redbreast sunfish were used. Eight sunfish were analyzed individually for mercury and PCBs at each site. At each site where fish were analyzed for metals other than mercury, four bluegill comprised the collection for metals analysis. Four largemouth bass (*Micropterus salmoides*) were collected from each pond site for PCB analysis, except at the K-1007-P5 pond where only two bass were available. (Four bluegill were used to supplement the collection.) Largemouth bass were chosen for screening PCB contamination in the ponds because this species is a lipid-rich game fish that is most likely to represent the maximum level of PCBs in pond fish. Eight channel catfish were collected from both PCK 6.9 and CRK 15.0 for PCB analysis as part of continued off-site monitoring conducted jointly by the Y-12, K-25, and ORNL BMAPs.

An attempt was made to restrict collections to individuals of a size likely to be taken by sport fisherman (>40 g for sunfish, >400 g for largemouth bass and channel catfish). Reference fish were obtained from Hinds Creek and from two sites upstream of DOE impacts: upper Poplar Creek (PCK 18.2) and Melton Hill Reservoir (MHR; catfish reference site).

Upon collection, sunfish from all sites were tagged with a unique four-digit tag wired to the lower jaw and placed on ice in a labelled ice chest; the chest was locked for transport to the laboratory. Upon return to the laboratory, each fish was weighed, measured (total length), skinned and fileted. For mercury determination, a 1- to 2-g portion of the anterior dorsal axial muscle filet was excised. A 5-g portion of the remaining tissue was used for analysis of PCBs and/or other metals. Largemouth bass and channel catfish (*Ictalurus punctatus*) were handled and processed as sunfish were, except that fish filets were frozen and ground three times in a hand-powered meat grinder before a 10- to 20-g sample was removed for PCB analysis. All samples were wrapped in heavy-duty aluminum foil, labelled and stored at -20°C in a locked freezer in Building 1505 at ORNL until delivered to the Analytical Services Organization (ASO) laboratory at ORNL.

To evaluate Mitchell Branch as a source of mercury to its own biota (see Sect. 4.1.3.1), blacknose dace (*Rhinichthys atratulus*, a common minnow of small streams in eastern North America) collected from an uncontaminated reference stream (Hinds Creek) were stocked in

cages and placed in Mitchell Branch (MIK 0.6). A similar evaluation of potential mercury sources was conducted concurrently at two sites on Bear Creek (BCK 4.5 and BCK 12.0). For comparison, caged blacknose dace were also placed in an uncontaminated tributary of the same Poplar Creek system [Pin Hook Creek (PHK 1.6)], in a known mercury-contaminated stream [East Fork Poplar Creek (EFK 23.4)], and in a laboratory tank with no food source.

Twelve blacknose dace (6-8 cm total length) were divided among three cages (four fish per cage) and placed at each site beginning in May 1992. Damaged cages at EFK 23.4 and BCK 11.0 necessitated that the monitoring period begin in August 1992 at these two sites. At the end of a 12-week exposure period, the muscle tissue (including a small amount of bone) of two blacknose dace from each cage, if possible, was analyzed for total mercury (n = 6 for each site) by ASO. Six resident blacknose dace from each site were also analyzed, as well as 12 fish from the reference stream, to obtain baseline values. To help assess blacknose dace condition during confinement, mortality and growth in length and weight were measured in dace over the 12-week exposure periods.

Asiatic clams were obtained in May 1991 from Bull Run Creek in Union County, Tennessee, and in June 1992 and June 1993 from Little Sewee Creek in Meigs County, Tennessee. After the clams were held for 24 h in the laboratory in dechlorinated process water, they were put in polypropylene cages that were held securely at each monitoring site. One set of clams from the reference stream was frozen immediately for analysis as a control. Each cage held approximately 50 clams, each of which contained 0.5–2 g (wet wt) of soft tissue. The clams were suspended in the stream for four weeks, at which time they were removed, placed on ice in a locked cooler, transported to the laboratory and deposited in a locked freezer in Building 1505 at ORNL. After the clams were frozen, the shells were removed and the frozen soft tissue was placed in a 20-ml glass vial. Duplicate composite samples weighing approximately 20 g were taken for organic priority pollutant analyses. The samples were later delivered to the ORNL ASO for analysis.

Total mercury determinations were conducted by cold vapor atomic adsorption spectrometry as described in EPA (1991). Previous studies (Southworth et al. 1994, Bloom 1992, Huckabee et al. 1979) have shown that the mercury content in fish muscle generally is greater than 90% methylmercury (the most toxic mercury species). Other metals were determined using inductively-coupled argon plasma emission spectroscopy or inductively coupled plasma/mass spectroscopy (EPA 1980, 1991). Organic priority pollutants were

analyzed by procedure PPB 12/83 (EPA 1984) in which the homogenized sample is extracted in methylene chloride, cleaned up using column chromatography, and analyzed by gas chromatography and mass spectrometry (GC/MS) or by gas chromatography with electron capture detectors (GC/ECD). Determinations of chlorinated pesticides were made by capillary column GC/ECD using a method based on EPA procedure PPB 12/83 (EPA 1984), which involves homogenization of the sample in anhydrous sodium sulfate, extraction with methylene chloride, cleanup using column chromatography, and GC/ECD.

Quality assurance was maintained using a combination of duplicate analyses (submission of two subsamples from the same individual fish); the analysis of biological reference standards; and the analysis of uncontaminated fish. Recoveries of representative organics were verified by spiking uncontaminated fish or clam samples with known amounts and analyzing them.

Statistical evaluations of data were made using SAS procedures and software (SAS 1985a, 1985b) for ANOVA, Tukey's multiple comparison test, and calculation of the mean, SD, standard error (SE), and CV. Tests for homogeneity of variance among various data groups were conducted using Levene's test on untransformed and $\log_e$-transformed variables (Sokal and Rohlf 1981). Comparisons were based on untransformed data unless Levene's test indicated that transformation was needed to meet assumptions of homogeneous variances. Dunnett's test was used to compare means of various groups with controls (Zar 1984). All comparisons were conducted using $p = 0.05$.

4.1.3 Results and Discussion

4.1.3.1 Mercury in Mitchell Branch fish

Sunfish

Mercury was clearly elevated in fish from Mitchell Branch compared with fish from the reference stream, averaging two to three times higher than the reference stream in each year from 1991 to 1993 (Table 4.1; Table C.1). Concentrations in Mitchell Branch fish were below EPA's interim screening value of 0.6μg/g for mercury in fish tissue (EPA 1993) and the FDA action level of 1.0 μg/g (FDA 1984b). However, five fish from Mitchell Branch exceeded the conservative Preliminary Guidance Value (PGV) for mercury of 0.42 μg/g (Travis et al. 1986).

Mean mercury concentrations in Mitchell Branch redbreast sunfish have remained stable (0.25–0.33 μg/g; Table 4.1) over the 1990–93 period. Average mercury concentration were

Table 4.1. Mercury concentrations (in micrograms per gram, wet weight) in bluegill (*Lepomis macrochirus*) and redbreast sunfish (*L. auritus*) collected from Mitchell Branch, Poplar Creek and the Clinch River, 1987-1993

Site[a]	Species	Date	Hg mean ±1 SE	Range	EPA exceedences[b]
PCK 18.2	Bluegill	March 1993	0.06 ± 0.01	0.04–0.12	0/8
PCK 10.4	Bluegill	November 1987	0.10 ± 0.03	0.05–0.16	0/8
		November 1988	0.08 ± 0.03	0.02–0.24	0/8
		December 1989	0.18 ± 0.01	0.04–0.62	1/8
PCK 8.2	Bluegill	November 1987	0.43 ± 0.06	0.21–0.69	2/8
		November 1988	0.41 ± 0.05	0.23–0.59	0/8
		November 1989	0.57 ± 0.07	0.20–0.88	4/8
		March 1990	0.36 ± 0.05	0.14–0.65	1/8
		January 1991	0.40 ± 0.08	0.07–0.71	1/8
		July 1992	0.38 ± 0.04	0.24–0.49	0/8
		March 1993	0.34 ± 0.05	0.15–0.55	0/8
MIK 0.2	Redbreast	May 1987	0.17 ± 0.01	0.15–0.18	0/7
		March 1988	0.12 ± 0.01	0.09–0.14	0/5
		March 1989	0.19 ± 0.01	0.12–0.22	0/8
		January 1990	0.27 ± 0.03	0.11–0.39	0/8
		January 1991	0.32 ± 0.05	0.18–0.56	0/8
		January 1992	0.25 ± 0.03	0.19–0.48	0/8
		March 1993	0.33 ± 0.03	0.16–0.46	0/8
PCK 6.9	Bluegill	June 1987	0.42 ± 0.05	0.24–0.65	1/8
		November 1987	0.43 ± 0.05	0.26–0.66	1/8
		November 1988	0.29 ± 0.05	0.09–0.54	0/8
		December 1989	0.62 ± 0.04	0.38–0.75	6/8
		March 1990	0.43 ± 0.04	0.30–0.68	1/8
		January 1991	0.37 ± 0.06	0.15–0.67	1/8
		July 1992	0.40 ± 0.07	0.27–0.69	2/8
		March 1993	0.36 ± 0.03	0.29–0.48	0/8
PCK 1.6	Bluegill	December 1987	0.16 ± 0.04	0.06–0.35	0/8
		November 1988	0.17 ± 0.02	0.06–0.24	0/8
		March 1993	0.25 ± 0.05	0.07–0.44	0/8
CRK 15.0	Bluegill	November 1987	0.14 ± 0.03	0.06–0.31	0/8
		November 1988	0.14 ± 0.03	0.02–0.28	0/8
		February 1991	0.20 ± 0.03	0.11–0.42	0/8
		May 1992	0.20 ± 0.07	0.09–0.71	1/8
		March 1993	0.18 ± 0.03	0.09–0.36	0/8
Hinds Creek	Redbreast/ Bluegill	1991-1993	0.11 ± 0.01	0.04–0.24	0/21

[a] PCK = Poplar Creek kilometer; MIK = Mitchell Branch kilometer; CRK = Clinch River kilometer.

[b] Number of samples exceeding the EPA screening value for Hg of 0.6 ppm (EPA 1993, *Guidance for Assessing Chemical Contaminant Data for Use in Fish Advisories*, Vol. 1, Fish Sampling and Analysis, U.S. Environmental Protection Agency, Washington, D.C.) divided by the total number of samples.

lower in fish collected prior to 1990; however, collections from 1987 through 1989 were of substantially smaller fish, averaging 5 and 26 g in each year respectively. Accumulation of mercury in fish tends to increase with body size, thus the larger fish obtained since 1990 (average weight greater than 40 g in all years) would be expected to contain higher concentrations of mercury than the extremely small fish collected in the previous years. When linear regression analysis of mercury vs weight for previously collected MIK 0.2 sunfish was used to estimate the concentration of mercury in a hypothetical 70-g fish from Mitchell Branch, a value of 0.33 μg/g was obtained (Smith et al. 1994). This value is very close to the average concentration in current collections, suggesting that mercury contamination has not changed substantially in Mitchell Branch from 1987 to 1993.

Blacknose Dace

Although sunfish collected from Mitchell Branch contained elevated concentrations of mercury, the proximity and accessibility of Poplar Creek, in which sunfish contained similar concentrations of mercury, made it uncertain that Mitchell Branch was the source of mercury to those fish. Placement of uncontaminated, caged sunfish into Mitchell Branch was considered as a means of determining the extent of biologically-available mercury within the system. However, past attempts at caging sunfish in streams on the Oak Ridge Reservation yielded limited success: sunfish were highly territorial and did not tolerate confinement, and the relatively large cages were susceptibable to damage and wash-out during storms.

Although monitoring sediment or water in such streams may give insight into the contamination problem, these components may not be reliable predictors of the extent of mercury contamination in biota in the system, hence the need for biological monitoring (Rada and Findley 1986, Phillips et al. 1987, Loar 1989, Suchanek et al. 1993). Blacknose dace (*R. atratulus*), were used as bioindicators of mercury contamination in Mitchell Branch and other nearby streams during the summer/fall of 1992. This species was chosen because (1) blacknose dace are common inhabitants of small creeks in east Tennessee and were abundant at all monitoring sites, (2) blacknose dace are omnivorous and adaptable in a caged environment, (3) mercury concentrations in muscle tissue of "forage" species such as blacknose dace may be more comparable to "sport fish" muscle concentrations (best suited for human risk analysis) than other biota such as macroinvertebrates or periphyton and (4) caged

blacknose dace rapidly accumulated mercury in upper East Fork Poplar Creek in previous studies (Loar 1993).

The mercury results from the caged dace experiment were analyzed by combining all individual fish data obtained for each site [n = 6 at all sites except EFK 23.4 (n = 3)]. Cage-to-cage variation in mercury concentrations within each site was low (except at EFK 23.4 where variability could not be evaluated); there was no significant difference in mercury concentrations in dace between cages at each site. Survival of caged blacknose dace after a 12-week exposure was ≥ 75% at all but EFK 23.4 (25%) and the laboratory tank (58%). A significant decrease in mean condition factor (wt/lgth^3 x 100) occurred in caged dace after a 12-week exposure at all sites except EFK 23.4 (Table 4.2). Although the mean condition factor of resident dace from the uncontaminated reference stream (Hinds Creek) was also significantly lower after the 12-week period, the magnitude of the decrease at some sites suggests that food availability may have been limited in the caged treatments.

Mean mercury concentrations in caged blacknose dace after 12 weeks' exposure were low (< 0.20 μg/g) at all sites monitored; however, concentrations of mercury in dace from Mitchell Branch, Bear Creek, and EFPC significantly exceeded those in dace from Hinds Creek, the baseline reference stream (Table 4.2). Caged blacknose dace from MIK 0.6 and from EFK 23.4 were also significantly higher in mercury than caged blacknose dace from an unimpacted tributary in the same watershed (PHK 1.6).

Mean mercury concentrations in resident blacknose dace from all monitored sites, including the unimpacted tributary, were significantly higher than that in fish from the baseline reference stream (log-transformed data; Table 4.2). Mercury concentrations in resident blacknose dace showed a site-to-site pattern similar to that of caged blacknose dace, but concentrations were significantly higher in resident blacknose dace than caged blacknose dace at all sites. As was the case with caged blacknose dace, mean mercury concentrations in resident blacknose dace were highest in Mitchell Branch and EFPC, with little difference between concentrations in Bear Creek fish and those from Pinhook Branch. The similar site-to-site pattern between caged and resident blacknose dace suggests that resident represent mercury exposure at the collection site. The difference in accumulation between

Table 4.2. Mean mercury concentrations and condition factors in blacknose dace *(Rhinichthys atratulus)*

Site	Mercury concentrations (μg/g, wet wt)		Condition factors (wt/vol.)	
	Caged dace	Resident dace	Before cage placement	After 12-week exposure
EFK 23.4	0.189 ± 0.029* (3)	0.367 ±0.025* (6)	0.92 ± 0.02 (12)	0.95 ± 0.06 (3)
MIK 0.6	0.144 ± 0.024* (6)	0.224 ±0.022* (6)	0.99 ± 0.03 (12)	0.82 ± 0.02 (12)
BCK 12.0	0.107 ± 0.007* (6)	0.190 ±0.015* (6)	0.93 ± 0.02 (12)	0.67 ± 0.04 (10)
BCK 4.5	0.104 ± 0.008* (6)	0.180 ±0.031* (6)	1.02 ± 0.02 (12)	0.90 ± 0.02 (9)
PHK 1.6	0.098 ± 0.012 (6)	0.162 ±0.022* (6)	1.01 ±0.03 (12)	0.92 ± 0.02 (12)
Lab Tank	0.080 ± 0.008 (6)	---	1.01 ± 0.02 (12)	0.76 ± 0.02 (7)
Hinds Creek (reference stream)	---	0.071 ±0.006 (12)	1.02 ± 0.01[a] (82)	0.93 ± 0.01[b] (70)

Note: Values are mean ± SE (sample size in parenthesis). Mercury values with asterisks (*) are significantly higher ($\alpha = 0.05$) than the value for Hinds Creek dace. EFK - East Fork Poplar Creek kilometer, MIK = Mitchell Branch kilometer, BCK = Bear Creek kilometer, PHK = Pinhook Branch kilometer.

[a]Mean condition factor of all dace collected in May 1992 from Hinds Creek.

[b]Mean condition factor of uncaged dace (i.e. resident fish) collected in August 1992 from Hinds Creek.

caged and resident fish was an expected consequence of limited food availability in the caged treatments, differences in the type of food ingested, and the short duration of exposure relative to time needed to attain steady state.

Evidence from both caged and resident reblacknose dace suggests that Mitchell Branch water and/or sediments are sources of mercury contamination to resident aquatic life, although the degree of mercury contamination in dace is low relative to that ordinarily found in fish from the upper reaches of East Fork Poplar Creek. Based on the results of the dace experiment, it appears likely that mercury levels in sunfish collected from Mitchell Branch are representative of the amount of mercury exposure at the collection site.

4.1.3.2 Mercury in Poplar Creek fish

Mercury concentrations were measured in bluegill from several sites in Poplar Creek and the Clinch River to assess the relative importance of various streams (Bear Creek, EFPC, Mitchell Branch, and White Oak Creek) to the system. Results for the Poplar Creek area are shown in Table 4.1; results for the entire reservation in winter 1992/1993 are shown in Fig. 4.1.

Bluegill were collected from a site upstream of DOE impacts on Poplar Creek (PCK 18.2) in 1993 as a comparison to the level of mercury contamination in fish downstream. This reference site is further upstream than a site used previously (PCK 10.4) for this purpose, and should be far enough upstream to not be influenced by water level changes in Watts Barr Reservoir. Mercury concentrations in bluegill from this site were typical of fish from reference streams (Table 4.1), indicating that neither upstream sources of mercury in Poplar Creek nor movement of fish between sites were probable explanations for the high mercury levels at PCKs 8.2 and 6.9, located just upstream (PCK 8.2) and downstream (PCK 6.9) of Mitchell Branch.

The highest mean mercury concentrations in sunfish from Poplar Creek continues to be from PCKs 8.2 and 6.9 (Table 4.1). During the 1991–93 period, no fish from these sites exceeded the FDA action level of 1 μg/g; however, a small number of fish (4/48) exceeded EPA's screening value of 0.6 μg/g, and 19 of 48 fish collected from PCKs 8.2 and 6.9 exceeded the more conservative PGV of 0.42 μg/g (Travis et al. 1986; Table C.1). Mean mercury concentrations in fish from these two sites has changed little between 1987 and 1993; mean values were approximately 0.3 to 0.5 μg/g at both sites in most years. There was, again, very little difference in mean mercury concentrations between the two sites (less than 0.03 μg/g difference in each year from 1991 to 1993). No increase in mercury concentrations in fish in Poplar Creek could be attributed to inputs from Mitchell Branch.

The high mercury levels in fish from PCKs 8.2 and 6.9 are not readily explainable by a one-source-hypothesis, such as input from EFPC only. Levels of mercury in sunfish from EFPC decreased in proportion to dilution of Oak Ridge Y-12 Plant discharges at sites along the length of EFPC (Loar 1993), yet mercury levels in fish from PCKs 8.2 and 6.9 were similar to those found in lower EFPC, despite an almost four-fold dilution of EFPC upon entering Poplar Creek. One potential source of this mercury is Bear Creek. Mercury contamination is evident in fish from lower Bear Creek, but the extent of contamination in fish

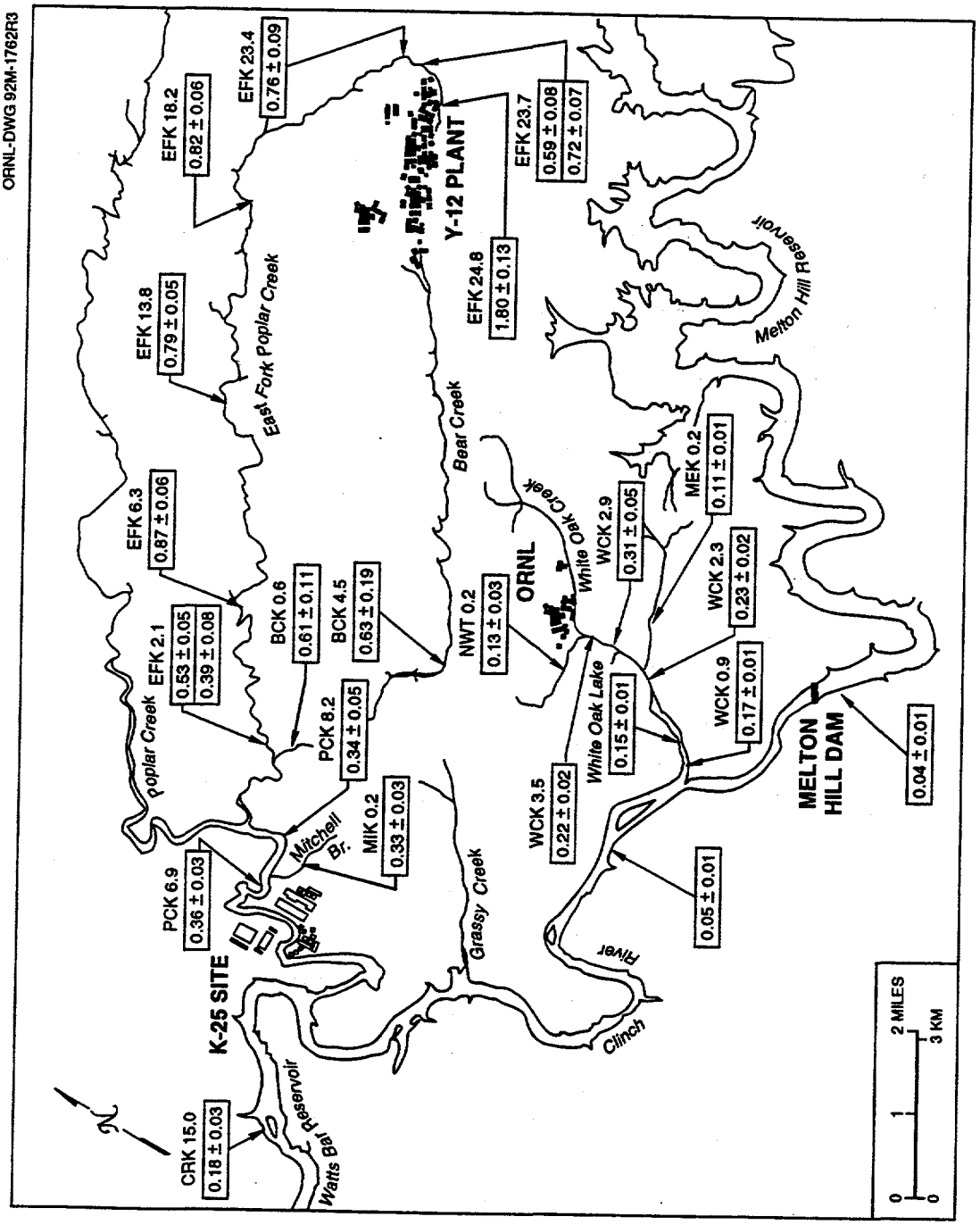

Fig. 4.1 Mean concentrations of mercury (μg/g wet wt., ± SE; n=8) in sunfish species (*Lepomis auritus, L. macrochirus, Ambloplites rupestris*) from waters on the DOE Oak Ridge Reservation.

does not appear to be large enough to explain the high levels seen in Poplar Creek fish (Fig. 4.1). It is possible that either changes occur in the environmental chemistry of mercury in lower Poplar Creek, enhancing the biological availability of water-borne or sediment-associated mercury, or that additional sources of mercury (e.g., Poplar Creek sediments) occur in the reach of Poplar Creek near the Oak Ridge K-25 Site.

Mercury contamination in fish at the most downstream site on Poplar Creek (PCK 1.6) was lower than at PCKs 6.9 or 8.2. Low concentrations were also found in fish in the Clinch River (CRK 15.0) downstream of the mouth of Poplar Creek. Mercury concentrations in fish at CRK 15.0 were elevated over concentrations in fish at sites on the Clinch River upstream of the mouth of Poplar Creek (Fig. 4.1). Mercury concentrations in sunfish from the Clinch River upstream of the Poplar Creek mouth were typical of those found in fish from the uncontaminated reference stream, Hinds Creek, and other uncontaminated streams in east Tennessee (Southworth et al. 1994). Although mercury contamination from Oak Ridge operations within the Poplar Creek watershed is detectable in Clinch River fish downstream of the mouth of Poplar Creek, the degree of contamination, in comparison with human health threshold limits, appears to be minimal.

In 1992/1993, the level of mercury contamination in bluegill from Poplar Creek sites near the K-25 Site was generally 20–30% of the level in fish from the middle to upper reaches of EFPC, and about half the level in Bear Creek fish (Loar 1993; Fig. 4.1). The mean Hg concentrations in fish at these two sites were comparable to those from Mitchell Branch, lowermost EFPC (EFK 2.1), and the most mercury-contaminated site on White Oak Creek at kilometer (WCK) 2.9. Mean mercury concentrations in fish from lowermost Poplar Creek (PCK 1.6) were similar to those of fish from the other sites on White Oak Creek proper [WCK 3.5, WCK 2.3, White Oak Lake (WOL), and WCK 0.9]. The mean mercury concentration in bluegill from the Clinch River (CRK 15.0) was similar to fish concentrations in WOL, White Oak Creek Embayment (WCK 0.9), and in two locations on tributaries of White Oak Creek: Melton Branch kilometer (MEK) 0.2 and Northwest Tributary kilometer (NTK) 0.2.

4.1.3.3 Metals screening

Despite abnormally high levels of some metals in Mitchell Branch water (Smith et al. 1993) and sediment (Ashwood et al. 1986), concentrations of metals (other than mercury) in

Mitchell Branch fish in all three sampling years (1991–1993) were similar to those found in fish from the reference stream (Table 4.3; Table C.1). In 1991 and 1992, only selenium, thallium, and zinc were found above the analytical detection limit. The selenium and thallium detections were from only one fish, and at concentrations only slightly higher than the detection limit. Improved detection limits in the 1993 analyses allowed explicit comparison of concentrations of additional metals. Mean concentrations of chromium, copper, nickel, selenium, and zinc in Mitchell Branch sunfish were quite similar to those observed in Hinds Creek bluegill in 1993 by TVA at one of their reference sites (Melton Hill Reservoir) (TVA 1985, 1986) and in fish in the National Contaminant Biomonitoring Program (Lowe et al. 1985).

Mean metal concentrations (other than mercury) in sunfish collected from sites in the vicinity of the K-25 Plant in 1993 were similar to those found in fish from Poplar Creek upstream of any DOE facilities (PCK 18.2) and to those in fish from Hinds Creek (HCK 12.7), the reference stream (Table 4.4). As with fish from Mitchell Branch, the concentrations of metals (Tables 4.3 and 4.4) in fish from near the K-25 Site were similar to other measures of local and national background concentrations (TVA 1985, 1986; Lowe et al. 1985). Mercury screening of fish from the three ponded tributaries of Poplar Creek (K-1007-P1, K-1007-P5, and K-710A) was conducted for the first time in spring 1993. Mercury concentrations in sunfish from the two K-1007 ponds were very similar to concentrations in fish from reference sites (PCK 18.2 and HCK 12.7). Although the mean mercury concentration in fish from the K-710A pond (0.12 μg/g) was about two times higher than found in fish from reference sites in the 1993 sampling year, past monitoring has shown that this level of mercury is within the range of reference site values (Ashwood et al. 1993, Southworth et al. in press).

A comparison of metal concentrations in fish from all K-25 sites with toxicological benchmarks or screening values indicates that fish from these sites pose little concern for use as human food (Tables 4.3, 4.4). The mean concentrations of all metals except arsenic and beryllium were at or below concentrations used by EPA and others to screen for levels of contamination that pose no threat for human consumption. EPA and PGV screening values for arsenic and beryllium are set below routine levels of detection due to the potential carcinogenity of these two contaminants; only arsenic was detected in these samples. Arsenic has a screening level lower than the concentration typical of fish from uncontaminated sites,

Table 4.3. Mean (± SE in parentheses) metal concentrations in redbreast sunfish (*Lepomis auritus*) from Mitchell Branch, 1991–1993

Reference and guidance values for these metals are given for comparison

	Mitchell Branch							
Metal	1991	1992	1993	Hinds Creek[a]	TVA[b]	USFWS[c]	PGV[d]	EPA[e]
Arsenic	<0.05	<0.05	0.05[f] (0.01)	<0.05	<0.03	0.16	0.0007	0.006
Beryllium	<0.003	<0.003	<0.003	<0.003	<1	—	0.004	0.0025
Cadmium	<0.34	<0.37	<0.02	<0.02	0.007 (0.003)	0.04	1.0	10.8
Chromium	<0.86	<0.93	0.08[g] (0.02)	<0.05	0.06	—	1.8	10,800
Copper	<0.86	<0.93	0.24 (0.01)	0.22 (0.01)	0.4 (0.3)	0.86	36	—
Lead	<0.86	<0.93	<0.02	<0.02	0.21 (0.33)	0.19	1.8	—
Mercury	0.32 (0.05)	0.25 (0.03)	0.33 (0.03)	0.06 (0.01)	<0.1	0.11	0.42	1.0
Nickel	<0.86	<0.93	0.06[g] (0.01)	<0.05	<0.1	—	5.2	2.15
Selenium	0.67[f] (0.08)	<0.93	0.41 (0.05)	0.25 (0.01)	0.7 (0.2)	0.46	12	5.4
Silver	<0.34	<0.37	<0.04	<0.04	<0.3	—	0.29	2.48
Thallium	0.02[f]	<0.02	<0.02	<0.02	<1.0	—	0.25	5.71
Uranium	<0.02	<0.003	<0.003	<0.003	—	—	—	—
Zinc	5.8 (0.22)	5.7 (0.19)	5.2 (0.2)	5.8 (0.07)	8.4 (2.6)	25.6	180	—

Note: Values expressed as micrograms per pram, wet weight. The mean for nonmercurous metals is based on four individual fish except at HCK 12.7 ($n = 3$). For mercury, $n = 8$ for Mitchell Branch in each year, $n = 6$ for Hinds Creek. See Chapter 8 for full citations of references listed below.

[a]Reference stream located in Anderson County, Tennessee.

[b]Mean concentrations of metals in fish collected by TVA from Melton Hill Reservoir ($n = 9$ for As, Cd, Cr, Ni, Ag,; $n = 2$ for others) TVA (1985).

[c]Mean concentrations of metals in fish collected for the National Contaminant Biomonitoring Program (NCBP) (Lowe et al. 1985).

[d]Preliminary Guidance Values used to screen for levels of contamination that pose no threat for human consumption (from Hoffman et al. 1984; Travis et al. 1986).

[e]EPA fish tissue criteria in 1990 (EPA 1990).

[f]One of four samples exceeded the detection limit.

[g]Three of four samples exceeded the detection limit.

Table 4.4. Mean (± SE in parentheses) metal concentrations in sunfish collected in the vicinity of the K-25 Site in spring 1993

Measurements expressed in micrograms per gram, wet weight

	Site									
	Tributaries				Poplar Creek				Clinch R[a]	Refer. Site[b]
Metal	K1007-P1	K1007-P5	K710A	MIK 0.2	PCK 1.6	PCK 6.9	PCK 8.2	PCK18.2	CRK15.0	HCK 12.7
Arsenic	0.06[c] (0.01)	<0.05	0.06 (0.01)	0.05[d] (0.01)	0.07[e] (0.01)	--	--	<0.05	--	<0.05
Beryllium	<0.003	<0.003	<0.003	<0.003	<0.003	--	--	<0.003	--	<0.003
Cadmium	<0.02	<0.02	0.02[d] (0.01)	<0.02	<0.02	--	--	<0.02	--	<0.02
Chromium	<0.05	0.05[d] (0.01)	<0.05	0.08[e] (0.02)	<0.07	--	--	<0.05	--	<0.05
Copper	0.17 (0.01)	0.17 (0.01)	0.19 (0.03)	0.24 (0.01)	0.20 (0.01)	--	--	0.16 (0.03)	--	0.22 (0.01)
Lead	<0.02	<0.02	<0.02	<0.02	<0.02	--	--	<0.02	--	<0.02
Mercury	0.09 (0.01)	0.05 (0.01)	0.12 (0.01)	0.33 (0.03)	0.25 (0.05)	0.36 (0.03)	0.34 (0.05)	0.06 (0.01)	0.18 (0.03)	0.06 (0.01)
Nickel	<0.05	<0.05	<0.05	0.06[e] (0.01)	0.06[d] (0.01)	--	--	<0.05	--	<0.05
Selenium	0.35 (0.01)	0.30 (0.03)	0.54 (0.04)	0.41 (0.05)	0.45 (0.03)	--	--	0.38 (0.03)	--	0.25 (0.01)
Silver	<0.04	<0.04	<0.04	<0.04	<0.04	--	--	<0.04	--	<0.04
Thallium	<0.02	<0.02	<0.02	<0.02	<0.02	--	--	<0.02	--	<0.02
Uranium	<0.003	<0.003	<0.003	<0.003	<0.003	--	--	<0.003	--	<0.003
Zinc	6.7 (0.08)	5.6 (0.03)	4.9 (0.4)	5.2 (0.2)	5.0 (0.7)	--	--	5.1 (0.7)	--	4.6 (0.2)

Note: Sunfish were bluegill (*Lepomis macrochirus*) at all sites except MIK 0.2 [redbreast sunfish (*Lepomis auritus*) were used at this site]. At each site where metals were analyzed, four individual fish comprise the mean, except at HCK 12.7 (m = 3). For mercury, eight individual fish were collected from each site except HCK 12.7 (n = 6).
MIK = Mitchell Branch kilometer, PCK = Poplar Creek kilometer, CRK = Clinch River kilometer and HCK = Hinds Creek kilometer. Numbered designations refer to the kilometer distance above the stream mouth (e.g. site MIK 0.2 is on Mitchell Branch 0.2 km upstream of the mouth).

[a]Clinch River

[b]Reference Site

[c]Two of four samples exceeded the detection limit. For samples reported below the detection limit, the detection limit value was used in calculating the mean.

[d]One of four samples exceeded the detection limit.

[e]Three of four samples exceeded the detection limit.

and also lower than the concentrations commonly found in other foodstuffs, such as fruits and vegetables (Bowen 1979).

4.1.3.4 PCBs in Mitchell Branch fish

Polychlorinated biphenyl concentrations were clearly elevated in fish from Mitchell Branch relative to fish from the reference stream, averaging at least an order of magnitude higher than the reference stream in each year from 1991 through 1993 (Table 4.5; Table C.2). A small number of individuals (3 of 40 redbreast sunfish) collected since 1989 contained PCB concentrations that exceeded the FDA tolerance level of 2.0 μg/g (FDA 1984a). The level of PCB contamination in Mitchell Branch fish is high for a relatively short-lived, low-lipid species such as redbreast sunfish; longer-lived older, fattier species such as channel catfish (*Ictaurus punctatus*), carp (*Cyprinus carpio*), or large-mouth bass (*Micropterus salmoides*), if present, would probably contain much higher levels. In general, the degree of PCB contamination in redbreast sunfish from Mitchell Branch was similar to the level of contamination found at the most highly contaminated sites on EFPC (EFKs 23.4, 23.7, 23.8) and Bear Creek (BCK 4.5) (Fig. 4.2).

Mean PCB concentrations in Mitchell Branch redbreast did not exhibit an increasing or decreasing trend over the 1989–1993 period. Average concentrations were highly variable temporally, ranging as low as 0.58 μg/g in 1991 to as high as 1.61 μg/g in 1989. The significance of Mitchell Branch as a source of PCB contamination is evident when comparing the levels of contamination in fish collected in Poplar Creek upstream and downstream of the mouth of Mitchell Branch. Mean PCB concentrations in Mitchell Branch redbreast sunfish were typically an order of magnitude higher than mean PCB concentrations in bluegill at PCKs 6.9 and 8.2 in each year (Table 4.5), despite the close proximity of Poplar Creek sites to Mitchell Branch. These results suggest that Mitchell Branch is a substantial source of PCBs to its resident biota, and Mitchell Branch fish were not likely to have obtained such high PCB concentrations elsewhere.

4.1.3.5 PCBs in Poplar Creek fish

PCB concentrations were measured in bluegill from several sites in Poplar Creek to assess the relative importance of various inputs to the system. PCB results for the Poplar Creek area are shown in Table 4.5; results for the entire reservation are shown in

Table 4.5. Polychlorinated biphenyl concentrations in redbreast sunfish (*Lepomis auritus*) and bluegill (*L. macrochirus*) collected from Mitchell Branch and Poplar Creek sites, 1987–1993

Site[a]	Species	Date	mean ± SE (Arochlor-1254 + 1260, µg/g, wet wt.)	Range (Arochlor-1254 + 1260, µg/g, wet wt.)
PCK 18.2	Bluegill	Mar. 1993	0.08 ± 0.02	0.03–0.14
PCK 10.4	Bluegill	Nov. 1987	0.06 ± 0.01	0.02–0.11
		Nov. 1988	0.05 ± 0.03	0.01–0.26
PCK 8.2	Bluegill	Nov. 1987	0.20 ± 0.03	0.11–0.36
		Nov. 1988	0.31 ± 0.07	0.08–0.59
		Mar. 1990	0.07 ± 0.02	0.02–0.15
		Feb. 1991	0.05 ± 0.02	0.01–0.10
		Jul. 1992	0.03 ± 0.01	0.01–0.05
		Mar. 1993	0.03 ± 0.01	0.01–0.05
MIK 0.2	Redbreast	Mar. 1989	1.61 ± 0.32	0.50–3.27
		Jan. 1990	0.80 ± 0.14	0.23–1.55
		Jan. 1991	0.58 ± 0.15	0.03–1.28
		Jan. 1992	1.34 ± 0.32	0.40–3.05
		Mar. 1993	0.76 ± 0.15	0.10–1.36
PCK 6.9	Bluegill	Jun. 1987	0.22 ± 0.06	0.05–0.53
		Nov. 1987	0.26 ± 0.04	0.14–0.47
		Nov. 1988	0.21 ± 0.05	0.08–0.40
		Mar. 1990	0.12 ± 0.01	0.07–0.23
		Feb. 1991	0.05 ± 0.02	0.01–0.10
		Jul. 1992	0.03 ± 0.01	0.01–0.07
		Mar. 1993	0.17 ± 0.09	0.03–0.81
PCK 1.6	Bluegill	Dec. 1987	0.17 ± 0.03	0.10–0.37
		Nov. 1988	0.19 ± 0.04	0.06–0.39
		Mar. 1993	0.11 ± 0.02	0.02–0.22

Note: $n = 8$ fish per site per year. The mean polychlorinated biphenyl (PCB) concentration in reference stream (Hinds Creek) sunfish, 1991–1992, was 0.03 ± 0.01 ($n = 40$). In 1993, the mean PCB concentration in Hinds Creek sunfish was higher, averaging 0.11 ± 0.04 ($n = 4$).

[a]PCK = Poplar Creek kilometer, MIK = Mitchell Branch kilometer.

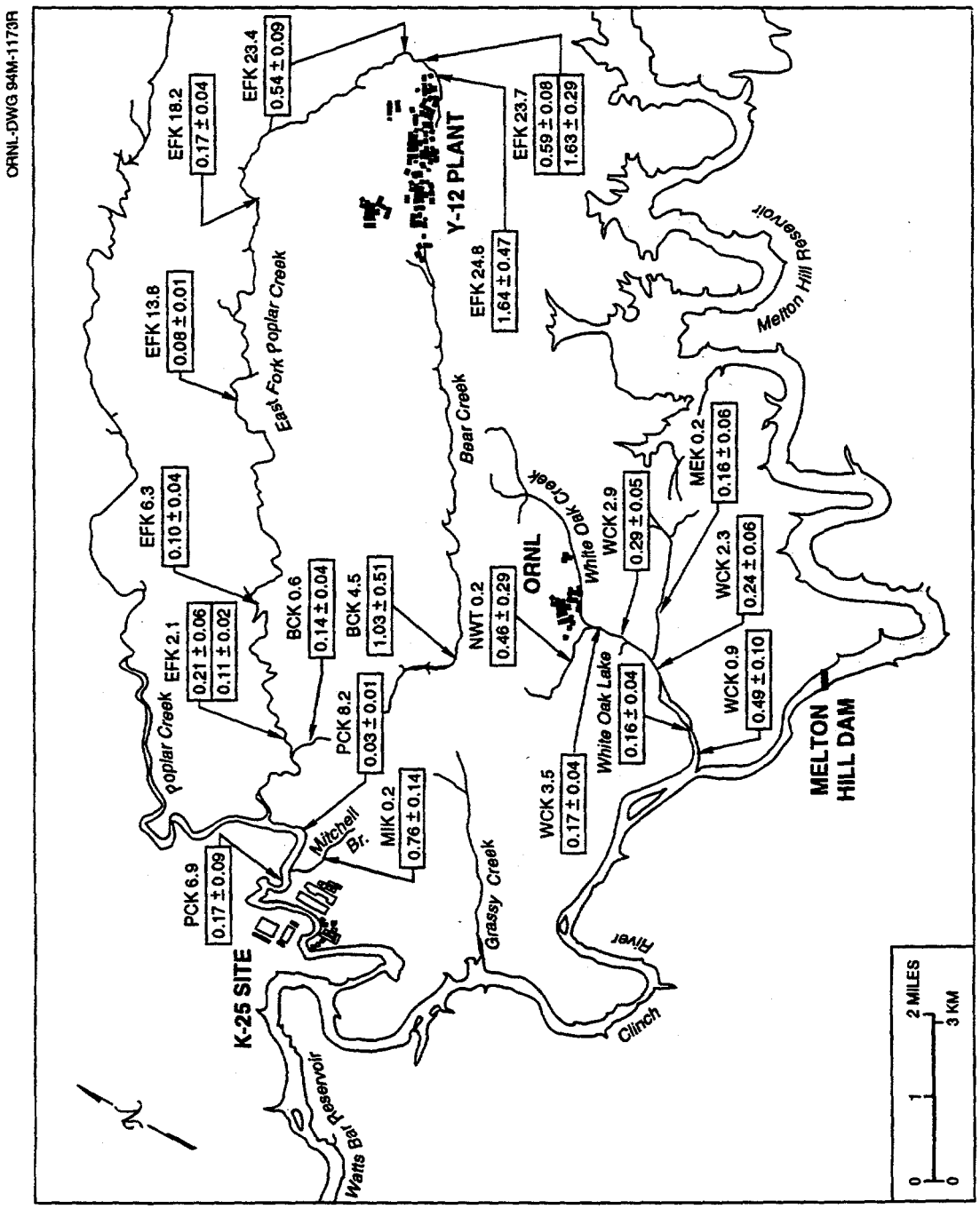

Fig. 4.2 Mean concentrations of PCBs (μg/g wet wt., ± SE; *n*=8) in sunfish species (*Lepomis auritus, L. macrochirus, Ambloplites rupestris*) from waters on the Department of Energy Oak Ridge Reservation. MIK = Mitchell Branch kilometer, PCK = Poplar Creek kilometer, EFK = East Fork Poplar Creek kilometer, BCK = Bear Creek kilometer, WCK = White Oak Creek kilometer, MEK = Melton Branch kilometer, NWT = Northwest Tributary, ORNL = Oak Ridge National Laboratory.

During the 1991–1992 period, PCB concentrations were near background in bluegill from PCK 6.9 and PCK 8.2 (Table 4.5). Prior to this period, PCB concentrations in Poplar Creek did not decline proportionally to the dilution of EFPC in Poplar Creek, but rather remained at levels typical of or higher than those of lower EFPC (Southworth 1990). Dilution or migration appeared to be more of a factor in 1991 and 1992, when PCB levels were substantially lower in Poplar Creek fish than in fish from the major PCB-contaminated tributaries to Poplar Creek (Mitchell Branch, Bear Creek, and EFPC).

In 1993, two additional sites on Poplar Creek were sampled, PCK 18.2 (upstream of all DOE impacts), and PCK 1.6 (downstream of all major K-25 Site discharges, including the 1007 ponds—see next section). The mean PCB concentration in PCK 18.2 fish was similar to reference stream concentrations reported in 1993, although this level (0.08 μg/g) is slightly higher than typically observed in reference fish in past years (0.03 μg/g; Table 4.5). Only the mean concentration in fish from PCK 6.9 (downstream of Mitchell Branch) was substantially higher than the PCK 18.2 reference site. Average PCB concentrations in fish at the two sites downstream of major K-25 Site inputs (PCKs 6.9 and 1.6) were higher (0.17 and 0.11 at PCK 6.9 and PCK 1.6 respectively) than upstream of the K-25 Site (PCK 8.2); however, the mean PCB concentration in 1993 at PCK 8.2 was anomalously low (0.03 μg/g). As has been the case previously (Smith et al. 1994), no relationship was obvious between PCB concentrations in fish in Poplar Creek and proximity to PCB-contaminated tributaries such as Mitchell Branch and K-1007 ponds outfall. Overall, the effect of the PCB-contaminated inputs from the K-25 Site to Poplar Creek appears to be undetectable in Poplar Creek sunfish.

In 1992–1993, the degree of PCB contamination in bluegill from the Poplar Creek sites near the Oak Ridge K-25 Site (PCKs 8.2, 6.9, and 1.6) was relatively low compared with other sites on the reservation (Fig. 4.2). The level of PCB contamination in Poplar Creek bluegill was most similar to levels in fish from lowermost EFPC, in rockbass from lower Bear Creek, and in fish from sites in the White Oak Creek drainage (MEK 0.2, WCK 3.5, and WOL).

Channel catfish have been collected annually since 1988 at PCK 6.9 and CRK 15.0 as part of an evaluation of the importance of inputs from DOE sources in contributing to PCB contamination in Watts Bar Reservoir. Results of this study are given in detail in Ashwood et al. 1994 and are only summarized here. In 1991 and 1992, PCB concentrations in catfish were, on average, higher at CRK 15.0 than in Poplar Creek—a change from previous

years and in 1993 (Table 4.6). Mean PCB concentrations in catfish at sites in Poplar Creek (PCK 6.9), the Clinch River near ORNL (CRK 32.2), and the K-25 Site (CRK 15.0) and did not differ significantly (p>0.05, ANOVA) in 1993. The results are consistent with the pattern seen in previous years. It is likely that movements of channel catfish and forage fish act to spread PCB contamination away from point sources; this results in a rather uniform distribution of PCB contamination in catfish throughout Watts Bar Reservoir. The mean PCB concentrations in channel catfish continue to justify the precautionary fish consumption advisory issued by the TDEC.

4.1.3.6 PCB Screening in pond fish

PCB screening of largemouth bass (*Micropterus salmoides*) was conducted in March and April 1993 from three ponded tributaries of Poplar Creek (K-1007-P1, K-1007-P5, and K-710A ponds; Fig. 2.1). Largemouth bass were chosen for PCB screening because they likely represent the worst-case level of PCB contamination in fish from the ponds. The mean PCB concentration (Arochlor 1254 + 1260) of four individually analyzed bass from the K-1007-P1 pond was 16.1 ± 9.0 μg/g, wet wt (mean ± SE) (Table 4.7). The high variability about the mean was due in large part to one very highly contaminated fish (42.9 μg/g). An independent reanalysis of archived muscle tissue from the same fish by Southwest Research Institute in San Antonio, Texas, confirmed the presence of high PCB concentrations. The mean PCB concentration for K-1007-P1 bass upon reanalysis (which included detectable concentrations of Arochlor-1248 as well as Arochlor-1254 and 1260) was 33.7 ± 5.6 SE. The mean PCB concentration in K-1007-P1 largemouth bass was higher than previously found in fish from the most PCB-contaminated sites on the ORR and is substantially higher than the FDA 2-μg/g tolerance level for PCBs of (FDA 1984a).

PCB concentrations in fish from the K-1007-P5 pond (located upstream of the P1 pond) were far lower (Table 4.7), suggesting that the source(s) to the P1 pond is not from the P5 vicinity. The mean PCB concentration of 0.03 ± 0.01 μg/g in four bluegill (*Lepomis macrochirus*) from the P5 pond was near background levels, and PCB concentrations in the two largemouth bass collected from the P5 pond were 0.13 μg/g and 1.21 μg/g. The mean PCB concentration in four largemouth bass from the K-710A pond was 0.36 ± 0.13 μg/g. The culvert at the downstream end of K-710A pond does not appear to be an effective barrier to water and/or fish exchanges, since water levels in the pond change with water level changes in

Table 4.6. Average concentrations of polychlorinated biphenyls and fraction of fish exceeding the FDA limit, for channel catfish from White Oak Creek embayment, the Clinch River, lower Poplar Creek (PCK), and Melton Hill Reservoir, 1986–1993

Measured in micrograms per gram, wet weight

	PCBs								Fraction over FDA limit							
Site	1986	1987	1988	1989	1990	1991	1992	1993	1986	1987	1988	1989	1990	1991	1992	1993
WCK 0.3	1.30	1.59	0.96	1.54	3.56	3.60	3.29	8.40	3/12	2/8	2/8	4/8	4/8	6/8	5/8	4/4
CRK 32.2	1.01	1.61	0.58	1.20	0.31	1.38	0.36	0.67	0/8	2/8	1/8	1/8	0/8	1/8	0/8	0/8
MHR	0.46	0.81	0.52	0.28	0.41	0.29	0.34	0.62	0/6	1/7	0/10	0/8	0/8	0/8	0/8	0/8
PCK 6.9	NS	NS	0.71	1.07	0.92	0.68	0.54	0.92	NS	NS	0/8	1/8	1/8	0/8	0/8	0/8
CRK 15.0	NS	NS	0.50	0.79	0.88	1.08	1.27	0.63	NS	NS	0/9	1/8	1/8	1/8	2/8	0/8

Note: NS = not sampled. WOK = White Oak Creek kilometer, CRK = Clinch River kilometer, MHR = Melton Hill Reservoir, PCK = Poplar Creek kilometer.

Table 4.7. Polychlorinated biphenyl concentrations (μg/g, wet wt.) in fish from ponds near the K-25 Site in spring 1993

Site	Species	Mean ± S.E.	Range	FDA[a] exceedences *n*
K-1007-P5	Bluegill	0.03 ± 0.01	0.01–0.04	0/4
	Largemouth bass	0.67 ± 0.54	0.13–1.21	0/2
K-1007-P1	Largemouth bass	16.1 ± 9.0	4.8–42.9	4/4
(Independent Reanalysis)		33.7 ± 5.6	20.5–47.8	4/4
K-710A	Largemouth bass	0.36 ± 0.13	0.14–0.74	0/4

[a]Number of samples exceeding the Food and Drug Administration threshold limit for polychlorinated biphenyls of 2.0 μg/g divided by the total number of samples (*n*).

Poplar Creek. Bass movement into the pond from the PCB-contaminated Poplar Creek seems likely.

The 1993 results of PCB tracking studies using bluegill in Poplar Creek suggest that the K-1007-P1 outfall to Poplar Creek is no greater a contributor of PCBs to fish in Poplar Creek than any of the upstream DOE inputs (see previous section). The mean PCB concentration in bluegill collected downstream of the K-1007-P1 discharge to Poplar Creek (PCK 1.6) was no higher than the mean PCB concentration in bluegill upstream of the discharge (PCK 6.9) (Table 4.5). Similar results were evident in bluegill collected in 1987 and 1988 (Table 4.5). Interpretation of such patterns is complicated by the unusual hydrodynamics of lower Poplar Creek (upstream hydroelectric generation causes daily flow reversals and flushing with Clinch River water) and the similarity of the lower creek to a reservoir, rather than a stream, habitat. If sunfish in lower Poplar Creek range more widely than typical stream sunfish, that, in combination with the complex hydrodynamic mixing, may tend to make the PCB contamination pattern in sunfish relatively homogeneous throughout the embayment, obfuscating any relationship to specific point sources.

4.1.3.7 Organics in caged clams

Caged clams held in Mitchell Branch for 4 weeks accumulated substantial concentrations of PCBs in all exposure periods from 1991 to 1993 (Table 4.8). Mean concentrations ranged from 1.4 μg/g in 1991 to 0.35 μg/g in 1993. Previous clam exposures in Mitchell Branch also resulted in substantial PCB accumulation (Smith et al. 1993, 1994). The degree of PCB

accumulation noted in clams was consistent with the high concentrations of PCBs found in redbreast sunfish over this period (Table 4.5).

PCB concentrations in clams decreased over the 3-year period; however, a similar decrease was not apparent in sunfish (Tables 4.5 and 4.8). PCB concentrations in sunfish represent a time integrated exposure of several months to more than a year, while the introduced clams only monitor a 1-month period. Concentrations in fish thus provide a more reliable indicator of temporal trends. The data collected to date show that Mitchell Branch contains significant PCB contamination, that is not rapidly increasing or decreasing.

Screening for organic contamination in clams placed at two pond sites (K-1007-P1 and K710A) and at lower Poplar Creek (PCK 1.6) detected elevated concentrations of PCBs. The high PCB concentrations in clams at K-1007-P1 (0.49 μg/g) corroborates the conclusions based on data from bass at this site. The average PCB concentration in clams from K-710A (0.26 μg/g) indicates that the pond is PCB contaminated; however, the free exchange of Poplar Creek water that occurs with water level changes in Watts Bar reservoir makes decoupling the two systems difficult. PCB contamination in the pond may be due to retention and resuspension of Poplar Creek sediment. The mean PCB concentration in four very large bass from 710A (a relatively low 0.36 μg/g) likely represents the maximum level of PCBs in fish from this system, and further monitoring of PCBs in this pond does not appear to be warranted. A slightly elevated average concentration of PCBs (0.09 μg/g) was found in clams placed at PCK 1.6 in comparison to the concentration in clams placed upstream of DOE discharges (PCK 18.2). This was consistent with fish data that indicated the presence of PCB contamination in Poplar Creek downstream from the mouth of EFPC.

Although the accumulation of PCBs by caged clams can be influenced by the presence of toxicants such as residual chlorine and by the abundance of particulate food in the water, the concentrations in clams can be viewed as providing an index of the concentration of waterborne PCBs (which is generally below the detection limit of analytical procedures routinely used to monitor PCBs in water at the DOE Oak Ridge facilities). A rough comparison of the mass flux of PCBs from Mitchell Branch and the K-1007-P1 discharges to the mass flux from upper EFPC can be made by comparing the products of PCB concentrations in clams times average flow at the site. Although only data from upper EFPC is available, the mass flux of PCBs in lower EFPC (with much greater flow) would be expected to be even greater than upper EFPC (despite expected lower PCB concentrations in

Table 4.8. Mean concentrations of organic compounds detected in duplicate samples of caged clams (*Corbicula fluminea*) at sites near the K-25 Site, 1991-1993

Measurements in micrograms per gram, wet weight

	Analytes[a]						
Sites by year	Total PCBs	DDT	Total chlordane	aldrin	Benzoic acid	Di-n-octyl phthalate	Bis(2-ethylhexyl) phthalate
1991							
MIK 0.2[b]	1.4	0.1	0.01	0.01	--	--	--
1992							
MIK 0.2	0.52	0.05	--	0.02	4.8[c]	--	--
1993							
MIK 0.2	0.35	--	0.006	--	--	--	20.4[d]
K-1007-P1	0.49	--	--	--	--	--	--
K-710A	0.26	--	0.004	--	--	2.0[d]	--
PCK 1.6	0.09	--	0.003	--	--	--	--
PCK 18.2	<0.04	--	0.002	--	--	--	5.4[d]

Note: Organic compounds not noted were below detection limits (see Appendix). DDT - dichlorodiphenyl-trichloroethane; PCB = polychlorinated biphenyls; MIK = Mitchell Branch kilometer; PCK = Poplar Creek kilometer.

[a]In most cases, mean values were calculated using **at least one** estimated concentration. Exceptions were values cited at MIK 0.2 in 1991 and the two bis(2-ethylhexyl) phthalate detections. Estimated concentrations were reported by the analytical laboratory when compounds were detected, but below the range of reliable quantitation. n = 2 clam composite samples for each site in each year unless otherwise noted.

[b]Only one clam composite sample was analyzed in 1991 at this site.

[c]Benzoic acid was detected in reference samples with this submission. Benzoic acid concentrations are likely to be an artifact of the analytical procedures.

[d]Compound was not detected in one of two duplicate samples. The detection limit value was used to calculate the mean.

clams downstream). A mass flux comparison yields values of 0.29 (0.49 μg/g × 26 L/s) for K-1007-P1, 0.36 (0.35 μg/g × 46 L/s) for Mitchell Branch, and 3.45 (0.69 μg/g × 219 L/s) for EFPC at the Y-12 Plant. PCB inputs to Poplar Creek from Mitchell Branch and the K-1007-P1 ponds, while not inconsequential, appear to be a small percentage of the total inputs from sources upstream from the K-25 Site.

Very low concentrations of other organic compounds were detected in clams placed in Poplar Creek and associated tributaries near the K-25 Site (Table C.3). With the exception of the K-1007-P1 site, chlordane was detected at all sites, including the reference site, but at concentrations far below EPA screening levels (EPA 1990) and near the limit of routine analytical detection. Two other pesticides, dichlorodiphenyl-trichloroethane (DDT) and aldrin, were also detected at levels near the detection limit in Mitchell Branch in 1991 and 1992 (Table 4.8). Aldrin was detected previously in Mitchell Branch (Smith et al. 1994). Benzoic acid, a common artifact of the analytical procedures, was detected in Mitchell Branch and in reference samples.

Two phthalate compounds were detected in clams from Mitchell Branch and the K-710A pond, but the detections should be considered non-confirmatory. Di-*N*-octyl phthalate was reported below the range of reliable quantitation in one of two clam samples from K-710A. Bis (2-ethylhexyl) phthalate was detected in one of two duplicate clam samples from Mitchell Branch, and in one of two duplicate samples from a reference site (PCK 18.2). Phthalates have been detected in clam samples previously placed in Mitchell Branch (Smith et al. 1993); however, these data must be regarded with caution since phthalates are common laboratory and sample contaminants.

4.1.4 Conclusions

Mercury and PCBs are the only contaminants that accumulate to above background levels in fish and clams at sites near the K-25 Site. Mercury bioaccumulation was observed in fish in Mitchell Branch and nearby sites on Poplar Creek. Mercury concentrations did not exceed the FDA limit, but some fish did exceed more conservative screening thresholds (EPA 1993). Evidence from the caged and resident blacknose dace experiment suggests that the mercury contamination in Mitchell Branch fish arises from a source within that watershed, rather than being a consequence of contaminated fish immigrating from nearby Poplar Creek. Based on the results of the blacknose dace experiment, it appears likely that the sunfish collected from

Mitchell Branch are representative of the degree of contaminant exposure at the collection site.

PCB concentrations were high in fish from two localized aquatic habitats at the K-25 Site (Mitchell Branch, K-1007-P1), but were much lower in fish from Poplar Creek in the vicinity of discharges from these sites. Some fish in Mitchell Branch contained PCB concentrations in excess of the 2-μg/g FDA action level. PCB screening of largemouth bass in the ponded tributaries at the K-25 Site revealed significant contamination in fish from the K-1007-P1 pond. Mitchell Branch and K-1007-P1 undoubtedly are sources of PCB contamination to Poplar Creek, but their significance as a source of PCB contamination to the biota in Poplar Creek and the Clinch River relative to other known sources (EFPC, White Oak Creek, Bear Creek, and Poplar Creek sediment) cannot be ascertained from a comparison of concentrations of PCBs in different species of fish from different habitats. Channel catfish from Poplar Creek continue to contain PCB concentrations similar to other sites in the Clinch River/Watts Bar system. PCB concentrations in caged clams that sample waterborne PCBs for a fixed time interval can be used to infer relative fluxes of PCBs from specific sources. Results of such a comparison indicate that chronic PCB fluxes to Poplar Creek from known K-25 Site sources are significant but are probably a small percentage of the total input from sources upstream from the K-25 Site.

Metals screening of fish from lower Poplar Creek, Mitchell Branch, and the pond sites revealed little evidence of contamination. Concentrations were either below detection limits or not substantially higher than reference values. Screening for organics in caged clams detected PCBs at high concentrations in Mitchell Branch and the K-1007-P1 pond consistent with the results of PCB monitoring in fish at those sites. Other organic compounds were detected at very low concentrations; most values were estimated concentrations below the quantification limit of the analytical method. An exception is the very high concentration of bis(2-ethylhexyl) phthalate detected in clams from Mitchell Branch. These data should be viewed with skepticism, however, since the compound was detected in only one of the two duplicates and was also detected at a substantial concentration in one reference site (PCK 18.2) sample but not its duplicate.

4.1.5 Future Studies

The most significant contaminant bioaccumulation problems near the K-25 Site are the high concentration of PCBs in fish in Mitchell Branch and K-1007-P1 and the possible export of PCBs from these two systems to Poplar Creek and Watts Bar Reservoir. A key objective of future studies will be to identify and characterize the PCB source(s) in Mitchell Branch and the K-1007 pond series. The results of the screening study in 1993 indicate that fish contaminant concentrations in the K-1007 pond approach or exceed commonly cited human health threshold limits; thus, monitoring will be initiated to more thoroughly track spatial and temporal changes in those contaminant levels. Caged clams will again be used to estimate the importance of Mitchell Branch and K-1007 inputs of PCBs to Poplar Creek relative to other sources.

In 1994, a survey of PCB contamination in fish in the remaining unsampled ponds in the K-1007 series will be initiated. In addition to routine sampling of sunfish in Mitchell Branch, caged clams may be tried in upper Mitchell Branch (upstream and downstream of SDs 170, 180, and 190) in an attempt to evaluate possible PCB source(s) in that system. Staff in the K-25 BMAP bioaccumulation task will continue to work closely with Y-12 BMAP staff in the development and application of passive monitors of PCB contamination in aquatic systems, which may eventually prove valuable in locating PCB sources within the K-25 Site storm sewer network.

Routine tracking of mercury and PCB contamination using sunfish will continue at sites on Poplar Creek to evaluate the changes over time in inputs of those contaminants to that system. Metals screening of fish from lower Poplar Creek, Mitchell Branch, and the pond sites revealed no evidence of contamination and no concerns with respect to human consumption of fish. Metals other than mercury will not be analyzed in fish filets in 1994. Whole body or specific tissue (liver, kidney) analyses for metals in fish may be needed in the future to address specific ecotoxicological questions at specific sites. Such future studies will be planned in response to a demonstrated need for such data.

Screening for organics in caged clams detected a very high concentration of bis (2-ethylhexyl) phthalate in Mitchell Branch, as well as low concentrations of aldrin over a 3-year period. Routine monitoring of organics will continue at this site. Only trace concentrations (below method quantitation limits) of a few organic compounds were detected

at the other sites monitored in the screening study; future monitoring, therefore, is not warranted.

4.2 CONTAMINANT ACCUMULATION IN TERRESTRIAL BIOTA

Biological monitoring programs typically focus on aquatic organisms because these organisms are directly exposed to waterborne contaminants. However, many terrestrial species obtain a substantial portion of their diet from aquatic sources; and some terrestrial species, especially waterfowl, spend a significant portion of their lifetime in the water.

Of contaminants that bioaccumulate and are concentrated as they pass up the foodchain, terrestrial predators, especially piscivorous animals, may receive a higher dose than would be predicted solely by measuring bioaccumulation in the aquatic system. Previous K-25 BMAP data indicate that two contaminants with a high bioaccumulation potential—mercury and PCBs—are found at elevated levels in Mitchell Branch and Poplar Creek. Thus, sensitive terrestrial organisms may be receiving a significant dose of these contaminants from K-25 Site sources.

In order to assess the potential magnitude of exposure within the terrestrial ecosystem, this task evaluated two terrestrial species known to bioaccumulate contaminants from the aquatic system. Three subtasks, two of which are extensions of reservation-wide monitoring efforts, were begun as 1-year trials to determine whether sufficient exposure exists to warrant further study.

4.2.1 Waterfowl Monitoring *(B. G. Blaylock, B. R. O'Neal and D. K. Cox)*

4.2.1.1. Introduction

Streams and ponds located on the ORR are used as resting areas for migrant waterfowl and as nesting sites for resident populations. Waterfowl using waste disposal ponds can accumulate contaminants by ingesting food or water. The accumulation of contaminants is greatly influenced by the feeding habits of the different species of waterfowl. Because of the mobility of waterfowl, they can be direct vectors of contaminants from aquatic systems on the ORR to humans off-site when they are harvested by hunters for food. As part of the ORNL BMAP, a study was initiated in 1987 to assess the waterfowl uptake of radionuclides and other contaminants from White Oak Lake and other waste disposal areas at ORNL.

The study was extended to include sites near the K-25 Site in late 1987. Because the resident Canada goose population has become well established on the ORR and geese have been found nesting at waste disposal ponds, the study was extended to monitor the local population and use these data to estimate the probability of a hunter harvesting a bird that had resided on the ORR. An annual leg-banding study, initiated in 1988, included placing throat collars with visible identification numbers on geese during the molting season. A census of the population is conducted on a weekly basis to estimate the population size and monitor the movement of individual geese.

4.2.1.2 Methods

Waterfowl census and Canada goose banding

Thirteen sites, including some inside the security area, are monitored on a weekly basis at the K-25 Site. During June 1993, in cooperation with the Tennessee Wildlife Resources Agency and the University of Tennessee Wildlife Society, adult and juvenile geese were rounded up to be fitted with leg bands. Adult geese were fitted with leg bands and white throat collars displaying black identification codes, and young birds (juveniles) were fitted with leg bands only. Codes on the throat collars are recorded each week during the waterfowl census to track the movement of individual geese.

Contaminants in Canada Geese

Waterfowl are collected periodically on the ORR for radiological and nonradiological analyses. In August 1993, fish collected from pond K-1007B at the K-25 Site were found to have high concentrations of PCBs. Concerns arose that Canada geese, which have been known to frequent the pond, may also have high concentrations of PCBs.

Five Canada geese, which had been banded and collared in 1989–1993 were collected near the K-1007B pond. Data from the waterfowl study showed that these geese had been observed at the K-25 Site for 1 to 5 years; although they were also observed at other areas. For example, one goose (C19T), which was collared in 1989 at the K-25 Site, was observed 41 times at the K-25 Site, 2 times at the Y-12 Plant, 1 time at the Oak Ridge Marina, and 1 time at the ORNL Swan Pond. The waterfowl data confirmed that these were resident geese. Five Canada geese were also collected below Norris dam. Breast and liver tissue were analyzed for PCBs. Breast, liver, and bone samples were analyzed for gamma-emitting

radionuclides (^{60}Co, ^{137}Cs, etc.). Selected samples will be analyzed for ^{99}Tc, ^{239}Pu, ^{240}Pu, ^{90}Sr, ^{235}U, and ^{238}U. Breast and liver will also be analyzed for cadmium, chromium, mercury, lead, and selenium.

4.2.1.3 Results and Discussion

Waterfowl census and Canada goose banding

The total number of Canada geese sighted at the K-25 Site from June 1993 to March 1994 is depicted in Fig. 4.3. For comparative purposes, the data for June 1992 to March 1993 are also given in Fig. 4.3. The population trends are generally the same, except during October–January 1993–1994, when an increase in the population occurred. The largest number of birds is usually seen during December when the local population is supplemented by migrants from the north. However, during February when the geese are seeking nesting sites, the birds are scattered, and the number of geese sighted at the K-25 Site decreases through the end of March, when the parents with their broods are evident and begin congregating near the ponds and streams associated with the K-25 Site.

During the June 1993 round up of Canada geese on the ORR and adjacent areas, 540 geese were captured. Many of the geese were recaptures and had leg bands and throat collars from previous years. Of the total geese captured, 273 were fitted with leg bands and throat collars. This is far fewer than the 500 geese that received throat collars and leg bands in 1992. During the round up, 43 geese were captured at the K-25 Site, 25 were recaptures and the remainder were fitted with throat collars and leg bands. This number, too, was smaller than the number usually captured at the K-25 Site; however, a large flock was on Poplar Creek and could not be captured. Data collected from the weekly census of geese are being used in a population model to estimate the probability that a hunter will harvest a goose that has lived on the ORR.

Contaminants in Canada geese

The five Canada geese collected near the K-1007B pond had been observed at the K-25 Site for 1 to 5 years. During this period, these geese were also observed at other areas. By having the census data we were able to establish the long term association of individual geese with the pond ecosystem.

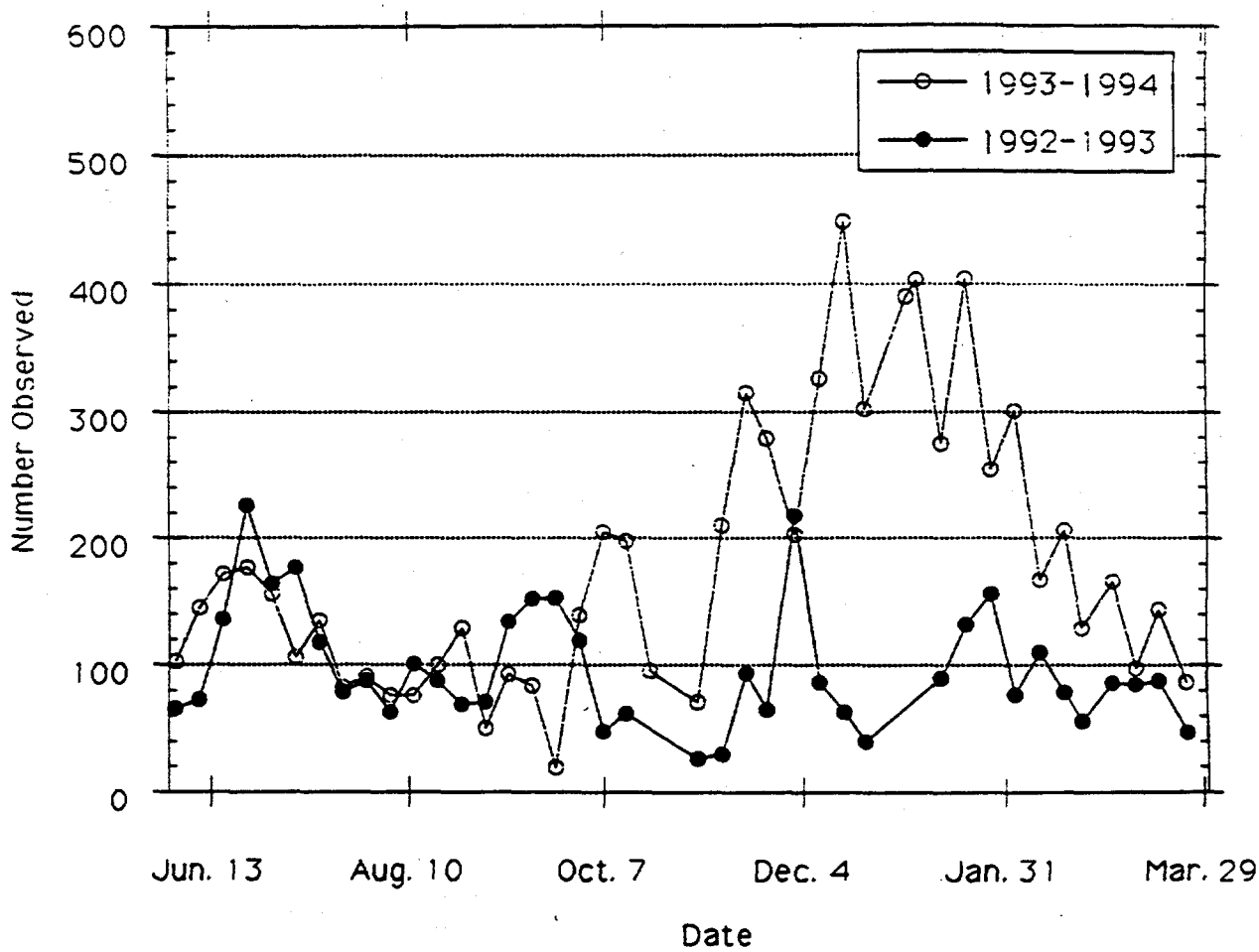

Fig. 4.3. Number of Canada geese observed at the K-25 Site during June 1993–March 1994, compared with numbers observed June 1992–March 1993.

Breast and liver tissue from these ten geese were analyzed for PCBs. All samples were below the PCB limits of detection (0.34 μg/g). Tissues from these geese were also counted for ^{137}Cs and other radionuclides by a germanium detector. Of the 20 samples examined, measurable quantities were found in only 2 liver samples (0.1 and 0.2 pCi/g) and 1 muscle sample (0.1 pCi/g) from the Clinch River and 1 liver sample (0.1 pCi/g) from the K-25 Site. Tissues from these geese are also being analyzed for other radionuclides and nonradiological contaminants.

4.2.1.4 Conclusions and recommendations

Very low to undetectable levels of PCBs and other contaminants occur in geese at the K-25 Site. Sufficient data have been collected to begin modeling of the Canada goose population. The census will be reduced to four sites (ORNL, the K-25 Site, the Y-12 Plant, and Melton Hill Dam). The Canada goose population at the K-25 Site will continue to be censused weekly. Contaminant levels will be monitored by collecting geese and other waterfowl annually for radiological and nonradiological analyses. Where contaminants are known to exist (ponds K-1007B and K-901A), additional samples will be taken and analyzed.

4.2.2 Raccoon Bioaccumulation Monitoring *(T. L. Ashwood)*

Raccoons consume a wide variety of prey and vegetation, some of which is from aquatic systems. Studies throughout North America have demonstrated the potential for raccoons to serve as indicator species for a wide range of contaminants including heavy metals, PCBs, organochlorine pesticides, and radionuclides (Ashwood 1993). During 1991, limited sampling of raccoons from the lower White Oak Creek watershed suggested that these animals could be used to monitor the bioavailability of ^{137}Cs to biota in the terrestrial ecosystem (Ashwood 1992). Based on this evidence, a long-term monitoring program was initiated using raccoons as ecological indicators of biologically available contamination—especially mercury and ^{137}Cs— in the terrestrial ecosystem.

This report presents data from the 1993 trapping of raccoons in the vicinity of the K-25 Site.

4.2.2.1 Methods

Raccoons were trapped using cage traps baited with sardines or canned cat food. Animals caught during the spring generally had little, if any, fat, so sampling was continued during the fall, when raccoons are building up fat reserves.

In all trapping operations, raccoons were anesthetized with Ketaset (ketamine hydrochloride solution) prior to handling. Weight, gender, and some physical dimensions were determined, and as much fur as possible (typically 0.5–2 g) was removed from the raccoon's tail. A biopsy sample of fat from the thigh area was removed for PCB analysis. Three raccoons in the K-25 Site area were fitted with radio collars for radiotracking to determine home range size and whether there was movement between areas upstream and downstream of the K-25 Site.

Fur samples were placed in plastic bags that were labeled with the sample identifier and collection date. Sample bags were generally kept frozen until rinsing or washing took place. Fat samples were placed in 20 mL scintillation vials and frozen immediately after collection.

Fur samples were washed using a 20-min mechanical agitation in a dilute solution of Prell® shampoo in deionized water (~250 mL) followed by three rinses in ~250 mL of deionized water. During each rinse, the hair and water were mechanically agitated for 20 min. A different, acid-cleaned bottle and fresh deionized water were used for each rinse. After the final rinse, each fur sample was air dried at 60°C for ≥16 h and then immediately sealed hermetically in a 93-cc plastic-lined aluminum can that had been previously labeled and tared. The sealed can was cooled for ≥30 min in a desiccator prior to the final weighing.

Samples were submitted to an external laboratory for trace metal (As, Cd, Hg, Pb, Se) and PCB analyses. Trace metals were analyzed by graphite furnace atomic absorption spectrometry (except Hg, which was analyzed by cold vapor atomic absorption). Preparation blanks of deionized water and shampoo were also analyzed for metals. Fat samples were extracted with methylene chloride, dried, and cleaned up using alumina and silica gel and gel permeation chromatography, and analyzed by a capillary column gas chromatograph coupled to an electron capture detector.

4.2.2.2 Results

In May 1993, seven raccoons were captured along the reach of East Fork Poplar Creek between Lambert Quarry and the confluence with Poplar Creek. During this period only two

raccoons were captured downstream of the K-25 Site along Poplar Creek. During September and October, an additional five raccoons were captured in each area.

No results of metals or PCB analyses were available at report time and will be presented in the quarterly report(s) following receipt and analysis of the data.

4.2.3 Whole-Body Analyses of Representative Prey Fish

Other K-25 bioaccumulation studies focus on bioaccumulation of mercury and PCBs in filets of sunfish because such samples are representative of human exposure to the contaminants and because sunfish can be used to assess differences in contaminant bioaccumulation among different sites. However, terrestrial predators tend to consume the entire fish rather than just the filet, so the whole-body burden of the fish is more representative of the dose received by these animals.

The purpose of this task is to determine whether representative prey fish from the K-25 Site are contaminated with PCBs and trace metals at levels of concentration that may be toxic to sensitive, piscivorous wildlife (e.g., mink). The original BMAP plan called for collection of stonerollers (*Campostoma anomalum*) from the K-1007-P1 pond and from Poplar Creek downstream from Mitchell Branch (Sect. 2.2.3.4 in Kszos et al. 1993). However, field researchers from other BMAP tasks discovered that stonerollers were not present at these sites in sufficient quantities to provide an adequate sample size. Therefore, gizzard shad (*Dorosoma cepedianum*) were selected as the representative prey species. Gizzard shad have relatively high concentrations of lipids, so they should accumulate PCBs, and they are an abundant prey fish.

Ten gizzard shad were collected by electrofishing at the K-1007-P1 pond (where largemouth bass contained high levels of PCBs) and near Mitchell Branch on Poplar Creek. The fish were frozen and homogenized. Representative aliquots were submitted to the Analytical Services Organization at ORNL for metals and PCB analyses. See Sect 4.1.2 for further details on methodology.

4.2.3.1 Results

Analytical results were not available at report time and will be presented in the quarterly report(s) following receipt and analysis of the data.

5. ASSESSMENT OF FISH HEALTH (TASK 3)

S. M. Adams, W. D. Crumby, and M. S. Greeley, Jr.

5.1 BIOINDICATORS SUBTASK *(S. M. Adams and W.D. Crumby)*

This report presents the results of the 1989–1992 bioindicator studies conducted in Mitchell Branch and three reference streams and also evaluates temporal changes in the health of fish populations in Mitchell Branch from 1989 to 1992. As a representative indicator of the health of the Mitchell Branch system, redbreast sunfish (*Lepomis auritus*) has been the single target fish species for this BMAP task since 1989. The redbreast sunfish is especially suited for assessing the ecological health of Mitchell Branch because of its relative abundance and position at the top of the food chain in this stream. Because of their position in the food chain, these fish are integrators of environmental conditions and may reflect, therefore, the overall health of the stream ecosystem.

In this study, an integrative approach has been taken to investigate holistic responses of fish to environmental stressors and to aid in the identification of causative agents or mechanisms that may be affecting the health of fish populations in Mitchell Branch.

Many ecotoxicological studies dealing with the biological effects of environmental stressors, such as contaminants, use measurements of one or more variables to evaluate the response of organisms to these stressors. Whereas single variable responses may reflect specific structural or functional attributes of an organism, usually at a particular level of biological organization, these single responses do not provide an integrated measure of the effects of stressors on organism health (Adams and Ryon, in press). If single variable measurements are analyzed separately, then the interrelationships among the observed variables are not considered in evaluating an organism's response to stress. Although the complexity of analysis and interpretation may be greater in multivariate analysis, statistical approaches that use integrated multivariate analysis take into account the interrelationships and associations that exist among variables. Therefore, multivariate approaches more accurately reflect the myriad of interactions that occur between the organism and the environment than do single variate approaches (Cairns and van der Schalie 1980, Capuzzo 1985, Heath 1987). The complexity of ecological systems, such as Mitchell Branch, dictate that a holistic approach be taken in evaluating the effects of environmental stressors in these systems.

5.1.1 Methods

5.1.1.1 Sampling procedures

Sampling was conducted during spring-summer periods of 1989–1992 in Mitchell Branch (above weir) and three reference streams, including Brushy Fork, located between Oliver Springs and Clinton (Anderson County, Tennessee); Hinds Creek, between Clinton and Norris (Anderson County); and Paint Rock Creek (Loudon County, Tennessee). At each site, redbreast sunfish (*L. auritus*) of all sizes and age groups were collected by electroshocking. Blood samples were taken from 12 to 15 of the larger male fish within 2 min after collection by puncturing the caudal vessels with a 20-gauge needle. Blood samples of ~0.7 mL were obtained from all fish by using unheparinized 3-mL vacutainers (Becton, Dickson, & Co.). Each tube was labeled with a fish identification number and placed on ice for transport to the laboratory. All fish were processed for population-level analysis by recording their lengths and weights and removing a sample of scales for age and growth information. Fish not used for the blood and bioindicator analysis were returned to the stream alive.

5.1.1.2 Analytical procedures

Total lengths and weights were recorded for fish transported from the field, and observations were also made on the general condition of the fish, such as presence or absence of disease, body and/or mouth sores, external parasites, and general appearance. Following sacrifice, the liver and spleen were removed from each fish for further analysis. A 100-mg section of liver was placed in a 20-mL scintillation vial with 5-mL of Bouin's fixative for histopathological analysis. The remaining viscera (minus liver and spleen) were excised from the body cavity, and their total weight was recorded after all food material was removed from the stomach and intestine. The liver- and visceral-somatic indices were calculated by dividing their respective weights by total body weight. The condition factor (K) of each fish was calculated as $K = 10^5 W/L^3$, where W is body weight (in grams) and L is total length (in centimeters).

Serum analysis

Blood collected in the unheparinized tubes was allowed to clot, transferred with Pasteur pipettes to 1.5-mL conical micro-centrifuge tubes labeled with the fish identification number,

and centrifuged for 3 min in a Beckman Microfuge. The clear supernatant (serum) was drawn off with clean pipettes and transferred to labeled 1-mL conical plastic tubes.

Indicators of organ dysfunction [creatinine, alanine aminotransferase (ALT), serum albumin, and blood urea nitrogen (BUN)] were analyzed following the methods described by Henry et al. (1974), Bergmeyer et al. (1978), Doumas et al. (1972), and Tiffany et al. (1972) respectively. Cholesterol was analyzed by the method of Allain et al. (1974), and triglycerides were analyzed by the procedure of Bucolo and David (1973). Total serum proteins were measured by the Biuret method (NCCLS 1979); the procedures for this assay are described in the Roche Diagnostic Systems (1986) information package. All of these methods are enzymatic assays; the reagents for each assay were obtained from Roche Diagnostic Systems. All serum assays were performed with an automated Centrifugal Fast Analyzer System (Cobas-Fara, Hoffman La Roche Instruments, Inc.). Calibrations were made by using the Roche serum calibrator as the standard and Moni-Trol-ES Level 1 and 2 (American Dade, Miami, Florida) as internal standards.

Histopathological analysis

The following histopathological analyses were performed by Dr. David Hinton of the School of Veterinary Medicine, University of California-Davis: (1) percentage of tissue occupied by parasites, (2) percentage of liver composed of necrotic parenchyma, (3) percentage of tissue composed of macrophage aggregates and (4) percentage of liver occupied by functional parenchyma. These analyses were performed by using methods described by Hinton and Couch (1984).

Detoxification Enzymes

Microsome isolation. Fish hepatic microsomes were prepared by differential centrifugation (McKee et al. 1983) with several modifications. Fish were sacrificed by severing the spinal cord, and the livers were immediately removed and blotted dry. Each liver sample was placed in ice-cold buffer (0.25 m*M* sucrose, 0.1 *M* tris, pH 7.4). The tissues were minced and then homogenized in 5 volumes of buffer with a motor-driven Potter-Elvehjem glass and Teflon homogenizer. The homogenates were centrifuged in a J-21B Beckman

centrifuge at 3000 *g*[*] for 10 min and at 10,000 *g* for 20 min. The resulting supernatants were then centrifuged at 105,000 *g* for 60 min in a Beckman L3-50 ultracentrifuge. Microsomal pellets were resuspended in 0.1 M tris buffer, 1 m*M* EDTA[**], and 20% glycerol at pH 7.4 by sonication for 10–15 s with a Braun Sonic 1510 at 50 W. All operations were performed at 0 to 4°C. The microsomes were frozen with liquid nitrogen and stored at -120°C; no significant change in 7-ethoxyresorufin O-deethylase (EROD) activity was detected after 6 months of storage at this temperature. The activity of fish microsomes stored under these conditions has been reported to be stable for 1 year (Forlin and Anderson 1985).

Enzyme assays. The activity of EROD was measured fluorimetrically at 30°C (Burke and Mayer 1974) and expressed as moles of resorufin per minute per milligram of microsomal protein. The final reaction buffer contained 80 m*M* HEPES buffer (pH 7.8), 5 m*M* magnesium acetate, 1.0 μM 7-ethoxyresorufin, 250 μM of reduced nicotinamide adenine dinucleotide phosphate (NADPH), and 100 μM EDTA. The concentration of total protein used in the enzyme assay ranged from 0.2 to 1.0 mg/mL, depending on the activity of the sample. Proteins were measured by the Bio-Rad (Richmond, California) reagent method (Bradford 1976) with a Centrifugal Fast Analyzer System in which bovine serum albumin was used as a standard. Cytochrome P-450 and cytochrome b_5 content were each measured by their characteristic oxidized and reduced spectra. Cytochrome P-450 samples were oxidized with carbon monoxide and reduced with sodium dithionite (Johannesen and DePierre 1978). Cytochrome b_5 was reduced with NADH, and the b_5 assays were conducted prior to the P-450 analysis (Stegeman et al. 1979). NADPH-cytochrome *c* reductase was assayed spectrophotometrically by the reduction of cytochrome *c* using an extinction coefficient of 21.1 cm/m*M*.

Population Evaluation

Scale samples were used to determine the age and to estimate the growth of redbreast sunfish in Mitchell Branch and the three reference streams. Approximately ten scales were removed from each fish using small metal forceps. Scales were taken from the left side of the

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[*] *An italic, lowercase "g" denotes the standard acceleration due to gravity (~9.8 m/s).*

[**] *EDTA = ethylene diaminetetrasetic acid.*

body near the tip of the pectoral fin and stored in 5.7 × 8.9 cm coin envelopes. Total length (in centimeters), weight (in grams), species, sampling site, date, and tag number (if applicable) were recorded on each envelope. Fish were returned to the stream alive after scales were obtained and the length and weight were recorded.

In the laboratory, scales from each fish were arranged in rows between two cellulose acetate slides. The slides were pressed together using a hand-cranked laboratory press (Wildlife Supply Co.), which left an impression of each scale on one of the slides. Scale impressions were stored with scale samples in corresponding coin envelopes.

Scales were analyzed using a video image digitizing system. This system includes a dissecting microscope with video camera attachment, a 25.4 cm black and white video monitor, and the video digitizer. The digitizer was connected to a personal computer to allow data transfer to the growth analysis program, ***Disbcal*** (Missouri Department of Conservation 1989).

At the beginning of each scale analysis session, a file was opened in the ***Disbcal*** program to store new scale data. Scale impressions were placed on the microscope stage and viewed on the video monitor. For each scale impression, the tag number, total length, weight, age, and sex (if known), were entered into ***Disbcal***. The distance from the focus to each annulus and to the scale edge was transmitted using the digitizer. Annuli were identified using the techniques outlined by Jearld (1983). A sample was excluded from analysis if scales exhibited abnormal form or if a large number of the scales had been lost and regenerated by the fish. Twenty percent of the scale samples from each site were read by a second individual for QA (verification) purposes.

Growth data were then analyzed using the files created during scale measurement. All back-calculations of fish length were made using the Fraser-Lee method (Fraser 1916 and Lee 1920), as follows:

$$L_i = a + \frac{(L_c - a)}{S_c} S_i, where$$

L_i = calculated length at age i;

= intercept of saale radius vs fish length regression;

L_c = length of fish at capture;

S_c = radius of scale at capture; and

S_i = scale measurement to annulus i.

Fish were divided into 1-cm size groups based on total length. Length frequency and age frequency histograms were developed for each population. Growth trends (based on total length at capture) were compared among sites for the years 1989–1992. For fish captured in 1992, a Tukey test (Zar 1984), was used to test for significant differences among sites in mean total length at capture ($p = 0.05$). Back-calculated length at age was also compared among sites for fish captured from 1990 to 1992.

5.1.1.3 Statistical procedures

To determine the integrated response of fish to the environmental conditions at each sampling site, bioindicator variables representative of five functional response groups were entered into a canonical discriminant analysis procedure (PROC CANDISC, SAS 1985b). Individual bioindicators were grouped into five functional categories representing indicators of (1) detoxification enzyme induction, (2) organ dysfunction, (3) histopathology, (4) overall fish health or condition, and (5) nutritional or feeding status. These functional groups reflect gradients of both ecological relevance and the time-course of a response to a stressor such as a contaminant (Adams 1990). Those variables in categories (1) and (2) are primarily short-term response indicators and have relatively low ecological relevance, whereas those in groups (3) through (5) are longer-term response indicators and are characterized by low toxicological but high ecological relevance (Adams 1990).

The canonical discriminant analysis procedure provides a two-dimensional graphic representation of the positions and orientations of the various responses of fish from each site relative to each other. In addition, this method derives a linear ranking of combinations of all variables and can identify statistically significant differences among treatment means even when single variables may not indicate such differences. A selection procedure for variables available in SAS (PROC STEPWISE, SAS 1985b) was used to identify the indicators that contributed most to the discrimination of fish health status among sites, based on the integrated parameters. This selection procedure for variables considered all possible combinations of the observed values and, for any specified subset size, selected those variables having the best discriminating power.

Procedures for canonical variate analysis are derived under the assumption that observations come from multivariate normal distributions with equal covariance matrices.

Using the Shapiro-Wilk statistic (Conover, 1980), the normality assumption was evaluated by testing (for each sample population) the marginal univariate normality of the distribution for each variable. Because of the large number of tests involved, the level of significance of each test was adjusted using a Bonferroni modification, to control the overall level of significance (Johnson and Wichern, 1992). If marginal abnormality was indicated for a variable, then it was transformed (usually with the log transformation) to achieve a more nearly normal marginal distribution. Although marginal normality does not guarantee multivariate normality, the abnormality of the marginal distribution frequently reflects multivariate abnormality. The homogeneity of the covariance matrices was tested using Bartlett's test for equal covariance matrices (Seal, 1964). This test is very sensitive to the normality of the variable distributions for each population (Johnson and Wichern, 1992).

Two levels of analyses were conducted to evaluate the effects of water quality and other environmental stressors on the health of fish in Mitchell Branch. The integrated bioindicator analysis was used to determine the overall health response of fish at both impacted and reference sites, and age and growth analyses were used to compare fish population health among streams.

5.1.2 Results and Discussion

5.1.2.1 Integrated bioindicator analysis

The integrated response of fish to the environmental conditions at each study site was evaluated by incorporating bioindicators representing five functional response categories in the multivariate analysis. This multivariate approach allows the health of fish populations at each sample site to be evaluated within a holistic or integrated framework. In addition, this is a useful approach for determining the effectiveness of remedial actions on aquatic ecosystem health.

Results of the canonical discriminant analysis procedure revealed several patterns relative to the health of the redbreast sunfish population in Mitchell Branch compared to the health of fish at the three reference sites. In all years (1989–1992) the integrated health response of fish collected from Mitchell Branch was distinct from that of sunfish collected from the reference sites (Figs. 5.1–5.4). The integrated response of Mitchell Branch fish for each year always

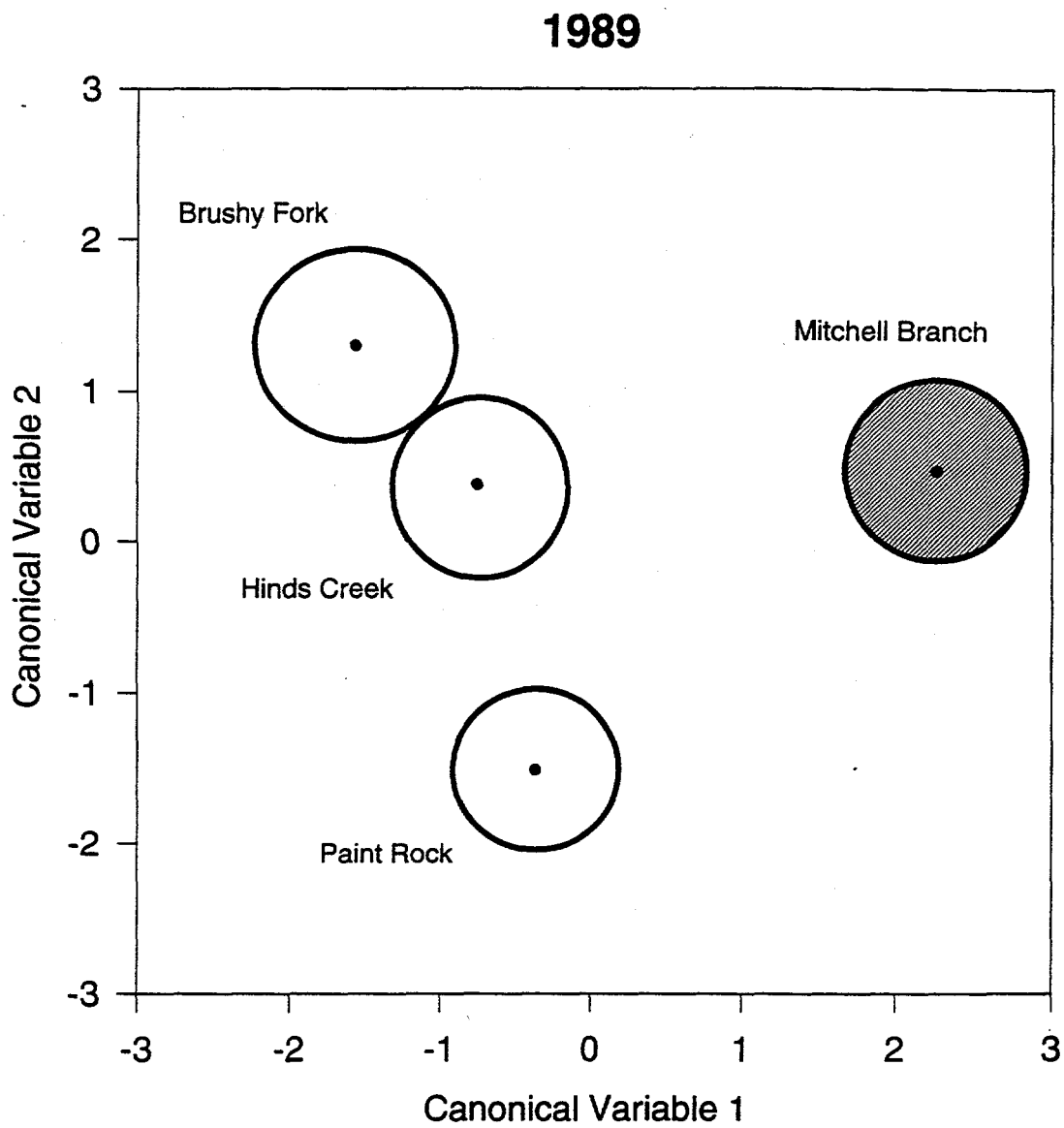

Fig. 5.1. Segregation of integrated health responses for male redbreast sunfish collected in 1989 at Mitchell Branch and three reference streams. Circles represent site means and the 95% confidence radii of the site-year means.

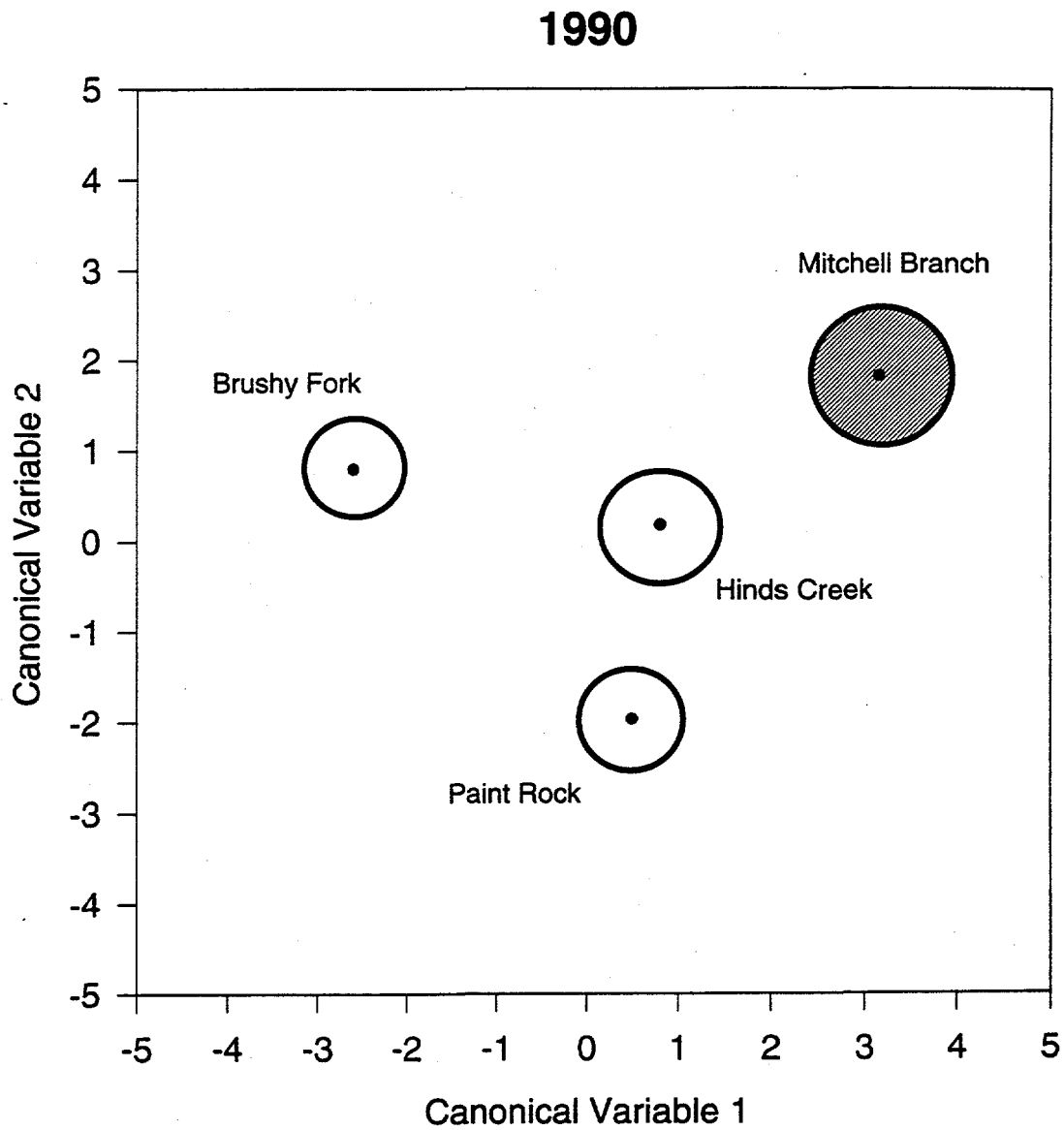

Fig. 5.2. Segregation of integrated health responses for male redbreast sunfish collected in 1990 at Mitchell Branch and three reference streams. Circles represent site means and the 95% confidence radii of the site-year means.

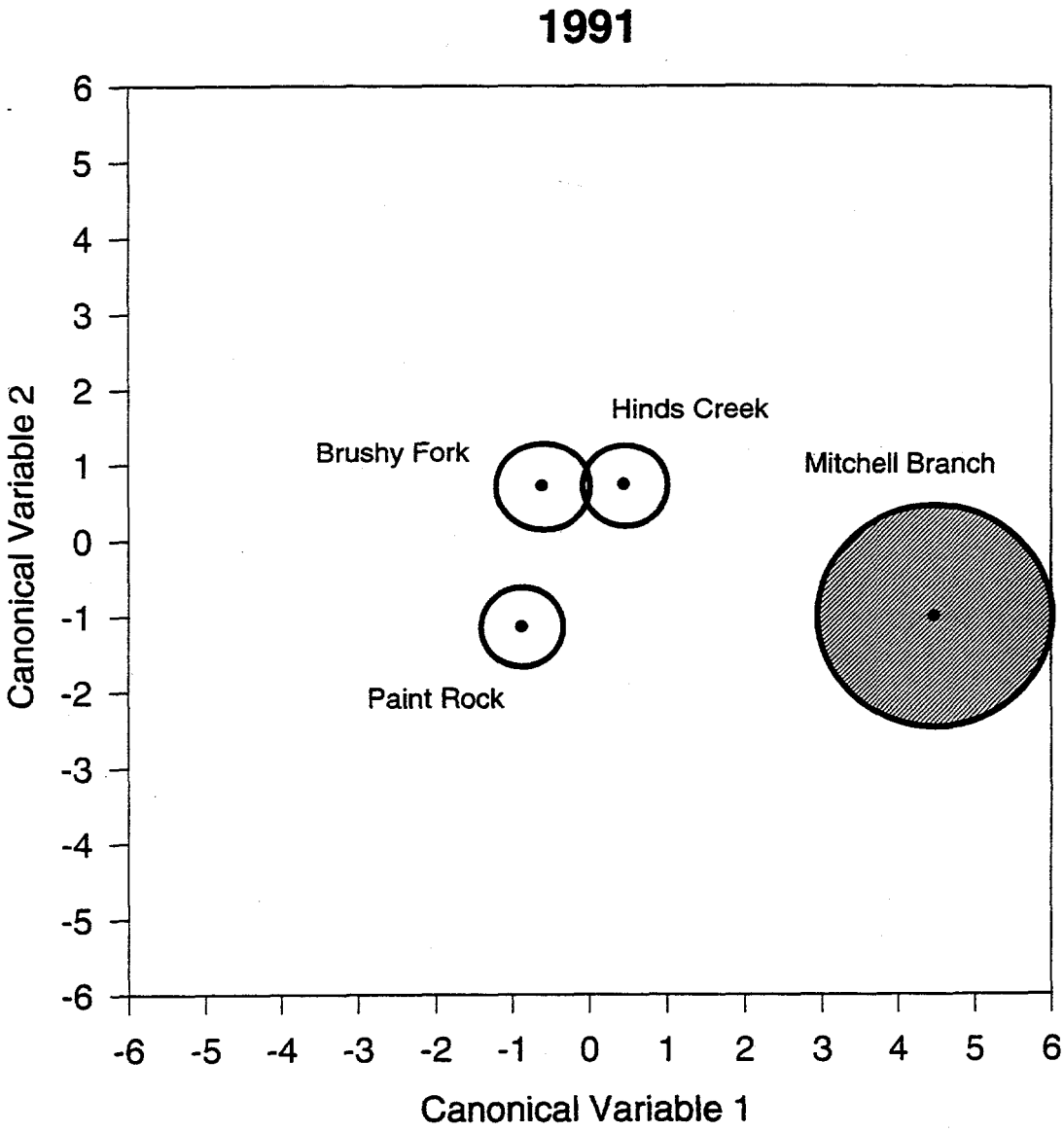

Fig. 5.3. Segregation of integrated health responses for male redbreast sunfish collected in 1991 at Mitchell Branch and three reference streams. Circles represent site means and the 95% confidence radii of the site-year means.

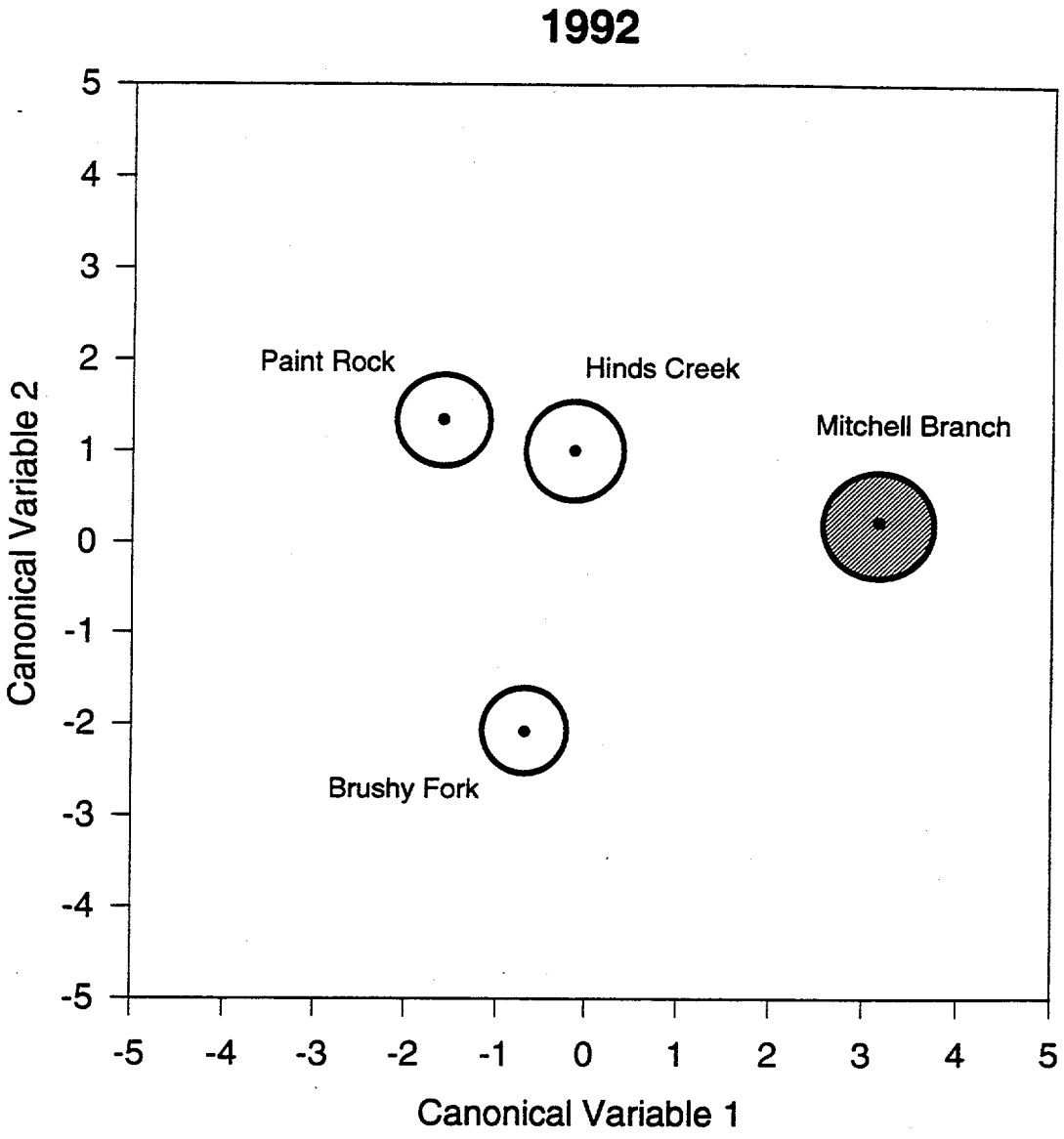

Fig. 5.4. Segregation of integrated health responses for male redbreast sunfish collected in 1992 at Mitchell Branch and three reference streams. Circles represent site means and the 95% confidence radii of the site-year means.

occurred on the right side of the discriminant function plot, while the reference site responses cluster at the middle on the left side of the discriminant plot (Figs. 5.1–5.4).

In general, there is a higher similarity in integrated responses among reference sites within each year than there is between the reference sites as a group and Mitchell Branch. In fact in 1989 and 1991, Brushy Fork and Hinds Creek were very similar in their overall response as indicated by the close proximity of the 95% confidence radii of the site means (Figs. 5.1 and 5.3). In all years except 1992, Paint Rock Creek tended to be rather dissimilar from the other two reference sites even though it still maintained its distinct dissimilarity to Mitchell Branch.

The fact that all three reference sites were relatively similar to each other in at least two of the study years (1989 and 1991) and fish from Mitchell Branch were consistently segregated from the reference groups for each of the 4 years provides convincing evidence that the Mitchell Branch fish are being differentially affected by environmental stressors in that system. As long as the integrated response of fish from Mitchell Branch can be distinguished statistically (see Table 5.1) from the reference site response group, this would indicate that fish in Mitchell Branch are in an unhealthy state compared with fish from the reference streams.

Table 5.1. Statistical linear distances (Mahalanobis distances) between the integrated bioindicator responses of fish collected from Mitchell Branch and each reference from 1989 to 1992

	Reference Sites			
	Brushy Fork	Hinds Creek	Paint Rock Creek	Mean
1989	9.4	15.5	10.8	11.9
1990	13	34	22	18.7
1991	20	29	29	26
1992	15	20	24	14.7
Mean	14.4	24.6	18.8	

Based on the relative positions of the integrated responses of Mitchell branch fish to the reference site fish over all years studied, there is little evidence to indicate that the health of Mitchell Branch fish has improved over this period (Figs. 5.1–5.4). Quantitative relationships between integrated site means can be demonstrated statistically with a measure of the

differences between the midpoints of these site means. The Mahalanobis statistic is a measure of the linear difference in the distances between site means. Table 5.1 shows the statistical distances between Mitchell Branch and each reference site and illustrates that since 1989 no clear pattern has emerged relative to the temporal change in health of Mitchell Branch fish. For sunfish from Brushy Fork and Paint Rock Creek, the statistical distance between their integrated site means and that of Mitchell Branch increased from 1989 to 1991 then declined in 1992. This pattern demonstrated that from 1989 to 1991 the health of sunfish from Mitchell Branch became increasingly different from that of fish from these two reference areas. In 1992, however, the linear distance decreased, indicating increasing similarity in health between the reference sites and Mitchell Branch from 1991 to 1992. When the mean for each year is calculated across all three reference sites, a similar pattern is noted; namely a progressive decrease in health of Mitchell Branch sunfish from 1989 to 1991 and an increased health status in 1992.

Activities to remediate the quality of surface waters at the K-25 Site have been ongoing since 1991. For example, since 1991 dechlorination of the major storm drains has occurred, which may account for the improved fish health seen in 1992. This action may explain, in part, the improvement in fish health observed after 1991.

5.1.2.2 Health indicator responses

For each year, the individual response variables that best discriminated among the health status of fish from the four sample sites were identified by a multivariate selection procedure. These variables were used to generate the integrated responses at sites shown in Figs. 5.1–5.4. Over all 4 years, a total of ten bioindicators were identified as being important in discriminating among site responses. Indicators from three of the functional response groups are represented, including those from the detoxification enzyme group (EROD, NADPH, and microsomal protein), organ dysfunction group (albumin, serum protein, blood urea nitrogen, and creatinine), and the condition indices (the visceral- and liver-somatic indices) (Table 5.2). In 1989, EROD, an indicator of contaminant exposure, was the variable most responsible for discriminating among sites, followed by two organ dysfunction indicators (albumin and blood urea nitrogen) and the liver somatic index.

Table 5.2. Ranking of bioindicator response variables that were significant in discriminating among the four sample sites each year

Bioindicator	1989	1990	1991	1992
EROD	1		1	6
NADPH		3		5
Albumin	2	2		
Serum Protein		1	2	
Microsomal Protein			3	3
Visceral Somatic Index		4		4
Liver-Somatic Index	4	5		2
Blood Urea Nitrogen	3	6		1
Condition Factor			4	
Creatinine				7

The activities or levels of liver detoxification enzymes are used to indicate exposure to various xenobiotics, such as polycyclic aromatic hydrocarbons (PAHs), polychlorinated biphenyls (PCBs), and pesticides (Payne and Penrose 1975). Many of these compounds commonly occur in industrial and municipal effluents and are accumulated within living tissues (Ahokas et al. 1976, Neff 1978, Brown et al. 1986). Of the detoxification enzymes measured, EROD has been shown to be the best indicator of toxicant exposure in fish collected from other contaminated systems (Adams et al. 1992a; Adams et al. 1992b). Serum albumin promotes the transport of insoluble or sparingly soluble compounds, such as fatty acids, bilirubin, and hormones, across the cell membranes. This protein is also involved with osmotic regulation and can be used as a reserve source of protein and amino acids. Blood urea nitrogen (BUN) is the major end product of protein nitrogen metabolism in mammals and is generally used in the medical profession to diagnose renal disorders. Fish, however, produce a more diverse array of compounds as nitrogenous waste including, but not limited to, ammonia and urea. In addition to being excreted by the kidneys, some ammonia and urea is also excreted through the gills. In diagnosing, therefore, the effects of stress on fish, abnormal levels of urea in the blood may serve as an important indicator of gill dysfunction. The liver-somatic index (LSI) reflects both short-term nutritional status and metabolic energy demands (Heidinger and Crawford 1977, Adams and McLean 1985). The LSI is also sensitive to

toxicant stress, and liver enlargement resulting from hyperplasia (increase in cell number), and hypertrophy (increase in cell size) has been reported in fish exposed to toxic compounds (Fletcher et al. 1982, Heath 1987, Addison 1984).

A different set of indicators were important, however, in 1990 for among-site discrimination. Surprisingly, EROD was not an important distinguishing variable in 1990. However, two organ dysfunction indicators, serum protein and albumin, were the most influential variables. After these two variables, a detoxification enzyme, NADPH, entered into the discrimination process followed by two condition indices, the visceral- and liver-somatic indices. The visceral-somatic index (VSI) provides information on energy stored as lipids in the mesenteries of the viscera. These lipids are important for long-term energy use and gonad maturation (Adams and McLean 1985). Additionally, the VSI generally reflects the level of total body lipids, which is used to indicate overall fat storage and general nutritional status of the fish.

Again, in 1991, a detoxification enzyme was the most influential discriminating variable followed in order by serum protein (which was the first most important variable in 1990), microsomal protein, and the condition factor. Microsomal protein is another indicator of detoxification enzyme induction in the organism and the condition factor is important for reflecting the overall condition of the organism. The condition factor (CDFC) is a generalized indicator of the overall health or plumpness of a fish and can reflect the integrated effect of both nutritional status and metabolic impairment caused by stress.

In 1992, as in 1990, detoxification enzymes played a reduced role in discrimination among sites. Even though microsomal protein, NADPH and EROD were ranked 3rd, 5th, and 6th, respectively, in importance, organ dysfunction and condition index variables were the most influential discrimitory variables.

It is evident from this analysis of health response indicators that there is no apparent consistency among years in the importance of variables relative to intra-year site discrimination (Table 5.2). Each year was characterized by a unique set of site discriminatory variables. In 1989 and 1991, EROD was the most important variable, but in 1990 and 1992 organ dysfunction variables headed the list. Albumin was important in 1989 and 1990 but not in 1991 and 1992. Serum protein was a significant variable in 1990 and 1991 but not in 1989 and 1992. Similarly, the condition indices were somewhat important in 1992 but not in 1989.

A reasonable interpretation of this inconsistent pattern in the importance of health response variables among years is that Mitchell Branch has been subjected to not only a variety of stressors over time but that the action of these stressors on the system may be episodic or periodic in nature. If, in fact, organisms in Mitchell branch are exposed to chronic consistent levels of the same contaminant or groups of contaminants, then we would expect to see a relatively consistent pattern in response variables across years. For example, relatively consistent patterns in bioindicator response variables have been observed in both East Fork Poplar Creek and White Oak Creek over the past several years even though the magnitude of the responses has declined somewhat over time (Hinzman et al. 1993; Ashwood et al. 1994). We might conclude, therefore, that the health of the fish populations in Mitchell Branch may be influenced primarily by episodic events that occur in that system, either by periodic releases of contaminants or by other unknown activities or events that occur within the Mitchell Branch watershed. In addition, the primary stressors involved in these episodic events may not be the same across time. Future studies should identify and confirm not only the effects of continued remedial activities on fish population health in Mitchell Branch but also the relative importance of episodic events in influencing the health status of important aquatic organisms.

5.1.2.3 Population-level analysis

Length-frequency histograms were used to evaluate the size distribution of redbreast sunfish in Mitchell Branch from 1989–1992 (Fig. 5.5). The structure and size of the redbreast sunfish population in Mitchell Branch has changed over the last several years. Only 25 redbreast sunfish were collected in 1989. All of these fish were larger than 11.0 cm, indicating an imbalance in the population. Reproductive impairment, recruitment failure and inadequate spawning substrate were suggested as possible reasons for the small number of redbreast sunfish present in 1989 (Smith et al. 1994). However, in subsequent years, the number of fish collected has increased and the size structure has become more evenly distributed. In 1992, 130 redbreast sunfish were collected, ranging in size from 6 to 22 cm. This distribution of sizes was considered balanced and was similar to the size distributions of redbreast sunfish found in the reference streams.

Age-frequency histograms were used to aid in the evaluation of year class strength (Fig. 5.6). The age structure of the population has remained relatively stable, with fish up to 5 years of age collected each year since 1989. However, fish older than age 3 seem to be less

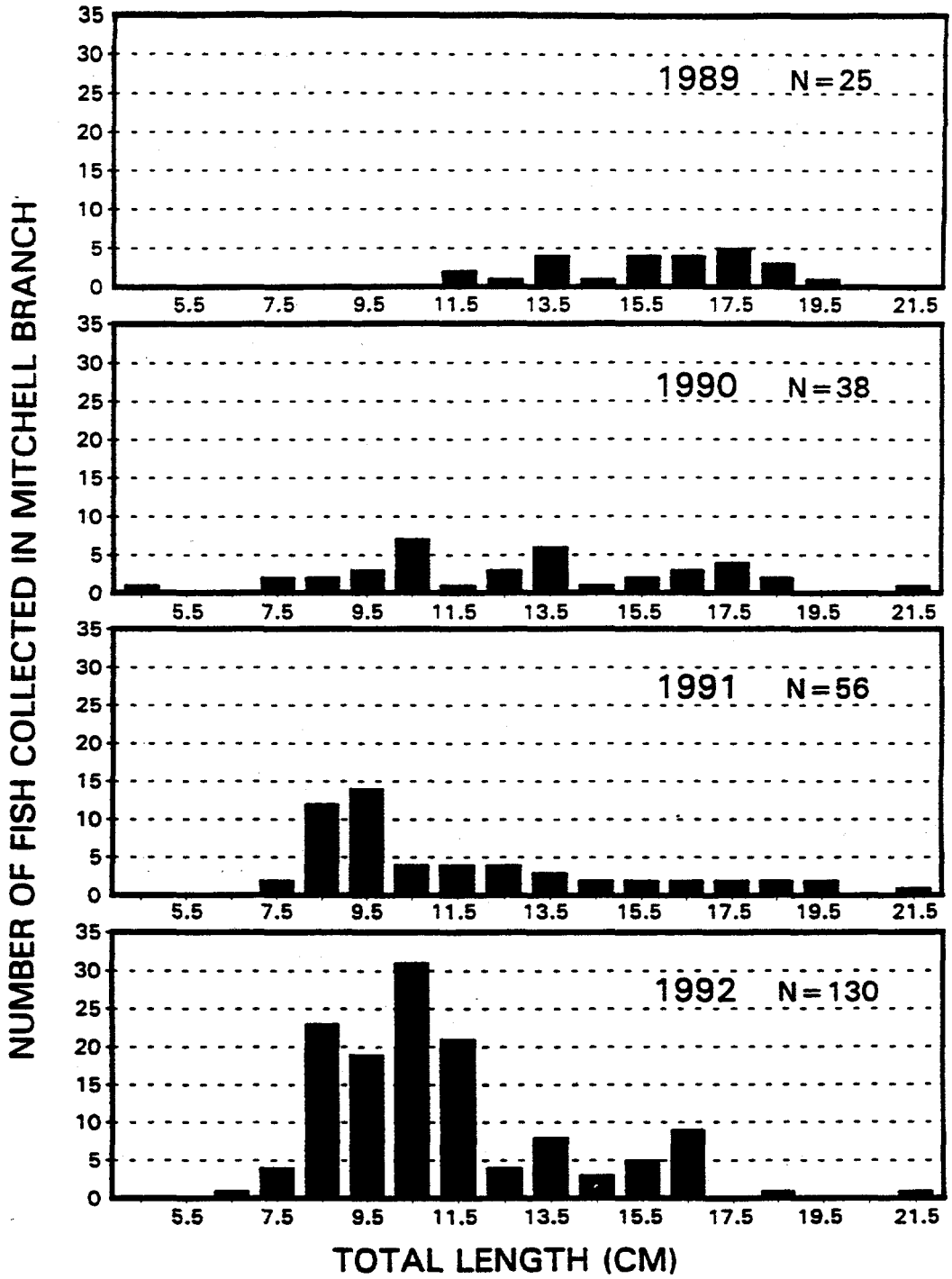

Fig. 5.5. Length frequency of redbreast sunfish collected in Mitchell Branch from 1989 to 1992.

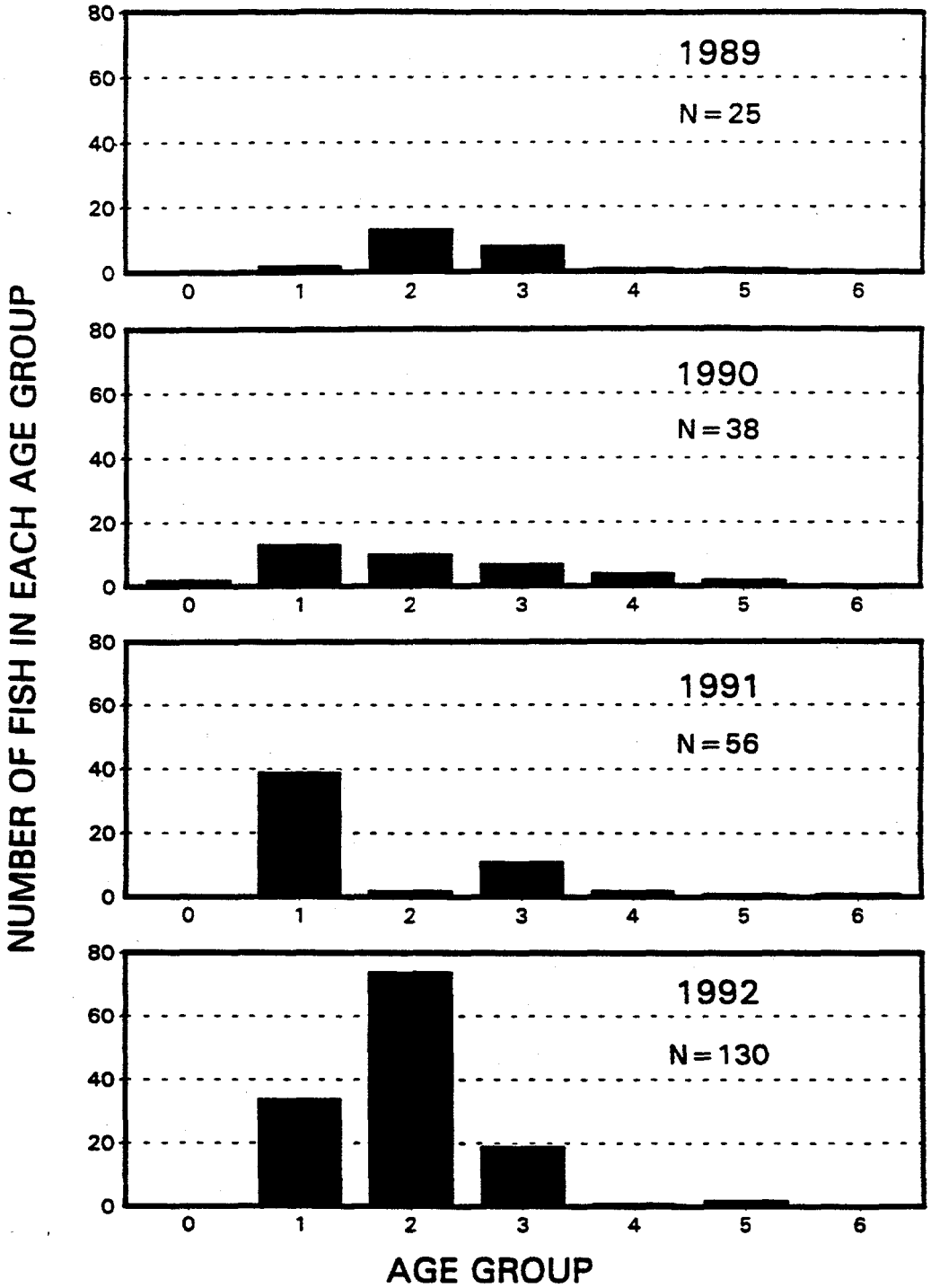

Fig. 5.6. Age frequency of redbreast sunfish collected in Mitchell Branch from 1989 to 1992.

common in Mitchell Branch than in the reference sites. In 1992, only 3 fish >age 3 were found in Mitchell Branch. In contrast, fish from the 1990 year class (age 2) made up 57% of the population, indicating an exceptionally successful spawning season in 1990. The silt and mud substrate of Mitchell Branch is apparently adequate for successful nesting of redbreast sunfish. Fluctuations in year class strength are more likely related to changes in seasonal flow patterns. Fluctuations in stream discharge can have a major effect on recruitment, growth, and survival of redbreast sunfish (Aho et al. 1986).

Growth Analysis

Total length (TL) of a particular age class at capture was used as an indicator of the cumulative growth history of redbreast sunfish from each site. Because fish at all sites were collected over a short period of time (7–10 days), any differences in mean TL at capture indicated differences in growth rates among sites. In 1989, redbreast sunfish in Mitchell Branch were exceptionally large when compared with reference site fish of the same age (Fig. 5.7). In each subsequent year, TL at capture decreased for Mitchell Branch redbreast sunfish, and size differences at age diminished relative to the reference sites. In 1992, there was no significant difference in TL at capture between fish from Mitchell Branch and Paint Rock Creek for any age group (Table 5.3).

Back-calculation of TL at annulus formation was also used to characterize growth trends from previous years. Sample sizes are increased in each age group when using this method because data from each sampling year are combined to calculate mean TL at age. The value of the intercept "a" that was used in back-calculations of fish length gave the best straight-line relationship for the regression of fish TL and scale radius (Carlander 1982). An intercept value of 24 mm was used in these calculations based on body-scale relationships from previous studies (Loar et al. 1993). The "a" of 24 mm was determined to be a good standard for use in growth analysis of redbreast sunfish populations in Mitchell Branch and the reference sites. Back-calculated lengths at age were generally larger for Mitchell Branch fish compared with reference site fish (Fig. 5.8). This was due to the higher growth rates that occurred before the fish population density increased in Mitchell Branch. In future years, back-calculated lengths for Mitchell Branch fish should decrease to reflect current growth rates, which are more similar to the reference sites.

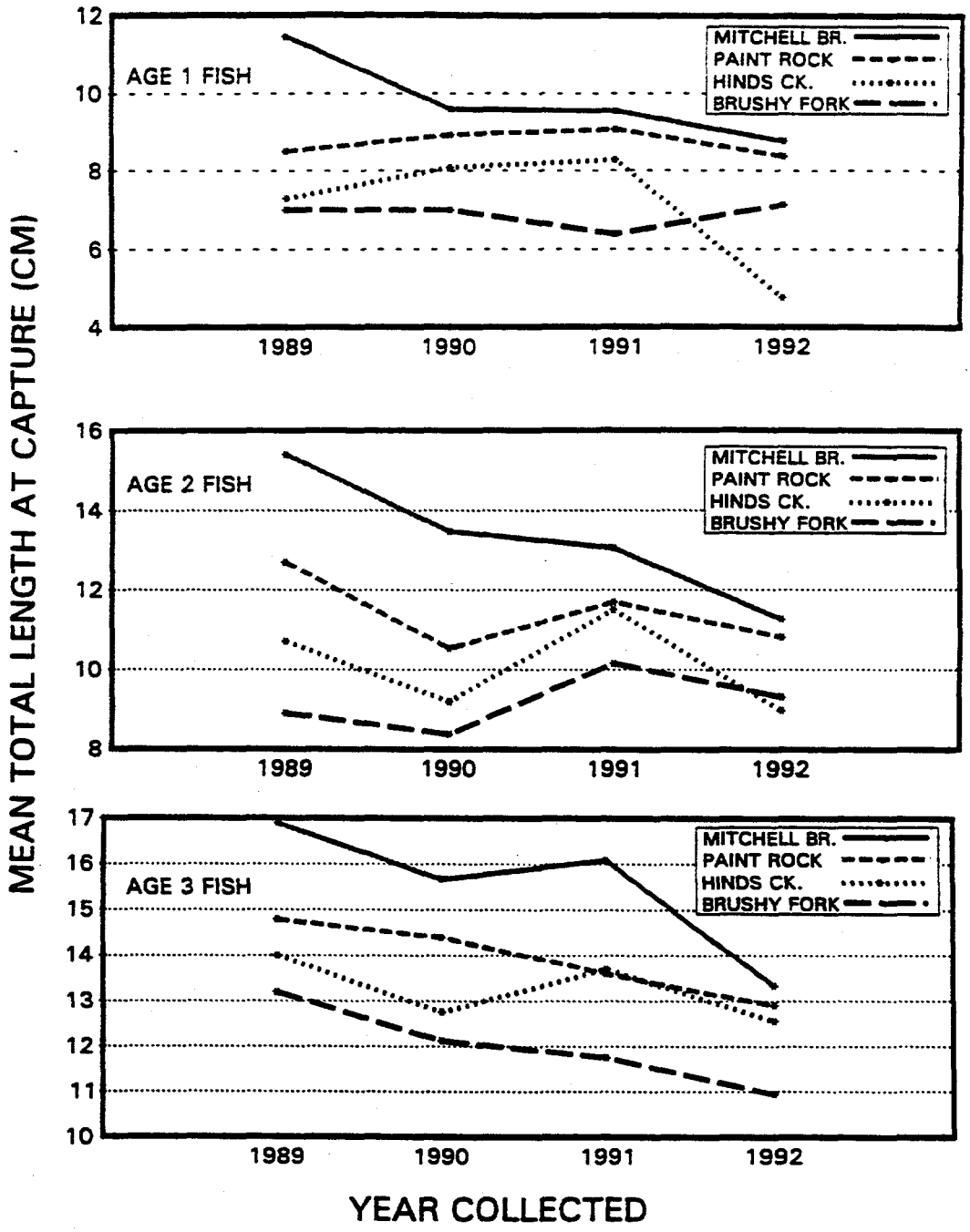

Fig. 5.7. Mean total length at capture (in centimeters) for redbreast sunfish collected in Mitchell Branch from 1989 to 1992.

Table 5.3. Comparison of mean total length of redbreast sunfish at capture

Means with different alphabetical characters are significantly different from each other ($p = 0.05$)

Site	N	Mean	Tukey test result
		Age 1	
Mitchell Branch	34	8.76	a
Paint Rock Creek	19	8.37	a
Hinds Creek	5	4.76	b
Brushy Fork	3	7.13	a
		Age 2	
Mitchell Branch	74	11.26	a
Paint Rock Creek	40	10.81	a
Hinds Creek	22	8.97	b
Brushy Fork	14	9.30	b
		Age 3	
Mitchell Branch	19	13.34	a
Paint Rock Creek	22	12.91	a
Hinds Creek	9	12.57	ab
Brushy Fork	48	10.94	b

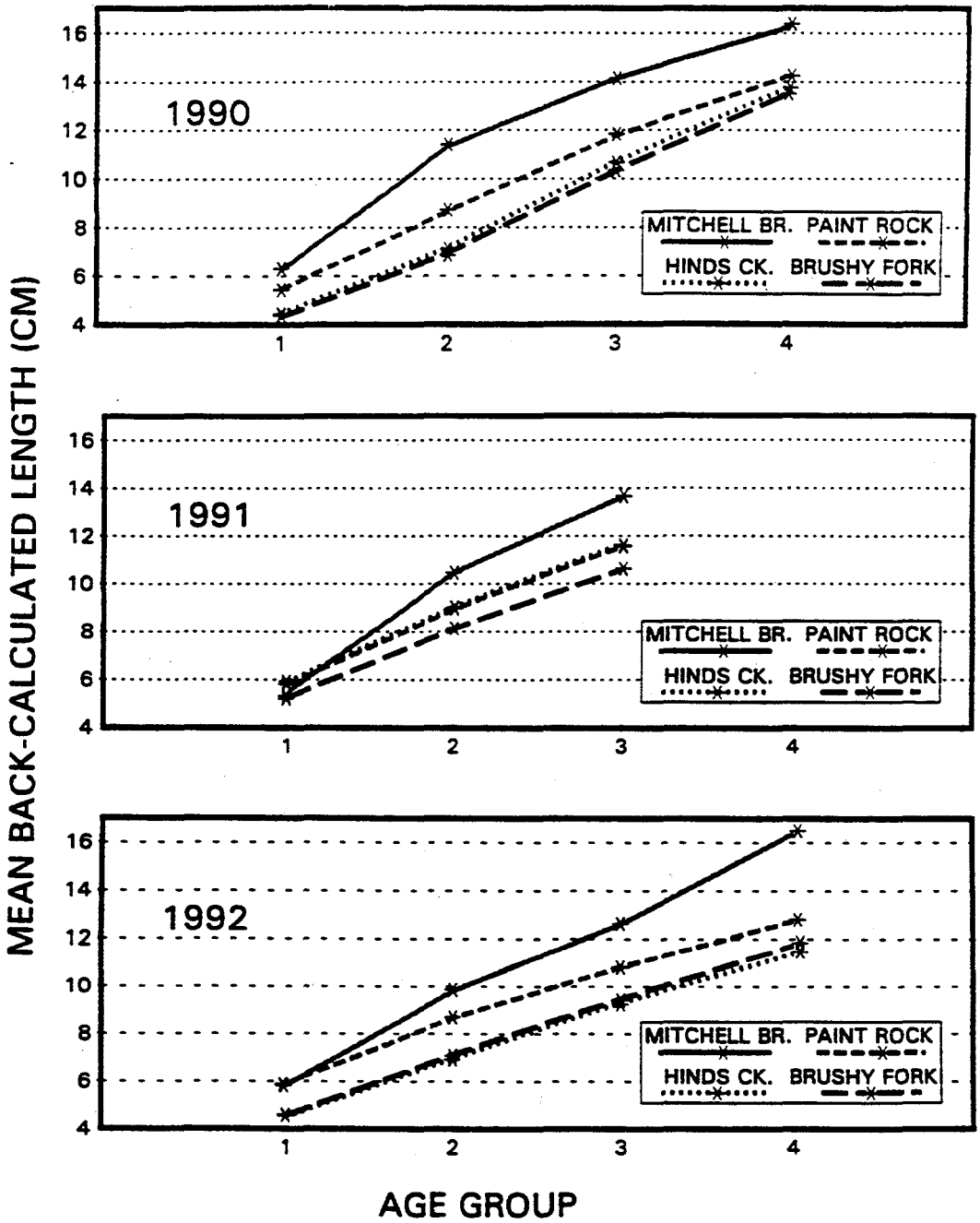

Fig. 5.8. Back-calculated length (in centimeters) at age for redbreast sunfish collected in Mitchell Branch from 1990 to 1992.

Growth is known to be affected by population density, food availability, habitat quantity and quality, or stream productivity. The interactions of these factors, among others, are the principal determinants of growth rates. The decline in TL at capture of Mitchell Branch redbreast sunfish from 1989 to 1992 was probably due to several variables. In 1989 fish abundance was low, resulting in decreased intra- and inter-specific competition for food and habitat. Growth rates were probably enhanced during this period of decreased competition. Adams et al. (1992a) found a similar pattern in growth of fish in the Pigeon River where competition was low. As the population density increased in Mitchell Branch, competition was probably increased, resulting in slower growth. Unlike the reference sites, Mitchell Branch may lack the necessary flow and substrate for good production of aquatic invertebrates. Studies by Smith (1994) in sections further upstream in Mitchell Branch indicate that the macroinvertebrate fauna is degraded in these areas. Instream conditions such as flow, food production and water quality may also have affected the growth of redbreast sunfish in Mitchell Branch as population numbers increased in later years.

5.2 REPRODUCTIVE EFFECTS SUBTASK *(M.S. Greeley, Jr.)*

5.2.1 Introduction

A major objective of the K-25 Site BMAP is to demonstrate that the NDPES effluent limitations for the site protect and maintain the classified use of Mitchell Branch for the growth and propagation of fish and other aquatic life. To directly examine the effects of K-25 Site activities on fish propagation in Mitchell Branch and Poplar Creek, a reproductive effects subtask was initiated in 1993 as part of the Fish Health Assessment Task of the new K-25 Site BMAP.

Fish reproduction involves a complex series of processes including gamete production, spawning and fertilization, and embryonic development and larval survival. This subtask assesses fish reproductive competence at a number of biological levels, including internal regulatory mechanisms (serum reproductive steroid concentrations), gamete quantity and quality (fecundity, oocyte atresia or death) and embryonic and early larval development (fish embryo-larval toxicity tests).

5.2.2 Methods

5.2.2.1 Fish sampling and processing

As with the bioindicator subtask, the redbreast sunfish is the primary sentinel species for the instream monitoring of fish reproductive competence in Mitchell Branch. Detailed descriptions of the study sites and basic fish processing procedures for the entire Fish Health Assessment Task are provided elsewhere in this report (Sect. 5.1). Female sunfish were collected by electroshocking from lower Mitchell Branch and three reference streams in spring 1993 immediately prior to the breeding season. Fish were bled for the measurement of circulating concentrations of reproductive hormones. Total body lengths and weights were recorded, and following sacrifice of the fish by spinal ligation, gonads were removed and weighed. A gonadal-somatic index (GSI) was determined for each fish: GSI = [gonad weight (g)/total body weight (g)] × 100. Weighed pieces of ovarian tissue were transferred to 20-mL glass scintillation vials containing 10-mL Karnovsky's fixative for quantitative analysis of ovarian condition and fecundity.

5.2.2.2 Gonadal assays

Morphometric reproductive analyses were conducted on the weighed subsamples of sunfish ovarian tissue by methods described in Hinzman et al. (1992). The developmental stages of oocytes were recorded, and the presence of any atretic (dead or nonviable) oocytes noted. Results were expressed as the number of oocytes of a certain size-class or stage per gram of fish body weight. The "batch" fecundity, or number of eggs to be released during the first spawn of the breeding season, was calculated from the number of oocytes present in the leading clutch of synchronously developing oocytes.

5.2.2.3 Radioimmunoassays

Radioimmunoassay (RIA) is a procedure for measuring minute amounts of a substance in biological samples. Radioimmunoassay techniques for measuring 17β-estradiol (E2) and testosterone (T) in redbreast sunfish plasma and serum were modified from Greeley et al. (1988). A detailed description of the RIA as conducted in this laboratory can be found in Hinzman et al. (1992). For quality control purposes, aliquots of plasma from previously assayed pools were run with each set of samples. Within-assay variation and between-assay variation were consistently less than 20%.

5.2.2.4 Fish embryo-larval toxicity tests

The Japanese medaka (*Oryzias latipes*), a small fish native to southeastern Asia, was employed to examine the effects of water from Mitchell Branch and Poplar Creek on fish embryo-larval development. This species has several advantages for use in laboratory developmental toxicity testing, including ease of maintenance in the laboratory, year-round availability of eggs, a long embryonic period with a resultant increase in sensitivity to many xenobiotics, and a clear embryo which facilitates observations of developmental abnormalities prior to hatching. Medaka are currently being employed in the BMAP for examining the effects on fish embryo-larval development of water from EFPC and WOC on the ORR, and in the Clinch River Environmental Restoration Program for examining the developmental toxicity of water from Poplar Creek and the Clinch River.

Procedures associated with the medaka embryo-larval test are described in detail elsewhere (Ashwood et al. 1993). Briefly, recently fertilized eggs from 20 to 30 female medaka were pooled, randomized, and then assigned to treatment groups that included grab samples of ambient water sources or laboratory controls. Embryos were examined daily under a dissecting microscope for developmental abnormalities and scored for survival. Water renewal and changes through the first 7 days of a test followed the protocol of the ESD Toxicology Laboratory (see Sec.3.1.2); thereafter, water changes were conducted every other day using water from the final grab sample that was stored and refrigerated in the dark. All embryos were monitored until death, to 48 hours post-hatch, or through 21 days post-fertilization. Study sites for medaka embryo-larval testing included several locations along Poplar Creek (see Sec. 3.1.2), as well as sites along Mitchell Branch upstream (MIK 1.43) and downstream (MIK 0.12) of the major K-25 Site discharges into this stream.

5.2.2.5 Statistical analysis

ANOVA was used to test for site effects related to the various reproductive parameters. If the ANOVA rejected a multisample null hypothesis of equal means, then Tukey's test (Zar 1984) was used to test for significant differences in response parameters at sites. The significance level for rejecting the hypothesis of equal means between sites was set at $p = 0.05$. Tests for homogeneity of variance of individual response variables between sites were conducted using Levene's test (Sokal and Rohlf 1981), an F-distribution test that compares the ratios of the variances from two independent sample populations. Results of the

medaka embryo-larval tests were examined by Chi-square analysis, with significant differences ($p = 0.05$) between sites determined by comparison of sample proportions as described in Daniel (1987).

5.2.3 Results and Discussion

5.2.3.1 Instream reproductive effects

Six reproductive indicators, representing several different levels of reproductive organization, were examined in female redbreast sunfish sampled from Mitchell Branch and three reference sites prior to the beginning of the 1993 breeding season (Table 5.4). The first of these parameters, the GSI, is a measure of the size of the sunfish ovary relative to body size and an indirect indicator of gamete quantity. The GSI indicated that the ovaries in sunfish from Mitchell Branch were significantly ($p < 0.05$) larger than the ovaries of fish from each of the three reference sites. Along with the GSI, determinations of fecundity and the abundance of vitellogenic or yolk-laden oocytes in ovaries serve as indicators of relative reproductive effort. Both fecundity and vitellogenic oocyte abundance were also significantly higher in fish from Mitchell Branch.

Atretic oocytes are indicators of gamete quality rather than quantity and may be one of the better reproductive indicators of an actual pathological condition. Many potential environmental contaminants, including PAHs (Krarup 1969, Mattison et al. 1983, Takizawa et al. 1984) and metals such as cadmium (Mattison et al. 1983), are known to damage developing oocytes. Relatively high incidences of oocyte atresia have been observed in redbreast sunfish from both upper EFPC below the Oak Ridge Y-12 Plant (Hinzman et al. 1992) and the Pigeon River, a stream heavily impacted by effluents from a paper and pulp mill (Adams et al. 1992). Abnormally high numbers of atretic oocytes have also been observed in fish undergoing acid-induced stress (McCormick et al. 1987). The incidence of oocyte atresia were only marginally higher in Mitchell Branch than in the reference sites (Table 5.4).

Two reproductive hormones, estradiol and testosterone, were measured in prespawning fish sampled from the study sites. The first of these two steroidal compounds, 17β-estradiol, has the critical function in female fish of regulating the production of yolk proteins by the liver for incorporation into vitellogenic oocytes. The primary role of testosterone in female fish is less clear, although it serves as a precursor in the production of 17β-estradiol and may also function in regulating spawning behavior (Greeley et al. 1988). The serum levels of one

Table 5.4. Comparision of reproductive bioindicators in female redbreast sunfish, *Lepomis auritus*, collected from Mitchell Branch and several reference streams at the beginning of the 1993 reproductive season.

Asterisks identify values at the reference sites which differ significantly ($\alpha = 0.05$) from Mitchell Branch

Site	Date	n	GSI[a]	Atretic[b] oocytes	Vitellogenic[b] oocytes	Fecundity[b]	Testosterone[c]	Estradiol[c]
Mitchell Branch	5/26/93	16	17.0±0.4	0.4±0.1	113.0±3.3	75.5±3.4	4.91±10.06	1.76±0.41
Hinds Creek	6/8/93	12	12.9±0.9*	0.2±0.2	77.7±5.0*	47.5±3.4*	6.59±2.52	4.13±0.71
Brushy Fork	6/7/93	16	9.9±0.6*	0.0±0.0*	63.6±3.9*	45.0±3.1*	8.00±1.65	6.12±1.39*
Paint Rock Creek	5/27/93	10	7.9±0.5*	0.3±0.1	59.5±3.1*	39.0±2.8*	2.88±0.85	2.63±0.40

[a] GSI = gonadal-somatic index
[b] Oocytes or eggs per gram of body weight
[c] Steroid concentration in ng per ml of serum

or both of these steroids have been shown to decrease in fish following laboratory or environmental exposure to a number of potential environmental pollutants, including PCBs (Sivarajah et al. 1978, Thomas 1990), aromatic hydrocarbons (Johnson et al. 1988), paper and pulp mill effluents (McMaster et al. 1991, Munkittrick et al. 1992), lead (Thomas 1990), cyanide (Ruby et al. 1993), organochlorine or organophosphorus pesticides (Singh and Singh 1987), and crude petroleum (Truscott et al. 1983). Plasma levels of both steroids tended to be lower at Mitchell Branch than the reference sites (Table 5.4), although only in the case of Brushy Fork and estradiol was there a significant difference between Mitchell Branch and any of the sites.

The integrated responses of the various reproductive indicators were examined by canonical discriminant analysis (Fig. 5.9). The overall reproductive condition of the redbreast sunfish population in Mitchell Branch was observed to differ significantly from the reproductive conditions of populations in each of the three reference sites, as shown both graphically (Fig. 5.9) and statistically (Table 5.5). Collectively, the results of both the integrative analysis and the individual reproductive responses, particularly the increased fecundity and abundance of vitellogenic oocytes in fish from Mitchell Branch, clearly demonstrate that, in the spring of 1993, redbreast sunfish in Mitchell Branch expended significantly more energy for reproduction than did the reference populations.

5.2.3.2. Embryo-larval tests for developmental toxicity

The successful propagation of fish populations requires that adults be capable of producing viable gametes and successfully completing the mating or spawning act and that offspring be able to develop and grow normally into subsequent reproductive cohorts. Beginning in the summer of 1993, water samples from several sites along Poplar Creek and two sites in Mitchell Branch were tested for their effects on fish developmental processes with a quarterly series of medaka embryo-larval tests (Table 5.6).

Embryos raised in control solutions had a suitably high survival rate during this series of toxicity tests (Table 5.6). Water samples from the headwaters of Mitchell Branch, upstream of any waste discharges from the K-25 Site, consistently yielded similarly benign results in the medaka embryo-larval tests. In contrast, water samples from lower Mitchell Branch (MIK 0.12) were nearly as consistent in their lethal effects on medaka embryos. In three out of the four tests conducted to date on water from these sites, embryo survival was significantly

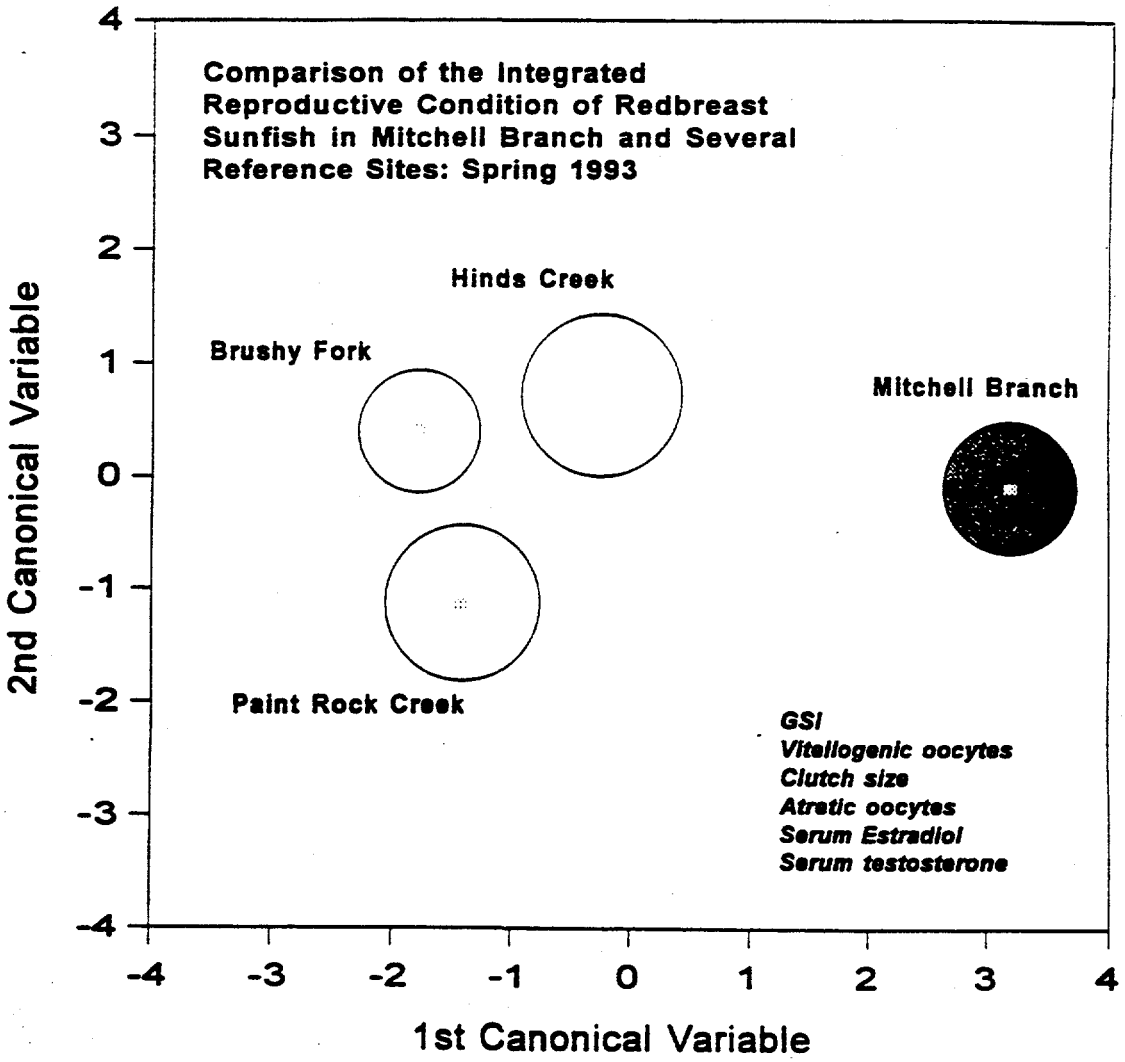

Fig. 5.9. Segregation of integrated reproductive responses of female redbreast sunfish collected in 1993 prior to the onset of the reproductive season. Circles represent site means and the reproductive approximate 95% confidence radii of that site-year means. Parameters contributing to the analysis are listed in the lower right portion of the figure. GSI = Gonadal-somatic index.

Table 5.5. Mahalanobis' distances and associated probability values for site-to-site comparisions of the integrated reproductive condition of female redbreast sunfish sampled from Mitchell Branch and various reference sites at the beginning of the 1993 reproductive season

Sites	Mitchell Branch	Hinds Creek	Brushy Fork	Paint Rock Creek
Mitchell Branch	0 $p<1.0000$	13.05 $p < 0.0001$	24.97 $p < 0.0001$	22.39 $p < 0.0001$
Hinds Creek	13.05 $p<0.0001$	0 $p < 1.000$	3.34 $p < 0.0290$	4.99 $p < 0.0080$
Brushy Fork	24.97 $p<0.0001$	3.34 $p < 0.0290$	0 $p < 1.000$	2.81 $p < 0.0461$
Paint Rock Creek	22.39 $p<0.0001$	4.99 $p < 0.0080$	2.81 $p < 0.0461$	0 $p < 1.000$

Table 5.6. Results of quarterly medaka embryo-larval toxicity tests of ambient water samples from Mitchell Branch and Poplar Creek, 1993-1994

		Survival (%)			
Sample	Conc. (%)	July 1993	October 1993	January 1994	April 1994
Control 1 (dH_2O)	100	91	83	92	96
Control 2 (ERM)	100	86	---	---	---
Control 3 (DMW)	100	---	---	96	87
MIK 1.43	100	96	100	100	100
MIK 0.12	100	23*	9*	72	28*
PCK 18.2	100	4*	40*	100	92
PCK 8.2	100	0*	20*	92	92
PCK 6.3	100	4*	8*	80	84
PCK 4.7	100	9*	20*	63*	92
PCK 1.6	100	76	12*	36*	84

Note: dH2O = distilled water, ERM = Medaka Embryo Rearing Medium, DMW = degassed mineral water, MIK = Mitchell Branch kilometer and PCK = Poplar Creek kilometer. Asterisks identify results that are significantly different from control 1 ($p = 0.05$)

($p < 0.05$) reduced as a result of exposure during development to water from this location. During the course of the fourth test, survival was also reduced, but not to the extent seen in other tests. These results clearly indicate that Mitchell Branch becomes toxic to the embryos of at least certain fish species during its passage through the K-25 Site.

The results of the medaka embryo-larval tests conducted on Poplar Creek are less consistent and more difficult to explain (Table 5.6). In the first test conducted for this project (in July 1993), embryo survival was extremely poor in water from all Poplar Creek sites, with the exception of PCK 1.6 near the confluence of Poplar Creek and the Clinch River. The extent to which activities at the K-25 Site contributed to the embryotoxic effects of water in lower Poplar Creek cannot be determined from this data, because water from the Poplar Creek reference site (PCK 18.2) located well upstream of any influences from either the K-25 Site or any other DOE facility proved in this test to be as or more toxic than water from sites closer to ORR. Potential upstream sources for toxic substances in the Poplar Creek system include wastes from a sewage treatment plant in Oliver Springs, Tennessee (which was undergoing major renovation and construction activities during the time period of these investigations), agricultural activities, domestic wastes, and urban runoff.

Similar results were obtained from a medaka embryo-larval test conducted in October 1993 (Table 5.6). In this instance, embryo survival was somewhat better at the instream reference site than during the previous July test and was further reduced at the study sites adjacent to or downstream of ORR. By January there was 100% survival of embryos raised in water from the upstream reference site, with significant toxicity developing only at the furthermost downstream study sites. The results of the latest medaka embryo-larval test, conducted in April 1994, indicate a near total absence of a toxic effect at any of the lower Poplar Creek study sites. Potential explanations for the highly variable results of medaka embryo-larval tests on water from Poplar Creek include intermittent toxic inputs from upstream sources such as farms, seasonal and weather-related climatic effects on water quality in the stream, and the complicating effects of irregular water flow into and out of Poplar Creek due to TVA practices for managing water levels in their reservoirs (including Watts Bar Lake) and generating electricity at nearby Melton Hill Dam.

5.2.4 Conclusions

Redbreast sunfish in Mitchell Branch appear to be reproductively competent, producing an over-abundance of apparently viable gametes and having a very high relative fecundity. However, it is this abnormally high expenditure of resources for gamete production that distinguishes the reproduction characteristics of the sunfish population in Mitchell Branch from reference populations (Fig. 5.9). The enhancement of gamete production in Mitchell Branch sunfish could be a stress response to xenobiotics in the diet or water, a result of nutrient enrichment in the stream, the consequence of altered temperature regimes, or the result of lessened competition from the absence of pollutant sensitive fish species. Most likely, a combination of several factors shares responsibility for the observed deviations from the apparent reproductive norm for this species.

The current results of an ongoing quarterly series of medaka embryo-larval tests of water from Mitchell Branch and Poplar Creek clearly indicate that lower Mitchell Branch is often very toxic to medaka embryos. This observation may well explain, at least in part, why fish species known to be relatively sensitive to pollutants have so far failed to establish breeding populations in the lower reaches of this stream. The results of embryo-larval tests conducted on water from Poplar Creek have been more variable and suggest additional sources of toxic inputs into the creek well upstream of the ORR.

5.2.5 Future Studies

Redbreast sunfish will continue to be sampled annually from lower Mitchell Branch to monitor the relative reproductive health of this population. Future studies along these lines will expand to include a closer analysis of the reproductive condition of male sunfish in this location and possibly include a reproductive examination of other fish species in the stream.

As a consequence of consistent findings of significant embryotoxic effects in water samples from the lower reaches of Mitchell Branch, medaka studies will be expanded to include additional reaches of the stream closer to the potential sources of the toxic effect. Similar experiments will also be conducted intermittently with the embryos of other fish species to address questions concerning the ecological relevance and applicability to other species of the medaka embryo-larval tests. In addition, studies are planned to identify the substance(s) in the stream water that are responsible for the observed embryotoxic effect.

6. INSTREAM MONITORING (TASK 4)

J.G. Smith and E. M. Schilling

The objectives of the instream ecological monitoring task are to (1) characterize spatial and temporal patterns in the distribution and abundance of the benthic macroinvertebrate and fish populations in Mitchell Branch and (2) document the effects of new pollution abatement facilities on community structure and function. This task consists of two components: (1) benthic macroinvertebrate community studies (Subtask 4a) and (2) fish population studies (Subtask 4b); results to date for these studies are presented in Sects. 6.1 and 6.2 respectively.

6.1 BENTHIC MACROINVERTEBRATES (*J. G. Smith*)

6.1.1 Introduction

During the initial 18 months (August 1986–January 1988) of the K-25 Site BMAP, results of the benthic macroinvertebrate community studies showed that, relative to the upstream reference site (MIK 1.43), the portion of Mitchell Branch immediately adjacent to the main K-25 Site was significantly impacted (Smith 1993; Smith 1994). Greatest impact was evident at the site (MIK 0.71) just downstream of SD 170 where several major taxonomic groups of invertebrates were conspicuously absent [e.g., Ephemeroptera, Plecoptera, Trichoptera (EPT)]. Some improvement in the condition of the macroinvertebrate community was evident at the two sites located downstream of MIK 0.71 (sites MIK 0.54 and MIK 0.45 located below SD 180 and SD 190 respectively), but major taxonomic groups were absent from these sites as well, thus, indicating these two sites were also impacted.

Studies of the benthic macroinvertebrate community continued after January 1988 with the specific objectives of (1) spatially evaluating the benthic macroinvertebrate community of Mitchell Branch for evidence of impacts associated with operations at the K-25 Site, (2) continuing efforts to help identify causal factors responsible for any observed adverse impacts and (3) documenting the response of the macroinvertebrate community to any new pollution abatement activities or remedial actions. This report summarizes the results obtained through April 1993.

6.1.2 Materials and Methods

The monthly sampling regime initiated in August 1986 continued through July 1988; after July 1988, the sampling frequency was reduced to once per quarter. Benthic macroinvertebrate samples were collected from six sites in Mitchell Branch through July 1992 (Fig. 2.1); then, beginning in October 1992, two sites were dropped (MIK 0.54 and MIK 0.86). Because of the close proximity of MIK 0.86 to MIK 0.78 and of MIK 0.54 to MIK 0.45, as well as the relative similarities of the benthic communities at these site-pairs, it was felt that any major changes in the invertebrate community should be detectable at the retained sites. Dropping MIK 0.54 is further justified because the effluent from SD 180, which is just upstream of MIK 0.54, is toxic less frequently than the effluent from SD 190, which is just upstream of MIK 0.45 (Sect. 3.2). With continued toxicity testing of effluents from SD 180 and SD 190, the retention of a benthic macroinvertebrate monitoring site at MIK 0.45, which is downstream of both storm drains, will allow the detection of major changes in Mitchell Branch that may be associated with discharges from either storm drain. Likewise, dropping MIK 0.86 is supported by the fact that there are no major effluent discharges upstream of MIK 0.78; thus, the existence of a single site between MIK 1.43 and SD 170 should allow the detention of any major changes should they occur in this portion of the stream. Also, although samples were collected monthly during the first 2 years of the BMAP and quarterly after the second year, only the samples collected during the spring (April) and fall (October) sampling periods after January 1988 were processed. The samples collected during the other sampling periods were archived and will be processed only if further resolution of the data is needed for future reports. Following this plan of action ensures the sample processing remains up to date without compromising the results from this task.

During each sampling period, three random benthic macroinvertebrate samples were collected from a riffle at each site with a Surber stream bottom sampler (0.09 m^2 or 1 ft^2; 363-μm-mesh net). Samples were placed into pre-labeled, glass jars and preserved in 80% ethanol; the ethanol was replaced with fresh ethanol within 1 week. Additional details of the field and associated quality assurance (QA) procedures followed in the collection and maintenance of these samples are given in Smith (1992).

Supplemental data on water quality and physical characteristics of each site were recorded at the time of sampling. Temperature, conductivity, dissolved oxygen, and pH were measured with an Horiba Model U-7 Water Quality Checker. Water depth, location within the riffle

area (distance from permanent headstakes on the stream bank), visual determination of relative current velocity (very slow, slow, moderate, or fast), and substrate type (visual determination), based on a modified Wentworth particle size scale (Loar et al. 1985), were recorded for each sample. All measurements/data for water quality and stream characteristics were obtained in accordance with established QA procedures (Smith 1992). The supplemental data were used only if needed for observational support.

Benthos samples were washed in the laboratory after being placed into a U. S. Standard No. 60-mesh (250-µm-mesh) sieve. Small aliquots of the sample were removed from the sieve, placed into a white tray containing water, and then the organisms were removed from the sample debris with forceps. This was repeated until the entire sample was sorted. Finally, the removed organisms were identified to the lowest practical taxon and enumerated. Further details of the laboratory procedures followed in order to process benthos samples may be found in Wojtowicz and Smith (1992).

Data were managed and analyzed on computer primarily with SAS software and procedures (SAS 1985a; SAS 1985b). Statistical analysis included both descriptive and parametric statistical techniques. Parametric procedures included ANOVA models on three responses: density (total number of organisms/0.1 m^2), total richness (total number of taxa/sample) and the total combined number of the EPT taxa (total number of EPT taxa/sample or EPT richness). These responses (density and total richness) were selected because they are a good, general indicator of the overall condition of a macroinvertebrate community and a more specific indication (EPT richness) of the condition of a macroinvertebrate community based on taxa that are generally intolerant of most types of pollution (Lenat 1988). Prior to performing the ANOVAs, observed values for each response were appropriately transformed [i.e., $\log_{10}(X+1)$ for density values, and the square root of X for total and EPT richness values, where X = the individual observed values for density, taxonomic richness, and EPT richness; Elliott 1977]. Statistical analysis with the ANOVAs ineluded two approaches that provided a step-wise evaluation of the data to try to detect temporal changes at sites previously identified as impacted (relative to MIK 1.43, the reference site), which may have been indicative of eithe improvements or further degradation. For each approach, a significance level of $p < 0.05$ was used. Although monthly data were available from the first 18 months of the BMAP, only the April (spring) and October (fall) data were used from this time period in order to provide a data set that was uniform with the data

available after this time period. Additionally, because MIK 0.54 and MIK 0.86 were limited to only 6 years of data (October 1986 through April 1992), all analyses that included these two sites were limited to data from the first 6 years only.

The first approach was a nested ANOVA model that analyzed the data for site differences (fixed effect) among the sampling years (fixed effect) with seasonal (i.e., April = spring and October = fall; random effect) differences nested within sampling year. This approach provided (1) an overall evaluation of the differences in mean responses across the sampled sites and sampling years, (2) an evaluation of the interaction between the site and sampling year effects, (3) an overall evaluation of the variation in the response from the spring to fall sampling period within a sampling year, and (4) a test of whether the variation in (3) changed significantly from site to site.

For the second approach, a simple linear regression was used to estimate site-specific linear trends for each response over the study period on season-specific data. For this approach, the slopes of the regression lines for all sites were compared by season for parallelism with an ANOVA. This allowed the data for the potentially impacted sites to be evaluated, relative to MIK 1.43, with respect to linear trends within each season that may have been indicative of persistent improvement (i.e., increasing values through time) or degradation (i.e., decreasing values through time). In addition, because the variability around the regression lines was not the same for all sites (an assumption of this technique), which reduced the ability to detect significant trends, the analysis was repeated for all possible pairwise comparisons of each downstream site with MIK 1.43.

6.1.3 Results

6.1.3.1 Taxonomic composition

A checklist of the benthic macroinvertebrates collected from each site on Mitchell Branch is presented in Appendix D, Table D.1. The collection of macroinvertebrates during only two seasons within each year over a 7-year period does not allow the compilation of a complete list of taxa from each site because the occurrence of many other taxa is limited to seasons of the year that were not sampled. However, the presence or absence of common taxa or major taxonomic groups from this limited number of sampling periods does provide useful information for assessing the overall well-being of the benthic macroinvertebrate community. Most major groups of macroinvertebrates typical of streams (e.g., Hynes 1970) were

represented in Mitchell Branch by one or several taxa during the 7 years of study, including the Oligochaeta (aquatic worms), Amphipoda (sideswimmers/scuds), Decapoda (crayfish), Hydracarina (mites), Bivalvia (mussels/clams), Ephemeroptera (mayflies), Plecoptera (stoneflies), Trichoptera (caddisflies), Odonata (dragonflies/damselflies), Megaloptera (dobsonfiles/fishflies), Coleoptera (beetles), and Diptera (true flies). Not only were spatial trends obvious in the occurrences of taxa within each of these major groups, temporal trends were obvious as well. Particularly notable was the absence or sporadic appearance of many of the taxa within certain groups at some sites downstream of MIK 1.43, especially from within groups such as the EPT (mayflies, stoneflies and caddisflies respectively). No ephemeropterans were collected at the three sites downstream of MIK 0.78 prior to the fifth year of sampling (i.e., October 1990), and the number of epemeropteran taxa present at MIK 0.78 and MIK 0.86 was considerably limited prior to the third year (i.e., October 1988). Plecopterans were not collected downstream of MIK 0.78 prior to the fifth year, and none were collected from MIK 0.54 during the entire study. At MIK 0.78 and MIK 0.86, most plecopteran taxa did not appear in samples until the second or third years. The few trichopterans sporadically collected from the three sites downstream of MIK 0.78 did not appear in the samples until after the third year, and most of the trichoptera taxa collected from MIK 0.78 and MIK 0.86 were not collected until the fourth year.

6.1.3.2 Abundance

Density. Total community density changed considerably through time at all sites (Fig. 6.1). Density at MIK 1.43 changed least during the study, with the highest (April 1989) and lowest (October 1986) values differing by a factor of 7.2×; a distinct pattern of seasonal changes was not obvious at this site. In contrast, densities at the five other sites exhibited much greater fluctuations during the study. The magnitude of changes occurring during the study at MIK 0.45 and MIK 0.54 were similar, where the highest (April 1990, both sites) and lowest (October 1990, both sites) density values differed by a factor of approximately 85×. Seasonal changes between these two sites, however, were different. At MIK 0.54 after April 1988, spring highs were generally followed by fall lows, while at MIK 0.45 no consistent seasonal pattern was evident. The magnitude of changes that occurred at MIKs 0.78 and 0.86 were also similar, with the highest (April 1989, both sites) and lowest (October 1986, both sites) density values differing by a factor of about 250×. These two sites showed distinct

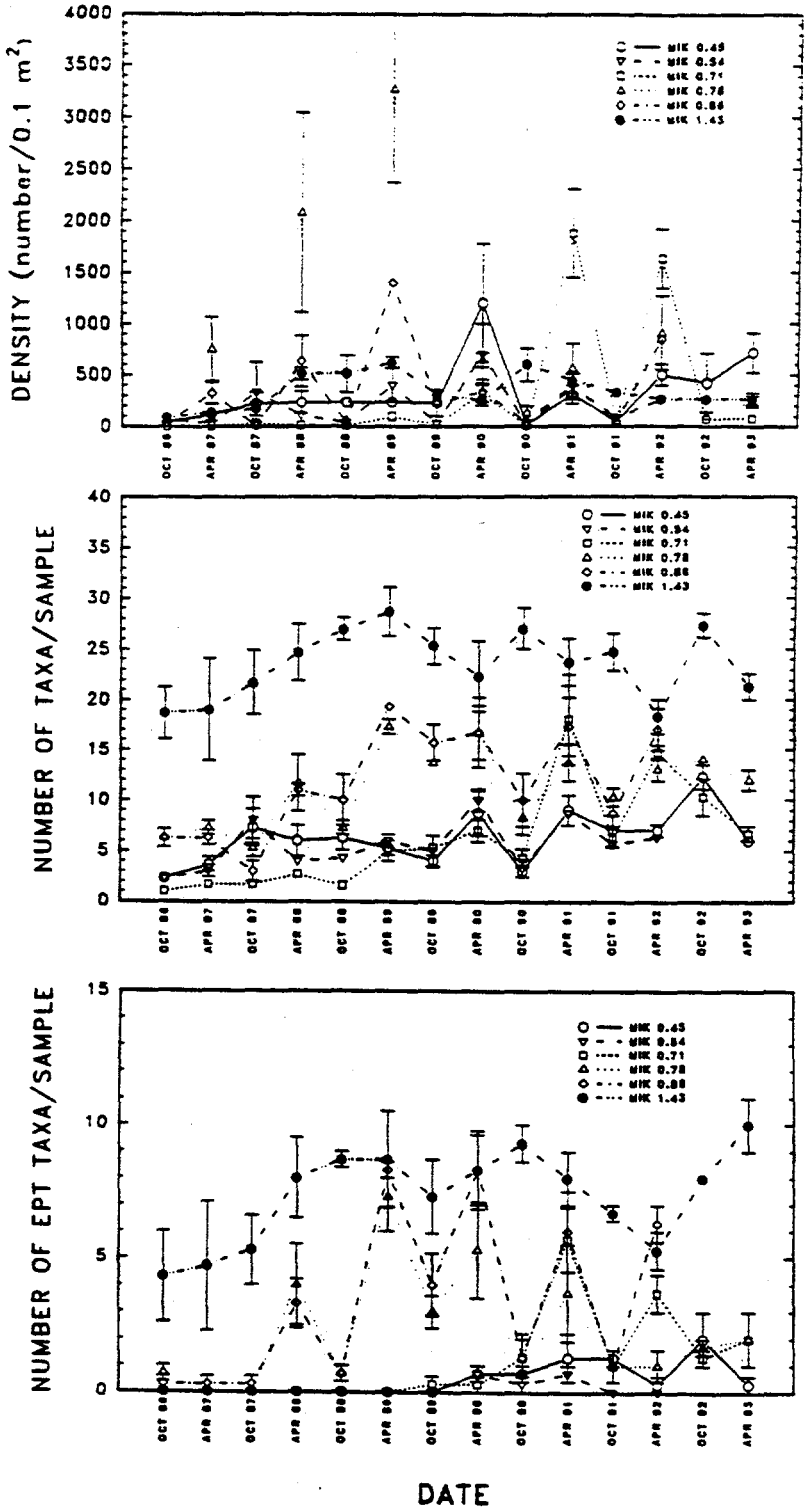

Fig. 6.1. Mean total density, mean total richness, and mean richness of the Ephemeroptera, Plecoptera, and Trichoptera (EPT richness) of the benthic macroinvertebrate communities in Mitchell Branch, October 1986–April 1993. Vertical bars represent ± SE. MIK = Mitchell Branch kilometer.

seasonal patterns in density, with spring highs followed by fall lows. The greatest change occurred at MIK 0.71 where the lowest (October 1986) and highest (April 1991) density values differed by a factor of 1338×. Density values at MIK 0.71 were generally highest during the spring sampling periods, although this pattern did not consistently hold.

Considerable spatial differences in mean density were also evident during each sampling period (Fig. 6.1). The lowest densities were most frequently observed at MIK 0.71 (9 of 14 sampling periods); however, densities were also higher at MIK 0.71 than at all other sites during 2 of the sampling periods (April 1991 and April 1992). The greatest difference (301.6×) among the sites between the highest (MIK 0.78) and lowest (MIK 0.71) values occurred in April 1987, and the least difference among the sites occurred in April 1990 when the factor of difference between the lowest (MIK 1.43) and highest (MIK 0.45) values was only 4.7×.

Seasonal changes among two or more of the sites were significantly different across 2 or more years whether the data were analyzed for all six sites from the first 6 years only or only 4 sites from all 7 years (Table E.1, Appendix E). The regression analyses further demonstrated that the long term trends of change occurring between two or more sites were different in both seasons (Table E.2). Pairwise comparisons of regression slopes indicated that only the patterns of change that occurred at MIK 0.54 and MIK 0.71 were significantly different from the pattern of change at MIK 1.43 (Table E.2; Figs. F.1 and F.2). The regression lines for MIK 0.54 and MIK 1.43 exhibited opposite trends in each season (Fig. F.1), while the regression lines for MIK 0.71 and MIK 1.43 exhibited opposite trends in the spring only (Fig. F.2).

Relative abundance. In general, the macroinvertebrate community at each site was composed predominantly of chironomids, oligochaetes, ephemeropterans, plecopterans, and/or trichopterans (Fig. 6.2). The chironomids were consistently one of the most abundant groups at all sites. Although considerable temporal fluctuations occurred in their relative abundances, they generally accounted for 20% or more of the total density at most sites during most sampling periods. A slight yet notable deviation from this trend occurred at MIK 0.71 where chironomids were absent from samples collected during two of the first three sampling periods. After the October 1988 sampling period, they accounted for at least 30% of the total density during the remainder of the study at MIK 0.71.

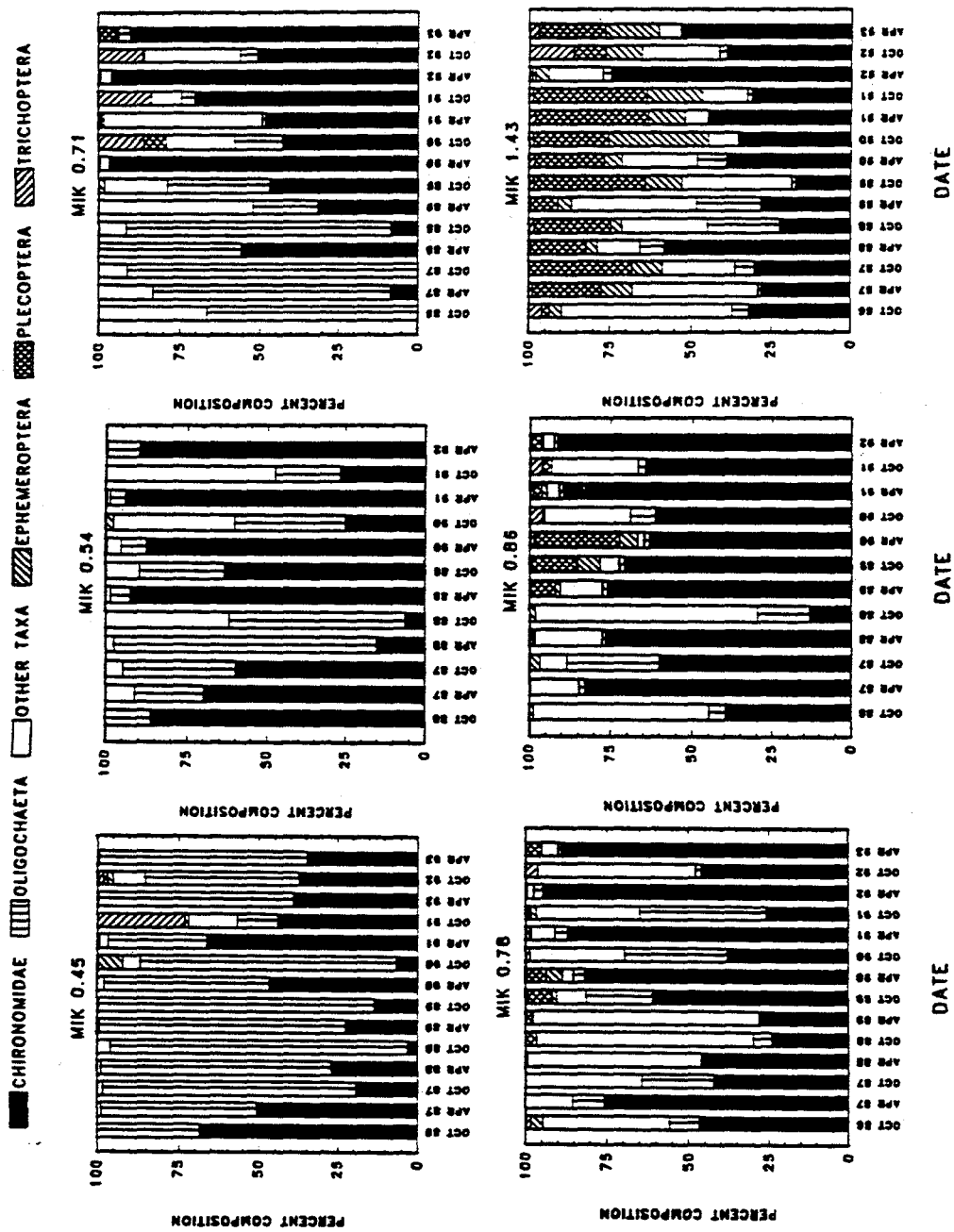

Fig. 6.2. Mean percent composition (i.e., percentage of total density) of selected benthic macroinvertebrate taxonomic groups in Mitchell Branch, October 1986–April 1993. MIK = Mitchell Branch kilometer.

The Oligochaeta were most prominent downstream of MIK 0.78, particularly at MIK 0.45 where they accounted for over 30% of the total density during almost every sampling period (Fig. 6.2). The relative abundances of the oligochaetes at MIK 0.54 were generally much less than at MIK 0.45 but they still usually accounted for more than 10% of the total density. At MIK 0.71 the oligochaetes constituted over 45% of the total density during the first five sampling periods; but after October 1988, they generally made up much less than 25% of the total density. At MIKs 0.78, 0.86, and 1.43, the relative abundance of the oligochaetes was highly variable, but they usually accounted for <10% of the total density.

Except at MIK 1.43, collections of Ephemeroptera, Plecoptera, and Trichoptera were sporadic, particularly downstream of MIK 0.78 where their individual or combined relative abundances rarely exceeded 1% of the total density (Fig. 6.2). Even though they were collected more frequently at MIK 0.78 and MIK 0.86 than at the three downstream most sites, the individual relative abundances of the ephemeropterans, plecopterans, and trichopterans usually did not exceed 5% of the total density, and their combined relative abundances usually did not exceed 10%. In contrast, the relative abundances of the Ephemeroptera and Plecoptera at MIK 1.43 usually exceeded 5% and often exceeded 20%. Furthermore, the combined relative abundance of the ephemeropterans, plecopterans, and trichopterans usually exceeded 30% of the total density at MIK 1.43.

6.1.3.3 Richness

Total richness. Total richness (i.e., total number of taxa/sample), like density, changed considerably through time at each site, although the magnitude of change was much less (Fig. 6.1). Also as for density, the greatest and least change in total richness occurred during the study at MIK 0.71 and MIK 1.43 respectively; the magnitude of difference between the highest (April 1991) and lowest (October 1986) values at MIK 0.71 was 18.0×, while the magnitude of difference between the highest (April 1989) and lowest (April 1992) values at MIK 1.43 was only 1.5×. The magnitude of differences between the highest and lowest total richness values during the study at the other four sites ranged from 2.7× at MIK 0.78 to 6.4× at MIK 0.86. Only MIK 0.78 and MIK 0.86 exhibited consistent and distinct seasonal patterns of change, with spring increases followed by declines in the fall.

Total richness remained distinctly higher at MIK 1.43 than at all other sites during all sampling periods (Fig. 6.1). The greatest difference between the highest and lowest values

occurred in October 1986 when total richness at MIK 1.43 was 18.7× higher than at MIK 0.71. The least difference among the sites occurred in April 1991 when the difference between the highest (MIK 1.43) and lowest (MIK 0.54) values was only 2.7×. A notable trend occurred in spatial differences after October 1988. Prior to April 1989, the difference between the highest and lowest values was never less than a factor of 9×, but after October 1988, the difference between the highest and lowest values never exceeded 9× and exceeded 5× only three times. Similarly, prior to April 1989 total richness at MIK 1.43 was more than 2× higher than at all other sites in all sampling periods, but after October 1988, the difference between MIK 1.43 and the sites with the next highest values exceeded 2× during only two sampling periods.

As was seen for density, the pattern of seasonal change in total richness exhibited among two or more sites was not the same across all years whether the data were analyzed for all six sites for the first 6 years only, or for only four sites for all 7 years (Table E.1). The regression analysis indicated that total richness changed differently at two or more sites during the spring sampling periods in the first 6 years and during all 7 years (Table E.2). In contrast, changes in total richness across all sites during the fall sampling periods were not different in the first 6 years, but over all 7 years a significant difference existed. Changes at MIK 0.71 were significantly different from those at MIK 1.43 over the entire 7-year study period in both the spring and fall, while those at MIK 0.45 or MIK 0.78 were not (Table E.2). Changes at MIK 0.54 and MIK 0.86 were significantly different from those at MIK 1.43 during the spring only; during this season, their regression lines indicated that richness increased, while at MIK 1.43 richness exhibited a slight decline (Table E.2; Fig. F.2).

EPT richness. The amount of temporal change exhibited in the combined EPT richness of the in Mitchell Branch during the study was substantial, particularly at the sites from MIK 0.86 downstream to MIK 0.45 (Fig. 6.1). EPT taxa were not collected from MIK 0.78 during two sampling periods (April 1987 and October 1987); from MIK 0.71 during all sampling periods from October 1986 through April 1989; from MIK 0.45 during all sampling periods from October 1986 through October 1989; and from MIK 0.54 during all sampling periods except April and October 1987. EPT taxa were collected in all sampling periods from MIK 0.86 and MIK 1.43; however, mean EPT richness at MIK 0.86 was much less than 1.0 during the first three sampling periods and frequently <2.0 during the remainder of the study.

Except in April 1992, EPT richness was higher at MIK 1.43 than at all other sites throughout the study (Fig. 6.1). Following the October 1987 sampling period, the amount of difference between MIK 1.43 and MIK 0.86 and MIK 0.78 followed a strong seasonal trend, with the largest differences generally occurring during the October sampling periods. At the remaining three sites, even after EPT taxa began to consistently occur, large differences (at least 4×) generally persisted between them and MIK 1.43.

As was seen for density and total richness, the pattern of seasonal change in EPT richness exhibited among two or more sites was not the same across all years whether the data were analyzed for the first 6 years only, or only four sites were analyzed for all 7 years (Table E.1). Analyses of the regression slopes indicated that no statistically significant differences existed in the pattern of change across all sites during the fall, whether evaluated for six sites during the first 6 years only, or for only four sites for all 7 years (Table E.2; Figs F.1 and F.2). Except for a marginally significant difference between MIK 1.43 and MIK 0.45, all pairwise comparisons supported the overall site analyses (Table E.2). These results suggest that none of the changes that occurred in EPT richness at MIK 1.43 differed markedly from those at the other five sites. During the spring however, analyses of the slopes across all sites indicated that significant differences existed (Table E.2; Figs. F.1 and F.2). Pairwise comparisons of the spring data indicated that the only site that changed differently from MIK 1.43 was MIK 0.71 (Table F.2; Fig. F.1). Although the regression lines suggest that there was an overall increase in EPT richness at both sites during the spring, the increase appeared to be more substantial at MIK 0.71 over the course of the study.

6.1.4 Discussion

The benthic macroinvertebrate community of Mitchell Branch within the main boundary of the K-25 Site continued to indicate the presence of degraded water quality through April 1993. Relative to the reference site, both total and EPT richness were clearly and persistently depressed at all sites from MIK 0.86 downstream to MIK 0.45. Furthermore, the macroinvertebrate community at these sites was predominantly composed of chironomids and/or oligochaetes; and the relative abundances of the Ephemeroptera and Plecoptera, and to a lessor extent the Trichoptera, were clearly depressed, a pattern often associated with streams having degraded water quality (e.g., Hynes 1960; Wiederholm 1984).

As observed during the first 18 months of the K-25 BMAP (Smith 1993, 1994), the macroinvertebrate community of Mitchell Branch appeared to be most severely impacted at the three sites downstream of MIK 0.78. This was particularly notable prior to the April 1990 sampling period when taxa within the Ephemeroptera, Plecoptera, and Trichoptera were virtually absent from these three sites. After October 1989, the differences in the invertebrate community among the five sites downstream of MIK 1.43 became less distinct, especially at MIKs 0.71, 0.78, and 0.86.

In addition to evaluating the condition of the macroinvertebrate community in Mitchell Branch spatially for evidence of area-specific impacts, the community at the impacted sites was also evaluated for evidence of temporal changes that might indicate recovery associated with improvements in water quality. The reference site was characterized by a relatively stable and diverse macroinvertebrate community that exhibited small fluctuations in density, total richness, and EPT richness between sampling periods as well as over the entire study, a characteristic of other reference sites on the DOE ORR as well (J. G. Smith, ESD/ORNL, unpublished data). Thus, although changes did occur at MIK 1.43 during the study, they were within a normal range.

In contrast, the impacted sites were characterized by substantial fluctuations in density, total richness, and EPT richness between most sampling periods and over the course of the study; this pattern has also been observed at other impacted stream sites on the ORR (J. G. Smith, ESD/ORNL, unpublished data). The most substantial change was observed at MIK 0.71, where, with the exception of EPT richness in the fall sampling periods, changes in density, total richness, and EPT richness were significantly different from those at MIK 1.43. Substantial and persistent increases in density, total richness, and EPT richness occurred at this site, particularly between October 1988 and October 1989, although total and EPT richness remained considerably depressed relative to MIK 1.43. Thus, although some recovery appeared to have occurred at this site, impacts were still evident.

At the other four sites as at MIK 0.71, the extent of fluctuations in density between sampling periods was substantial, but only the changes at MIK 0.54 were statistically significant from those at MIK 1.43. However, the existence of opposite trends of change at MIK 0.54 over the spring (i.e., density increased) and fall (i.e., density decreased) sampling periods, in addition to the large fluctuations between sampling periods, suggests that no persistent and marked changes occurred in density at this site during the study. Similarly, the

absence of trends of change at the other three sites that were significantly different from those of MIK 1.43, as well as the large fluctuations between sampling periods, suggests that densities at these three sites made no marked and persistent changes either.

Even though the only significant trends of change exhibited in total richness were at MIK 0.54 and MIK 0.86 vs MIK 1.43 during the spring, and changes in EPT richness were at MIK 0.45 vs MIK 1.43 during the fall; MIK 0.78 exhibited persistent and substantial changes in these two parameters as well. Most changes were evident in total richness by April 1988 or April 1989 and in EPT richness by October 1989. The most notable changes among these four sites occurred in EPT richness at MIKs 0.54 and 0.45. Prior to October 1989, EPT taxa were virtually absent from these two sites; after this period these taxa were usually represented by at least a few individuals during the remaining sampling periods, although their densities remained low. These changes suggest that some recovery occurred at all four sites, with the greatest amount of recovery occurring at MIKs 0.45 and 0.54. However, as at MIK 0.71, persistently low values for total and EPT richness along with continued low relative abundances of the generally pollution intolerant EPT taxa (e.g., Lenat 1988), suggests that impact was continuing at these sites.

The period of greatest change observed at the three sites downstream of MIK 0.78 coincided closely with an improvement in water quality that was demonstrated by ambient toxicity tests after 1988 (Kszos 1994). A delayed response in the invertebrate community following the apparent improvements in water quality in Mitchell Branch would be expected, because the rate of recolonization of a depauperate community would depend primarily upon drift of organisms from unimpacted or lesser impacted areas further upstream (Williams and Hynes 1976, 1977).

Because no major operational changes occurred at the K-25 Site prior to the end of 1988 that would have affected any site upstream of MIK 0.54, and because some recovery appeared to occur at MIKs 0.78 and 0.86 after this period, the most likely explanation for the apparent improvement in lower Mitchell Branch after 1988 would be that of increased flow and dilution in the stream as a result of increased rainfall (Table 2.1; Hinzman and Loar 1994). The lack of a more significant response at MIKs 0.54 and 0.45 may be due to one or more of the following factors. (1) Prior to mid-1988, the invertebrate community at MIK 0.71 was considerably more degraded than at either MIK 0.54 or MIK 0.45 (Smith 1993, Smith 1994). Equal improvements in water quality would allow some recovery to occur at all affected sites,

but since greater degradation originally existed at MIK 0.71, the magnitude of recovery would be greater at this site. (2) Impacts as a result of discharges from SDs 180 and 190 may have suppressed recovery at MIKs 0.45 and 0.54. (3) Habitat (i.e., substrate for attachment) at MIKs 0.45 and 0.54 was less suitable than at MIK 0.71. The substrate at MIK 0.71 was generally composed of a mixture of gravel, sand, and silt with some rubble, while the substrate at MIKs 0.45 and 0.54 was generally composed of a mixture of small gravel, sand, and silt (J. G. Smith, ESD/ORNL, personal observation). In general, a mixture of rubble, gravel, and sand serves as a more suitable substrate for a greater variety of invertebrates, while small gravel, sand, and silt are preferred more by oligochaetes and some chironomids (Hynes 1970). This may partially explain the predominance of oligochaetes and chironomids at MIKs 0.45 and 0.54, but excessive amounts of organic matter may favor these taxa as well (e.g., Hynes 1960).

Several potential factors contributing to the degraded macroinvertebrate community downstream of MIK 1.43 were previously identified and discussed by Smith (1994). These will likely continue to be important factors: siltation/sedimentation (J. G. Smith, ESD/ORNL, personal observation), elevated temperatures (Section 2.2.3), and the presence of at least periodically toxic conditions (Kszos 1993, 1994; Section 3), as suggested by the frequently low density values at some sites, particularly during the fall. The presence of periodically toxic conditions (or the episodic release of toxicants) is also suggested by the predominance of taxa (e.g., Chirononidae, Oligochaeta) that have short generation times; are capable of producing several generations each year; and/or always have a reproducing population present, thus increasing the probability that a resistent or unaffected stage will be present during a toxic excursion. Potential toxicants included chlorine (downstream of MIK 0.78) and possibly one or more metals or organic compounds. Flows were augmented from effluent discharges and flow regimes were altered by effluent discharges and changes in surface water runoff due to the lack of riparian vegetation in the vicinity of the stream. In addition to altering the flow regime, the lack of riparian vegetation also contributes to alterations of the stream's thermal regime by greater solar heating. The presence of enriched conditions was suggested by the periodically high macroinvertebrate densities at most of the impacted sites.

6.1.5 Summary and Conclusions/Recommendations

Further studies of the benthic macroinvertebrate community after January 1988 indicated that the water quality of Mitchell Branch within the main K-25 Site boundary continued to be significantly degraded. However, some recovery in the macroinvertebrate community was evident at all impacted sites, most notably after 1988, indicating that some improvement has occurred in water quality. Recovery was greatest at MIK 0.71, the site previously identified as the most severely degraded. Because recovery began prior to any significant operational changes at the K-25 Site and soon after the end of a drought (i.e., after the end of 1988), the improvements observed in the macroinvertebrate community may be caused by increased dilution of toxicants from associated increased instream flows and an increase in minimal flows, both of which are associated with the increase in rainfall. Continued impacts were most likely associated with operations of the K-25 Site that include a combination of factors such as siltation/sedimentation, elevated temperatures, the presence of at least periodically toxic conditions, altered stream flow and thermal regimes, and nutrient enrichment.

As a means of potentially increasing the amount and rate of recovery of the benthic macroinvertebrate community in Mitchell Branch, it is recommended that the riparian zone be expanded as much as feasible. It is also recommended that the strategic placement of rubble in the stream be considered as a means of habitat improvement and stability downstream of SD 170.

6.1.6 Future Studies

Monitoring of the benthic macroinvertebrate community in Mitchell Branch will continue as described in the BMAP Plan for the K-25 Site (Kszos et al. 1993). This will include the quarterly collection of samples from only four sites (MIKs 0.45, 0.71, 0.78, and 1.43). Only the samples collected during the spring and fall sampling periods will be processed, while those collected during the summer and winter will be stored and processed only if necessary (Kszos et al. 1993). In addition to the use of MIK 1.43 as a reference site, other appropriate reference sites used for other biological monitoring programs on the ORR will also be incorporated into future data analyses. This will provide a more accurate assessment of the expected range of reference conditions in the Oak Ridge area.

Because similar perturbations affect Mitchell Branch and streams near ORNL and the Y-12 Plant, studies will be conducted over the next 5 years in conjunction with these other

BMAPs to try to more accurately identify the causal factors associated with the degraded biotic communities in these streams. These studies may include but not necessarily be limited to a determination of the importance of elevated temperatures and excessive sedimentation to macroinvertebrates.

6.2 FISH COMMUNITY ASSESSMENT *(E. M. Schilling)*

6.2.1 Introduction

Fish population and community studies can also 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 (see Karr et al. 1986 and Karr 1987, 1991) and are relevant to assessment of the biotic integrity of streams such as Mitchell Branch. For example, fish communities include several trophic levels and species that are at or near the end of food chains. Consequently, they potentially integrate the direct effects of water quality and habitat changes on primary producers (periphyton) and consumers (benthic invertebrates) that are utilized for food. Because of these trophic interrelationships, the well-being of fish populations has often been used as an index of water quality (e.g., Weber 1973; Greeson et al. 1977; Karr et al. 1986). Statements about the condition of the fish community are often better understood by the general public than statements about the condition of diatoms or benthic macroinvertebrates (Karr 1981).

The initial objectives of the instream fish monitoring task (Subtask 4b of the BMAP, as described in Loar et al. 1992a) were (1) to characterize spatial and temporal patterns in the distribution and abundance of fishes in Mitchell Branch and (2) to document any possible effects of new pollution abatement facilities on fish community structure and function.

6.2.2 Materials and Methods

Quantitative sampling of the fish population was conducted in Mitchell Branch by electrofishing biannually in fall 1990 through fall 1993. Four sites in Mitchell Branch [MIKs 0.45, 0.54, 0.71 and 0.78] were sampled from fall 1990 through fall 1992 (Fig. 2.2). Beginning spring 1993, only MIKs 0.45 and 0.71 were sampled. Similar quantitative samples were made at two nearby reference sites (GCK 2.4 and SCK 2.2) for comparisons of population parameters. Grassy Creek was sampled in fall 1990 through fall 1992 and Scarboro Creek was sampled spring 1991 through fall 1993. Stream conditions have declined

at Grassy Creek and, therefore, Scarboro Creek was added as a new reference site beginning in spring 1991. The resulting data were used to (1) determine species composition, (2) estimate population size (numbers and biomass per unit area), (3) determine length-frequency, and (4) determine annual production. The lengths of the sampling reaches ranged from 30 to 54 m at Mitchell Branch and from 36 to 60 m at the reference sites (Table 6.1). Lengths of the sampling reaches in Mitchell Branch and Grassy Creek were similar to those in 1988 through spring 1990. (Ryon 1994b).

6.2.2.1 Field sampling procedures

All stream sampling was conducted using one Smith-Root Model 15A backpack electrofisher. The unit has a self-contained, gasoline-powered generator capable of delivering up to 1200 V of pulsed direct current. A pulse frequency of 90 to 120 Hz was used, and the output voltage was adjusted to the optimal value (generally 300–500 V) based on the specific conductance of the water. A circular (ring) electrode at the end of a fiberglass anode pole was fitted with a nylon net (0.64-cm mesh) to allow the electrofisher operator to collect stunned 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 two- to five-person sampling team electrofished the site in an upstream direction on three consecutive passes. Depending upon the turbidity of the water, consecutive passes could not always be made immediately. Rather, fish were processed after each pass to allow sufficient time for the water to clear before another pass was initiated. Stunned fish were collected and stored, by pass, in buckets with small holes to allow fresh water to pass through the buckets during further sampling. When possible, aerators were used in buckets to reduce mortality.

Following electrofishing, fish were anesthetized with MS-222 (tricaine methanesulfonate), identified, measured to the nearest 0.1 cm (total length) and weighed on Pesola spring scales to the nearest 0.1 g (for fish less than 100 g) or gram (for fish greater than 100 g). Individuals were recorded by 1-cm size classes and species. For the fall 1990 and spring 1991 sampling, after 25 individuals of a species-size class were measured and weighed, additional members of that size class were only measured. Beginning in fall 1991, 10 individuals of a species-size class were measured and weighed, and additional members of that size class were only measured. Later, length-weight regressions were determined using Railsback et al. (1989)

Table 6.1. Length, mean width, mean depth, surface area, and pool to riffle ratio of fish sampling sites in Mitchell Branch and in two reference streams, Grassy Creek and Scarboro Creek, for each sampling date

Site[a] (km)	Date	Length (m)	Mean width (m)	Mean Depth (cm)	Surface area (m^2)	Pool-riffle ratio
MIK 0.45	09/25/90	35	1.9	12.0	67	1.9
MIK 0.45	04/29/91	30	1.8	10.7	54	14.0
MIK 0.45	11/04/91	30	1.5	9.8	45	14.0
MIK 0.45	05/13/92	31	1.8	14.0	56	P[b]
MIK 0.45	11/10/92	30	1.8	14.3	54	14.0
MIK 0.45	04/19/93	31	1.8	11.3	56	2.1
MIK 0.45	10/15/93	33	1.5	7.3	50	3.1
MIK 0.54	09/25/90	40	1.5	9.3	60	2.1
MIK 0.54	04/29/91	39	1.2	8.2	47	0.5
MIK 0.54	11/04/91	38	1.0	8.4	38	2.5
MIK 0.54	05/12/92	39	1.2	8.1	47	1.3
MIK 0.54	11/10/92	39	1.2	8.9	47	1.6
MIK 0.71	09/25/90	50	1.3	10.3	65	1.0
MIK 0.71	05/01/91	54	1.0	8.7	54	0.9
MIK 0.71	11/05/91	34	0.9	4.8	31	1.1
MIK 0.71	05/13/92	39	1.0	6.2	39	0.6
MIK 0.71	11/10/92	38	1.1	6.8	42	0.7
MIK 0.71	04/19/93	38	1.6	12.5	61	0.7
MIK 0.71	10/15/93	38	1.2	9.4	46	1.7
MIK 0.78	09/25/90	45	1.0	9.0	45	1.4
MIK 0.78	05/01/91	44	1.0	11.3	44	2.1
MIK 0.78	11/05/91	47	1.0	8.0	47	4.9
MIK 0.78	05/13/92	48	1.1	9.9	53	1.4
MIK 0.78	11/10/92	47	1.1	7.6	52	1.2
GCK 2.4	10/25/90	36	1.5	8.5	54	P
GCK 2.4	03/28/91	60	2.1	13.0	126	0.8
GCK 2.4	11/01/91	42	1.6	10.0	67	P
GCK 2.4	04/08/92	57	1.7	7.3	97	0.6
GCK 2.4	12/09/92	59	1.1	8.0	65	0.6
SCK 2.2	05/08/91	38	2.2	15.1	84	0.5
SCK 2.2	11/18/91	36	2.0	16.0	72	2.0
SCK 2.2	04/23/92	34	2.1	17.7	71	2.4
SCK 2.2	12/14/92	41	1.8	15.8	74	1.1
SCK 2.2	05/18/93	40	1.9	13.8	76	1.0
SCK 2.2	10/28/93	40	1.7	14.5	68	1.0

[a]MIK = Mitchell Branch kilometer; GCK = Grassy Creek kilometer; SCK = Scarboro Creek kilometer.
[b]P = Stream was a continuous pool; no riffle present.

to estimate weights of unweighed fish. Other data recorded (if possible to determine) included sex, reproductive state, whether the fish was unusually plump or emaciated, deposition (i.e., dead, or retained for laboratory identification and reference collection), and presence of any abnormalities (e.g., external parasites, skeletal deformities, etc.). After processing fish from all passes, the fish were allowed to recover fully from the anesthesia and were returned to the stream. Any additional mortality that occurred as a result of processing was noted at that time.

In addition to data on individual fish, selected physical and chemical parameters were measured, and sampling effort was recorded. An Horiba Model U-7 battery-powered field sampler was used to measure conductivity, pH, water temperature, and dissolved oxygen content. An HF Instruments Model DRT-15 turbidimeter was used to measure turbidity. The duration of the electrofishing effort was recorded and a visual estimate was made of percentage cloud cover. Following completion of fish sampling, the length, width, and depth of the sampling reach were measured at each site.

6.2.2.2 Data analysis

Population size

Species population estimates were obtained using the three-pass removal method of Carle and Strub (1978). Biomass was estimated by multiplying the population estimate by the mean weight of the individuals. To calculate density and biomass per unit area, total numbers and biomass were divided by the water surface area (in square meters) of the study reach. For each sampling date, surface area was estimated by multiplying the length of the sampling reach by the mean width based on measurements taken at 5-m intervals (Table 6.1). These data were compiled and analyzed by a comprehensive Fortran 77 program developed by Railsback et al. (1989).

Length-frequency analyses

The population structures of the most abundant species were examined by length frequencies generated by the Fortran program. Length data for the selected species were separated into 1- or 2-cm size classes, depending on the maximum size of the species. These frequencies indicated whether the population included young and adult individuals and if any unusual mortality had affected a size class (Anderson and Gutreuter 1983).

Annual production

Annual production was estimated using a size-frequency method (Garman and Waters 1983). Production was calculated for each site (where data was available) from spring 1990 to spring 1991, spring 1991 to spring 1992, and spring 1992 to spring 1993. Production values were calculated from estimated biomass and population estimates. Annual production estimates for Mitchell Branch were compared with published values and those calculated for other streams on the ORR.

6.2.3 Results and Discussion

6.2.3.1 Species richness and composition

A total of eight species were collected in the quantitative surveys of the four sites in Mitchell Branch from fall 1990 through fall 1993 (Table 6.2). In comparison, a total of 6 species were found at Grassy Creek in 5 surveys from fall 1990 through fall 1992; and a total of 16 species were found in 6 surveys of Scarboro Creek from spring 1991 through fall 1993. Species richness ranged from 0 to 6 species at Mitchell Branch, 3 to 5 species at Grassy Creek, and from 7 to 13 species at Scarboro Creek for the sampling periods (Table 6.3).

The fish community in Mitchell Branch in fall 1990 and spring 1991 (Tables 6.4 and 6.5) consisted of five species, central stoneroller (*Campostoma anomalum*), blacknose dace (*Rhinichthys atratulus*), creek chub (*Semotilus atromaculatus*), redbreast sunfish (*Lepomis auritus*), and green sunfish (*L. cyanellus*). The blacknose dace, creek chub, and redbreast sunfish are species that appear to be insensitive to many habitat and water quality stresses and would be considered tolerant species in an impact assessment methodology such as the Index of Biotic Integrity (Karr et al. 1986). Sampling in January 1988 through fall 1989 revealed the absence of fish at MIKs 0.45, 0.54, and 0.71 (Ryon 1994b). There were no fish collected in fall 1990 from MIKs 0.45, 0.54, or 0.71. The fish community at MIK 0.78 in fall 1990 and spring 1991 was similar to the fish community in January 1988 through April/May 1989 (Ryon 1994b) with the exception of the presence of the central stoneroller and green sunfish.

Three additional species, banded sculpin (*Cottus carolinae*), green sunfish (*Lepomis cyanellus*), and bluegill (*L. macrochirus*), were found infrequently during sampling from fall 1991 through fall 1993 (Tables 6.6 through 6.10) at MIK 0.78. These species had not previously been collected in Mitchell Branch since sampling began in September 1986 (Ryon 1993 and 1994b).

Table 6.2. Fish species composition and occurrence (number of sampling periods when found) in Mitchell Branch, Grassy Creek, and Scarboro Creek, fall 1990 through fall 1993

	Sites[a]					
Species	MIK 0.45 (7[b])	MIK 0.54 (5)	MIK 0.71 (7)	MIK 0.78 (5)	GCK 2.4 (5)	SCK 2.2 (6)
Cyprinidae						
Central stoneroller (*Campostoma anomalum*)	2[c]	1	4	2	-	6
Spotfin shiner (*Cyprinella spiloptera*)	-	-	-	-	-	3
Striped shiner (*Luxilus chrysocephalus*)	-	-	1	1	1	-
Bluntnose minnow (*Pimephales notatus*)	-	-	-	-	-	4
Fathead minnow (*Pimephales promelas*)	-	-	-	-	-	1
Blacknose dace (*Rhinichthys atratulus*)	5	3	6	5	5	6
Creek chub (*Semotilus atromaculatus*)	6	4	6	-	5	4
Catostomidae						
White sucker (*Catostomus commersoni*)	-	-	-	-	4	-
Northern hogsucker (*Hypentelium nigricans*)	-	-	-	-	-	2
Ictaluridae						
Yellow bullhead (*Ameiurus natalis*)	-	-	-	-	-	4
Poeciliidae						
Western mosquitofish (*Gambusia affinis*)	-	-	-	-	-	1
Cottidae						
Banded sculpin (*Cottus carolinae*)	-	-	-	3	2	6
Centrarchidae						
Redbreast sunfish (*Lepomis auritus*)	1	-	2	4	-	4
Green sunfish (*Lepomis cyanellus*)	-	-	-	1	2	6
Bluegill (*Lepomis macrochirus*)	-	-	-	1	-	4
Hybrid sunfish (*Lepomis spp.*)	-	-	-	-	-	1
Largemouth bass (*Micropterus salmoides*)	-	-	-	-	-	2
Percidae						
Logperch (*Percina caprodes*)	-	-	-	-	-	4
Number of species (N)	4	3	5	7	6	16

[a]MCK = Mitchell Branch kilometer; GCK = Grassy Creek kilometer; SCK = Scarboro Creek kilometer.

[b]Numbers represent the number of sampling periods for each site.

[c]Numbers represent the number of sampling periods that a given species was collected at the site and a 'hyphen' indicates that the species was not collected.

Table 6.3. Total fish density, total biomass, and species richness in Mitchell Branch and two reference streams; Grassy Creek and Scarboro Creek, for fall 1990-1993 and spring 1991-1993

	Sites[b]					
Sampling periods[a]	MIK 0.45	MIK 0.54	MIK 0.71	MIK 0.78	GCK 2.4	SCK 2.2
Fall 1990						
Total density (individuals/m^2)	NF[c]	NF	NF	4.20	3.41	NS[d]
Total biomass (g/m^2)				6.00	5.80	
Species richness				3	3	
Spring 1991						
Total density	0.04	0.02	0.35	2.09	0.41	2.15
Total biomass	0.22	0.36	2.30	6.77	1.84	11.41
Species richness	1	1	2	4	4	7
Fall 1991						
Total density	6.00	5.00	3.71	7.10	4.35	6.19
Total biomass	12.35	11.40	5.82	14.50	5.63	26.55
Species richness	2	2	2	4	4	13
Spring 1992						
Total density	0.59	0.38	2.62	2.99	1.51	2.75
Total biomass	3.00	0.89	8.34	10.35	2.31	25.60
Species richness	3	2	3	5	3	11
Fall 1992						
Total density	3.43	3.77	4.07	8.89	4.37	4.91
Total biomass	5.53	7.94	9.97	23.93	4.00	32.21
Species richness	3	3	3	6	5	9
Spring 1993						
Total density	2.41	NS	4.53	NS	NS	4.04
Total biomass	7.38		40.37			42.23
Species richness	2		5			7
Fall 1993						
Total density	4.48	NS	6.29	NS	NS	7.56
Total biomass	4.83		22.58			48.33
Species richness	3		4			11

[a]The months sampled in the spring and fall were: September 1990 for Mitchell Branch; October 1990 for Grassy Creek; April and May 1991 for Mitchell Branch; May 1991 for Scarboro Creek; March 1991 for Grassy Creek; November 1991 for Mitchell Branch, Grassy Creek, and Scarboro Creek; May 1992 for Mitchell Branch; April 1992 for Grassy Creek and Scarboro Creek; November 1992 for Mitchell Branch; December 1992 for Grassy Creek and Scarboro Creek; April 1993 for Mitchell Branch; May 1993 for Scarboro Creek; October 1993 for Mitchell Branch and Scarboro Creek.

[b]MCK=Mitchell Branch kilometer; GCK=Grassy Creek kilometer; SCK=Scarboro Creek kilometer.

[c]NF=No fish taken in sample.

[d]NS=Not sampled.

Table 6.4. Fish densities (number of individuals per square meter) and biomass (grams per square meter) in Mitchell Branch and in two reference streams, Grassy Creek and Scarboro Creek, for fall 1990

	Density (Biomass)					
	Sites[a]					
Species	MIK 0.45	MIK 0.54	MIK 0.71	MIK 0.78	GCK 2.4	SCK 2.2
Cyprinidae	NF[b]	NF	NF	-	-	NS[c]
Central stoneroller	-	-	-	0.18 (1.45)	-	-
Blacknose dace	-	-	-	2.58 (1.50)	2.54 (2.11)	-
Creek chub	-	-	-	1.44 (3.05)	0.83 (3.25)	-
Cottidae						
Banded sculpin	-	-	-	-	0.04 (0.44)	-
Number of species	0	0	0	3	3	NS
Total density	0	0	0	4.20	3.41	-
Total biomass	0	0	0	6.00	5.80	-

[a]MIK = Mitchel Branch kilometer; GCK = Grassy Creek kilometer; SCK = Scarboro Creek kilometer.

[b]NF = No fish taken in sample.

[c]NS = Not sampled.

Table 6.5. Fish densities and biomass in Mitchell Branch and in two reference streams, Grassy Creek and Scarboro Creek, for spring 1991

	Density (number of individuals/m^2) (Biomass) (g/m^2)					
	Sites[a]					
Species	MIK 0.45	MIK 0.54	MIK 0.71	MIK 0.78	GCK 2.4	SCK 2.2
Cyprinidae						
Central stoneroller	-	-	-	-	-	0.16 (2.00)
Blacknose dace	-	-	0.11 (0.43)	1.16 (1.57)	0.20 (0.19)	0.52 (1.92)
Creek chub	0.04 (0.22)	0.02 (0.36)	0.24 (1.87)	0.86 (4.26)	0.18 (1.10)	0.04 (0.08)
Catostomidae						
White sucker	-	-	-	-	0.01 (0.36)	-
Northern hogsucker	-	-	-	-	-	0.02 (0.69)
Cottidae						
Banded sculpin	-	-	-	-	0.02 (0.19)	1.25 (5.68)
Centrarchidae						
Redbreast sunfish	-	-	-	0.02 (0.12)	-	-
Green sunfish	-	-	-	0.05 (0.82)	-	0.12 (0.66)
Percidae						
Logperch	-	-	-	-	-	0.04 (0.38)
Number of species	1	1	2	4	4	7
Total density	0.04	0.02	0.35	2.09	0.41	2.15
Total biomass	0.22	0.36	2.30	6.77	1.84	11.41

[a]MIK = Mitchell Branch kilometer; GCK = Grassy Creek kilometer; SCK = Scarboro Creek kilometer.

Table 6.6. Fish densities and biomass in Mitchell Branch and in two reference streams, Grassy Creek and Scarboro Creek, for fall 1991

	Density (as number of individuals/m^2) (Biomass) (as g/m^2)					
	Sites[a]					
Species	MIK 0.45	MIK 0.54	MIK 0.71	MIK 0.78	GCK 2.4	SCK 2.2
Cyprinidae						
Central stoneroller	-	-	-	-	-	1.53 (8.65)
Spotfin shiner	-	-	-	-	-	0.69 (1.70)
Bluntnose minnow	-	-	-	-	-	0.71 (0.89)
Blacknose dace	1.84 (2.73)	1.55 (1.67)	1.58 (1.24)	3.60 (3.63)	3.24 (2.15)	0.61 (2.59)
Creek chub	4.16 (9.62)	3.45 (9.73)	2.13 (4.58)	3.40 (10.03)	0.72 (2.54)	0.06 (0.41)
Catostomidae						
White sucker	-	-	-	-	0.37 (0.77)	-
Poeciliidae						
Western mosquitofish	-	-	-	-	-	0.01 (0.01)
Cottidae						
Banded sculpin	-	-	-	0.04 (0.16)	-	1.58 (5.98)
Centrarchidae						
Redbreast sunfish	-	-	-	0.06 (0.68)	-	0.17 (0.85)
Green sunfish	-	-	-	-	0.02 (0.17)	0.46 (3.91)
Bluegill	-	-	-	-	-	0.29 (1.00)
Hybrid sunfish	-	-	-	-	-	0.01 (0.10)
Largemouth bass	-	-	-	-	-	0.01 (0.06)
Percidae						
Logperch	-	-	-	-	-	0.06 (0.40)
Number of species	2	2	2	4	4	13
Total density	6.00	5.00	3.71	7.10	4.35	6.19
Total biomass	12.35	11.40	5.82	14.50	5.63	26.55

[a]MIK = Mitchell Branch kilometer; GCK = Grassy Creek kilometer; SCK = Scarboro Creek kilometer.

Table 6.7. Fish densities and biomass in Mitchell Branch and in two reference streams, Grassy Creek and Scarboro Creek, for spring 1992

	Density (as number of individuals/m^2) (Biomass) (as g/m^2)					
	Sites[a]					
Species	MIK 0.45	MIK 0.54	MIK 0.71	MIK 0.78	GCK 2.4	SCK 2.2
Cyprinidae						
Central stoneroller	-	-	0.03 (0.58)	-	-	0.87 (10.00)
Striped shiner	-	-	-	0.04 (0.15)	-	-
Bluntnose minnow	-	-	-	-	-	0.04 (0.10)
Blacknose dace	0.23 (0.67)	0.34 (0.83)	2.23 (6.06)	1.38 (2.88)	1.01 (0.94)	0.37 (1.21)
Creek chub	0.34 (1.95)	0.04 (0.06)	0.36 (1.70)	1.51 (6.96)	0.39 (1.08)	0.01 (0.10)
Catostomidae						
White sucker	-	-	-	-	0.11 (0.29)	-
Northern hogsucker	-	-	-	-	-	0.07 (4.96)
Ictaluridae						
Yellow bullhead	-	-	-	-	-	0.01 (0.18)
Cottidae						
Banded sculpin	-	-	-	0.04 (0.24)	-	1.00 (5.36)
Centrarchidae						
Redbreast sunfish	0.02 (0.38)	-	-	0.02 (0.12)	-	0.03 (0.19)
Green sunfish	-	-	-	-	-	0.30 (3.10)
Bluegill	-	-	-	-	-	0.04 (0.25)
Percidae						
Logperch	-	-	-	-	-	0.01 (0.15)
Number of species	3	2	3	5	3	11
Total density	0.59	0.38	2.62	2.99	1.51	2.75
Total biomass	3.00	0.89	8.34	10.35	2.31	25.60

[a]MIK = Mitchell Branch kilometer; GCK = Grassy Creek kilometer; SCK = Scarboro Creek kilometer.

Table 6.8. Fish densities and biomass in Mitchell Branch and in two reference streams, Grassy Creek and Scarboro Creek, for fall 1992

	Density (as number of individuals/m^2) (Biomass) (as g/m^2)					
	Sites[a]					
Species	MIK 0.45	MIK 0.54	MIK 0.71	MIK 0.78	GCK 2.4	SCK 2.2
Cyprinidae						
Central stoneroller	0.11 (0.39)	0.60 (2.37)	0.50 (1.58)	3.06 (10.86)	-	2.12 (17.40)
Spotfin shiner	-	-	-	-	-	0.18 (0.44)
Striped shiner	-	-	-	-	0.02 (0.25)	-
Bluntnose minnow	-	-	-	-	-	0.61 (1.05)
Blacknose dace	2.43 (3.23)	1.70 (2.20)	1.88 (2.37)	2.60 (2.29)	3.85 (1.87)	0.30 (1.81)
Creek chub	0.89 (1.91)	1.47 (3.37)	1.69 (6.02)	3.15 (9.83)	0.42 (1.33)	-
Catostomidae						
White sucker	-	-	-	-	0.06 (0.48)	-
Ictaluridae						
Yellow bullhead	-	-	-	-	-	0.05 (0.87)
Cottidae						
Banded sculpin	-	-	-	0.02 (0.23)	-	1.31 (6.13)
Centrarchidae						
Redbreast sunfish	-	-	-	0.04 (0.41)	-	0.01 (0.07)
Green sunfish	-	-	-	-	0.02 (0.07)	0.30 (4.30)
Bluegill	-	-	-	0.02 (0.31)	-	0.03 (0.14)
Number of species	3	3	3	6	5	9
Total density	3.43	3.77	4.07	8.89	4.37	4.91
Total biomass	5.53	7.94	9.97	23.93	4.00	32.21

[a]MIK = Mitchell Branch kilometer; GCK = Grassy Creek kilometer; SCK = Scarboro Creek kilometer.

Table 6.9. Fish densities and biomass in Mitchell Branch and two reference streams, Grassy Creek and Scarboro Creek, for spring 1993

	Density[a] (Biomass)[b]					
	Sites[c]					
Species	MIK 0.45	MIK 0.54	MIK 0.71	MIK 0.78	GCK 2.4	SCK 2.2
Cyprinidae		NS[d]		NS	NS	
Central stoneroller	-	-	1.08 (8.78)	-	-	2.49 (31.76)
Striped shiner	-	-	0.02 (0.21)	-	-	-
Blacknose dace	1.32 (2.54)	-	1.79 (5.26)	-	-	0.47 (2.30)
Creek chub	1.09 (4.84)	-	1.61 (25.51)	-	-	0.03 (0.32)
Ictaluridae						
Yellow bullhead	-	-	-	-	-	0.01 (0.96)
Cottidae						
Banded sculpin	-	-	-	-	-	0.75 (3.94)
Centrarchidae						
Redbreast sunfish	-	-	0.03 (0.61)	-	-	-
Green sunfish	-	-	-	-	-	0.16 (1.69)
Percidae						
Logperch	-	-	-	-	-	0.13 (1.26)
Number of species	2	NS	5	NS	NS	7
Total density	2.41		4.53			4.04
Total biomass	7.38		40.37			42.23

[a]Density expressed in number of individuals per square meter.
[b]Biomass, in parentheses, expressed in grams per square meter.
[c]MIK = Mitchell Branch kilometer; GCK = Grassy Creek kilometer; SCK = Scarboro Creek kilometer.
[d]NS = Not sampled.

Table 6.10. Fish densities and biomass in Mitchell Branch and two reference streams, Grassy Creek and Scarboro Creek, for fall 1993

	Density[a] (Biomass)[b]					
	Sites[c]					
Species	MIK 0.45	MIK 0.54	MIK 0.71	MIK 0.78	GCK 2.4	SCK 2.2
Cyprinidae		NS[d]		NS	NS	
Central stoneroller	0.06 (0.22)	-	1.48 (6.41)	-	-	3.46 (33.95)
Spotfin shiner	-	-	-	-	-	0.10 (0.21)
Bluntnose minnow	-	-	-	-	-	1.88 (3.28)
Fathead minnow	-	-	-	-	-	0.02 (0.03)
Blacknose dace	3.18 (2.85)	-	2.00 (1.82)	-	-	0.38 (2.69)
Creek chub	1.24 (1.76)	-	2.74 (14.26)	-	-	-
Ictaluridae						
Yellow bullhead	-	-	-	-	-	0.02 (0.11)
Cottidae						
Banded sculpin	-	-	-	-	-	0.68 (2.41)
Centrarchidae						
Redbreast sunfish	-	-	0.07 (0.09)	-	-	0.03 (0.28)
Green sunfish	-	-	-	-	-	0.28 (3.06)
Bluegill	-	-	-	-	-	0.69 (2.27)
Largemouth bass	-	-	-	-	-	0.02 (0.04)
Number of species	3	NS	4	NS	NS	11
Total density	4.48		6.29			7.56
Total biomass	4.83		22.58			48.33

[a]Density expressed in number of individuals per square meter.
[b]Biomass, in parentheses, expressed in grams per square meter.
[c]MIK = Mitchell Branch kilometer; GCK = Grassy Creek kilometer; SCK = Scarboro Creek kilometer.
[d]NS=Not sampled

The variability in species number and composition in Mitchell Branch, Grassy Creek, and Scarboro Creek may be related to a combination of their proximity to Poplar Creek or Melton Hill Reservoir, impacts to Mitchell Branch by the K-25 Site facilities, and possible barriers in Mitchell Branch to upstream movement of fish. Grassy Creek is located approximately twice the distance from an embayment as either Mitchell Branch or Scarboro Creek and the dominant species, blacknose dace, creek chub, and banded sculpin, are common to small headwaters streams. The closeness of Scarboro Creek to Melton Hill Reservoir allows species to be found in Scarboro Creek when both are near impoundment waters that are atypical for streams of that size. The absence of fish species in Mitchell Branch compared with Scarboro Creek, when both are near impoundment waters, may be the result of adverse impacts to the stream. There has been an increase in the number of species present in Mitchell Branch since sampling began in September 1986 (Ryon 1993 and 1994b). The absence of northern hogsucker (*Hypentelium nigricans*) and yellow bullhead (*Ameiurus natalis*) in Mitchell Branch may reflect past extermination and the prevention of recolonization from Poplar Creek. The logperch (*Percina caprodes*) has been found at SCK 2.2 but not in Mitchell Branch. This is a species that can migrate from the embayments to the streams.

Fish have been absent or occurred in small numbers at MIKs 0.45, 0.54, and 0.78 from September/October 1986 through September 1990 (Ryon 1993 and 1994b). The presence of fish and the increase in species diversity at these sites through fall 1993 suggests improved water quality.

6.2.3.2 Density and biomass

Population surveys of Mitchell Branch and the two reference sites were conducted twice a year (spring and fall), and the data were used to estimate species density and biomass for each period. The total estimated density and biomass at each site for each sampling period are presented in Table 6.3. Values for individual species are given in Tables 6.4 through 6.10.

Total fish densities have exhibited similar trends throughout the sampling periods in Mitchell Branch. Total fish density within the four sampling sites was highest at MIK 0.78 for each sampling period from fall 1990 through fall 1992. Total density was highest at MIK 0.71 in 1993 (MIK 0.78 was not sampled). Total density was lower in the spring compared with fall at all sites.

Total fish densities have increased at MIK 0.45 and MIK 0.71 from spring 1991 through spring 1993 and at MIK 0.54 and MIK 0.78 from spring 1991 to spring 1992. In contrast, density decreased from fall 1991 to fall 1992 at MIK 0.45 and MIK 0.54.

Total fish densities in Mitchell Branch were similar to values for the two reference sites. The total fish densities increased for each spring to spring and fall to fall period at the reference sites

The blacknose dace and the creek chub have been the dominant species in terms of density in Mitchell Branch throughout the sampling periods. Beginning in fall 1992, the central stoneroller greatly increased in density and was also a dominant species at MIKs 0.71 and 0.78. The central stoneroller was also one of the more abundant species present in Scarboro Creek but was absent in Grassy Creek.

In general, biomass in Mitchell Branch and the reference sites paralleled the trends observed in population densities. Biomass has generally increased at each site in Mitchell Branch between the spring sampling periods and between the fall sampling periods. Biomass has decreased at MIK 0.45 from fall 1991 through fall 1993 yet spring biomass has increased steadily throughout the same time period. Biomass also decreased slightly at MIK 0.54 from fall 1991 to fall 1992. The stoneroller became a dominant species in terms of biomass at MIK 0.71 in fall 1992.

6.2.3.3 Length-frequency

Length-frequency histograms represent the overall distribution of size classes of individuals within a population. The expected distribution would include a greater proportion of smaller individuals, with numbers decreasing as size increases. Length-frequency distributions can be used to demonstrate trends for a given species or trends among species at the same site. Length-frequency histograms were constructed for the most abundant species, blacknose dace, creek chub, and central stoneroller, collected in Mitchell Branch (Appendix G, Figs. G.1 through G.11) for the sampling periods of fall 1990 through fall 1993.

Length-frequency distributions were developed for the central stoneroller occurring in Mitchell Branch from fall 1992 through fall 1993 (Figs. G.1 through G.3). The central stoneroller was absent from fall 1990 through fall 1991 at three sites (MIKs 0.45, 0.54, and 0.71) sampled in Mitchell Branch. The central stoneroller was present in fall 1990 at MIK 0.78 but absent in 1991. The central stoneroller was represented by only a few

individuals in fall 1992 and 1993 at MIK 0.45 and fall 1992 at MIK 0.54, but these do not represent stable populations (Fig. G.1). The central stoneroller was represented in 1992 at MIK 0.71 by a small number of individuals and was not indicative of a stable population at MIK 0.71 (Fig. G.2). The central stoneroller exhibited the expected length-frequency distribution of a normal population for spring and fall 1993 at MIK 0.71.

The central stoneroller was represented by a few individuals in fall 1990 at MIK 0.78 (Fig. G.3). The central stoneroller was absent from this site in spring and fall 1991 and spring 1992. The central stoneroller population was represented by a large number of intermediate sized individuals in fall 1992.

Length-frequency histograms were developed for blacknose collected in Mitchell Branch from fall 1990 through fall 1993 (Figs. G.4 through G.7). Blacknose dace exhibited the expected length-frequency distribution of a normal population for the fall 1991, 1992, and 1993 and spring 1993 sampling periods at MIK 0.45 (Fig. G.4). Blacknose dace were absent in fall 1990 and spring 1991 at MIK 0.45. Blacknose dace were present in low numbers at MIK 0.45 in spring 1992 but did not represent a normal population.

Blacknose dace exhibited the expected length-frequency distribution of a normal population for the fall 1991 and 1992 at MIK 0.54 (Fig. G.5). Blacknose dace were absent from MIK 0.54 in fall 1990 and spring 1991. Blacknose dace were present in low numbers at MIK 0.54 in spring 1992, but still had a length-frequency distribution of a normal population.

Blacknose dace exhibited the expected length-frequency distribution of a normal population for the fall 1991, 1992, and 1993 at MIK 0.71 (Fig. G.6). Blacknose dace were absent in fall 1990. Blacknose dace were present in low numbers at MIK 0.71 in spring 1991 but did not have a distribution of a normal population. Large numbers of the smaller size classes (3–3.9 and 4–4.9) of blacknose dace were lacking at MIK 0.71 in spring 1992 and 1993. This may indicate over winter mortality or emigration from the site.

Blacknose dace exhibited the expected length-frequency distribution of a normal population for fall 1990, 1991, and 1992, and spring 1991 at MIK 0.78 (Fig. G.7). The majority of the blacknose dace population in spring 1992 was represented by relatively equal numbers of fish in three length classes, 3–3.9, 4–4.9, and 5–5.9 cm and was not representative of a normal population.

The creek chub exhibited the expected length-frequency distribution of a normal population for fall 1991 and 1993 and spring 1993 at MIK 0.45 (Fig. G.8). Creek chub were

absent at MIK 0.45 in fall 1990. The creek chub population in fall 1992 was represented by relatively equal numbers of fish in the 4–5.9 and 6–7.9 cm size class. The creek chub was present in low numbers at MIK 0.45 in spring 1991 and 1992.

The creek chub exhibited the expected length-frequency distribution of a normal population for fall 1991 and 1992 at MIK 0.54 (Fig. G.9). Fish were absent from MIK 0.54 in fall 1990. The creek was represented by a few individuals for both spring 1991 and 1992 and did not represent a stable population.

The creek chub exhibited the expected length-frequency distribution of a normal population for fall 1991, 1992, and 1993 at MIK 0.71. The creek chub was absent at MIK 0.71 in fall 1990 and represented by individuals in the intermediate size class in spring 1991 and 1992 (Fig. G.10). In spring 1993, the numbers of creek chub present at MIK 0.71 had increased substantially from spring 1992 but did not represent a normal population.

The creek chub exhibited the expected length-frequency distribution of a normal population for fall 1990, and spring and fall of 1991 and 1992 at MIK 0.78 (Fig. G.11).

6.2.3.4 Annual production

Annual production was calculated for spring 1990 to spring 1991, spring 1991 to spring 1992, and spring 1992 to spring 1993. Total production and production for selected species is given in Table 6.11.

In general, production increased each year for both individual species and the whole community at the four sites sampled in Mitchell Branch. Two exceptions were a slight decline in production of creek chub at MIK 0.71 and banded sculpin at MIK 0.78 in the 1991–1992 sampling period. Two other exceptions were that production of creek chub at MIK 0.54 and green sunfish at MIK 0.78 remained unchanged between sampling periods.

Total production of the four sampling sites in Mitchell Branch was greatest at MIK 0.78 for any given sampling period. Total production at MIK 0.78 was greater than at Grassy Creek for all periods. This was also the case for 1987 through 1990 (Ryon 1994b). Total production at MIKs 0.45 and 0.54 was much lower than at MIKs 0.78 or 0.71. This may be due to stresses to the stream below the upstream locations.

Total production in Mitchell Branch was less than in Scarboro Creek for each sampling period. This may be due to the fact that Scarboro Creek contains many more species of fish, and biomass values were generally greater than at Mitchell Branch.

Table 6.11. Annual production rates[a] of fish in Mitchell Branch, Grassy Creek, and Scarboro Creek from spring 1990 to spring 1993

Species	Production period	Sites[b]					
		MIK 0.45	MIK 0.54	MIK 0.71	MIK 0.78	GCK 2.4	SCK 2.2
Central stoneroller	1990–1991			0.00 ± 0.00	-0.10 ± 0.03		
	1991–1992			-0.15	0.00 ± 0.00		1.85
	1992–1993			1.29			18.22
Blacknose dace	1990–1991	0.00 ± 0.00	0.00 ± 0.00	-0.09 ± 0.04	1.40 ± 0.10	0.48 ± 0.07	
	1991–1992	0.11 ± 0.04	0.18	2.21	2.30 ± 0.14	0.28	0.74 ± 0.09
	1992–1993	2.34 ± 0.10		3.34			0.06 ± 0.11
Creek chub	1990–1991	-0.02 ± 0.01	-0.05	0.36	2.74 ± 0.47	0.62	
	1991–1992	-0.15 ± 0.04	-0.05	0.19 ± 0.07	4.32 ± 0.40	0.47	-0.01
	1992–1993	0.84 ± 0.07		4.90 ± 0.23			-0.04
Banded sculpin	1990–1991				0.00 ± 0.00	-0.16 ± 0.03	
	1991–1992				-0.06 ± 0.03	-0.05 ± 0.02	2.80 ± 0.19
	1992–1993						1.48 ± 0.17
Green sunfish	1990–1991				-0.12 ± 0.01		
	1991–1992				-0.12 ± 0.01		0.99 ± 0.08
	1992–1993						1.13 ± 0.15
Total[c]	1990–1991	-0.02	-0.05	0.27	3.91	0.83	
	1991–1992	-0.10	0.13	2.25	6.37	0.67	5.81
	1992–1993	3.12		9.41			20.23

[a]Measured in grams, wet weight, per square meter per year, ± SE.

[b]MIK = Mitchell Branch kilometer; GCK = Grassy Creek kilometer; SCK = Scarboro Creek kilometer.

[c]Totals include miscellaneous species (striped shiner, redbreast sunfish, white sucker, bluntnose minnow, yellow bullhead, northern hogsucker, bluegill and logperch) not listed in table.

Annual production at MIK 0.71 and MIK 0.78 was similar to values for other streams on the ORR (Table 5.5 in Ryon 1994a). Annual production values at MIK 0.45 and MIK 0.54 were generally less than those in other streams on the ORR.

6.2.4 Fish kills

From June 1990 through December 1993, two fish kills were reported in Mitchell Branch. On November 5, 1990, dead fish were reported in Mitchell Branch downstream of the railroad crossing. Surveys were initiated from the railroad bridge crossing downstream to the confluence with Poplar Creek. A total of 21 surveys were conducted from November 5, 1990, through December 26, 1990. Dead fish were found on the first ten surveys conducted. A total of 12 creek chub, 2 western mosquitofish (*Gambusia affinis*), 227 redbreast sunfish, 7 unidentified sunfish, 1 salamander, and 1 crayfish were found dead during the surveys. No cause for the fish kill was determined.

The second fish kill was reported on August 31, 1992. Four surveys were made from August 31, 1992, through September 3, 1992. One dead of each of the following were found on the initial survey: creek chub, blacknose dace, and crayfish. This appeared to be a rather small isolated kill. No cause for the kill was determined.

6.2.5 Conclusions

Data on the fish populations in Mitchell Branch have demonstrated that the stream has received stress from K-25 plant operations discharges in the past. Mitchell Branch had more species of fish compared with the reference stream, Grassy Creek, but was also missing common fish species found in Scarboro Creek and other area streams. The lack of these species may have been caused by past extermination and the prevention of recolonization from Poplar Creek and possibly by continued disturbances to the stream.

Fish population data from Mitchell Branch have suggested that improvements in water quality have occurred. There has been an increase in the number of fish species present in the fall 1990 through fall 1993 period compared with earlier years. In general, total fish density, biomass, and annual production values have increased in Mitchell Branch. Length-frequency distributions appear to have more stable populations of fish in Mitchell Branch in 1993 in comparison with earlier years. Continued remedial actions and a decrease in perturbations to

the stream may allow for more species to colonize the stream and populations to become more stabilized.

6.2.6 Future Studies

Routine, quantitative monitoring of fish density, biomass, richness, and population size structure will be conducted twice annually (spring and fall) at MIKs 0.45 and 0.71.

Because immigration into Mitchell Branch from Poplar Creek is occurring, further qualitative sampling will be conducted to assess immigration. This sampling may be a cooperative effort with sampling for the bioaccumulation task.

7. SUMMARY OF STREAM HEALTH

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One of the goals of the K-25 Site BMAP is to determine whether new pollution abatement facilities and other remedial actions have had an effect on the aquatic biota of Mitchell Branch. Table 2.2 lists some of the K-25 Site activities that probably had a positive impact on water quality in Mitchell Branch. In addition, the end of the drought in 1988 was a significant natural phenomena that influenced the results of the biological monitoring. The number and timing of both remedial activities and natural events makes it difficult to associate any one activity with a specific biological response; toxicity monitoring is the only method capable of detecting changes in water quality immediately following an abatement activity. The difficulty in discriminating the effects of single events highlights the need for long-term monitoring. Using this approach, the K-25 Site BMAP has documented the positive results of some abatement activities in nearly every task. The BMAP also shows that although improvements have been documented, the biota in Mitchell Branch remain impacted.

Ecological improvement or recovery has been documented in the toxicity monitoring and instream monitoring tasks. The K-25 Site has successfully eliminated the acute toxicity of water from Mitchell Branch to fathead minnows and *Ceriodaphnia* in laboratory tests. In 1990, water from MIK 0.71 downstream to MIK 0.45 was acutely toxic. In 1991, 1992, and 1993, no acute toxicity was observed. However, the results of both the Mitchell Branch and storm drain toxicity tests indicate that the effluent from SD 170 and SD 190 continues to adversely impact biota.

Likewise, instream monitoring of the benthic macroinvertebrate and fish communities has demonstrated recovery of these communities since 1989. Recovery of the benthic macroinvertebrate populations occurred after October 1989 from MIK 0.78 downstream to MIK 0.45 with the greatest recovery occurring at MIK 0.45 and MIK 0.54. These changes generally coincided with improvements in water quality, as documented by ambient toxicity tests conducted after 1988. In 1990, the appearance of pollution-intolerant groups of aquatic insects (e.g., mayflies and stoneflies) downstream of MIK 0.78 indicated improved conditions. Although conditions have improved, the most recent data (April 1993) indicate that the benthic

macroinvertebrate community remains impacted, as documented by a depression of total and EPT richness and substantial fluctuations in density, total richness, and EPT richness.

Similar changes have also been noted in the fish community of Mitchell Branch. Prior to fall 1990, fish were virtually absent from MIK 0.78 downstream to MIK 0.45. In spring 1990, the presence of fish from MIK 0.78 downstream to MIK 0.45 indicated improved conditions. Since 1991, three species that had not previously been collected were found at MIK 0.71 and MIK 0.78, and there has been a general increase in fish density, biomass, and annual production from MIK 0.78 downstream to MIK 0.45. In 1993, the length-frequency distributions of fish were indicative of a more stable population compared with earlier years. Similar to the benthic macroinvertebrate results, the fish data show there has been recovery, but the communities differ from those in the reference streams and are indicative of degraded water quality.

Two BMAP tasks have documented only slight improvement or no change in status. The assessment of fish health, which uses an integrated approach to investigate the holistic responses of fish to environmental stressors, showed a decrease in fish health from 1989 to 1991 but some improvement in 1992. The increase in growth and change in size distribution of the redbreast sunfish population at MIK 0.12 since 1990 is a documented improvement. Although there are indications of improvement, the integrated health responses of the Mitchell Branch fish remain dissimilar from the health responses of reference fish. This dissimilarity suggests that the Mitchell Branch population, although healthier in 1992 than in previous years, remains stressed. Based on measures of reproductive health, the sunfish collected in spring 1993 from Mitchell Branch were clearly different from reference site fish. Toxicity tests with medaka indicated that Mitchell Branch is often toxic to embryos.

The bioaccumulation of contaminants in aquatic biota has not changed over the past several years. Levels of mercury and PCBs have remained stable in sunfish from 1990 to 1993. Some fish in Mitchell Branch have had PCB concentrations that exceed the FDA tolerance limit of 2 μg/g. The results of mercury and PCB monitoring of fish prior to 1992 did not show clearly whether or not Mitchell Branch was a source of PCBs or mercury. Evidence collected in 1992 from caged and resident fish suggests that the source of mercury contamination in Mitchell Branch fish arises from a source within that watershed.

While BMAP has demonstrated that improvements have occurred in water quality at the K-25 Site, it has also shown that aquatic communities and the health of fish in Mitchell

Branch are dissimilar from reference sites. Moreover, the BMAP tasks document that the water quality of Mitchell Branch does not fully support the growth and propagation of fish and other aquatic life, a classified use of the stream. The K-25 Site has addressed many pollution sources (e.g., the removal of chlorinated discharges); and, with additional abatement actions, further ecological recovery is expected. The challenge now is to use BMAP to identify those additional sources of impact to biota and then develop appropriate remedial actions.

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APPENDIX A

WATER QUALITY DATA FOR MITCHELL BRANCH

Table A.1. Mean concentrations of National Pollutant Discharge Elimination System parameters detected in the water at K–1700 (Mitchell Branch kilometer 0.12)
Measured in mg/L unless otherwise noted

Parameter	Mean (range)				Reference value
	1990	1991	1992	1993	
1,1,1–Trichloroethane (μg/L)	2.23 (<0.4–5)				
1,1,–Dichloroethane (μg/L)	4.38 (<0.6–5)				
1,2,–Dichloroethane (μg/L)	5.74 (<5–36)				
1,2 Dichloroethene				0.011 (0.006–0.022)	0.07[a]
Aluminum	0.288 (<0.032–2.9)	0.287 (0.031–3.5)	<0.2697 (<0.1–3.5)		1.0
Alkalinity				91.75 (64–155)	
Beryllium		0.0011 (0.0003–0.003)			
Bromodichloromethane	4.51 (<0.8–5)				
Cadmium	0.002 (<0.002–0.004)	0.0026 (0.002–0.049)			
Calcium Hardness				81.5 (59–130)	
Chemical Oxygen Demand	5.87 (<5–28)	6.46 (<5–36)	<7.035 (<5–152)	8.5 (5–14)	
Chloride				17 (4–39.1)	250[b]
Chlorine				0.045 (0.01–0.1)	
Chloroform (μg/L)	4.19 (<0.5–13)				
Chromium	0.01 (<0.01–0.029)	0.011 (0.01–0.055)	<0.0104 (<0.01–0.012)		0.08
Color				495 (490–500)	
Conductivity				271.5 (180–404)	
Dissolved Oxygen				8.82 (7.6–11.5)	5.0[c]
Dissolved Solids	306.26 (88–892)	218.9 (74–1080)	211.8 (98–2700)	196 (130–262)	500[b]
Flow (L/s)	35.49	31.48 (2.77–325.26)	28.85 (11.39–176.98)	?	
Fluoride	0.186 (<0.1–0.9)	0.1863 (0.1–0.9)	<0.17 (<0.1–0.3)	0.2 (0.2–0.2)	4.0[a]
Iron				0.6775 (0.39–0.82)	0.3[b]

Table A.1 (continued)

Parameter	Mean (range)				Reference value
	1990	1991	1992	1993	
Lead	0.004 (<0.004–0.02)	0.00487 (0.004–0.026)		0.0058 (0.004–0.011)	0.082
Manganese				0.185 (0.11–0.27)	0.05[b]
Mercury	<0.0002 (<0.0002–0.00032)	0.00021		0.0001 (0 – 2e-04)	0.0024
Methylene chloride (μg/L)	4.789 (<1–5)				
Nickel				0.0445 (0.028–0.05)	1.4
Nitrate nitrogen	0.383 (<0.2–1.8)	0.3515 (0.04–1.13)	0.637 (<0.05–5.1)	0.48 (0.29–0.62)	10.0[a]
Oil and grease		2.16 (2–10.98)	<2.69 (<2–35.66)		15.0
pH	6.8–8.1	6.95–8.5	7.731(7.0–8.4)	7.68 (7.3–8)	6.5–8.5
Phosphate				0.14 (0.1–0.17)	
Potassium				2.025 (1.5–2.4)	
Selenium		0.0051 (0.005–0.013)			
Silicon	6.1 (6.1–6.1)				
Silver		0.011 (0.01–0.077)			
Sodium				13.575 (2.8–40)	
Sulfate				25 (16–39)	250[b]
Suspended Solids	7.266 (<1–147)	6.4 (1–1080)	<8.086 (<1–225)	7.75 (1–12)	50.0
Temperature	16.24 (1.2–28.1)		16.82 (5.6–25.5)	15.54 (8.2–22.7)	30.5
Tetrachloroethene (μg/L)	4.65 (<0.6–5)				
Toluene (μg/L)	4.93 (<0.7–5)				
Trans–1,2–dichloroethane		25.08 (<5–50)			
Trans–1,2–dichloroethene	27.01 (<2–53)				
Trichloroethelene (μg/l)		41.773 (<5–69)			
Trichloroethene	0.033 (<0.002–0.060)			0.0138 (0.007–0.027)	0.807[d]
Turbidity (NTU)[e]	9.851 (1–290)	6.91 (0.85–72)	8.13 (2.2–80)		

Table A.1 (continued)

Parameter	Mean (range)				Reference value
	1990	1991	1992	1993	
Vinyl chloride (μg/L)	7.218 (<2–12)				
Zinc	0.023 (<0.0057–0.29)		<0.0252 (<0.02–0.092)	0.0347 (0.02–0.056)	0.117

Note: All reference values are Tennessee Water Quality Standards for Fish and Aquatic Life, unless otherwise noted. Table compiled from Kornegay et al., 1991, 1992, 1993, 1994 (see Chapter 8 for full citations).

[a]Tennessee Primary Drinking Water Maximum Contaminant Level.

[b]Tennessee Secondary Drinking Water Maximum Contaminant Level.

[c]Minimum value.

[d]Tennessee Water Quality Standard for Recreation.

[e]Nephelometric turbidity units.

Table A.2. Mean concentrations of National Pollutant Discharge Elimination System parameters detected in the water at K-1007-B Pond
Measured in mg/L unless otherwise noted

Parameter	Mean (range)				Reference value
	1990	1991	1992	1993	
Alkalinity				96.25 (75-108)	
Calcium Hardness				73.50 (48.00-91.00)	
Chemical Oxygen	8.649 (<4.5–23)	8.939 (<5–21)	<7.5244 (<5–22)	16.50 (5.00-40.00)	
Chloride				10.05 (9.40-11.00)	250[a]
Chlorine				0.08 (0.04-0.12)	
Chromium	0.01 (<0.01–0.037)	0.0106 (0.01–0.04)	<0.01 (<0.01–0.0105)		
Color				498 (490-500)	
Conductivity				338.00 (216.00-588.00)	
Dissolved Oxygen	9.52 (4.3–20)		9.5608 (5.1–18)	9.85 (7.50-11.50)	5.0[b]
Dissolved Solids				157.50 (138.00-186.00)	500[a]
Flow (L/s)		86.3 (15.02–2037.9)	45.5 (3.9–635.2)		
Fluoride	0.132 (<0.1–0.2)	0.1217 (<0.01–1)	<0.1244 (<0.1–0.2)	0.10 (0.10-0.10)	4.0[c]
Iron				0.33 (0.12-0.61)	0.3[a]
Manganese				0.06 (0.04-0.11)	0.05
Nickel				0.04 (0.05-0.01)	1.40
Nitrate				0.23 (0.10-0.45)	10.0[c]
Oil and Grease		2.03 (<2–3.5)	<2.259 (<0.1–7.53)		15.0
pH (standard units)	7.1–9.3	7–9.5	8.388 (7.3–9.2)	8.58 (7.80-9.00)	6.5–8.5
Potassium				2.18 (1.90-2.90)	
Sodium				4.55 (4.00-4.90)	
Sulfate				22.75 (20.00-28.00)	250[a]
Suspended Solids	9.328 (<1–20)	10.144 (1–24)	8.9512 (4–25)	12.75 (5.00-24.00)	
Temperature (°C)	18.198 (4.8–29.8)			19.43 (6.30-30.30)	30.50
Total Phosphate as				0.12 (0.06-0.17)	
Uranium			0.001 (0.001–0.001)		
Zinc				0.02 (0.02-0.03)	0.12

Note: All reference values are Tennessee Water Quality Standards for Fish and Acquatic Life, unless otherwise noted. Table compiled from data in Kornegay et al., 1991, 1992, 1993 and 1994 (see Chapter 8 for full citations).

[a]Tennessee Secondary Drinking Water Maximum Contaminant Level

[b]Minimum value.

[c]Tennessee Primary Drinking Water Maximum Contaminant Level.

Table A.3. Mean concentrations of National Pollutant Discharge Elimination System parameters detected in the water at the Central Neutralization Facility (K-1407-J basin)
Expressed as mg/L unless otherwise noted

Parameter	Mean (range)				Reference value[a]
	1990	1991	1992	1993	
Total toxic organics	<0.145 (<0.005-2.123)				
1,1,1-Trichloroethane	0.00531 (<0.0005-0.033)			<0.0005(<0.0005-<0.0005)	
1,1-Dichloroethane	0.004912 (<0.0007-0.006)				
1,1-Dichloroethene	0.005 (<0.0009-0.021)		<0.0046 (<0.0012-0.006)		
1,2-Dichloropropane			<0.005 (<0.0007-6)		
2,4-Dichlorophenol	0.00984 (<0.001-0.01)				
4-Nitrophenol (μg/L)	52.17 (50-200)				
Acetone				<0.01(<0.01-<0.01)	
Acetonitrile				<0.01(<0.01-<0.01)	
Aluminum	0.205 (<0.02-2.8)	0.205 (0.095-1.9)			
Ammonia nitrogen	0.281 (<0.14-1.7)	0.201 (<0.2-0.25)			
Anthracene (μg/L)	9.931 (<6-10)				
Aroclor-1254			<0.0009 (<0.005-0.00019)		
Arsenic	0.005 (<0.005-0.011)		<0.0038 (<0.002-0.004)		
Barium	0.099 (<0.0066-0.1)	0.098 (0.022-0.33)			
Benzene	0.00489 (<0.0008-0.005)			<0.0007(<0.0007-<0.0007)	
Benzidine (μg/L)		10.43 (<10-40)			
Benzo(a)anthracene (μg/L)		3.6 (10.43-<40)			
Bis(2-ethylhexyl)phthalate	9.982 (<3-1.2)				
Boron	0.084 (<0.004-1.2)	0.367 (0.019-1.6)			
Bromide		2.39 (1-10)			
Bromoform				<0.0019(<0.0019-<0.0019)	
Bromodichloromethane	0.0047 (<0.0005-0.006)	0.004996 (<0.002-0.007)	<0.0044 (<0.0017-0.007)		
Butylbenzylphthalate			<0.0087 (<0.0021-0.005)		
Cadmium		<0.0021 (<0.002-0.0057)		<0.002(0.003 -<0.002)	0.69
Carbontetrachloride				<0.0006(<0.0006-<0.0006)	0.5
Chemical Oxygen Demand	16.28 (<5-89)	<15.5 (<5-343)	<5.4687 (<5-11)	<8(31 -<1)	250[b]

Table A.3 (continued)

Parameter	Mean (range)				Reference value[a]
	1990	1991	1992	1993	
Chloride	153.08 (20-883)	626.21 (<20-3090)	309.96 (35-1927)	524 (2051 -75)	39479
Chlorine, total residual	0.044 (<0.02-0.5)	<0.072 (<0.02-0.22)	0.0316 (0.01-0.09)	<0.05(0.13 -<0.05)	0.14
Chlorobenzene			<0.0049 (<0.0004-0.001)		
Chloroethane			<0.0088 (<0.0017-0.01)		
Chlorodibromomethane				<0.0008(<0.0008-<0.0008)	
Chloroform	0.00438 (<0.0003-0.031)	<0.00586 (<0.001-0.052)	<0.0063 (<0.0045-0.043)	0.0031 (0.0055 -0.0018)	0.5
Chromium	0.01 (<0.01-0.05)	0.014 (<10-0.1)	<0.0137 (<0.01-0.017)	<0.01(0.02 -<0.01)	2.77
Copper	0.011 (<004-0.1)	<0.014 (<0.004-0.081)	<0.0103 (<0.004-0.04)	<0.007(0.011 -<0.004)	2.15
Cyanide colorimetric	0.081 (<0.002-0.1)	<0.11 (<0.1-0.33)	<0.124 (<0.1-0.14)		
Di-n-butylphthalate			<0.0074 (<0.0024-0.004)		
Dichlorobromomethane				<0.0017(<0.0017-<0.0017)	
Dissolved solids (μg/L)	658.3 (182-2394)	2077.2 (260-7322)	1671.475 (498-3864)		500[b]
Ethylbenzene	0.00495 (<0.0008-0.005)			<0.0005(<0.0005-<0.0005)	
Fluoranthene (μg/L)	9.948 (<8-10)				
Fluoride	1.452 (0.2-11)	13.2 (0.2-71)			4.0[c]
Flow, L/s			5.12 (0-27.16)	4.82 (0–19.27)	
Iron	0.552 (<0.05-9.9)	0.68 (0.099-3.3)	0.2789 (0.06-0.8)		0.3[b]
Kjeldahl nitrogen	1.244 (<0.6-3)	1.08 (0.84-2.74)			
Lead	0.005 (<0.004-0.03)	<0.0046 (<0.004-0.016)	<0.004 (<0.002-0.0072)	<0.004(<0.004-<0.004)	0.69
Magnesium	12.84 (8-17)	17.36 (8.9-47)			
Manganese	0.049 (<0.0089-0.28)	0.34 (<0.01-0.342)	<0.0328 (<0.01-0.079)		0.3[b]
MBAS		<0.203 (<0.2-0.3)			
Mercury	<0.001 (<0.0002-0.0045)				
Methylethylketone				<0.01(<0.01-<0.01)	
Methylene chloride	0.00616 (<0.0004-0.09)	0.00566 (<0.002-0.019)	<0.0042 (0.0012-0.011)	<0.0012(<0.0012-<0.0012)	
Naphthalene				<0.0011(<0.0016-<0.0009)	
Nickel	0.071 (<0.024-0.5)		<0.0618 (<0.01-0.15)	<0.07(0.1 -<0.05)	3.98
Nitrate nitrogen		<2.02 (<0.2-23)			10.0[c]

Table A.3 (continued)

Parameter	Mean (range)				Reference value[a]
	1990	1991	1992	1993	
Oil and grease	2.021 (<2-3.79)	<2.3 (<2-14.5)	<2.533 (<2-12.4)	<2.1(3.8 -<2)	30
PCB				<0.0005(<0.0005-<0.0005)	0.0001
PCB (Aroclor-1250) (μg/L)		1.01 (<1-1.4)			
PCB (Aroclor-1254) (μg/L)	0.984 (<0.27-1)				
pH (standard units)	7.89 (7.0-9.3)	6.7-8.9	7.9451 (7.3-8.9)	(6.0-8.7)	6.0-9.0
Phenanthrene (μg/L)	9.913 (<5-10)				
Phenol (μg/L)	10.379 (<6-20)				
Phenols (μg/L)	0.07 (<0.01-2)	<0.07 (0.03-1.5)			
Phosphorus	1.536 (<0.2-15)	<3.06 (<0.2-3.7)			
Pyrene (μg/L)	10.069 (<9-15)				
Selenium	0.005 (<0.004-0.0057)				
Silver	0.01 (<0.006-<0.01)	<0.011 (<0.01-0.06)		0.01 (0.02 -<0.01)	0.43
Sulfate	234.239 (49-576)	553.23 (135-1500)	426.88 (140-1000)		250[b]
Sulfide		<1.02 (<1-2)			
Sulfite		<1.38 (<0.1-2)			
Total suspended solids	7.097 (<1-576)	7.18 (0.1-36)	4.7 (<1-46)	4 (36 -1)	40
Temperature (°C)	20.285 (0-34.1)		19.59 (8.5-32.4)	23.3 (35.3 -9.4)	[d]
Tetrachloroethene (μg/L)	5.101 (<0.6-59)				
Tetrachloroethylene				<0.0004(<0.0004-<0.0004)	0.7
Tin	0.01 (<0.01-0.013)	<0.02 (<0.01-0.05)			
Titanium	0.004 (<0.003-0.068)	<0.0053 (<0.003-0.03)			
Toluene	0.0048 (<0.0008-0.006)	<0.00503 (<0.005-0.011)		<0.0006(<0.0006-<0.0006)	
Total organic carbon	5.12 (2-15)	<4.05 (<1-19)			
Total phosphate	4.03 (0.5-21)	9.32 (0.6-92)			
Total toxic organics				0.0038 (0.0064 -0.0022)	2.13
Trans-1,2-dichloroethene (μg/L)	8.169 (<1-64)	<5.04 (<5-8)			
Trans-1,2-dichloropropene	5.024 (<0.08-460)				
Trichloroethene	0.0174 (<0.0008-0.00223)		<0.0042 (<0.00145-0.007)		

Table A.3 (continued)

Parameter	Mean (range)				Reference value[a]
	1990	1991	1992	1993	
Trichloroethylene				<0.0035(0.0041 -<0.0009)	0.5
Uranium	0.239 (0.032-2.23)		0.1158 (0.017-0.43)		
Vinyl chloride	0.09768 (<0.001-0.01)		<0.0096 (<0.0015-0.002)	<0.0015(<0.0015-<0.0014)	0.2
Zinc	0.033 (<0.02-0.31)	<0.028 (<0.002-0.28)	<0.0276 (<0.02-0.37)	<0.03(0.056 -<0.02)	2.61
bis(2-Ethylhexyl)phthalate			<0.0086 (<0.0025-0.025)		

Note: Table compiled from data in Kornegay et al. 1991, 1992, 1993, 1994 (see Chapter 8 for full citations).
[a]NPDES permit limit unless otherwise noted.
[b]Tennessee Secondary Drinking Water Maximum Contaminant Level.
[c]Tennessee Primary Drinking Water maximum Contaminant Level.
[d]Effluent must not cause the temperature of receiving stream to exceed 30.5°C.

Table A.4. Mean concentrations of National Pollutant Discharge Elimination System parameters detected in the water at K-1700-E/F ponds
Expressed as mg/L unless otherwise noted

Parameter	Mean (range)	
	1990	1991
Arsenic	0.005 (<0.005–0.0065)	
Cadmium	0.002 (<0.002–0.004)	
Chromium	0.01 (<0.01–0.011)	
Copper	0.024 (<0.004–0.21)	0.0242 (0.00733–0.037)
Flow		0.13947 (0–0.6554)
Iron	0.364 (<0.05–1.9)	0.745 (0.2–2.4)
Lead	0.004 (<0.004–0.0067)	0.0044 (0.004–0.008)
Manganese	0.134 (0.023–1.1)	0.181 (0.06–0.54)
Nickel	0.198 (<0.041–2.6)	0.086 (<0.05–0.239)
Oil and grease	2.024 (<2–3.52)	
PCB (Aroclor 1254) (μg/L)		1.107 (<1.0–2.6)
pH (standard units)	3.7–9.6	6.4–9.0
Selenium		0.006 (0.005–0.02)
Silver	0.009 (<0.006–0.01)	
Sulfate	649.47 (75–1440)	1007.25 (76–1660)
Suspended solids	8.95 (1–42)	8.286 (76–1660)
Temperature (°C)	19.3 (3.5–38.3)	
Thorium (pCi/L)	5.88 (1.05–10.7)	
Zinc	0.22 (<0.006–0.12)	0.025 (0.02–0.047)

Note: Table compiled from data in Kornegay et al. 1991, and 1992 (See Chapter 8 for full citation).

Table A.5. National Pollutant Discharge Elimination System Compliance at K-1700 at Mitchell Branch kilometer 0.12, K-1700-B pond, K-1407-E/F ponds, and K-1407-J basin, 1990–1993

Parameter	K1700	K-1700-B	K-1407-E/F	K-1407-J
		1990[a]		
Aluminum	7			
Chromium		1		
Dissolved oxygen		1		
Iron			4	
		1991[a]		
Aluminum	7			
Chemical oxygen Demand	1	5		
Chromium		1		
Dissolved oxygen	2	2		
PCB		1		
Iron		4		
Suspended solids		1		
		1992[a]		
Aluminum	4			
Chemical oxygen Demand		1		
Oil & grease	1			
		1993[b]		
Iron	4	2		
Manganese	4	1		
pH		3		

Note: Compliance was 100% at K-1407-J.
[a]Number of noncompliances for this year.
[b]Number of values exceeding reference (see Tables A.1-A.4) for this year.

Table A.6. Monthly means (±1 SD) and range (number of days of record) of water temperature (°C) at Mitchell Branch kilometer (MIK) 0.50 just below the outfall of storm drain 190 and at two reference sites, Grassy Creek kilometer (GCK) 2.4 and Scarboro Creek kilometer (SCK) 2.2

Sampling period		MIK 0.50		GCK 2.4	
Year	Month	Mean	Range	Mean	Range
1987	April	17.7 (2.1)	12.1–24.7 (20)	13.2 (2.4)	6.8–19.5 (20)
	May	22.2 (2.2)	17.1–29.6 (31)	16.0 (2.0)	9.9–19.7 (31)
	June	23.5 (3.9)	11.9–41.2 (30)	17.8 (1.4)	13.3–21.7 (30)
	July	25.3 (6.4)	16.8–41.4 (5.5)	18.9 (1.2)	15.7–31.1 (28)
	August	NA[a]	NA	20.1 (1.0)	16.8–22.0 (31)
	September	NA	NA	16.9 (2.0)	12.0–19.9 (30)
	October	NA	NA	9.5 (2.2)	4.9–15.9 (31)
	November	NA	NA	7.4 (2.4)	3.1–11.3 (30)
	December	12.8 (1.2)	10.4–16.4 (12)	6.0 (1.8)	3.4–10.7 (12)
1988	January	9.9 (1.4)	5.6–13.4 (25)	3.0 (2.7)	−(6.8)–4.3 (25)
	February	11.4 (1.7)	7.6–16.5 (29)	4.5 (2.4)	0.6–10.1 (29)
	March	13.1 (2.1)	8.3–21.8 (31)	7.9 (3.9)	−(3.6)–18.0 (31)
	April	16.6 (1.9)	12.3–21.9 (30)	12.4 (2.7)	6.1–19.7 (30)
	May	20.1 (2.0)	14.4–24.6 (31)	14.9 (2.8)	7.4–20.9 (31)
	June	24.3 (1.9)	19.1–29.5 (30)	18.3 (3.0)	11.4–25.3 (30)
	July	25.3 (1.9)	19.6–32.2 (26)	20.8 (2.1)	14.7–25.6 (26)
	August	26.7 (1.2)	23.1–29.9 (26)	21.3 (1.4)	16.8–24.1 (26)
	September	23.9 (1.3)	20.4–28.3 (30)	18.3 (2.0)	12.7–23.5 (30)

Table A.6 (continued)

Sampling period		MIK 0.50		GCK 2.4	
Year	Month	Mean	Range	Mean	Range
	October	18.3 (2.3)	13.6–24.9 (31)	10.3 (3.7)	2.6–21.4 (31)
	November	14.7 (1.5)	10.8–19.1 (30)	8.8 (2.0)	4.1–13.1 (30)
	December	11.3 (1.2)	5.8–14.5 (31)	3.7 (2.7)	−(0.2)–10.3 (31)
1989	January	10.6 (1.3)	5.7–14.2 (20)	7.2 (1.8)	2.5–10.7 (20)
	February	10.9 (2.6)	5.9–16.6 (16)	7.3 (2.2)	1.7–13.8 (28)
	March	NA	NA	10.2 (2.5)	4.9–18.1 (31)
	April	NA	NA	12.1 (2.5)	6.0–18.4
	May	19.5 (1.7)	15.1–24.2 (18)	14.2 (2.4)	9.0–20.9 (26)
	June	20.2 (1.3)	17.5–25.4 (30)	16.7 (1.0)	14.1–20.8 (30)
	July	21.4 (1.7)	19.4–27.8 (31)	17.6 (1.1)	14.4–20.1 (31)
	August	23.3 (0.9)	21.0–27.3 (28)	17.8 (1.5)	13.1–21.5 (28)
	September	NA	NA	17.5 (1.8)	12.8–20.9 (30)
	October	NA	NA	13.1 (2.8)	7.1–18.5 (31)
	November	NA	NA	10.3 (2.0)	5.3–14.3 (30)
	December	NA	NA	4.9 (2.4)	0.4–9.8 (28)
1990	January	10.5 (1.0)	8.0–12.5 (12)	7.5 (1.6)	2.9–11.1 (31)
	February	11.6 (1.5)	7.7–16.7 (28)	9.4 (1.7)	4.6–12.8 (28)
	March	13.1 (1.8)	9.2–18.1 (31)	10.7 (2.1)	5.9–16.6 (31)
	April	15.7 (2.6)	9.8–24.1 (30)	12.7 (1.8)	10.0–16.6 (2)

Table A.6 (continued)

Sampling Period		MIK 0.50		GCK 2.4	
Year	Month	Mean	Range	Mean	Range
	May	18.3 (2.0)	13.2–23.1 (29)	14.7 (1.5)	10.4–19.2 (29)
	June	22.1 (2.0)	17.6–31.1 (30)	16.4 (1.3)	12.2–20.8 (30)
	July	24.1 (1.3)	21.4–28.0 (31)	18.2 (1.1)	15.3–22.0 (31)
	August	24.2 (1.2)	20.8–28.8 (31)	18.9 (1.2)	16.1–23.3 (31)
	September	22.7 (1.9)	19.2–29.1 (26)	17.1 (2.6)	11.0–20.3 (28)
	October	19.1 (1.8)	15.3–23.6 (31)	12.7 (3.2)	5.2–18.3 (31)
	November	14.9 (1.4)	11.0–18.5 (30)	7.8 (1.7)	4.0–12.9 (30)
	December	12.1 (1.7)	4.6–15.6 (31)	7.5 (2.4)	3.7–13.1 (30)
1991	January	10.4 (1.5)	6.4–14.4 (31)	7.8 (2.1)	1.9–11.0 (31)
	February	9.9 (2.1)	3.0–15.3 (28)	7.6 (2.2)	2.3–12.1 (28)
	March	12.6 (2.0)	8.5–18.5 (31)	10.5 (2.2)	5.6–15.8 (31)
	April	16.3 (1.8)	10.5–21.3 (28)	13.3 (2.0)	8.5–17.8 (16)
	May	20.5 (2.1)	14.5–26.5 (31)		
	June	23.1 (1.2)	20.2–26.5 (30)		
	July	24.4 (0.8)	22.3–28.8 (31)		
	August	24.7 (0.6)	22.9–27.5 (30)		
	September	23.2 (2.0)	18.1–26.4 (30)		
	October	19.4 (1.5)	15.2–22.7 (31)	SCK 2.2	
				Mean	Range
	November	14.4 (1.8)	10.8–18.6 (30)	11.0 (2.2)	6.4–15.9 (30)

Table A.6 (continued)

Sampling Period		MIK 0.50		SCK 2.2	
Year	Month	Mean	Range	Mean	Range
	December	12.6 (1.5)	9.8–17.3 (12)	11.6 (1.0)	9.0–15.0 (29)
1992	January	NA	NA	10.6 (1.2)	7.8–13.2 (31)
	February	NA	NA	10.5 (1.2)	6.5–13.3 (29)
	March	NA	NA	10.7 (0.8)	8.5–12.3 (10)
	April	NA	NA	13.9 (1.9)	10.4–18.1 (7)
	May	19.0 (5.8)	8.6–40.1 (31)	15.5 (1.5)	11.6–19.6 (31)
	June	21.5 (1.7)	12.5–28.9 (30)	NA	NA
	July	23.8 (1.2)	21.0–26.9 (31)	NA	NA
	August	24.2 (5.2)	16.1–45.0 (30)	17.4 (1.0)	15.0–19.6 (18)
	September	22.7 (5.2)	10.0–42.7 (30)	17.1 (1.1)	12.6–19.6 (30)
	October	15.8 (5.1)	5.7–39.2 (31)	13.8 (1.5)	9.0–16.8 (31)
	November	14.4 (1.6)	10.5–19.9 (30)	11.7 (1.8)	8.0–16.8 (30)
	December	11.6 (0.9)	8.5–13.5 (20)	9.8 (1.3)	6.3–13.1 (25)
1993	January	11.1 (1.1)	8.0–13.9 (31)	11.1 (0.9)	8.0–13.5 (31)
	February	10.6 (2.2)	5.6–15.7 (28)	10.0 (1.3)	6.3–13.3 (28)
	March	10.8 (2.1)	5.2–17.1 (30)	11.0 (1.7)	5.6–15.6 (30)
	April	14.0 (2.0)	10.0–19.0 (29)	13.3 (1.5)	10.5–17.4 (28)
	May	19.0 (1.9)	14.6–24.4 (31)	15.2 (1.4)	11.8–20.2 (31)
	June	21.8 (1.7)	16.8–25.4 (30)	16.6 (1.2)	13.5–19.1 (30)

Table A.6 (continued)

Sampling Period		MIK 0.50		SCK 2.2	
Year	Month	Mean	Range	Mean	Range
	July	24.1 (1.4)	21.9–27.7 (19)	18.3 (1.1)	15.7–21.1 (29)
	August	NA	NA	18.2 (1.1)	15.4–21.0 (31)
	September	19.9 (1.7)	16.0–22.5 (9)	17.0 (1.7)	11.8–22.3 (30)
	October	17.7 (2.3)	12.8–22.1 (20)	13.8 (1.8)	9.5–18.0 (30)
	November	13.4 (1.7)	9.1–17.7 (30)	11.2 (1.7)	8.2–15.3 (30)
	December	10.2 (1.6)	6.8–14.1 (31)	10.2 (1.4)	6.4–13.3 (31)
1994	January	7.3 (1.6)	2.0–10.7 (30)	9.2 (1.4)	4.1–11.6 (30)
	February	9.3 (1.7)	5.2–14.1 (28)	10.8 (1.3)	6.0–13.9 (28)
	March	11.2 (1.6)	7.7–15.8 (31)	11.8 (1.3)	7.9–16.3 (31)

[a]NA = no data available.

Note: Data were obtained with Ryan Tempmentor digital temperature recorders with values recorded every 20 min, April–June, 1987 and every 60 min beginning July 1987.

APPENDIX B

RESULTS OF AMBIENT TOXICITY TESTS AND WATER QUALITY MEASUREMENTS, JANUARY 1990 THROUGH JANUARY 1994

Table B.1. Summary (mean ± SD) of water chemistry analyses conducted during toxicity tests of ambient waters from Mitchell Branch, Poplar Creek and Clinch River

Sample[a]	Date	pH	Alkalinity (mg/L as $CaCO_3$)	Hardness (mg/L as $CaCO_3$)	Conductivity (µS/cm)
MIK 1.43	Jan. 1990	7.5 (0.2)	48.3 (5.5)	64.3 (5.5)	110.1 (15.3)
MIK 1.00		7.8 (0.1)	71.7 (15.2)	93.7 (13.4)	174.0 (24.7)
MIK 0.71		7.8 (0.1)	81.9 (23.9)	125.7 (26.5)	268.6 (101.9)
MIK 0.54		7.9 (0.1)	91.7 (21.6)	123.4 (31.6)	319.1 (122.3)
MIK 0.45		7.9 (0.1)	95.0 (25.4)	145.1 (43.9)	333.6 (126.6)
MIK 0.12		7.9 (0.1)	102.6 (33.1)	148.6 (37.3)	357.6 (140.3)
MIK 1.43	Mar. 1990	7.6 (0.1)	64.1 (6.2)	72.6 (12.0)	125.3 (14.0)
MIK 1.00		7.9 (0.1)	96.9 (5.5)	110.6 (10.4)	200.0 (10.1)
MIK 0.71		7.9 (0.1)	114.0 (7.8)	150.7 (26.6)	350.3 (121.9)
MIK 0.54		8.0 (0.1)	123.8 (9.3)	160.9 (33.7)	371.6 (119.3)
MIK 0.45		7.9 (0.1)	129.4 (9.4)	179.7 (29.2)	392.4 (110.8)
MIK 0.12		7.9 (0.1)	143.6 (9.5)	193.4 (31.2)	420.7 (94.9)
MIK 1.43	May 1990	7.6 (0.2)	71.6 (8.6)	79.7 (11.7)	147.3 (21.0)
MIK 1.00		7.9 (0.2)	107.1 (17.5)	120.3 (19.4)	222.4 (37.2)
MIK 0.71		7.9 (0.1)	119.1 (22.1)	159.7 (40.3)	359.9 (123.2)
MIK 0.54		8.0 (0.2)	125.1 (25.7)	169.7 (41.4)	369.6 (119.7)
MIK 0.45		7.9 (0.1)	124.6 (34.9)	173.4 (48.8)	373.6 (135.9)
MIK 0.12		7.9 (0.1)	137.0 (41.9)	188.0 (59.2)	415.0 (145.5)
MIK 1.43	July 1990	8.1 (0.2)	111.6 (19.1)	114.3 (4.8)	203.0 (3.7)
MIK 1.00		8.0 (0.1)	144.4 (15.9)	169.1 (7.4)	321.1 (15.1)
MIK 0.71		8.1 (0.1)	125.9 (4.40	158.9 (4.7)	336.7 (21.2)
MIK 0.54		8.1 (0.1)	128.3 (6.9)	167.4 (5.9)	355.4 (28.0)
MIK 0.45		8.1 (0.1)	122.6 (7.1)	164.3 (8.0)	344.3 (18.7)
MIK 0.12		8.0 (0.1)	134.1 (9.9)	193.1 (28.1)	433.4 (115.3)
MIK 1.43	Sep. 1990	8.0 (0.1)	111.4 (4.1)	123.7 (9.6)	215.4 (8.3)
MIK 1.00		7.9 (0.1)	163.7 (11.8)	192.6 (14.8)	351.4 (20.7)
MIK 0.71		8.1 (0.1)	120.3 (4.2)	156.6 (5.3)	333.7 (12.7)
MIK 0.54		8.1 (0.1)	125.6 (4.2)	172.6 (24.9)	383.9 (112.0)
MIK 0.45		8.0 (0.1)	122.4 (4.0)	171.6 (24.8)	378.4 (98.5)
MIK 0.12		8.0 (0.1)	130.7 (3.9)	179.4 (13.5)	394.0 (56.2)
MIK 1.43	Nov. 1990	7.7 (0.2)	100.7 (13.3)	106.9 (11.8)	193.4 (10.6)
MIK 1.00		7.8 (0.1)	135.9 (25.6)	154.9 (27.8)	302.3 (43.6)
MIK 0.71		8.0 (0.1)	127.6 (17.9)	156.9 (18.0)	364.3 (50.2)
MIK 0.54		8.0 (0.1)	134.3 (20.0)	175.1 (21.0)	385.4 (70.4)
MIK 0.45		8.0 (0.1)	132.4 (20.3)	171.4 (21.6)	368.9 (51.4)

Table B.1 (continued)

Sample[a]	Date	pH	Alkalinity (mg/L as $CaCO_3$)	Hardness (mg/L as $CaCO_3$)	Conductivity (µS/cm)
MIK 0.12		8.0 (0.1)	136.1 (21.9)	186.3 (47.1)	454.3 (150.8)
MIK 1.43	Jan. 1991	7.5 (0.2)	62.4 (3.5)	78.3 (6.3)	138.0 (5.6)
MIK 1.00		7.8 (0.1)	97.1 (2.9)	121.1 (9.2)	211.4 (9.5)
MIK 0.71		7.9 (0.1)	118.0 (10.1)	155.1 (13.4)	313.1 (41.8)
MIK 0.54		8.0 (0.1)	126.7 (12.1)	172.3 (15.6)	340.3 (46.3)
MIK 0.45		8.1 (0.1)	135.9 (15.0)	184.6 (21.4)	374.9 (43.9)
MIK 0.12		8.1 (0.1)	144.3 (26.6)	251.1 (149.3)	557.3 (431.8)
MIK 1.43	Mar. 1991	7.6 (0.1)	56.6 (2.4)	64.9 (3.4)	147.6 (19.5)
MIK 1.00		7.8 (0.1)	76.9 (20.4)	90.9 (15.6)	203.9 (34.4)
MIK 0.71		7.8 (0.10	96.7 (20.6)	113.4 (24.7)	262.7 (56.5)
MIK 0.54		7.8 (0.1)	104.6 (25.2)	122.3 (29.9)	286.4 (65.2)
MIK 0.45		7.8 (0.1)	105.9 (36.5)	129.9 (44.6)	295.9 (96.6)
MIK 0.12		7.8 (0.2)	101.9 (47.9)	148.6 (52.9)	349.6 (161.5)
MIK 1.43	May 1991	7.8 (0.1)	95.4 (2.4)	119.4 (32.9)	199.3 (2.2)
MIK 1.00		7.9 (0.2)	130.7 (2.8)	160.6 (38.6)	284.1 (9.8)
MIK 0.71		7.9 (0.1)	147.4 (7.4)	180.0 (12.7)	375.0 (14.9)
MIK 0.54		7.9 (0.1)	151.3 (8.9)	188.6(7.2)	401.3 (13.4)
MIK 0.45		7.9 (0.1)	169.3 (63.6)	181.7 (7.7)	384.4 (13.6)
MIK 0.12		8.0 (0.1)	149.3 (13.9)	194.0 (14.1)	399.1 (29.6)
MIK 1.43	July 1991	7.7 (0.2)	108.9 (2.8)	112.7 (4.2)	201.3 (7.2)
MIK 1.00		7.8 (0.2)	151.0 (25.3)	162.6 (27.6)	294.6 (40.7)
MIK 0.71		7.8 (0.2)	126.0 (20.4)	165.7 (27.7)	336.7 (56.7)
MIK 0.54		7.8 (0.2)	125.6 (22.4)	162.0 (29.8)	333.1 (58.0)
MIK 0.45		7.7 (0.2)	120.4 (28.4)	159.7 (36.2)	326.1 (74.2)
MIK 0.12		7.7 (0.3)	135.7 (3.7)	176.9 (5.2)	360.1 (15.9)
MIK 1.43	Sep. 1991	7.7 (0.2)	121.7 (6.4)	135.7 (13.0)	224.4 (14.3)
MIK 1.00		--- (---)	---- (---)	---- (---)	---- (---)
MIK 0.71		7.8 (0.1)	114.0 (5.0)	155.7 (4.2)	337.1 (6.3)
MIK 0.54		7.8 (0.1)	119.0 (4.0)	164.9 (11.6)	327.9 (16.4)
MIK 0.45		7.8 (0.1)	119.3 (4.0)	158.9 (10.7)	322.4 (11.8)
MIK 0.12		7.7 (0.1)	127.0 (2.8)	176.0 (15.0)	344.1 (17.9)
MIK 1.43	Nov. 1991	7.6 (0.2)	97.7 (18.2)	112.6 (21.1)	191.4 (26.5)
MIK 1.00		7.6 (0.2)	134.1 (32.2)	169.0 (34.60	307.9 (51.8)
MIK 0.71		7.7 (0.2)	131.9 (25.2)	176.9 (24.7)	344.3 (58.4)
MIK 0.54		7.7 (0.2)	139.0 (28.0)	187.4 (30.4)	356.0 (56.5)
MIK 0.45		7.8 (0.2)	132.4 (22.9)	178.0 (25.6)	347.3 (50.7)

Table B.1 (continued)

Sample[a]	Date	pH	Alkalinity (mg/L as $CaCO_3$)	Hardness (mg/L as $CaCO_3$)	Conductivity (µS/cm)
MIK 0.12		7.7 (0.2)	137.3 (34.2)	186.0 (42.6)	353.0 (81.8)
MIK 1.43	Jan. 1992	7.7 (0.1)	61.9 (6.8)	83.1 (19.7)	123.9 (13.8)
MIK 1.00		8.0 (0.1)	92.7 (8.1)	106.6 (13.2)	192.6 (15.0)
MIK 0.71		8.0 (0.1)	112.6 (5.7)	136.6 (14.6)	265.1 (17.0)
MIK 0.54		8.0 (0.1)	123.6 (6.8)	151.7 (10.4)	289.4 (20.2)
MIK 0.45		8.0 (0.1)	127.9 (6.4)	165.4 (14.8)	311.4 (21.6)
MIK 0.12		8.0 (0.1)	139.9 (7.4)	178.0 (9.4)	334.1 (22.1)
MIK 1.43	Mar. 1992	7.7 (0.1)	59.1 (2.0)	70.9 (9.9)	126.0 (4.7)
MIK 1.00		8.0 (0.1)	89.7 (3.3)	102.0 (6.2)	196.3 (6.5)
MIK 0.71		8.0 (0.1)	108.0 (4.7)	133.1 (5.5)	265.4 (9.3)
MIK 0.54		8.0 (0.1)	118.9 (4.9)	150.0 (6.9)	296.4 (14.9)
MIK 0.45		8.0 (0.1)	121.9 (3.5)	156.9 (10.3)	310.7 (14.1)
MIK 0.12		8.0 (0.1)	129.6 (11.8)	161.7 (17.1)	324.6 (27.4)
MIK 1.43	May 1992	8.1 (0.1)	95.1 (6.2)	108.0 (8.2)	176.6 (11.6)
MIK 1.00		8.1 (0.1)	132.7 (7.2)	149.7 (3.9)	255.9 (14.8)
MIK 0.71		8.1 (0.1)	130.1 (8.3)	173.4 (7.4)	329.7 (17.2)
MIK 0.54		8.1 (0.1)	139.7 (4.8)	184.3 (7.8)	348.1 (16.1)
MIK 0.45		8.1 (0.1)	129.6 (6.1)	177.4 (8.0)	339.7 (19.5)
MIK 0.12		8.0 (0.2)	138.3 (5.8)	185.7 (4.7)	353.7 (23.5)
MIK 1.43	July 1992	8.1 (0.1)	114.1 (4.6)	120.9 (8.1)	205.7 (10.5)
MIK 1.00		8.2 (0.1)	160.9 (12.7)	175.1 (11.1)	310.6 (16.2)
MIK 0.71		8.1 (0.1)	128.7 (5.2)	171.1 (9.9)	341.1 (21.8)
MIK 0.54		8.2 (0.1)	136.1 (6.0)	175.1 (8.1)	356.1 (20.3)
MIK 0.45		8.1 (0.1)	125.6 (3.3)	168.3 (6.4)	350.1 (14.7)
MIK 0.12		8.0 (0.2)	127.4 (17.6)	168.0 (26.3)	348.1 (50.2)
MIK 1.43	Sep. 1992	8.1 (0.2)	117.6 (4.7)	127.1 (8.6)	223.3 (11.7)
MIK 1.00		8.1 (0.2)	159.4 (35.3)	182.6 (40.6)	336.0 (57.5)
MIK 0.71		8.1 (0.1)	133.3 (20.9)	176.6 (32.9)	360.7 (59.0)
MIK 0.54		8.1 (0.1)	138.4 (24.4)	183.4 (39.0)	364.4 (69.5)
MIK 0.45		8.1 (0.1)	139.4 (29.1)	189.7 (46.2)	381.4 (83.0)
MIK 0.12		8.0 (0.1)	148.0 (40.8)	189.1 (59.1)	392.1 (105.5)
MIK 1.43	Nov. 1992	8.0 (0.2)	103.7 (4.5)	112.3 (9.3)	207.0 (5.9)
MIK 1.00		8.1 (0.1)	139.4 (21.7)	163.7 (24.4)	314.1 (48.0)
MIK 0.71		8.1 (0.1)	131.7 (16.3)	168.9 (23.9)	341.7 (41.9)
MIK 0.54		8.1 (0.2)	139.3 (19.8)	174.0 (26.8)	361.6 (56.4)
MIK 0.45		8.1 (0.1)	146.7 (27.7)	193.1 (40.6)	388.4 (70.3)

Table B.1 (continued)

Sample[a]	Date	pH	Alkalinity (mg/L as $CaCO_3$)	Hardness (mg/L as $CaCO_3$)	Conductivity (µS/cm)
MIK 1.43	Feb. 1993	7.7 (0.2)	51.4 (7.5)	70.3 (12.0)	115.6 (14.8)
MIK 1.00		7.9 (0.1)	71.7 (14.5)	93.4 (15.1)	165.9 (24.4)
MIK 0.71		8.0 (0.1)	86.3 (20.3)	121.4 (15.4)	222.1 (41.1)
MIK 0.54		8.1 (0.1)	95.6 (22.8)	123.4 (24.6)	227.7 (52.7)
MIK 0.45		8.1 (0.2)	100.7 (28.4)	136.0 (31.1)	261.6 (65.0)
MIK 0.12		8.1 (0.2)	110.0 (32.8)	147.4 (42.4)	279.7 (72.5)
MIK 0.12		8.1 (0.3)	164.1 (18.7)	209.4 (22.2)	420.9 (51.3)
MIK 1.43	Apr. 1993	7.8 (0.3)	60.4 (2.9)	78.3 (6.8)	130.3 (4.4)
MIK 0.78		8.0 (0.2)	102.9 (5.8)	133.1 (18.1)	219.1 (27.2)
MIK 0.71		8.1 (0.1)	101.3 (5.3)	140.6 (19.1)	250.7 (31.0)
MIK 0.54		8.1 (0.1)	111.1 (6.3)	145.4 (9.7)	281.9 (7.4)
MIK 0.45		8.1 (0.1)	117.4 (8.6)	172.9 (28.4)	310.6 (9.3)
MIK 0.12		8.2 (0.1)	125.4 (11.4)	178.0 (38.5)	331.3 (14.0)
PCK 18.2		7.9 (0.1)	46.3 (4.5)	84.0 (6.0)	166.3 (6.4)
PCK 8.2		8.0 (0.1)	57.3 (4.9)	92.7 (3.1)	187.3 (18.8)
PCK 6.9		8.1 (0.1)	60.3 (6.5)	130.7 (68.9)	197.3 (16.0)
PCK 4.7		8.0 (0.1)	66.3 (9.9)	106.0 (15.1)	209.7 (24.0)
PCK 1.6		8.0 (0.1)	64.0 (7.2)	120.7 (34.2)	205.3 (13.3)
CRK 15.0		8.0 (0.1)	101.7 (5.8)	150.0 (19.1)	254.3 (7.8)
MIK 1.43	July 1993	8.1 (0.1)	110.0 (2.2)	125.4 (14.2)	205.0 (5.2)
MIK 0.78		8.0 (0.1)	192.3 (9.1)	215.7 (25.7)	386.4 (25.2)
MIK 0.71		8.2 (0.1)	125.0 (5.4)	181.1 (18.0)	350.4 (15.8)
MIK 0.54		8.1 (0.1)	135.6 (6.8)	184.3 (12.3)	370.9 (1.5)
MIK 0.45		8.0 (0.1)	141.9 (4.1)	192.3 (10.1)	398.9 (8.0)
MIK 0.12		7.9 (0.1)	145.4 (3.8)	208.3 (17.3)	410.6 (6.1)
PCK 18.2		8.0 (0.1)	140.3 (4.5)	176.7 (13.6)	325.3 (4.7)
PCK 8.2		8.1 (0.1)	128.0 (3.0)	183.3 (26.6)	335.3 (3.5)
PCK 6.9		8.2 (0.1)	127.3 (2.1)	183.3 (16.2)	329.3 (19.4)
PCK 4.7		8.2 (0.2)	122.0 (2.0)	174.7 (31.3)	335.7 (7.6)
PCK 1.6		8.2 (0.2)	113.7 (3.5)	147.3 (7.0)	268.0 (28.2)
CRK 15.0		8.1 (0.2)	112.0 (2.0)	158.7 (23.7)	253.3 (15.0)
MIK 1.43	Oct. 1993	8.0 (0.1)	117.4 (4.1)	129.7 (17.9)	253.3 (62.7)
MIK 0.78		7.9 (0.1)	184.1 (6.4)	206.0 (23.2)	418.9 (81.6)
MIK 0.71		8.0 (0.2)	123.9 (30.6)	173.7 (42.9)	358.1 (106.3)
MIK 0.54		8.0 (0.1)	131.3 (31.6)	167.4 (38.7)	374.7 (110.8)
MIK 0.45		7.9 (0.1)	139.7 (40.6)	196.6 (44.0)	397.0 (129.5)

Table B.1 (continued)

Sample[a]	Date	pH	Alkalinity (mg/L as $CaCO_3$)	Hardness (mg/L as $CaCO_3$)	Conductivity (µS/cm)
MIK 0.12		7.8 (0.1)	152.9 (11.1)	207.1 (17.3)	433.7 (78.3)
PCK 18.2		7.8 (0.1)	140.7 (3.8)	180.0 (22.7)	337.0 (70.9)
PCK 8.2		7.8 (0.1)	132.0 (3.5)	192.7 (32.1)	375.0 (71.4)
PCK 6.9		7.7 (0.1)	129.0 (4.4)	189.3 (26.1)	376.3 (76.5)
PCK 4.7		7.8 (0.0)	124.7 (4.9)	175.3 (28.1)	311.7 (57.7)
PCK 1.6		8.2 (0.1)	123.3 (3.1)	165.3 (15.0)	326.0 (65.5)
CRK 15.0		8.0 (0.2)	120.0 (3.6)	158.0 (29.9)	296.0 (7.0)
MIK 1.43	Jan. 1994	7.8 (0.2)	45.7 (8.8)	61.4 (7.6)	118.7 (17.1)
MIK 0.78		7.9 (0.1)	82.0 (15.1)	100.0 (15.1)	225.9 (44.4)
MIK 0.71		8.0 (0.1)	86.0 (10.6)	112.6 (13.8)	257.0 (36.2)
MIK 0.54		8.1 (0.1)	96.9 (13.6)	130.6 (15.8)	295.0 (52.0)
MIK 0.45		8.1 (0.1)	102.6 (13.5)	140.3 (22.0)	333.1 (86.3)
MIK 0.12		8.0 (0.1)	114.3 (18.1)	147.1 (30.3)	341.9 (47.2)
PCK 18.2		7.4 (0.2)	41.0 (16.0)	73.3 (20.1)	172.3 (39.1)
PCK 8.2		7.7 (0.3)	47.7 (15.8)	91.3 (32.0)	193.3 (50.7)
PCK 6.9		7.7 (0.3)	53.7 (16.5)	86.7 (19.0)	202.0 (49.0)
PCK 4.7		7.6 (0.4)	55.3 (17.6)	84.7 (23.0)	208.3 (49.5)
PCK 1.6		7.5 (0.3)	60.0 (14.0)	90.0 (22.5)	226.3 (56.5)
CRK 15.0		7.8 (0.2)	101.3 (5.5)	124.0 (6.9)	259.7 (8.4)

[a]MIK = Mitchell Branch kilometer, PCK = Poplar Creek kilometer, CRK = Clinch River kilometer.

Table B.2. Results of toxicity tests of ambient samples from Mitchell Branch, Poplar Creek and Clinch River. Asterisks (*) indicate concentrations that are significantly different from control ($\alpha = 0.05$)

		Fathead Minnow		*Ceriodaphnia dubia*	
Sample[a]	Date	Mean survival (%)	Mean (SD) growth (mg/fish)	Survival (%)	Mean (SD) offspring/surviving female
Control	Jan. 1990	97.5	0.42 (0.04)	80	20.8 (4.1)
MIK 1.43		50.0	0.34 (0.01)	90	19.0 (5.5)
MIK 1.00		60.0	0.39 (0.02)	100	19.9 (1.6)
MIK 0.71		100.0	0.40 (0.02)	90	15.4 (6.8)
MIK 0.54		97.5	0.39 (0.04)	90	14.0 (6.8)
MIK 0.45		100.0	0.38 (0.05)	100	17.8 (1.7)
MIK 0.12		90.0	0.39 (0.05)	100	19.6 (3.8)
Control	Mar. 1990	92.5	0.58 (0.04)	100	17.6 (5.0)
MIK 1.43		12.5*	0.58 (—)	90	20.2 (2.7)
MIK 1.00		17.5*	0.50 (0.06)	80	21.8 (4.6)
MIK 0.71		90.0	0.62 (0.03)	20*	10.0 (7.1)*
MIK 0.54		87.5	0.56 (0.12)	70	9.3 (4.9)*
MIK 0.45		95.0	0.61 (0.04)	0*	— (—)
MIK 0.12		72.5	0.67 (0.04)	90	13.0 (7.7)
Control	May 1990	100.0	0.50 (0.08)	100	21.9 (3.6)
MIK 1.43		82.5	0.50 (0.09)	90	24.0 (4.0)
MIK 1.00		55.0*	0.59 (0.09)	100	28.1 (3.7)
MIK 0.71		95.0	0.56 (0.07)	100	28.5 (4.0)
MIK 0.54		95.0	0.64 (0.02)	100	22.6 (6.6)
MIK 0.45		95.0	0.56 (0.08)	100	25.3 (6.8)
MIK 0.12		100.0	0.60 (0.08)	80	19.0 (8.4)
Control	July 1990	97.5	0.58 (0.06)	70	10.9 (5.9)
MIK 1.43		50.0	0.56 (0.07)	100	28.5 (6.2)
MIK 1.00		87.5	0.59 (0.08)	100	32.2 (9.5)
MIK 0.71		95.0	0.61 (0.07)	0*	— (—)
MIK 0.54		92.5	0.67 (0.08)	0*	— (—)
MIK 0.45		90.0	0.67 (0.08)	0*	— (—)
MIK 0.12		92.5	0.65 (0.08)	100	26.0 (4.7)
Control	Sep. 1990	100.0	0.57 (0.07)	90	39.1 (10.6)
MIK 1.43		92.5	0.53 (0.05)	100	42.8 (10.9)
MIK 1.00		92.5	0.56 (0.09)	90	45.2 (5.0)

Table B.2 (continued)

		Fathead Minnow		*Ceriodaphnia dubia*	
Sample[a]	Date	Mean survival (%)	Mean (SD) growth (mg/fish)	Survival (%)	Mean (SD) offspring/surviving female
MIK 0.71		---	--- (---)	---	--- (---)
MIK 0.54		100.0	0.62 (0.07)	0*	--- (---)
MIK 0.45		0.0*	--- (---)	0*	--- (---)
MIK 0.12		100.0	0.62 (0.08)	100	32.2 (12.2)
Control	Nov. 1990	75.0	0.59 (0.04)	100	28.0 (2.4)
MIK 1.43		92.5	0.58 (0.03)	90	26.1 (2.1)
MIK 1.00		55.0*	0.62 (0.14)	100	27.8 (3.9)
MIK 0.71		0.0*	--- (---)	0*	--- (---)
MIK 0.54		---	--- (---)	0*	--- (---)
MIK 0.45		0.0*	--- (---)	0*	--- (---)
MIK 0.12		80.0	0.60 (0.05)	0*	--- (---)
Control	Jan. 1991	100.0	0.54 (0.06)	100	25.0 (2.4)
MIK 1.43		17.5*	0.49 (---)	90	31.1 (5.6)
MIK 1.00		20.0*	0.68 (0.09)	100	27.6 (2.1)
MIK 0.71		67.5	0.55 (0.03)	100	27.6 (2.8)
MIK 0.54	Jan. 1991	70.0	0.58 (0.06)	100	26.6 (2.6)
MIK 0.45		72.5	0.65 (0.07)	100	27.8 (2.6)
MIK 0.12		70.0	0.62 (0.01)	100	25.8 (1.4)*
Control	Mar. 1991	82.5	0.41 (0.04)	100	39.3 (2.1)
MIK 1.43		15.0*	0.40 (0.15)	100	38.4 (5.2)
MIK 1.00		60.0	0.37 (0.08)	100	36.6 (2.2)
MIK 0.71		50.0	0.29 (0.14)	100	27.0 (7.0)
MIK 0.54		57.5	0.56 (0.13)	100	29.5 (5.5)
MIK 0.45		60.0	0.38 (0.24)	90	34.3 (3.6)
MIK 0.12		72.5	0.37 (0.06)	100	34.0 (8.7)
Control	May 1991	97.5	0.56 (0.08)	100	43.0 (2.8)
MIK 1.43		47.5	0.62 (0.05)	100	44.3 (7.5)
MIK 1.00		65.0	0.55 (0.05)	90	41.3 (4.2)
MIK 0.71		80.0	0.51 (0.12)	90	46.9 (3.4)
MIK 0.54		70.0	0.54 (0.08)	100	46.3 (9.4)
MIK 0.45		75.0	0.54 (0.09)	100	43.7 (8.3)
MIK 0.12		82.5	0.47 (0.09)	100	44.9 (7.1)
Control	July 1991	95.0	0.58 (0.01)	100	36.7 (5.4)
MIK 1.43		55.0	0.62 (0.04)	80	40.4 (4.7)

Table B.2 (continued)

Sample[a]	Date	Fathead Minnow		*Ceriodaphnia dubia*	
		Mean survival (%)	Mean (SD) growth (mg/fish)	Survival (%)	Mean (SD) offspring/surviving female
MIK 1.00		92.5	0.57 (0.02)	80	32.0 (3.7)
MIK 0.71		95.0	0.62 (0.05)	100	39.9 (7.2)
MIK 0.54		92.5	0.59 (0.03)	90	37.6 (12.7)
MIK 0.45		92.5	0.58 (0.12)	90	38.9 (7.9)
MIK 0.12		97.5	0.63 (0.09)	90	33.3 (5.2)
Control	Sep. 1991	100.0	0.51 (0.02)	100	26.8 (8.6)
MIK 1.43		97.5	0.60 (0.06)	80	20.0 (2.9)
MIK 1.00		---	--- (---)	---	--- (---)
MIK 0.71		92.5	0.65 (0.02)	80	17.5 (2.4)
MIK 0.54		95.0	0.56 (0.03)	100	19.3 (9.6)
MIK 0.45		97.5	0.60 (0.03)	100	24.8 (2.6)
MIK 0.12		87.5	0.55 (0.01)	100	14.5 (7.4)
Control	Nov. 1991	87.5	0.39 (0.13)	100	27.9 (4.7)
MIK 1.43		---	--- (---)	90	11.0 (5.7)*
MIK 1.00		---	--- (---)	90	11.1 (3.9)*
MIK 0.71		80.0	0.54 (0.15)	100	17.5 (3.4)
MIK 0.54		70.0	0.51 (0.03)	100	17.7 (3.7)
MIK 0.45		80.0	0.51 (0.07)	90	15.8 (2.0)
MIK 0.12		62.5	0.51 (0.06)	100	18.3 (4.2)
Control	Jan. 1992	80.0	0.59 (0.05)	90	31.1 (3.1)
MIK 1.43		75.0	0.55 (0.05)	100	30.3 (4.9)
MIK 1.00		57.5	0.61 (0.03)	100	31.5 (5.2)
MIK 0.71		92.5	0.58 (0.07)	100	25.7 (4.4)*
MIK 0.54		72.5	0.58 (0.08)	100	24.3 (8.9)*
MIK 0.45		80.0	0.58 (0.09)	100	28.3 (3.1)
MIK 0.12		72.5	0.58 (0.11)	100	27.0 (4.6)
Control	Mar. 1992	55.0	0.62 (0.04)	80	27.4 (5.9)
MIK 1.43		37.5	0.59 (0.03)	100	31.7 (2.7)
MIK 1.00		62.5	0.65 (0.13)	100	31.1 (5.2)
MIK 0.71		67.5	0.58 (0.05)	100	26.2 (7.4)
MIK 0.54		65.0	0.50 (0.04)	100	27.3 (3.8)
MIK 0.45		77.5	0.59 (0.08)	100	31.4 (4.7)
MIK 0.12		72.5	0.60 (0.17)	90	31.0 (4.0)
Control	May 1992	100.0	0.57 (0.03)	80	32.0 (2.7)

Table B.2 (continued)

		Fathead Minnow		*Ceriodaphnia dubia*	
Sample[a]	Date	Mean survival (%)	Mean (SD) growth (mg/fish)	Survival (%)	Mean (SD) offspring/surviving female
MIK 1.43		55.0	0.79 (0.06)	100	29.0 (2.3)
MIK 1.00		60.0	0.79 (0.04)	90	32.7 (6.9)
MIK 0.71		87.5	0.68 (0.03)	80	15.9 (5.7)*
MIK 0.54		75.0	0.73 (0.12)	90	15.2 (4.5)*
MIK 0.45		87.5	0.69 (0.01)	90	18.8 (8.1)*
MIK 0.12		82.5	0.62 (0.08)	100	19.9 (8.4)*
Control	July 1992	95.0	0.40 (0.03)	100	18.7 (5.6)
MIK 1.43		82.5	0.41 (0.06)	90	23.1 (3.6)
MIK 1.00		95.0	0.36 (0.04)	90	23.1 (4.8)
MIK 0.71		90.0	0.39 (0.03)	100	14.4 (4.6)*
MIK 0.54		92.5	0.38 (0.06)	100	18.7 (4.6)
MIK 0.45		92.5	0.45 (0.04)	100	21.7 (3.5)
MIK 0.12		100.0	0.45 (0.04)	100	17.2 (4.1)*
Control	Sep. 1992	100.0	0.48 (0.02)	100	28.0 (4.9)
MIK 1.43		97.5	0.57 (0.03)	100	30.6 (2.9)
MIK 1.00		95.0	0.60 (0.03)	100	27.7 (8.3)
MIK 0.71		97.5	0.64 (0.05)	0*	--- (---)
MIK 0.54		100.0	0.57 (0.02)	50*	11.0 (3.9)*
MIK 0.45		97.5	0.52 (0.06)	100	23.8 (5.0)
MIK 0.12		95.0	0.49 (0.07)	100	28.5 (7.3)
Control	Nov. 1992	92.5	0.64 (0.03)	100	33.8 (3.5)
MIK 1.43		82.5	0.66 (0.09)	100	31.2 (5.4)
MIK 1.00		85.0	0.65 (0.01)	100	30.8 (5.7)
MIK 0.71		72.5	0.65 (0.05)	60*	7.5 (4.3)*
MIK 0.54		90.0	0.56 (0.02)	100	13.1 (6.9)*
MIK 0.45		95.0	0.54 (0.02)	100	12.3 (8.0)*
MIK 0.12		87.5	0.59 (0.07)	100	26.8 (8.1)
Control	Feb. 1993	97.5	0.61 (0.11)	100	22.2 (5.1)
MIK 1.43		90.0	0.60 (0.04)	100	21.4 (4.8)
MIK 1.00		97.5	0.60 (0.04)	90	21.7 (3.3)
MIK 0.71		87.5	0.62 (0.06)	100	21.2 (6.9)
MIK 0.54		97.5	0.58 (0.03)	100	18.8 (3.4)
MIK 0.45		97.5	0.62 (0.03)	100	20.9 (3.9)
MIK 0.12		97.5	0.60 (0.03)	100	21.2 (4.7)

Table B.2 (continued)

		Fathead Minnow		*Ceriodaphnia dubia*	
Sample[a]	Date	Mean survival (%)	Mean (SD) growth (mg/fish)	Survival (%)	Mean (SD) offspring/surviving female
Control	Apr. 1993	100.0	0.42 (0.06)	90	39.6 (11.6)
MIK 1.43		54.5	0.63 (0.07)	80	31.1 (4.6)
MIK 0.78		75.2	0.52 (0.09)	100	34.4 (13.7)
MIK 0.71		93.8	0.56 (0.07)	20*	34.0 (2.8)
MIK 0.54		90.6	0.51 (0.05)	60	33.5 (7.0)
MIK 0.45		90.6	0.53 (0.05)	60	33.3 (7.3)
MIK 0.12		92.5	0.50 (0.06)	90	27.6 (12.6)
Control	Apr. 1993	100.0	0.42 (0.06)	90	24.6 (10.2)
PCK 18.2		85.0	0.38 (0.03)	100	26.3 (7.5)
PCK 8.2		80.0	0.43 (0.04)	100	27.7 (6.7)
PCK 6.9		90.0	0.39 (0.02)	90	27.9 (10.7)
PCK 4.7	Apr. 1993	97.5	0.41 (0.04)	80	24.5 (9.4)
PCK 1.6		100.0	0.40 (0.03)	100	26.8 (4.7)
CRK 15.0		100.0	0.38 (0.03)	100	30.1 (3.3)
Control	July 1993	92.5	0.25 (0.08)	100	23.2 (5.4)
MIK 1.43		80.0	0.33 (0.02)	90	22.1 (5.0)
MIK 1.00		75.0	0.32 (0.13)	90	27.7 (3.2)
MIK 0.71		77.5	0.29 (0.03)	80	20.0 (6.1)
MIK 0.54		80.0	0.27 (0.09)	90	26.4 (6.0)
MIK 0.45		85.0	0.31 (0.11)	100	29.8 (3.3)
MIK 0.12		67.5	0.23 (0.06)	100	23.2 (5.4)
Control	July 1993	90.0	0.33 (0.06)	80	25.3 (7.9)
PCK 18.2		80.0	0.32 (0.09)	100	30.8 (6.4)
PCK 8.2		80.0	0.38 (0.04)	100	31.0 (11.8)
PCK 6.9		65.0	0.35 (0.08)	80	33.2 (5.3)
PCK 4.7		72.5	0.37 (0.05)	90	33.8 (3.9)
PCK 1.6		67.5	0.42 (0.10)	100	31.7 (6.1)
CRK 15.0		90.0	0.30 (0.06)	100	32.3 (6.5)
Control	Oct. 1993	87.5	0.39 (0.07)	100	28.5 (3.9)
MIK 1.43		35.0*	0.47 (0.09)	100	31.7 (6.4)
MIK 0.78		80.0	0.57 (0.10)	90	31.1 (5.4)
MIK 0.71		82.5	0.43 (0.03)	100	23.3 (4.1)*
MIK 0.54		72.5	0.45 (0.06)	100	30.3 (5.2)
MIK 0.45		65.0	0.50 (0.07)	90	30.9 (6.2)

Table B.2 (continued)

		Fathead Minnow		*Ceriodaphnia dubia*	
Sample[a]	Date	Mean survival (%)	Mean (SD) growth (mg/fish)	Survival (%)	Mean (SD) offspring/surviving female
MIK 0.12		57.5	0.53 (0.08)	100	30.9 (5.0)
Control	Oct. 1993	87.5	0.39 (0.07)	100	29.4 (1.7)
PCK 18.2		70.0	0.54 (0.11)	100	32.8 (4.8)
PCK 8.2		65.0	0.42 (0.09)	100	35.3 (6.2)
PCK 6.9		87.5	0.46 (0.05)	100	31.6 (4.8)
PCK 4.7		62.5	0.51 (0.10)	100	33.0 (3.3)
PCK 1.6		60.0	0.39 (0.08)	100	32.7 (3.0)
CRK 15.0		82.5	0.51 (0.09)	100	31.2 (4.2)
Control	Jan. 1994	100.0	0.42 (0.03)	100	26.6 (2.9)
MIK 1.43		72.5	0.47 (0.02)	100	21.7 (4.7)
MIK 0.78		92.5	0.45 (0.08)	100	21.2 (2.40
MIK 0.71		97.5	0.44 (0.05)	90	20.9 (2.4)
MIK 0.54		85.0	0.49 (0.04)	100	21.0 (3.7)
MIK 0.45		95.0	0.46 (0.05)	100	18.1 (6.1)
MIK 0.12		95.0	0.44 (0.04)	90	20.0 (2.6)
Control	Jan. 1994	100.0	0.41 (0.05)	100	18.4 (3.4)
PCK 18.2		82.5	0.45 (0.04)	90	12.0 (4.7)
PCK 8.2		77.5	0.48 (0.12)	100	15.4 (2.1)
PCK 6.9		75.0	0.55 (0.09)	90	13.3 (3.8)
PCK 4.7		82.5	0.54 (0.06)	90	17.3 (4.0)
PCK 1.6		75.0	0.56 (0.06)	100	18.6 (3.6)
CRK 15.0		95.0	0.48 (0.02)	100	18.2 (1.8)

[a]MIK = Mitchell Branch kilometer, PCK = Poplar Creek kilometer, CRK = Clinch River kilometer.

APPENDIX C

RESULTS OF QUALITY ASSURANCE/QUALITY CONTROL ANALYSES OF FISH SAMPLES

Table C.1. Mercury and other metals in fish from Mitchell Branch and nearby reaches of Poplar Creek and the Clinch River, 1991–1993

Measurements in micrograms per grams, wet weight unless otherwise indicated

Site[a]	Date	Spp.[b]	Sex	Tag	Wgt. (g)	Lgt. (cm)	Hg	As	Cd	Cr	Cu	Ni	Pb	Se	Tl	Zn
PCK 8.2	01/14/91	BLUGIL	M	5605	56.0	14.0	0.47	.	.	.	.	.	.	.	.	.
PCK 8.2	01/14/91	BLUGIL	M	5606	53.2	14.5	0.53	.	.	.	.	.	.	.	.	.
PCK 8.2	01/14/91	BLUGIL	M	5607	46.5	14.1	0.28	.	.	.	.	.	.	.	.	.
PCK 8.2	01/14/91	BLUGIL	F	5609	64.5	15.5	0.71	.	.	.	.	.	.	.	.	.
PCK 8.2	02/11/91	BLUGIL	M	5644	69.5	17.4	0.62	.	.	.	.	.	.	.	.	.
PCK 8.2	02/11/91	BLUGIL	M	5645	87.4	17.1	0.07	.	.	.	.	.	.	.	.	.
PCK 8.2	02/11/91	BLUGIL	M	5646	64.2	15.5	0.30	.	.	.	.	.	.	.	.	.
PCK 8.2	02/11/91	BLUGIL	F	5647	49.4	14.8	0.24	.	.	.	.	.	.	.	.	.
PCK 6.9	01/14/91	BLUGIL	F	5600	95.8	17.0	0.67	.	.	.	.	.	.	.	.	.
PCK 6.9	01/14/91	BLUGIL	F	5601	52.1	14.5	0.57	.	.	.	.	.	.	.	.	.
PCK 6.9	01/14/91	BLUGIL	M	5602	52.0	14.5	0.15	.	.	.	.	.	.	.	.	.
PCK 6.9	01/14/91	BLUGIL	M	5603	78.0	16.1	0.32	.	.	.	.	.	.	.	.	.
PCK 6.9	01/14/91	BLUGIL	F	5604	46.9	14.0	0.42	.	.	.	.	.	.	.	.	.
PCK 6.9	02/11/91	BLUGIL	M	5640	63.8	15.6	0.24	.	.	.	.	.	.	.	.	.
PCK 6.9	02/11/91	BLUGIL	M	5641	54.7	14.5	0.21	.	.	.	.	.	.	.	.	.
PCK 6.9	02/11/91	BLUGIL	M	5643	51.9	15.0	0.40	.	.	.	.	.	.	.	.	.
MIK 0.2	01/18/91	REDBRE	F	5610	41.5	14.0	0.56	.	.	.	.	.	.	.	.	.
MIK 0.2	01/18/91	REDBRE	M	5612	30.4	12.2	.	.	.	.	.	.	.	.	.	5.9
MIK 0.2	01/18/91	REDBRE	F	5614	26.9	11.7	0.27	.	.	.	.	.	.	.	.	.
MIK 0.2	01/18/91	REDBRE	M	5617	22.4	10.5	0.20	.	.	.	.	.	.	.	.	.
MIK 0.2	01/18/91	REDBRE	F	5618	18.3	10.2	0.18	.	.	.	.	.	.	.	.	.
MIK 0.2	01/18/91	REDBRE	M	5386	93.1	17.6	0.29	.	.	.	.	.	.	.	0.02	5.1
MIK 0.2	01/18/91	REDBRE	F	5387	71.0	16.0	0.45	.	.	.	.	.	.	0.73	.	6.0
MIK 0.2	01/18/91	REDBRE	F	5388	63.6	15.9	0.35	.	.	.	.	.	.	.	.	6.0
MIK 0.2	01/18/91	REDBRE	F	5389	22.9	11.1	0.23	.	.	.	.	.	.	.	.	.

Table C.1 (continued)

Site[a]	Date	Spp.[b]	Sex	Tag	Wgt. (g)	Lgt. (cm)	Hg	As	Cd	Cr	Cu	Ni	Pb	Se	Tl	Zn
CRK 15.0	02/12/91	BLUGIL	M	5620	70.9	16.4	0.22	.	.	.	.	.	.	.	.	.
CRK 15.0	02/12/91	BLUGIL	M	5621	84.5	17.1	0.42	.	.	.	.	.	.	.	.	.
CRK 15.0	02/12/91	BLUGIL	M	5622	99.1	17.2	0.15	.	.	.	.	.	.	.	.	.
CRK 15.0	02/12/91	BLUGIL	M	5623	68.8	16.5	0.22	.	.	.	.	.	.	.	.	.
CRK 15.0	02/12/91	BLUGIL	M	5624	96.0	17.8	0.11	.	.	.	.	.	.	.	.	.
CRK 15.0	02/12/91	BLUGIL	F	5625	54.8	15.1	0.19	.	.	.	.	.	.	.	.	.
CRK 15.0	02/12/91	BLUGIL	M	5626	67.0	16.5	0.15	.	.	.	.	.	.	.	.	.
CRK 15.0	02/12/91	BLUGIL	M	5627	54.9	15.1	0.15	.	.	.	.	.	.	.	.	.
HINDSCR	11/14/90	REDBRE	M	5768	66.3	15.8	0.15	.	.	.	.	.	.	.	.	.
HINDSCR	11/14/90	REDBRE	M	5769	90.5	17.6	0.06	.	.	.	.	.	.	.	.	.
HINDSCR	11/14/90	REDBRE	F	5794	47.8	14.4	0.10	.	.	.	.	.	.	.	.	.
HINDSCR	11/14/90	REDBRE	M	5795	46.8	14.0	0.04	.	.	.	.	.	.	.	.	5.0
HINDSCR	11/14/90	REDBRE	M	5796	53.3	14.4	0.08	.	.	.	.	.	.	.	.	.
HINDSCR	11/14/90	REDBRE	M	5797	52.7	15.0	0.06	.	.	.	.	.	.	.	.	.
HINDSCR	11/14/90	REDBRE	F	5798	49.0	14.5	0.16	.	.	.	.	.	.	.	.	.
HINDSCR	11/14/90	BLUGIL	M	5760	87.9	17.1	0.08	.	.	.	.	.	.	.	.	.
HINDSCR	11/14/90	BLUGIL	M	5761	126.4	19.1	0.08	.	.	.	.	.	.	.	.	.
HINDSCR	11/14/90	BLUGIL	M	5762	144.2	19.4	0.09	.	.	.	.	.	.	.	.	.
HINDSCR	11/14/90	BLUGIL	F	5763	69.1	15.7	0.10	.	.	.	.	.	.	.	.	.
HINDSCR	11/14/90	BLUGIL	F	5764	56.6	14.9	0.13	.	.	.	.	.	.	.	.	.
HINDSCR	11/14/90	BLUGIL	F	5765	58.9	15.5	0.08	.	.	.	.	.	.	.	.	.
HINDSCR	11/14/90	REDBRE	M	5766	73.1	16.8	0.08	.	.	.	.	.	.	.	.	.
MIK 0.2	01/27/92	HYBRID	M	3270	110.9	18.8	0.19	.	.	.	.	.	.	.	.	.
MIK 0.2	01/27/92	REDBRE	M	3271	46.6	14.5	0.28	.	.	.	.	.	.	.	.	5.8
MIK 0.2	01/27/92	REDBRE	M	3272	61.5	15.2	0.21	.	.	.	.	.	.	.	.	5.2
MIK 0.2	01/27/92	REDBRE	M	3273	63.3	17.1	0.48	.	.	.	.	.	.	.	.	5.7
MIK 0.2	01/27/92	REDBRE	M	3274	19.6	10.5	0.21	.	.	.	.	.	.	.	.	.
MIK 0.2	01/27/92	REDBRE	M	3275	16.6	10.2	0.22	.	.	.	.	.	.	.	.	6.1
MIK 0.2	01/27/92	REDBRE	M	3276	21.1	11.3	0.21	.	.	.	.	.	.	.	.	.
MIK 0.2	01/27/92	REDBRE	M	4078	21.8	11.0	0.20	.	.	.	.	.	.	.	.	.

Table C.1 (continued)

Site[a]	Date	Spp.[b]	Sex	Tag	Wgt. (g)	Lgt. (cm)	Hg	As	Cd	Cr	Cu	Ni	Pb	Se	Tl	Zn
HINDSCR	01/21/92	REDBRE	F	3298	39.6	14.0	0.23	.	.	.	.	.	.	.	.	.
PCK 8.2	07/16/92	BLUGIL	M	3990	97.3	16.3	0.41	.	.	.	.	.	.	.	.	.
PCK 8.2	07/16/92	BLUGIL	M	3991	70.1	15.9	0.42	.	.	.	.	.	.	.	.	.
PCK 8.2	07/16/92	BLUGIL	F	3992	87.8	16.4	0.45	.	.	.	.	.	.	.	.	.
PCK 8.2	07/16/92	BLUGIL	M	3993	91.0	16.8	0.44	.	.	.	.	.	.	.	.	.
PCK 8.2	07/16/92	BLUGIL	M	3994	58.2	14.4	0.24	.	.	.	.	.	.	.	.	.
PCK 8.2	07/16/92	BLUGIL	F	3995	101.0	17.2	0.49	.	.	.	.	.	.	.	.	.
PCK 8.2	07/16/92	BLUGIL	M	3996	58.0	14.1	0.31	.	.	.	.	.	.	.	.	.
PCK 8.2	07/16/92	BLUGIL	M	3997	50.0	13.8	0.24	.	.	.	.	.	.	.	.	.
PCK 6.9	07/16/92	BLUGIL	F	3980	89.0	16.5	0.64	.	.	.	.	.	.	.	.	.
PCK 6.9	07/16/92	BLUGIL	M	3981	86.6	16.2	0.27	.	.	.	.	.	.	.	.	.
PCK 6.9	07/16/92	BLUGIL	M	3982	65.9	14.2	0.36	.	.	.	.	.	.	.	.	.
PCK 6.9	07/16/92	BLUGIL	M	3983	80.9	15.5	0.42	.	.	.	.	.	.	.	.	.
PCK 6.9	07/16/92	BLUGIL	M	3984	90.7	16.9	0.43	.	.	.	.	.	.	.	.	.
PCK 6.9	07/16/92	BLUGIL	M	3985	115.0	17.8	0.05	.	.	.	.	.	.	.	.	.
PCK 6.9	07/16/92	BLUGIL	M	3987	75.7	15.4	0.33	.	.	.	.	.	.	.	.	.
PCK 6.9	07/16/92	BLUGIL	F	3989	58.4	14.5	0.69	.	.	.	.	.	.	.	.	.
CRK 15.0	05/14/92	BLUGIL	M	3552	84.3	16.5	0.09	.	.	.	.	.	.	.	.	.
CRK 15.0	05/14/92	BLUGIL	M	3553	69.7	15.9	0.13	.	.	.	.	.	.	.	.	.
CRK 15.0	05/14/92	BLUGIL	M	3554	89.5	16.7	0.12	.	.	.	.	.	.	.	.	.
CRK 15.0	05/14/92	BLUGIL	F	3555	69.5	15.4	0.18	.	.	.	.	.	.	.	.	.
CRK 15.0	05/14/92	BLUGIL	M	3556	74.0	15.8	0.10	.	.	.	.	.	.	.	.	.
CRK 15.0	05/14/92	BLUGIL	M	3557	76.1	15.0	0.10	.	.	.	.	.	.	.	.	.
CRK 15.0	05/14/92	BLUGIL	M	3558	68.7	15.5	0.13	.	.	.	.	.	.	.	.	.
CRK 15.0	05/14/92	BLUGIL	F	3559	54.3	14.9	0.71	.	.	.	.	.	.	.	.	.
MIK 0.2	03/11/93	REDBRE	M	4330	72.6	16.0	0.42	.	.	.	.	.	.	.	.	.
MIK 0.2	03/11/93	REDBRE	M	4331	64.1	15.4	0.28	.	.	.	.	.	.	.	.	.
MIK 0.2	03/11/93	REDBRE	M	4332	64.1	15.4	0.29	.	.	.	.	.	.	.	.	.
MIK 0.2	03/11/93	REDBRE	M	4333	65.2	15.5	0.28	.	.	.	.	.	.	.	.	.
MIK 0.2	03/11/93	REDBRE	F	4334	55.5	14.5	0.46	.	.	.	.	.	.	.	.	.

Table C.1 (continued)

Site[a]	Date	Spp.[b]	Sex	Tag	Wgt. (g)	Lgt. (cm)	Hg	As	Cd	Cr	Cu	Ni	Pb	Se	Tl	Zn
MIK 0.2	03/11/93	REDBRE	M	4335	65.3	14.8	0.16	.	.	.	.	.	.	.	.	.
MIK 0.2	03/11/93	REDBRE	M	4336	63.0	15.1	.	.	.	0.11	0.21	0.07	.	0.36	.	5.3
MIK 0.2	03/11/93	REDBRE	M	4337	49.4	14.5	0.37	.	.	0.11	0.26	0.07	.	0.55	.	4.6
MIK 0.2	03/11/93	REDBRE	F	4338	61.5	14.3	0.37	.	.	.	0.26	.	.	0.35	.	5.3
MIK 0.2	03/11/93	REDBRE	M	4379	47.1	13.3	.	0.05	.	0.05	0.23	0.06	.	0.39	.	5.7
K1007B[c]	03/11/93	BLUGIL	M	4380	68.3	15.8	0.04	.	.	0.06	0.16	.	.	0.31	.	6.0
K1007B	03/11/93	BLUGIL	M	4381	50.5	14.9	0.07	.	.	.	0.17	.	.	0.28	.	6.3
K1007B	03/11/93	BLUGIL	M	4382	69.1	16.0	0.04	.	.	.	0.16	.	.	0.25	.	5.0
K1007B	03/11/93	BLUGIL	M	4383	55.2	15.1	0.04	.	.	.	.	.	.	.	.	.
K1007B	03/11/93	BLUGIL	M	4384	109.8	18.0	0.09	.	.	.	0.17	.	.	0.37	.	5.0
K1007B	03/11/93	BLUGIL	M	4385	75.7	166.4	0.05	.	.	.	.	.	.	.	.	.
K1007B	03/11/93	BLUGIL	M	4386	54.3	15.1	0.05	.	.	.	.	.	.	.	.	.
K1007B	03/11/93	BLUGIL	M	4387	62.2	15.6	0.03	.	.	.	.	.	.	.	.	.
701A[c]	03/25/93	BLUGIL	M	4320	81.7	16.1	0.12	0.06	0.03	0.06	0.28	.	.	0.49	.	4.8
701A	03/25/93	BLUGIL	M	4321	73.6	15.7	0.15	0.06	.	0.11	0.19	.	.	0.56	.	5.1
701A	03/25/93	BLUGIL	M	4322	119.8	17.9	0.14	0.05	.	.	0.13	.	.	0.65	.	5.7
701A	03/25/93	BLUGIL	M	4323	56.5	15.0	0.11	0.08	.	.	0.14	.	.	0.46	.	3.8
701A	03/25/93	BLUGIL	M	4324	67.5	15.3	0.09	.	.	.	.	.	.	.	.	.
701A	03/25/93	BLUGIL	M	4328	79.2	16.4	0.10	.	.	.	.	.	.	.	.	.
701A	03/25/93	BLUGIL	M	4329	85.2	16.5	0.14	.	.	.	.	.	.	.	.	.
701A	03/25/93	BLUGIL	M	4315	71.9	15.6	0.10	.	.	.	.	.	.	.	.	.
K1007B	04/02/93	BLUGIL	M	4420	83.3	16.1	0.06	.	.	.	0.20	.	.	0.38	.	4.5
K1007B	04/02/93	BLUGIL	M	4421	77.8	17.0	0.09	0.05	.	.	0.15	.	.	0.34	.	7.1
K1007B	04/02/93	BLUGIL	M	4422	98.1	17.7	0.09	0.07	.	.	0.16	.	.	0.34	.	7.4
K1007B	04/02/93	BLUGIL	M	4423	74.1	16.3	0.10	.	.	.	0.17	.	.	0.33	.	7.9
K1007B	04/02/93	BLUGIL	M	4424	61.9	14.9	0.08	.	.	.	.	.	.	.	.	.
K1007B	04/02/93	BLUGIL	M	4425	69.9	15.9	0.09	.	.	.	.	.	.	.	.	.
K1007B	04/02/93	BLUGIL	F	4426	58.0	15.1	0.08	.	.	.	.	.	.	.	.	.
K1007B	04/02/93	BLUGIL	M	4427	72.5	15.8	0.10	.	.	.	.	.	.	.	.	.

Table C.1 (continued)

Site[a]	Date	Spp.[b]	Sex	Tag	Wgt. (g)	Lgt. (cm)	Hg	As	Cd	Cr	Cu	Ni	Pb	Se	Tl	Zn
PCK 6.9	03/09/93	BLUGIL	M	4301	104.3	17.4	0.29	.	.	.	.	.	.	.	.	.
PCK 6.9	03/09/93	BLUGIL	M	4302	103.7	16.8	0.47	.	.	.	.	.	.	.	.	.
PCK 6.9	03/09/93	BLUGIL	F	4303	79.2	16.3	0.30	.	.	.	.	.	.	.	.	.
PCK 6.9	03/09/93	BLUGIL	F	4304	89.1	16.8	0.38	.	.	.	.	.	.	.	.	.
PCK 6.9	03/09/93	BLUGIL	M	4305	58.3	15.0	0.34	.	.	.	.	.	.	.	.	.
PCK 6.9	03/09/93	BLUGIL	M	4306	96.9	16.7	0.32	.	.	.	.	.	.	.	.	.
PCK 6.9	03/09/93	BLUGIL	M	4307	82.8	16.7	0.48	.	.	.	.	.	.	.	.	.
PCK 6.9	03/09/93	BLUGIL	M	4308	53.1	14.4	0.30	.	.	.	.	.	.	.	.	.
PCK 1.6	03/09/93	BLUGIL	M	4358	113.8	18.1	0.08	0.09	.	0.13	0.18	.	.	0.40	.	6.9
PCK 1.6	03/09/93	BLUGIL	M	4359	85.8	16.7	0.07	0.07	.	0.05	0.19	0.08	.	0.49	.	3.9
PCK 1.6	03/09/93	BLUGIL	M	4360	71.4	15.7	0.44	.	.	.	0.19	.	.	0.38	.	4.2
PCK 1.6	03/09/93	BLUGIL	M	4361	70.3	15.5	0.44	0.08	.	.	0.22	.	.	0.52	.	5.0
PCK 1.6	03/09/93	BLUGIL	M	4362	66.1	15.2	0.19	.	.	.	.	.	.	.	.	.
PCK 1.6	03/09/93	BLUGIL	M	4363	107.8	17.0	0.33	.	.	.	.	.	.	.	.	.
PCK 1.6	03/09/93	BLUGIL	M	4364	97.8	17.0	0.19	.	.	.	.	.	.	.	.	.
PCK 1.6	03/09/93	BLUGIL	F	4369	54.5	14.8	0.29	.	.	.	.	.	.	.	.	.
PCK 18.2	03/29/93	BLUGIL	M	4480	121.3	17.6	0.04	.	.	.	0.15	.	.	0.38	.	4.8
PCK 18.2	03/29/93	BLUGIL	M	4481	109.6	16.6	0.05	.	.	.	0.23	.	.	0.44	.	5.4
PCK 18.2	03/29/93	BLUGIL	M	4482	92.1	17.0	0.12	.	.	.	0.11	.	.	0.30	.	6.7
PCK 18.2	03/29/93	BLUGIL	M	4483	103.7	17.9	0.07	.	.	.	0.15	.	.	0.40	.	3.4
PCK 18.2	03/29/93	BLUGIL	M	4484	93.9	16.0	0.05	.	.	.	.	.	.	.	.	.
PCK 18.2	03/29/93	BLUGIL	F	4485	54.8	14.3	0.04	.	.	.	.	.	.	.	.	.
PCK 18.2	03/29/93	BLUGIL	M	4487	48.8	13.4	0.05	.	.	.	.	.	.	.	.	.
PCK 18.2	03/29/93	BLUGIL	F	4489	49.2	13.4	0.05	.	.	.	.	.	.	.	.	.
PCK 8.2	03/09/93	BLUGIL	M	4370	89.4	15.9	0.32	.	.	.	.	.	.	.	.	.
PCK 8.2	03/09/93	BLUGIL	M	4371	74.7	16.0	0.31	.	.	.	.	.	.	.	.	.
PCK 8.2	03/09/93	BLUGIL	M	4372	71.9	15.7	0.33	.	.	.	.	.	.	.	.	.
PCK 8.2	03/09/93	BLUGIL	M	4373	77.2	16.5	0.17	.	.	.	.	.	.	.	.	.
PCK 8.2	03/09/93	BLUGIL	M	4374	154.0	20.1	0.39	.	.	.	.	.	.	.	.	.
PCK 8.2	03/09/93	BLUGIL	M	4375	74.5	16.2	0.51	.	.	.	.	.	.	.	.	.
PCK 8.2	03/09/93	BLUGIL	F	4376	70.2	16.0	0.55	.	.	.	.	.	.	.	.	.
PCK 8.2	03/09/93	BLUGIL	.	4377	60.1	14.5	0.15	.	.	.	.	.	.	.	.	.

Table C.1 (continued)

Site[a]	Date	Spp.[b]	Sex	Tag	Wgt. (g)	Lgt. (cm)	Hg	As	Cd	Cr	Cu	Ni	Pb	Se	Tl	Zn
CRK 15.0	03/09/93	BLUGIL	M	4350	82.8	16.0	0.23	.	.	.	.	.	.	.	.	.
CRK 15.0	03/09/93	BLUGIL	M	4351	79.7	16.5	0.36	.	.	.	.	.	.	.	.	.
CRK 15.0	03/09/93	BLUGIL	M	4352	102.4	17.6	0.16	.	.	.	.	.	.	.	.	.
CRK 15.0	03/09/93	BLUGIL	M	4353	77.9	15.9	0.09	.	.	.	.	.	.	.	.	.
CRK 15.0	03/09/93	BLUGIL	M	4354	62.3	15.5	0.17	.	.	.	.	.	.	.	.	.
CRK 15.0	03/09/93	BLUGIL	M	4355	103.2	17.5	0.12	.	.	.	.	.	.	.	.	.
CRK 15.0	03/09/93	BLUGIL	M	4356	55.2	14.9	0.12	.	.	.	.	.	.	.	.	.
CRK 15.0	03/09/93	BLUGIL	M	4357	62.0	15.8	0.20	.	.	.	.	.	.	.	.	.
HINDSCR	05/05/93	REDBRE	M	4468	53.9	12.5	0.04	.	.	.	.	.	.	.	.	.
HINDSCR	05/05/93	BLUGIL	M	4472	53.1	14.0	0.09	.	.	.	.	.	.	.	.	.
HINDSCR	05/05/93	BLUGIL	M	4473	68.0	15.0	0.04	.	.	.	.	.	.	.	.	.
HINDSCR	05/05/93	REDBRE	M	4499	39.2	12.2	0.05	.	.	.	.	.	.	.	.	.
HINDSCR	05/05/93	BLUGIL	M	4540	77.9	15.7	0.10	.	.	.	.	.	.	.	.	.
HINDSCR	05/05/93	BLUGIL	M	4541	60.4	14.7	0.04	.	.	.	.	.	.	.	.	.
HINDSCR	05/05/93	REDBRE	M	4588	48.5	13.7	0.04	.	.	.	.	.	.	.	.	.
HINDSCR	05/05/93	REDBRE	F	4589	46.0	13.0	0.05	.	.	.	.	.	.	.	.	.

[a]MIK = Mitchell Branch kilometer, PCK = Poplar Creek kilometer, CRK = Clinch River kilometer, HINDSCR = Hinds Creek.
[b]Species: BLUGIL = Bluegill (*Lepomis macrochirus*); REDBRE = redbreast sunfish (*L. auritus*).
[c]Ponded tributaries of Poplar Creek.

Table C.2. Concentrations of polychlorinated biphenyls (PCBs) in resident fish from Mitchell Branch and nearby reaches of Poplar Creek and the Clinch River, 1991–1993

Measurements given in micrograms per gram, wet weight unless otherwise indicated

Site[a]	Date	Spp.[b]	Sex	Tag	Wgt. (g)	Lgt. (cm)	∑ PCB	1254[c]	1260[d]
PCK 8.2	01/14/91	BLUGIL	M	5605	56.0	14.0	0.10	0.02	0.08
PCK 8.2	01/14/91	BLUGIL	M	5606	53.2	14.5	0.10	0.04	0.06
PCK 8.2	01/14/91	BLUGIL	M	5607	46.5	14.1	0.10	0.03	0.07
PCK 8.2	01/14/91	BLUGIL	F	5609	64.5	15.5	0.02	<0.01	0.02
PCK 8.2	02/11/91	BLUGIL	M	5644	69.5	17.4	0.01	<0.01	0.01
PCK 8.2	02/11/91	BLUGIL	M	5645	87.4	17.1	<0. 01	<0.01	<0.01
PCK 8.2	02/11/91	BLUGIL	M	5646	64.2	15.5	0.02	<0.01	0.02
PCK 8.2	02/11/91	BLUGIL	F	5647	49.4	14.8	0.02	0.01	0.02
PCK 6.9	01/14/91	BLUGIL	F	5600	95.8	17.0	<0.01	<0.01	<0.01
PCK 6.9	01/14/91	BLUGIL	F	5601	52.1	14.5	0.07	0.02	0.05
PCK 6.9	01/14/91	BLUGIL	M	5602	52.0	14.5	0.01	<0.01	0.01
PCK 6.9	01/14/91	BLUGIL	M	5603	78.0	16.1	0.05	0.02	0.03
PCK 6.9	01/14/91	BLUGIL	F	5604	46.9	14.0	0.07	<0.01	0.07
PCK 6.9	02/11/91	BLUGIL	M	5640	63.8	15.6	<0.01	<0.01	<0.01
PCK 6.9	02/11/91	BLUGIL	M	5641	54.7	14.5	0.04	<0.01	0.04
PCK 6.9	02/11/91	BLUGIL	M	5643	51.9	15.0	0.08	0.02	0.06
MIK 0.2	01/18/91	REDBRE	F	5610	41.5	14.0	0.34	0.26	0.08
MIK 0.2	01/18/91	REDBRE	M	5612	30.4	12.2	0.52	0.42	0.10
MIK 0.2	01/18/91	REDBRE	F	5614	26.9	11.7	1.07	0.80	0.27
MIK 0.2	01/18/91	REDBRE	M	5617	22.4	10.5	0.79	0.62	0.17
MIK 0.2	01/18/91	REDBRE	M	5386	93.1	17.6	0.40	0.35	0.05
MIK 0.2	01/18/91	REDBRE	F	5387	71.0	16.0	0.03	0.03	0.01
MIK 0.2	01/18/91	REDBRE	F	5388	63.6	15.9	0.22	0.18	0.04
MIK 0.2	01/18/91	REDBRE	F	5389	22.9	11.1	1.28	1.04	0.24

Table C.2 (continued)

Site[a]	Date	Spp.[b]	Sex	Tag	Wgt. (g)	Lgt. (cm)	∑ PCB	1254[c]	1260[d]
HINDSCR	11/14/90	REDBRE	M	5767	74.2	17.0	0.07	0.07	<0.01
HINDSCR	11/14/90	REDBRE	M	5769	90.5	17.6	<0.01	<0.01	<0.01
HINDSCR	11/14/90	REDBRE	F	5794	47.8	14.4	0.01	0.01	<0.01
HINDSCR	11/14/90	REDBRE	M	5796	53.3	14.4	0.02	0.02	<0.01
HINDSCR	11/14/90	REDBRE	M	5797	52.7	15.0	0.01	0.01	<0.01
HINDSCR	11/14/90	REDBRE	F	5798	49.0	14.5	<0.01	<0.01	<0.01
HINDSCR	11/14/90	BLUGIL	M	5760	87.9	17.1	0.01	0.01	<0.01
HINDSCR	11/14/90	BLUGIL	M	5761	126.4	19.1	<0.01	<0.01	<0.01
HINDSCR	11/14/90	BLUGIL	M	5762	144.2	19.4	<0.01	<0.01	<0.01
HINDSCR	11/14/90	BLUGIL	F	5763	69.1	15.7	0.01	0.01	<0.01
HINDSCR	11/14/90	BLUGIL	F	5764	56.6	14.9	<0.01	<0.01	<0.01
HINDSCR	11/14/90	BLUGIL	F	5765	58.9	15.5	0.05	0.05	<0.01
MIK 0.2	01/27/92	HYBRID	M	3270	110.9	18.8	1.74	1.21	0.53
MIK 0.2	01/27/92	REDBRE	M	3271	46.6	14.5	1.02	0.21	0.81
MIK 0.2	01/27/92	REDBRE	M	3272	61.5	15.2	0.40	0.23	0.17
MIK 0.2	01/27/92	REDBRE	M	3273	63.3	17.1	3.05	<0.01	3.05
MIK 0.2	01/27/92	BLUGIL	M	3277	51.2	14.3	1.91	1.56	0.35
MIK 0.2	01/27/92	BLUGIL	M	3278	27.4	11.6	0.74	0.42	0.32
MIK 0.2	01/27/92	BLUGIL	M	3279	33.2	12.7	1.43	0.68	0.75
MIK 0.2	01/27/92	BLUGIL	F	4079	29.6	12.5	0.42	0.12	0.30
HINDS CR	01/21/92	REDBRE	M	3290	100.9	18.5	<0.01	<0.01	<0.01
PCK 8.2	07/16/92	BLUGIL	M	3990	97.3	16.3	<0.01	<0.01	<0.01
PCK 8.2	07/16/92	BLUGIL	M	3991	70.1	15.9	0.02	<0.01	0.02
PCK 8.2	07/16/92	BLUGIL	F	3992	87.8	16.4	0.07	0.02	0.05
PCK 8.2	07/16/92	BLUGIL	M	3993	91.0	16.8	0.03	<0.01	0.03
PCK 8.2	07/16/92	BLUGIL	M	3994	58.2	14.4	0.05	0.01	0.04
PCK 8.2	07/16/92	BLUGIL	F	3995	101.0	17.2	0.03	0.01	0.02
PCK 8.2	07/16/92	BLUGIL	M	3996	58.0	14.1	0.02	<0.01	0.02
PCK 8.2	07/16/92	BLUGIL	M	3997	50.0	13.8	0.03	0.02	0.01

Table C.2 (continued)

Site[a]	Date	Spp.[b]	Sex	Tag	Wgt. (g)	Lgt. (cm)	∑ PCB	1254[c]	1260[d]
PCK 6.9	07/16/92	BLUGIL	F	3980	89.0	16.5	0.02	<0.01	0.02
PCK 6.9	07/16/92	BLUGIL	M	3981	86.6	16.2	0.02	0.01	0.01
PCK 6.9	07/16/92	BLUGIL	M	3982	65.9	14.2	0.05	0.02	0.03
PCK 6.9	07/16/92	BLUGIL	M	3983	80.9	15.5	0.08	0.01	0.07
PCK 6.9	07/16/92	BLUGIL	M	3984	90.7	16.9	0.09	0.02	0.07
PCK 6.9	07/16/92	BLUGIL	M	3985	115.0	17.8	0.07	0.04	0.03
PCK 6.9	07/16/92	BLUGIL	M	3987	75.7	15.4	0.06	<0.01	0.06
PCK 6.9	07/16/92	BLUGIL	F	3989	58.4	14.5	0.06	0.06	<0.01
MIK 0.2	03/11/93	REDBRE	M	4330	72.6	16.0	0.91	0.70	0.21
MIK 0.2	03/11/93	REDBRE	M	4331	64.1	15.4	1.25	0.63	0.62
MIK 0.2	03/11/93	REDBRE	M	4332	64.1	15.4	0.49	0.32	0.17
MIK 0.2	03/11/93	REDBRE	M	4333	65.2	15.5	0.77	0.60	0.17
MIK 0.2	03/11/93	REDBRE	F	4334	55.5	14.5	0.10	0.06	0.04
MIK 0.2	03/11/93	REDBRE	M	4335	65.3	14.8	1.36	0.95	0.41
MIK 0.2	03/11/93	REDBRE	M	4337	49.4	14.5	0.66	0.11	0.55
MIK 0.2	03/11/93	REDBRE	F	4338	61.5	14.3	0.57	0.33	0.24
K-1007B[e]	03/11/93	BLUGIL	M	4382	69.1	16.0	0.04	<0.01	0.04
K-1007B	03/11/93	BLUGIL	M	4383	55.2	15.1	0.03	0.03	<0.01
K-1007B	03/11/93	BLUGIL	M	4384	109.8	18.0	0.02	0.02	<0.01
K-1007B	03/11/93	BLUGIL	M	4385	75.7	16.6	0.01	0.01	<0.01
PCK 6.9	03/09/93	BLUGIL	M	4301	104.3	17.4	0.81	0.50	0.31
PCK 6.9	03/09/93	BLUGIL	M	4302	103.7	16.8	0.09	0.01	0.08
PCK 6.9	03/09/93	BLUGIL	F	4303	79.2	16.3	0.07	0.03	0.04
PCK 6.9	03/09/93	BLUGIL	F	4304	89.1	16.8	0.03	<0.01	0.03
PCK 6.9	03/09/93	BLUGIL	M	4305	58.3	15.0	0.04	0.02	0.02
PCK 6.9	03/09/93	BLUGIL	M	4306	96.9	16.7	0.15	<0.01	0.15
PCK 6.9	03/09/93	BLUGIL	M	4307	82.8	16.7	0.10	<0.01	0.10
PCK 6.9	03/09/93	BLUGIL	M	4308	53.1	14.4	0.10	<0.01	0.10

Table C.2 (continued)

Site[a]	Date	Spp.[b]	Sex	Tag	Wgt. (g)	Lgt. (cm)	∑ PCB	1254[c]	1260[d]
PCK 1.6	03/09/93	BLUGIL	M	4358	113.8	18.1	0.02	0.02	<0.01
PCK 1.6	03/09/93	BLUGIL	M	4359	85.8	16.7	0.22	0.09	0.13
PCK 1.6	03/09/93	BLUGIL	M	4360	71.4	15.7	0.08	0.05	0.03
PCK 1.6	03/09/93	BLUGIL	M	4361	70.3	15.5	0.08	0.06	0.02
PCK 1.6	03/09/93	BLUGIL	M	4362	66.1	15.2	0.11	0.03	0.08
PCK 1.6	03/09/93	BLUGIL	M	4363	107.8	17.0	0.11	0.04	0.07
PCK 1.6	03/09/93	BLUGIL	M	4364	97.8	17.0	0.15	0.01	0.14
PCK 1.6	03/09/93	BLUGIL	F	4369	54.5	14.8	0.12	0.05	0.07
PCK 18.2	03/29/93	BLUGIL	M	4480	121.3	17.6	0.10	0.02	0.08
PCK 18.2	03/29/93	BLUGIL	M	4481	109.6	16.6	0.03	0.02	0.01
PCK 18.2	03/29/93	BLUGIL	M	4482	92.1	17.0	0.06	0.06	<0.01
PCK 18.2	03/29/93	BLUGIL	M	4483	103.7	17.9	0.04	0.04	<0.01
PCK 18.2	03/29/93	BLUGIL	M	4484	93.9	16.0	0.03	0.01	0.02
PCK 18.2	03/29/93	BLUGIL	F	4485	54.8	14.3	0.14	0.14	<0.01
PCK 18.2	03/29/93	BLUGIL	M	4487	48.8	13.4	0.13	0.13	<0.01
PCK 18.2	03/29/93	BLUGIL	F	4489	49.2	13.4	0.09	<0.01	0.09
PCK 8.2	03/09/93	BLUGIL	M	4370	89.4	15.9	0.02	<0.01	0.02
PCK 8.2	03/09/93	BLUGIL	M	4371	74.7	16.0	0.04	<0.01	0.04
PCK 8.2	03/09/93	BLUGIL	M	4372	71.9	15.7	0.05	0.02	0.03
PCK 8.2	03/09/93	BLUGIL	M	4373	77.2	16.5	0.03	<0.01	0.03
PCK 8.2	03/09/93	BLUGIL	M	4374	154.0	20.1	0.04	<0.01	0.04
PCK 8.2	03/09/93	BLUGIL	M	4375	74.5	16.2	0.04	<0.01	0.04
PCK 8.2	03/09/93	BLUGIL	F	4376	70.2	16.0	0.02	0.01	0.01
PCK 8.2	03/09/93	BLUGIL	.	4377	60.1	14.5	<0.01	<0.01	<0.01
701A[e]	03/25/93	LMBASS	F	4310	2604.0	53.8	0.74	0.13	0.61
701A	03/25/93	LMBASS	M	4448	1544.0	45.5	0.31	0.16	0.15
701A	03/25/93	LMBASS	F	4419	1413.0	43.7	0.14	0.06	0.08
701A	03/25/93	LMBASS	F	4418	2165.0	50.2	0.24	0.07	0.17

Table C.2 (continued)

Site[a]	Date	Spp.[b]	Sex	Tag	Wgt. (g)	Lgt. (cm)	∑ PCB	1254[c]	1260[d]
K-1007B[e]	04/02/93	LMBASS	M	4400	1846.0	50.5	8.76	4.91	3.76
K-1007B	04/02/93	LMBASS	M	4401	2636.0	53.5	42.92	25.42	17.50
K-1007B	04/02/93	LMBASS	F	4402	1141.0	41.5	8.17	5.38	2.79
K-1007B	04/02/93	LMBASS	F	4403	1997.0	48.0	4.76	2.95	1.81
K-1007B	03/11/93	LMBASS	F	4389	1460.0	45.0	0.13	<0.01	0.13
K-1007B	03/11/93	LMBASS	F	4388	1698.0	47.4	1.21	0.49	0.72
HINDSCR	05/05/93	REDBRE	M	4469	54.5	13.6	0.05	0.05	<0.01
HINDSCR	05/05/93	BLUGIL	M	4473	68.0	15.0	0.09	<0.01	0.09
HINDSCR	05/05/93	BLUGIL	M	4540	77.9	15.7	0.21	0.21	<0.01
HINDSCR	05/05/93	BLUGIL	M	4541	60.4	14.7	0.07	<0.01	0.07
HINDSCR	05/05/93	REDBRE	M	4588	48.5	13.7	0.03	0.02	0.01
HINDSCR	05/05/93	REDBRE	F	4589	46.0	13.0	0.11	0.11	<0.01
HINDSCR	11/30/92	COCARP	M	3769	2737.0	58.7	0.16	0.07	0.09
HINDSCR	11/30/92	COCARP	F	4108	5171.0	71.0	0.05	<0.01	0.05

[a]MIK = Mitchell Branch kilometer, PCK = Poplar Creek kilometer, CRK = Clinch River kilometer, HINDSCR = Hinds Creek.

[b]BLUGIL - bluegill; REDBRE - redbreast sunfish; LMBASS = large-mouth bass.

[c]1254 - PCB-1254 (Arochlor 1254) in fish axial muscle; in micrograms per pram, wet weight.

[d]1260 - PCB-1260 (Arochlor 1260) in fish axial muscle, in micrograms per gram, wet weight.

[e]Ponded tributaries of Poplar Creek.

Table C.3. Detection limits of organic compounds (μg/g, wet wt)

Compound	Detection limit
CAPILLARY COLUMN GC/MS[a]	
Phenol	<0.1
Bis(2-chloroethylether)	<0.1
2-chlorophenol	<0.1
1,3-Dichlorobenzene	<0.1
1,4-Dichlorobenzene	<0.1
Benzyl Alcohol	<0.1
1,2-Dichlorobenzene	<0.1
2-Methylphenol	<0.1
Bis(2-Chlorodisopropyl) ether	<0.1
4-Methylphenol	<0.1
N-Nitroso-di-*N*-propylamine	<0.1
Hexachloroethane	<0.1
Nitrobenzene	<0.1
Isophorone	<0.1
2-Nitrophenol	<0.1
2,4-Dimethylphenol	<0.1
Benzoic acid	<0.1
Bis(2-Chloroethoxy) methane	<0.1
2,4-Dichlorophenol	<0.1
1,2,4-Trichlorobenzene	<0.1
Naphthalene	<0.1
4-Chloroaniline	<0.1
Hexachlorobutadiene	<0.1
4-Chloro-3-methylphenol	<0.1
2-Methylnaphthalene	<0.1
Hexachlorocyclopentadiene	<0.1
2,4,6-Trichlorophenol	<0.1
2,4,5-Trichlorophenol	<0.1
2-Chloronaphthalene	<0.1
2-Nitroaniline	<0.5
Dimethylphthalate	<0.1
Acenaphthalene	<0.1
3-Nitroaniline	<0.5
Acenaphthene	<0.1
2,4-Dinitrophenol	<0.5
4-Nitrophenol	<0.5
Dibenzofuran	<0.1
2,4-Dinitrotoluene	<0.1
2,6-Dinitrotoluene	<0.1
Diethylphthalate	<0.1
4-Chlorophenol-phenylether	<0.1
Fluorene	<0.1
4-Nitroaniline	<0.5
4,6-Dinitro-2-methylphenol	<0.5
N-Nitrosodiphenolamine	<0.1
4-bromophenol-phenylether	<0.1
Hexachlorobenzene	<0.1
Pentachorophenol	<0.5
Phenanthrene	<0.1
Anthracene	<0.1
Di-*N*-butylphthalate	<0.1
Fluoranthene	<0.1
Pyrene	<0.1
Butylbenzylphthalate	<0.1

Table C.3 (continued)

Compound	Detection limit
3,3-Dichlorobenzene	<0.1
Benz[*a*]anthracene	<0.1
Bis(2-ethylhexyl)phthalate	<0.1
Chrysene	<0.1
Di-*N*-octylphthalate	<0.1
Benzo[*b*]fluoranthene	<0.1
Benzo[*k*]fluoranthene	<0.1
Benzo[*a*]pyrene	<0.1
Indeno(1,2,3-*cd*)pyrene	<0.1
Dibenz[*a,h*]anthracene	<0.1
Benzo[*g,h,i*]perylene	<0.1
CAPILLARY COLUMN GC/ECD[b]	
Alpha-bhc	<0.001
Beta-bhc	<0.001
Delta-bhc	<0.001
Gamma-bhc	<0.001
Heptachlor	<0.001
Aldrin	<0.001
Heptachlor epoxide	<0.001
Endosulfan I	<0.001
Dieldrin	<0.002
4,4'-DDE	<0.002
Endrin	<0.002
Endosulfan II	<0.002
4,4'-DDD	<0.002
Endosulfan Sulfate	<0.002
4,4'-DDT	<0.002
Endrin ketone	<0.002
Methoxychlor	<0.012
Alpha chlordane	<0.012
Gamma chlordane	<0.012
Toxaphene	<0.023

[a]GC/MS - gas chromatography/mass spectrometry.
[b]GC/ECD = gas chromatography/electron capture detector.

APPENDIX D

CHECKLIST OF BENTHIC MACROINVERTEBRATE TAXA FROM MITCHELL BRANCH, OCTOBER 1986 THROUGH APRIL 1993

Table D.1. Checklist of benthic macroinvertebrates collected from Mitchell Branch, October 1986–April 1993

Taxon	Site[a,b] MIK 0.45	MIK[c] 0.54	MIK 0.71	MIK 0.78	MIK[c] 0.86	MIK 1.43
Turbellaria						
Tricladida						
Planariidae	4,5,6,7	2,3,5	4,5,6,7	2,3,4,5,6,7	3,4,5,6	2,3,4,5,6,7
Nematoda	2,3,4,5,6,7	2,3,4,5	3,4,5,6	1,2,3,4,5,6	1,2,3,4,5,6	3,4
Annelida						
Hirudinea?	2,3,4,5,6,7	3,5	4	3	3	-
Oligochaeta	1,2,3,4,5,6,7	1,2,3,4,5,6	1,2,3,4,5,6,7	1,2,3,4,5,6,7	1,2,3,4,5,6	1,2,3,4,5,6,7
Crustacea						
Isopoda						
Caecidotia	-	-	-	-	-	1,2,3,4,5,6
Lirceus	-	-	-	-	-	1,2,3,4,5,6,7
Amphipoda						
Gammaridae						
Crangonyx	-	-	4,5,6	4,5,6,7	3,4,5,6	1,2,3,4,5,6,7
Decapoda	-	-	-	-	1	-
Cambaridae	-	-	-	4,6,7	2,4,5,6	2,3,4,6,7
Hydracarina	6	-	5,6	6	3,6	2,3,5,6,7
Insecta						
Ephemeroptera						
Baetidae	-	5	4	6	-	1,2,5
Baetis	5,6	-	5,6	1,2,3,4,5	1,2,3,4,5,6	1,2,3,4,5,6,7
Cloeon	-	5	5	4,6	-	1,2,7
Pseudocloeon	5	-	5	3,4,5	3,4	-
Ephemerelidae						
Ephemerella	7	-	-	-	4	-
Eurylophella	7	5	-	4	3,4	1,2,3,4,6,7

Table D.1 (continued)

Taxon	Site[a,b] MIK 0.45	MIK[c] 0.54	MIK 0.71	MIK 0.78	MIK[c] 0.86	MIK 1.43
Ephemeroptera (continued)						
Ephemeridae						
Ephemera	-	-	-	-	-	1,4,5,6,7
Heptageniidae	-	-	-	3	-	-
Nixe	-	-	-	3	3	-
Stenonema	5	-	-	-	-	-
Leptophlebiidae	-	-	-	-	-	1,3
Habrophlebiodes	7	-	5,7	3,4	3,4,5	1,2,3,4,5,6,7
Habrophlebiodes?	-	-	-	-	-	5
Leptophlebia?	-	-	-	2	-	-
Paraleptophlebia	-	-	-	3,4	3,4	2,3,4,5,7
Paraleptophelbia?	-	-	-	-	-	3
Siphlonuridae						
Ameletus	-	-	-	2,3	3,4	2,3,4,5
Odonata						
Anisoptera						
Aeshnidae						
Boyeria vinosa	-	4	5,6	5	-	-
Cordulegastridae						
Cordulegaster	-	-	-	-	-	1,3,4,5,7
Gomphidae	-	-	-	-	-	1,2,4,5,6
Gomphus	-	6	-	-	-	-
Stylogomphus albistylus	-	-	-	7	-	3,4,5,6,7
Libellulidae						
Perithemis	-	-	-	4	-	-
Zygoptera						
Calopterygidae						
Calopteryx	7	-	7	4,5,6,7	4	4,5,6,7

Table D.1 (continued)

Taxon	Site[a,b] MIK 0.45	MIK[c] 0.54	MIK 0.71	MIK 0.78	MIK[c] 0.86	MIK 1.43
Odonata (continued)						
Coenagrionidae	-	-	2,7	-	-	7
Argia	1,2,3,4,7	2,3,4,5,6	4,5,6,7	2,5,6	4,5	4
Enallagma	-	2	-	-	-	-
Ischnura	3	4	-	5	-	-
Plecoptera	-	-	-	-	1	1,2,3,7
Capniidae	-	-	-	-	-	1
Allocapnia	7	-	4,5,7	3,4,6,7	3,4,6	2,3,4,5,6,7
Capniidae/Leuctridae	-	-	-	-	6	1
Plecoptera (continued)						
Leuctridae	-	-	-	-	2	-
Leuctra	5,6,7	-	5,6,7	2,3,4,5	3,4,5,6	2,3,4,5,6,7
Nemouridae						
Amphinemura	-	-	5,6,7	2,3,4,5,6,7	2,3,4,5,6	1,2,3,4,5,6,7
Peltoperlidae						
Tallaperla	-	-	-	-	-	7
Perlidae	-	-	-	-	-	1,2,3,4,7
Eccoptura xanthenes	-	-	5	-	3,4,5,6	4,5,6,7
Perlesta	-	-	7	5	2,3,5,6	2,3,4,5,7
Perlodidae	-	-	-	-	-	2,4,5,6
Clioperla clio	-	-	-	-	-	3,4,5
Isoperla	-	-	5	2	2,5,6	3,4
Perlidae/Perlodidae	-	-	6	-	4,5,6	2,3,4,5,7
Taeniopterygidae						
Taeniopteryx	5	-	-	-	-	-

Table D.1 (continued)

Taxon	Site[a,b] MIK 0.45	MIK[c] 0.54	MIK 0.71	MIK 0.78	MIK[c] 0.86	MIK 1.43
Megaloptera						
Corydalidae						
Nigronia fasciatus	-	-	-	-	-	1,2,4,5,6
Sialidae						
Sialis	-	-	-	-	-	2,7
Trichoptera	-	-	-	-	-	1,2
Glossosomatidae						
Agapetus	-	-	-	4	-	4,7
Glossosoma	-	-	-	-	-	7
Goeridae						
Goera	-	-	-	-	-	5
Hydropsychidae						
Cheumatopsyche	4,6,7	-	5,6,7	2,3,4,5,6,7	2,3,4,5,6	5
Diplectrona modesta	-	-	-	4	4	1,3,4,5,6,7
Hydroptilidae						
Hydroptila	-	-	5	4,5,6,7	-	-
Ochrotrichia	-	-	-	-	4	-
Ochrotrichia?	-	-	7	-	-	-
Oxyethira	4	4	-	4	-	-
Leptoceridae						
Triaenodes	-	-	-	-	-	1
Linmephilidae						
Hydatophylax	-	-	-	-	-	1
Neophylax	-	-	5	-	-	3,5,7
Pycnopsyche	-	-	-	-	-	1,2,3,4,5,6,7
Pycnopsyche?	-	-	-	-	-	3,5,7
Molannidae						
Molanna	-	-	-	-	-	3,4,6,7

Table D.1 (continued)

Taxon	Site[a,b] MIK 0.45	MIK[c] 0.54	MIK 0.71	MIK 0.78	MIK[c] 0.86	MIK 1.43
Trichoptera (continued)						
Philopotamidae						
Chimarra	-	-	-	1,4	4,5	5,6,7
Polycentropodidae						
Polycentropus	-	-	5	-	-	-
Psychomyiidae						
Lype diversa	-	-	-	-	-	1,4,5,6
Rhyacophilidae						
Rhyacophila	-	-	-	-	-	2
Coleoptera						
Dryopidae						
Helichus	-	-	-	-	4	1,4,5
Dytiscidae						
Hydroporus	-	-	-	-	-	3
Elmidae						
Dubiraphia	6	6	7	6	-	2,3,7
Optioservus	6	-	6,7	2,5,6,7	3,4,5,6	1,2,3,4,5,6,7
Stenelmis	4	-	3,4,6,7	1,2,3,4,5,6,7	1,2,3,4,5,6	1,2,3,4,5,6,7
Haliplidae						
Peltodytes	-	-	-	3	-	-
Hydrophilidae	-	-	-	-	-	2
Tropisternus	-	4	-	-	-	-
Psephenidae						
Psephenus herricki	7	-	7	-	-	-
Ptilodactylidae						
Ancytarsus bicolor	-	-	-	-	-	1
Staphylinidae	-	-	1	1	-	-

Table D.1 (continued)

Taxon	Site[a,b] MIK 0.45	MIK[c] 0.54	MIK 0.71	MIK 0.78	MIK[c] 0.86	MIK 1.43
Diptera	-	-	-	-	6	1
Ceratopogonidae	2,3,4,5,6,7	2,4,5,6	2	1,3,4,6,7	1,3,4,5,6	1,2,3,4,5,6,7
Atrichopogon	-	4	-	-	3	3
Ceratopogonidae?	-	-	5	-	-	-
Chironomidae	2,3,4,5,6,7	2,3,4,5,6	3,4,5,6,7	2,3,4,5,6,7	2,3,4,5,6	2,3,4,5,6,7
Chironominae	2	2	-	-	-	2
Chironomini	1,2,3,4,5,6,7	1,2,3,4,5,6	3,4,5,6,7	1,2,3,4,5,6,7	1,3,4,5,6	1,2,3,4,5,6,7
Tanytarsini	1,2,3,4,5,6,7	1,2,3,4,5,6	4,5,6,7	2,3,4,5,6,7	1,2,3,4,5,6	1,2,3,4,5,6,7
Diamesinae	-	-	-	1	1	-
Orthocladiinae	1,2,3,4,5,6,7	1,2,3,4,5,6	1,2,3,4,5,6,7	1,2,3,4,5,6,7	1,2,3,4,5,6	1,2,3,4,5,6,7
Podonominae	-	-	-	-	-	2,3
Tanypodinae	1,2,3,4,5,6,7	2,3,4,5,6	2,4,5,6	1,2,3,4,5,6,7	1,2,3,4,5,6	1,2,3,4,5,6,7
Dixidae						
Dixa	-	-	-	5	-	1,2,3,4,5,6
Dixella	-	-	-	-	-	3,6
Dolicopodidae	-	-	3	-	-	1,3
Dolicopodidae?	-	-	-	-	-	2
Empididae						
Hemerodromia	2,3,6,7	2,3,6	6,7	1,4,6,7	4,5,6	3,4,5,6,7
Empididae?	-	-	-	-	-	1
Muscidae	-	-	5	-	-	1
Psychodidae	-	-	-	-	-	2
Pericoma	-	-	5	-	3	-
Simuliidae	1,2	1,4	4,6	1	1	1
Simulium	2,3,4,5,7	2,3,4,5,6	2,3,4,5,6,7	2,3,4,5,6,7	2,3,4,5,6	2,3,4,5,6
Stratiomyidae						
Caloparyphus	-	-	-	-	3	-
Stratiomys	4	-	-	-	-	-

Table D.1 (continued)

Taxon	Site[a,b] MIK 0.45	MIK[c] 0.54	MIK 0.71	MIK 0.78	MIK[c] 0.86	MIK 1.43
Diptera (continued)						
Tabanidae	-	-	-	-	-	1
Chrysops	-	-	-	7	-	2,3,4,5,6,7
Tabanus	-	-	-	-	-	3,5,6,7
Tipulidae	-	2	-	-	1,2	1,7
Hexatoma	-	-	-	-	-	2,3,4,5,6
Limnophila	-	-	-	7	-	2,3,4,5,6,7
Limnophila?	-	-	-	-	-	2
Limonia	-	-	5	3	-	-
Ormosia	-	4	-	-	-	-
Pilaria	-	-	-	-	6	3,7
Pseudolimnophila	-	-	-	-	-	3,4,5,6,7
Tipula	2,7	-	5	1,3,5,6,7	1,2,3,4,5,6	1,2,3,4,5,6,7
Mollusca						
Gastropoda						
Ancylidae						
Ferrissia	-	-	-	7	-	2,3,5,6,7
Lymnaeidae	-	-	-	1	-	-
Pseudosuccinea columella	6	4	7	-	-	-
Pseudosuccinea?	-	-	4,5	-	3	-
Physidae						
Physella	7	4	1,7	2,5	2,3	-
Planorbidae						
Heliosoma?	-	-	-	-	3	-

Table D.1 (continued)

Taxon	Site[a,b] MIK 0.45	MIK[c] 0.54	MIK 0.71	MIK 0.78	MIK[c] 0.86	MIK 1.43
Bivalvia						
Sphaeriidae	-	-	-	-	-	2,3,4,5,6,7
Pisidium	7	-	-	-	-	1,2,3,4,7
Sphaerium	-	-	-	-	-	1,3,6

[a]MIK = Mitchell Branch kilometer.

[b]The numbers associated with each taxon and site indicate the sample year(s) within which the taxon was collected at least once: 1=October 1986 and April 1987; 2=October 1987 and April 1988; 3=October 1988 and April 1989; 4=October 1989 and April 1990; 5=October 1990 and April 1991; 6=October 1991 and April 1992; and 7=October 1992 and April 1993. A blank indicates that a lower level of classification (e.g., family, genus, or species) was possible at one or more sites, and a dash (-) indicates that the taxon was not collected or that the taxon was identified to lower level at one or more sites.

[c]Samples were not collected from MIK 0.54 and MIK 0.86 during the seventh year of the study.

APPENDIX E

F-VALUES AND P-VALUES FOR NESTED ANOVAS AND REGRESSION ANALYSES

Table E.1. F-values and *p*-values for nested analysis of variance using the response variables density, total richness, and EPT[a] richness, Mitchell Branch, October 1986–April 1993

Source of variation	df[b] Num	df[b] Den	Density F-value	Density *p*-value	Total richness F-value	Total richness *p*-value	EPT richness F-value	EPT richness *p*-value
				All Sites[c]				
Site	5	30	4.47	0.0037	54.22	0.0001	34.98	0.0001
Year	5	6	0.36	0.8599	2.16	0.1883	2.06	0.2018
Season(Year)	6	30	7.70	0.0001	4.39	0.0027	3.12	0.0171
Site*Year	25	30	0.65	0.8662	1.22	0.2980	1.25	0.2736
Site*Season(Year)	30	144	8.51	0.0001	3.72	0.0001	3.74	0.0001
				Four Sites[d]				
Site	3	21	7.16	0.0017	89.12	0.0001	56.41	0.0001
Year	6	7	0.58	0.7348	2.28	0.1522	3.68	0.0559
Season(Year)	7	21	3.71	0.0091	2.74	0.0346	1.37	0.2692
Site*Year	18	21	0.83	0.6485	1.45	0.2037	1.47	0.1987
Site*Season(Year)	21	112	9.13	0.0001	3.87	0.0001	3.22	0.0001

[a]EPT = Ephemeroptera, Plecoptera, and Trichoptera

[b]df = degrees of freedom; Num = numerator; Den = Denominator

[c]Analysis included all six sites but included data from the first 6 years only (October 1986–April 1992).

[d]Analysis included Mitchell Branch kilometers 0.45, 0.71, 0.78, and 1.43 only but included data from all 7 years (October 1986–April 1993).

Table E.2. F-values and *p*-values for regression analyses testing for equal slopes across sites using the response variables density, total richness, and EPT[a] richness, Mitchell Branch, October 1986–April 1993

Source of variation	df[b] Num	df[b] Den	Density F-value	Density *p*-value	Total richness F-value	Total richness *p*-value	EPT richness F-value	EPT richness *p*-value
				Six Years[c,d]				
All sites								
Fall	5	96	4.15	0.0019	1.80	0.1209	1.12	0.3538
Spring	5	96	20.01	0.0001	7.58	0.0001	3.01	0.0144
MIK 0.54 vs MIK 1.43								
Fall	1	32	8.02	0.0079	0.97	0.3314	2.50	0.1234
Spring	1	32	4.57	0.0402	4.96	0.0330	0.00	0.9913
MIK 0.86 vs MIK 1.43								
Fall	1	32	0.20	0.6570	0.31	0.5794	0.00	0.9557
Spring	1	32	0.05	0.8276	6.41	0.0165	3.74	0.0621
				All Years[e]				
All sites								
Fall	3	76	3.64	0.0163	3.67	0.0158	1.80	0.1549
Spring	3	76	12.76	0.0001	6.17	0.0008	3.06	0.0333
MIK 0.45 vs MIK 1.43								
Fall	1	38	1.64	0.2080	0.52	0.4753	4.14	0.0488
Spring	1	38	3.19	0.0822	2.35	0.1337	0.04	0.8491
MIK 0.71 vs MIK 1.43								
Fall	1	38	5.40	0.0255	15.90	0.0003	2.89	0.0972
Spring	1	38	14.72	0.0004	13.02	0.0009	5.52	0.0241
MIK 0.78 vs MIK 1.43								
Fall	1	38	0.39	0.5369	1.60	0.2137	0.00	0.9544
Spring	1	38	3.97	0.0537	1.39	0.2452	0.26	0.6133

[a]EPT = Ephemeroptera, Plecoptera, and Trichoptera
[b]df = degrees of freedom; Num = numerator; Den = Denominator
[c]MIK = Mitchell Branch kilometer.
[d]Analysis included all six sites but included data from the first 6 years only (October 1986–April 1992).
[e]Analysis included Mitchell Branch kilometers 0.45, 0.71, 0.78, and 1.43 only but included data from all 7 years (October 1986–April 1993).

APPENDIX F

SEASONAL REGRESSION LINES FOR DENSITY, TOTAL RICHNESS, AND EPT RICHNESS

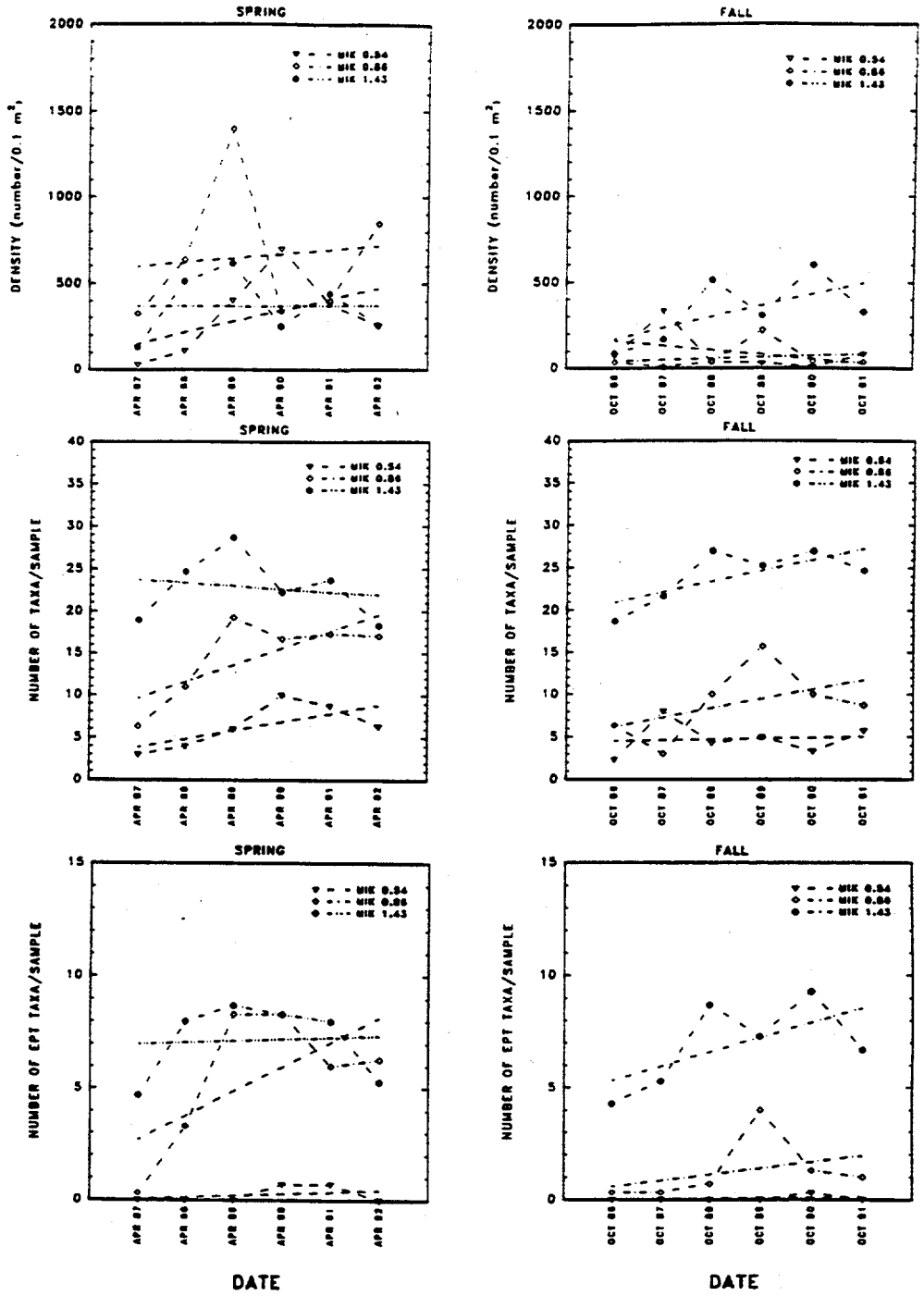


Fig. F.1. Seasonal regression lines for total density, total richness, and richness of the Ephemeroptera, Plecoptera, and Trichoptera (EPT richness) of the benthic macroinvertebrate communities in Mitchell Branch, October 1986–April 1992. Estimates of error (i.e., standard error) are available in Fig. 6.1. MIK = Mitchell Branch kilometer. Spring = April sampling periods; Fall = October sampling periods.

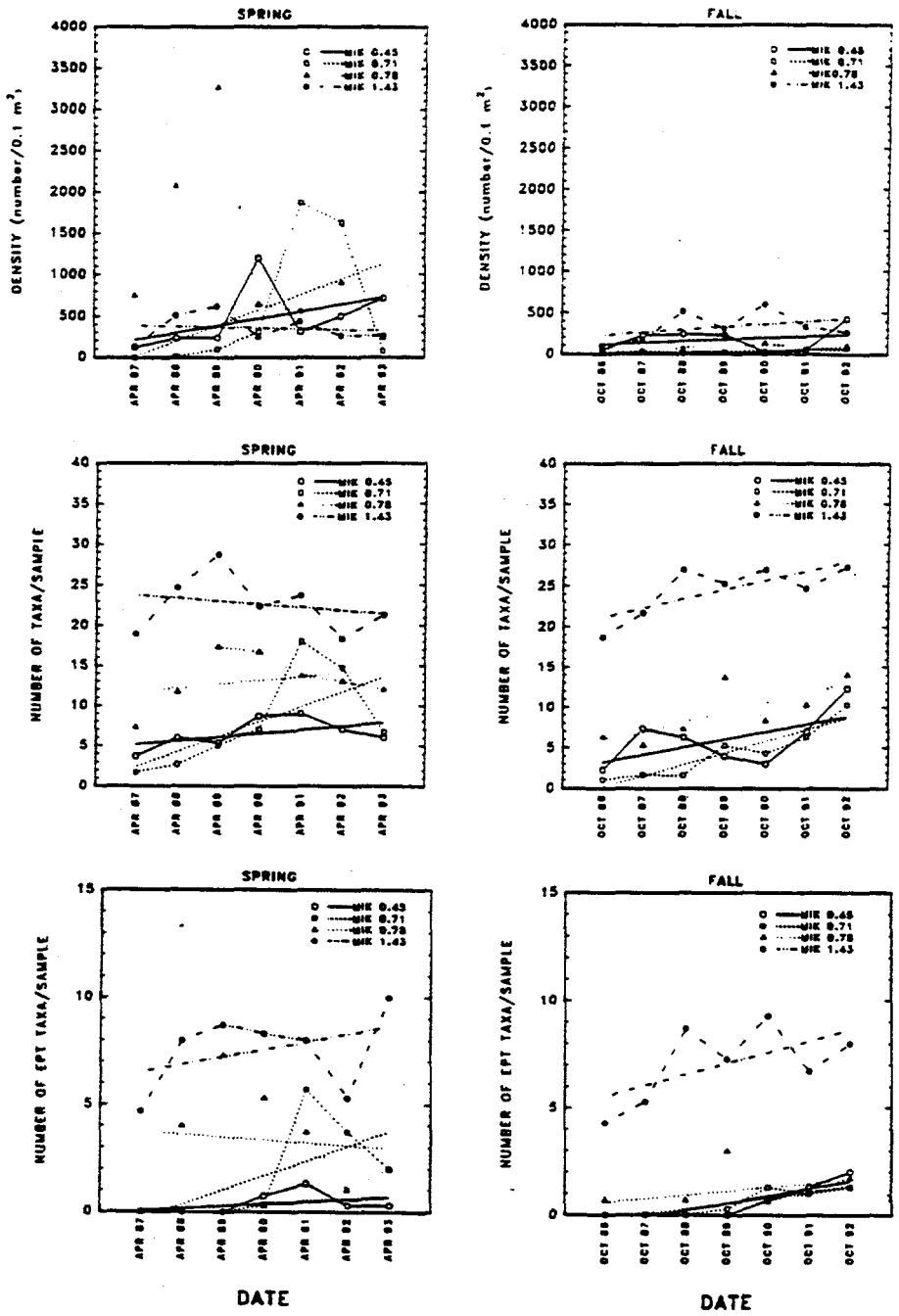


Fig. F.2. Seasonal regression lines for total density, total richness, and richness of the Ephemeroptera, Plecoptera, and Trichoptera (EPT richness) of the benthic macroinvertebrate communities in Mitchell Branch, October 1986–April 1993. Estimates of error (i.e., standard error) are available in Fig. 6.1. MIK = Mitchell Branch kilometer. Spring = April sampling periods; Fall = October sampling periods.

APPENDIX G

LENGTH FREQUENCY DISTRIBUTION OF FISHES IN MITCHELL BRANCH

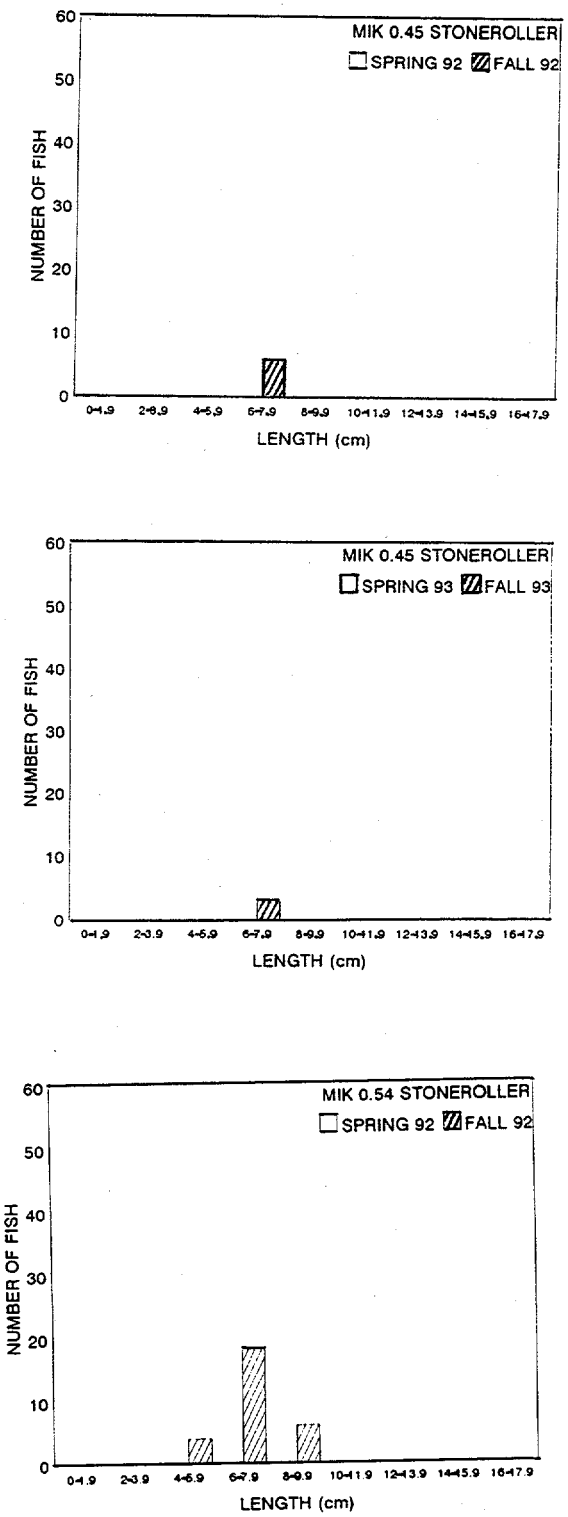
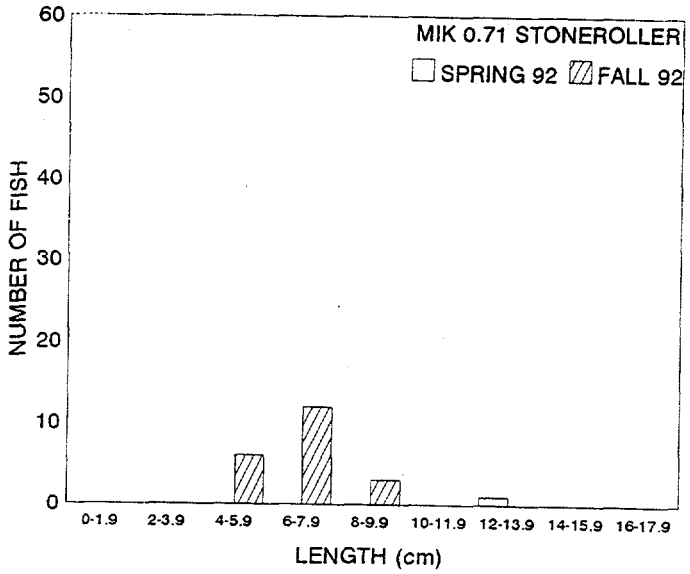


Fig. G.1. Length-frequency distribution of estimated populations of central stoneroller in Mitchell Branch (MIK) from spring 1992 through fall 1993.



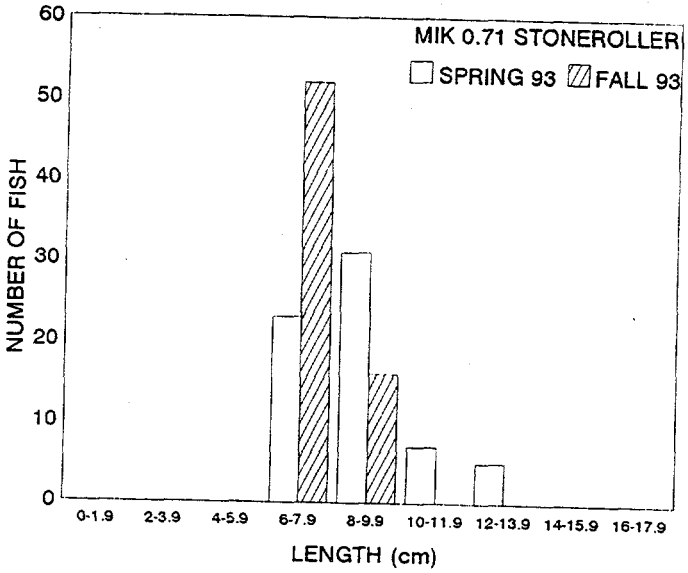
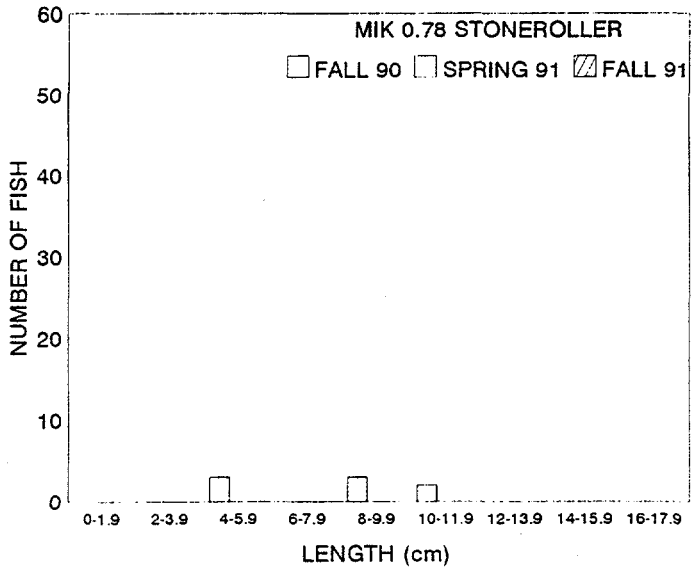


Fig. G.2. Length-frequency distribution of estimated populations of central stoneroller in Mitchell Branch (MIK) from spring 1992 through fall 1993.



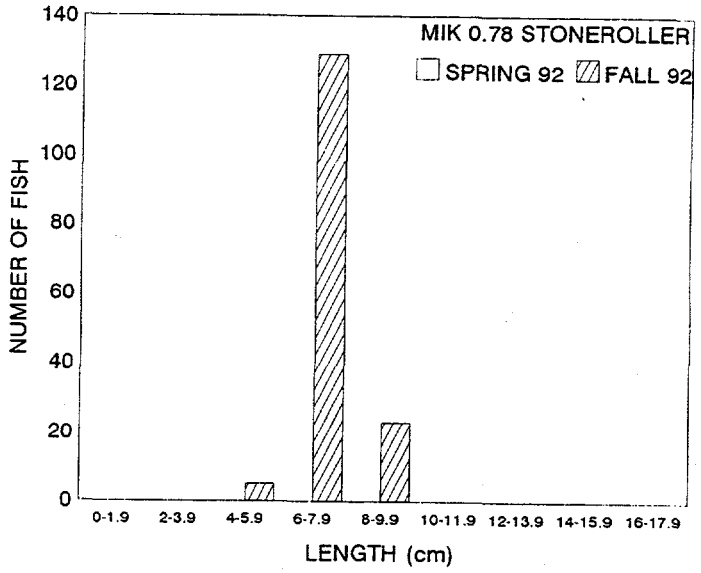
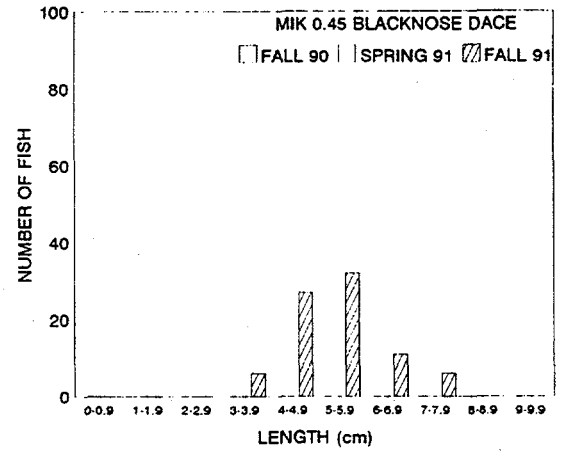
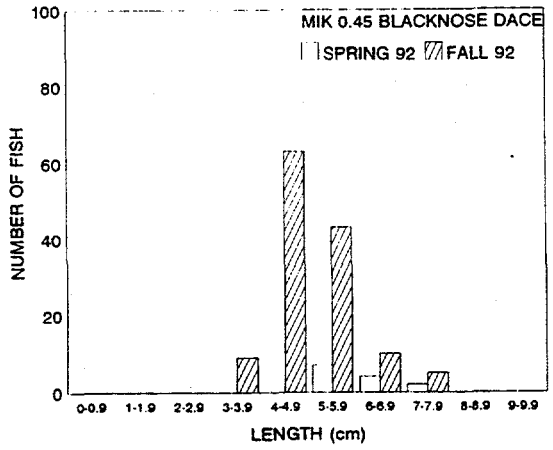


Fig. G.3. Length-frequency distribution of estimated populations of central stoneroller in Mitchell Branch (MIK) from fall 1990 through fall 1992.





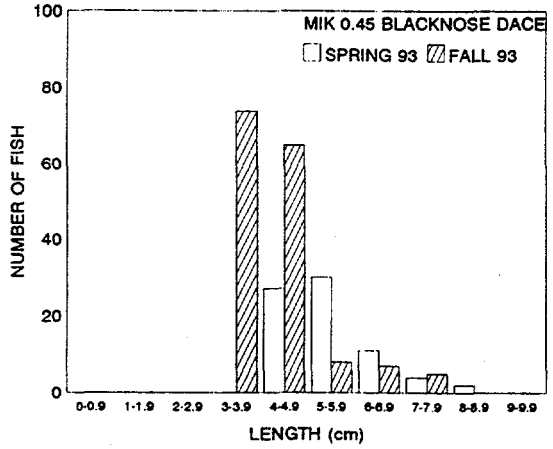
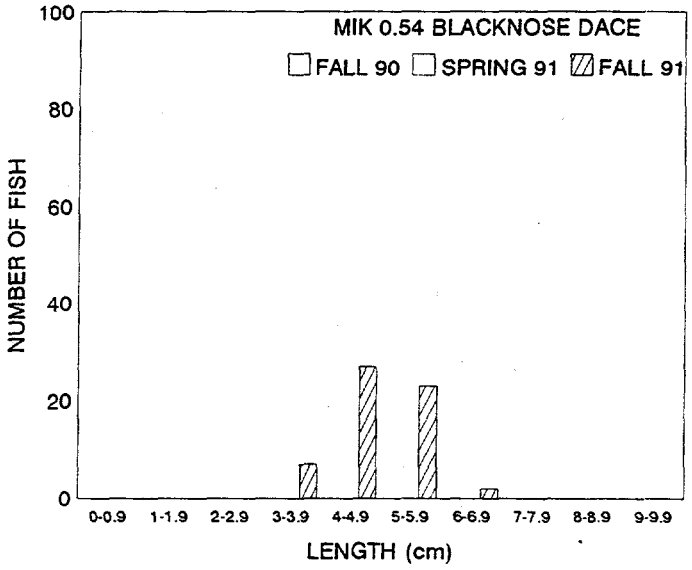


Fig. G.4. Length-frequency distribution of estimated populations of blacknose dace (BNDACE) in Mitchell Branch (MIK) from fall 1990 through fall 1993.



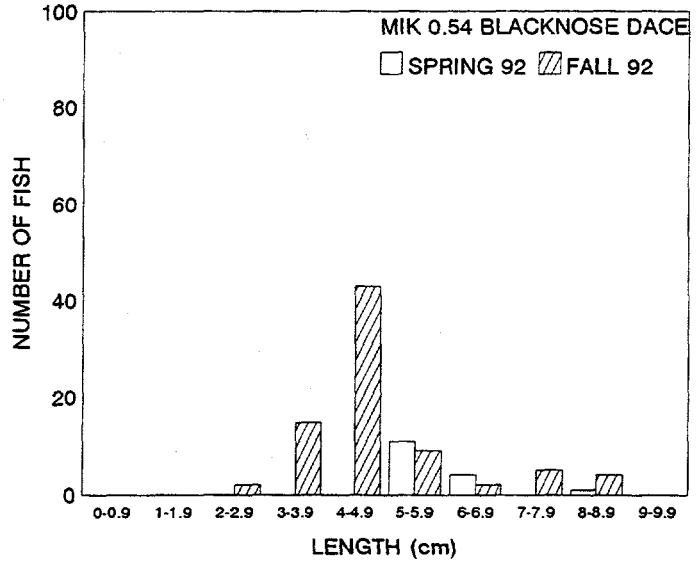


Fig. G.5. Length-frequency distribution of estimated populations of blacknose dace (BNDACE) in Mitchell Branch (MIK) from fall 1990 through fall 1992.

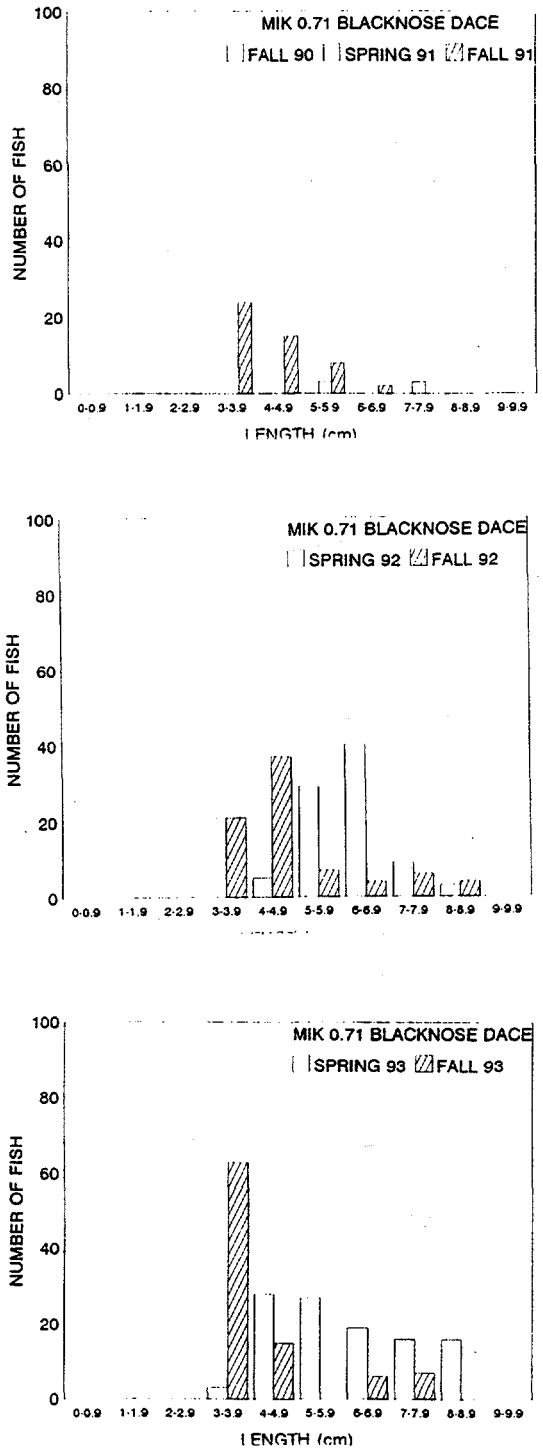
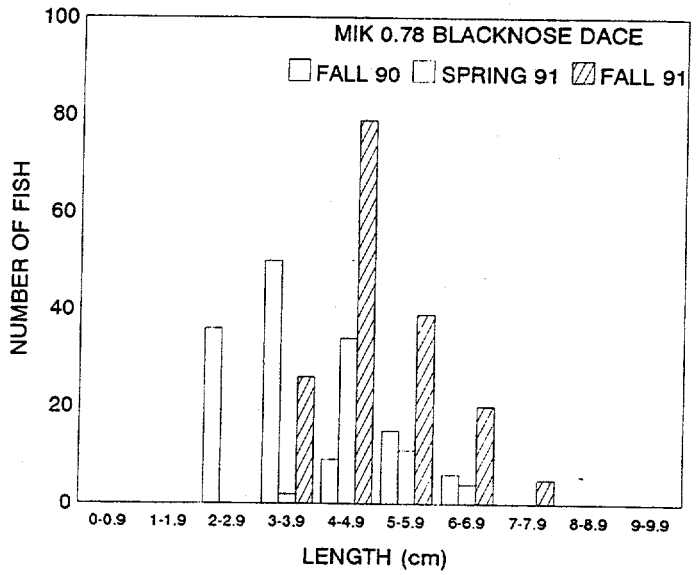


Fig. G.6. Length-frequency distribution of estimated populations of blacknose dace (BNDACE) in Mitchell Branch (MIK) from fall 1990 through fall 1993.



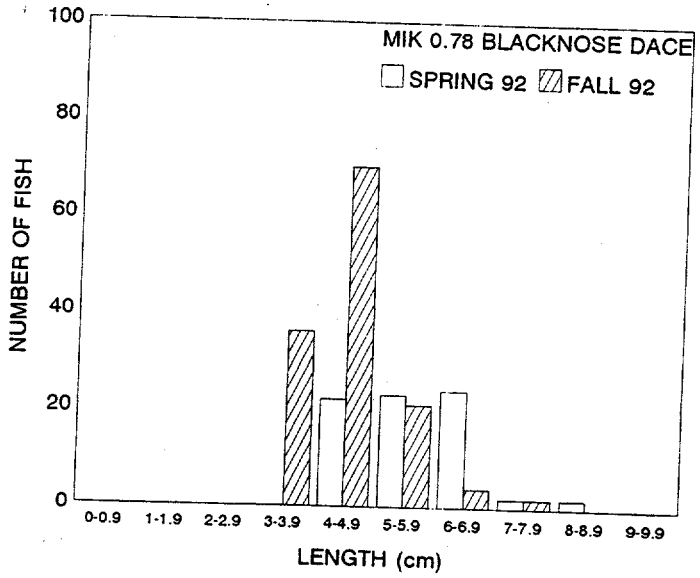


Fig. G.7. Length-frequency distribution of estimated populations of blacknose dace (BNDACE) in Mitchell Branch (MIK) from fall 1990 through fall 1992.

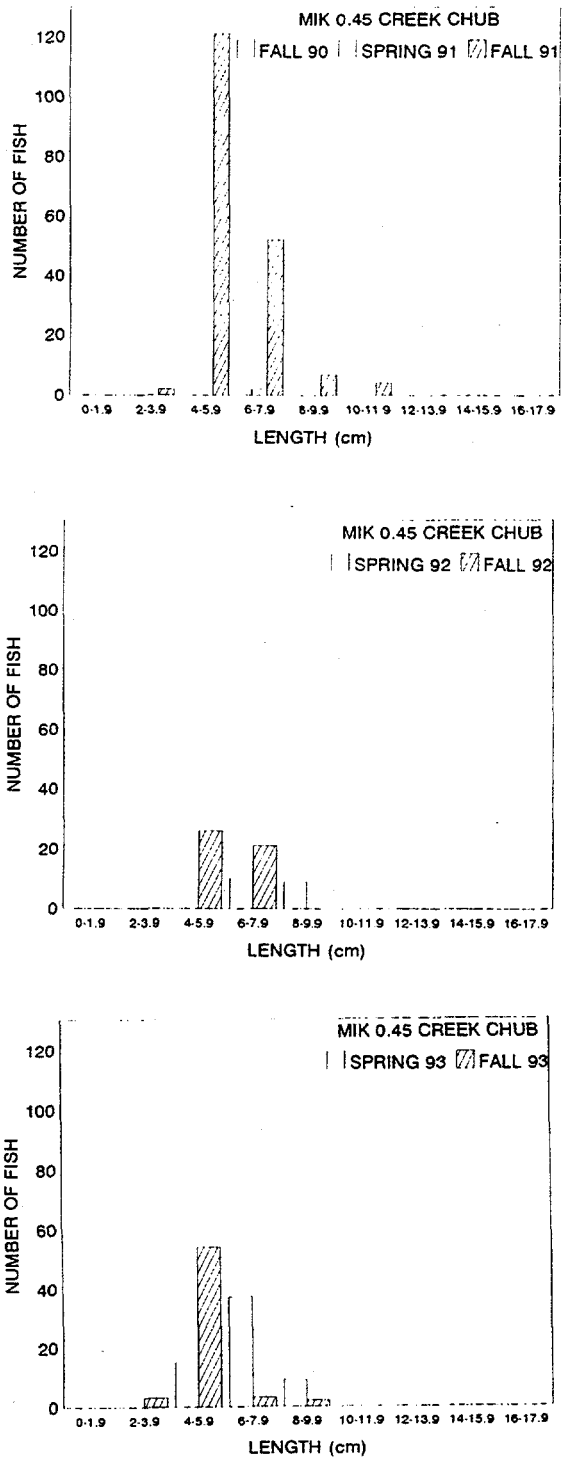
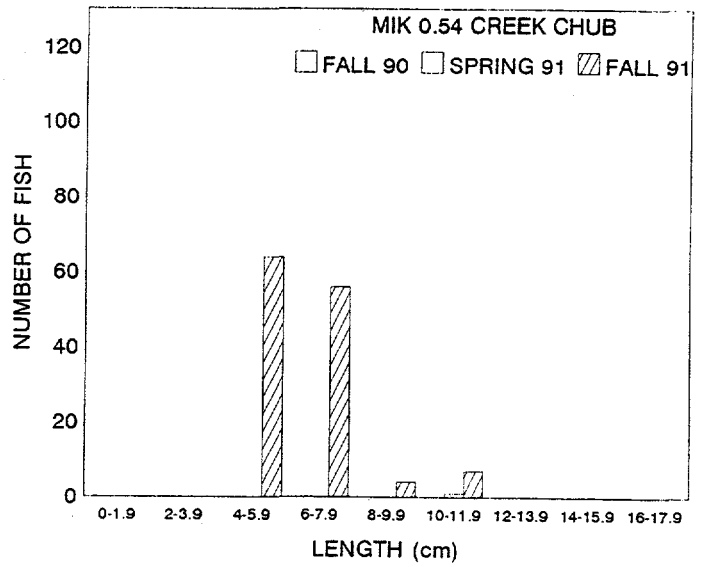


Fig. G.8. Length-frequency distribution of estimated populations of creek chub in Mitchell Branch (MIK) from fall 1990 through fall 1993.



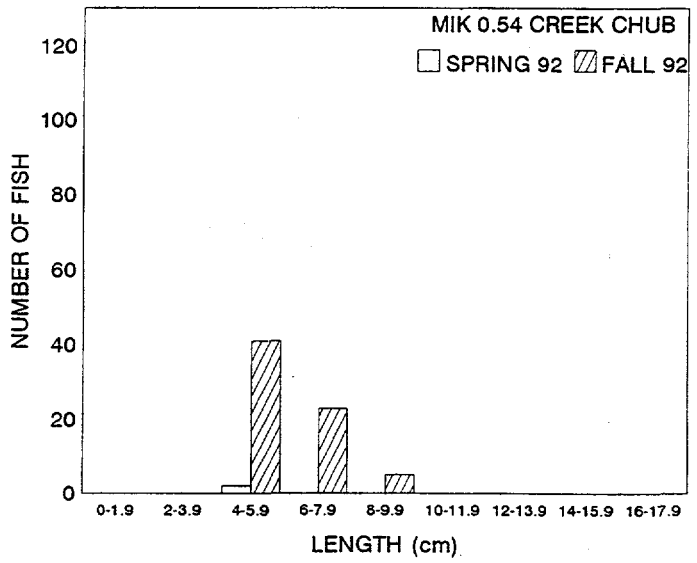


Fig. G.9. Length-frequency distribution of estimated populations of creek chub in Mitchell Branch (MIK) from fall 1990 through fall 1992.

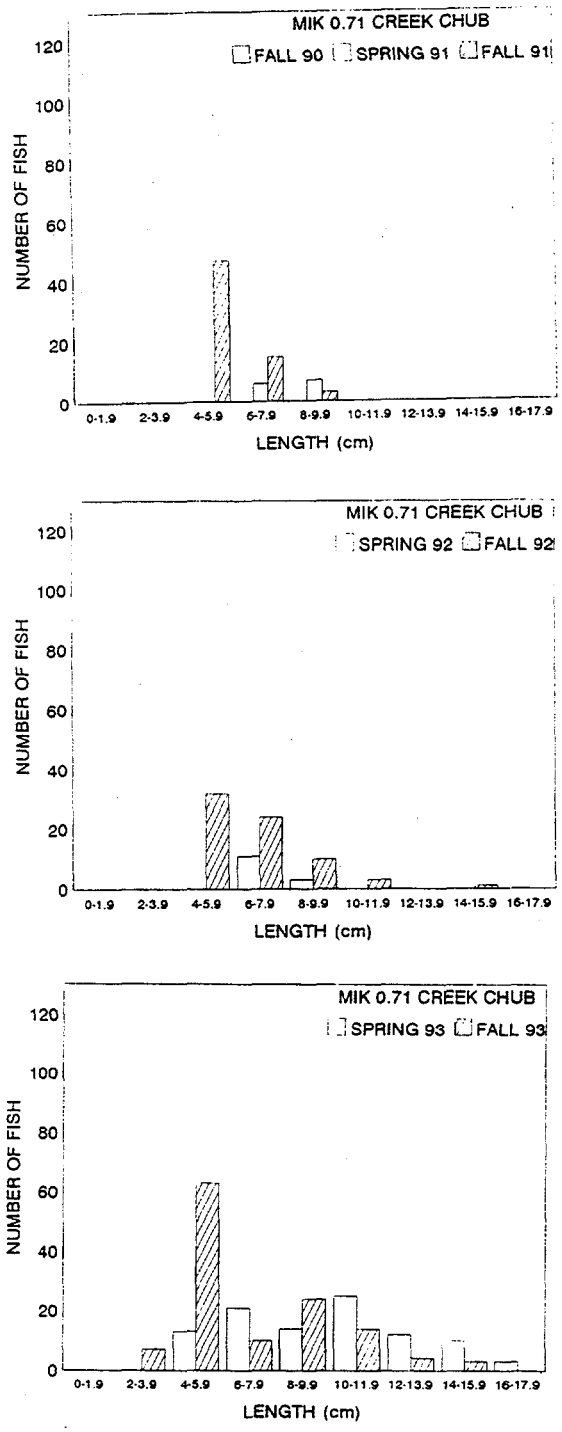
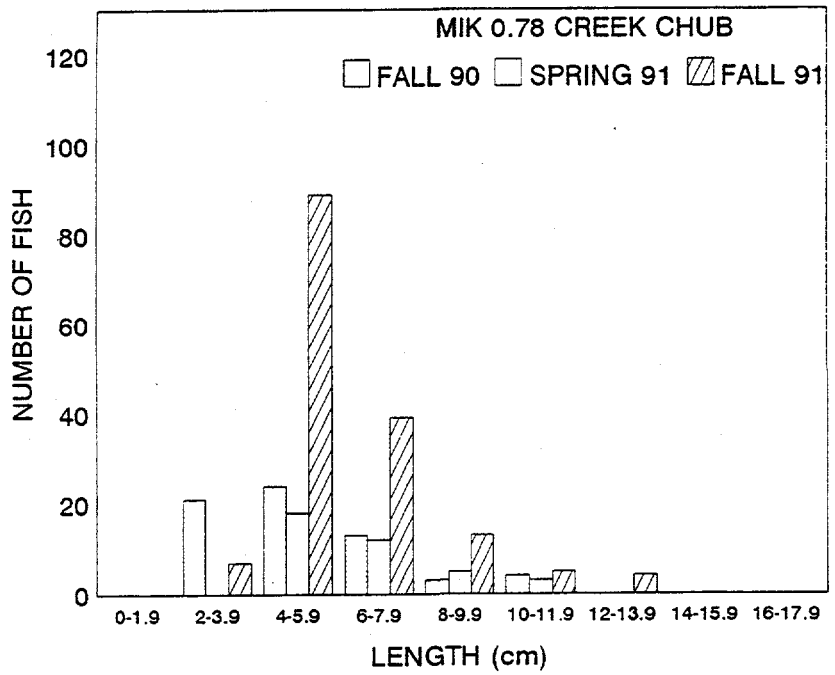


Fig. G.10. Length-frequency distribution of estimated populations of creek chub in Mitchell Branch (MIK) from fall 1990 through fall 1993.



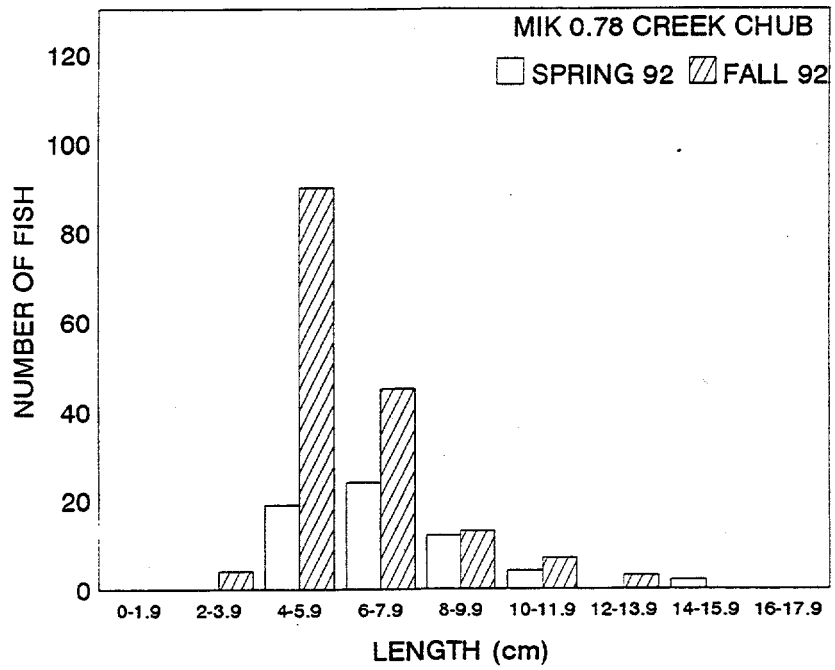


Fig. G.11 Length-frequency distribution of estimated populations of creek chub in Mitchell Branch (MIK) from fall 1990 through fall 1992.

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