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**ECOLOGICAL EFFECTS OF CONTAMINANTS  
IN MCCOY BRANCH, 1991-1993**

**M. G. Ryon**

L. A. Baron	E. M. Schilling
J. M. Bricker	J. G. Smith
B. A. Carrico	G. R. Southworth
C. T. Garten	A. J. Stewart
R. P. Hoffmeister	L. F. Wicker
M. J. Peterson	

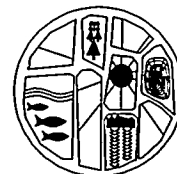
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Environmental Sciences Division

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Editor  
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Contributors

L. A. Baron <sup>1</sup>	E. M. Schilling
J. M. Bricker <sup>2</sup>	J. G. Smith
B. A. Carrico <sup>3</sup>	G. R. Southworth
C. T. Garten	A. J. Stewart
R. P. Hoffmeister <sup>3</sup>	L. F. Wicker
M. J. Peterson	

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<sup>1</sup>McLaren/Hart Environmental Engineering Corp., Warren, New Jersey

<sup>2</sup>University of Alabama, Birmingham, Alabama

<sup>3</sup>JAYCOR, Inc.

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Environmental Restoration Program  
Y-12 Plant  
Oak Ridge, Tennessee 37831

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OAK RIDGE NATIONAL LABORATORY  
Oak Ridge, Tennessee 37831  
managed by  
LOCKHEED MARTIN ENERGY RESEARCH CORP.  
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## ACRONYMS

ACD	Analytical Chemistry Division
A:C	alkalinity to conductivity
A:H	alkalinity to hardness
ALAD	aminolevulinic acid dehydratase
ANOVA	analysis of variance
BMAP	Biological Monitoring and Abatement Program
CRDL	contract required detection limit
CV	coefficient of variation
DC	direct current
DEM	Department of Environmental Management
DMW	diluted mineral water
DO	dissolved oxygen
DOE	Department of Energy
EFK	East Fork Poplar Creek kilometer
EFPC	East Fork Poplar Creek
EPA	U.S. Environmental Protection Agency
EPT	Ephemeroptera, Plecoptera, and Trichoptera richness
ESD	Environmental Sciences Division, ORNL
FCAP	Filled coal ash pond
FDA	U.S. Food and Drug Administration
GCK	Grassy Creek kilometer
GC/ECD	gas chromatography/electron capture detector
GC/MS	gas chromatography/mass spectrophotometry
GFAA	Graphic furnace atomic absorption
HESA	Department of Health, Safety, and Environmental Affairs
HMK	Hansard Mill Branch kilometer
HSRD	Health and Safety Research Division
ICP/MS	inductively coupled plasma mass spectrometry
LCT	Life-Cycle Test
L:D	light to dark
LOEC	lowest observed-effect level
MCK	McCoy Branch kilometer
MDL	minimum detection limit
NOEC	no observed-effect concentration
NPDES	National Pollutant Discharge Elimination System
ORNL	Oak Ridge National Laboratory
ORR	Oak Ridge Reservation
P	proportion of the population
PAH	polycyclic aromatic hydrocarbons
P/B	production/biomass
PCB	polychlorinated biphenyl
PGV	preliminary guidance values
PPM	parts per million

QA	quality assurance
RCRA	Resource Conservation and Recovery Act
RFI	RCRA Facilities Investigation
RI	remedial investigation
RPD	relative percent difference
RQ	Rogers Quarry
SAS	Statistical Analysis System
SCK	Scarboro Creek kilometer
SD	standard deviation
SE	standard error
SWRI	Southwest Research Institute
TDC	Tennessee Department of Conservation
TL	total length
TVA	Tennessee Valley Authority
X	population mean
WCK	White Oak Creek kilometer
WOC	White Oak Creek

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## EXECUTIVE SUMMARY

The 1984 Hazardous and Solid Waste Amendments to the Resource Conservation and Recovery Act (RCRA) required assessment of all current and former solid waste management units. Following guidelines under RCRA and the Comprehensive Environmental Response, Compensation, and Liability Act (CERCLA), a remedial investigation (RI) was required of the Y-12 Plant for their filled coal ash pond (FCAP) and associated areas on McCoy Branch. The RI process was initiated and assessments were presented (CDM 1993). Because the disposal of coal ash in the ash pond, McCoy Branch, and Rogers Quarry was not consistent with the Tennessee Water Quality Act, several remediation steps were implemented between 1986 and 1994 for McCoy Branch to address disposal problems.

The required ecological risk assessments of McCoy Branch watershed included provisions for biological monitoring of the watershed. The objectives of the biological monitoring were to (1) document changes in biological quality of McCoy Branch after completion of a pipeline bypassing upper McCoy Branch and further, after termination of all discharges to Rogers Quarry, (2) provide guidance on the need for additional remediation, and (3) evaluate the effectiveness of implemented remedial actions. The data from the biological monitoring program may also determine whether the goals of protection of human health and the environment, as identified by the State of Tennessee, of McCoy Branch are being accomplished.

## BACKGROUND

In connection with the coal ash disposal, McCoy Branch has effectively been segmented into four basic areas: (1) the headwater sections of McCoy Branch and a large coal ash pond (FCAP) created by a 19-m earthen dam; (2) a section of free-flowing stream that flows from under the dam downstream into Rogers Quarry; (3) Rogers Quarry, a deep, steep-sided, water-filled quarry; and (4) a section of free-flowing stream that extends from Rogers Quarry into an embayment area of Melton Hill Reservoir. Prior to May 1990, the major source of water for McCoy Branch was the coal-ash slurry pumped from the Y-12 Steam Plant. Initially (1955), the slurry was piped to the upper reaches of the branch and was contained behind the earthen dam. As the ash filled in the area behind the dam (1965), the slurry moved by overflow to Rogers Quarry; at first directly through McCoy Branch and then in 1989 via a pipeline. In 1986, the Y-12 Steam Plant switched to a higher grade of washed coal, increasing efficiency and decreasing the total volume of coal utilized. The Steam Plant was converted in the winter of 1988 to use natural gas as the primary fuel type. The targeted yearly ratio of energy derived from gas compared to that from coal is approximately 5 to 1; a higher percentage of coal was used in winter months. A dry vacuum system was installed in May 1990 to collect dry fly ash. Fly ash is now transported to a landfill, and all sluice water discharge to Rogers Quarry has been terminated. By January 1994, a bottom ash dewatering system was installed and functional at the steam plant. The wet bottom ash is sent through the system and the ash residue is trucked to a landfill. The wash down water generated by this process is recycled in the system for future wash down use. The last actual discharge of wet bottom ash to Rogers Quarry occurred in July 1993.

Previous biological monitoring of McCoy Branch was conducted from 1989 through 1990 and included evaluations of instream toxicity, bioaccumulation (in Rogers Quarry), fish community, and benthic community parameters (Ryon et al. 1992). This biomonitoring indicated that McCoy Branch was receiving low to moderate stress. The instream toxicity studies produced only limited effects on fathead minnow and *Ceriodaphnia* survival and fecundity in 7-d laboratory tests. *In situ* snail studies indicated that more stress was seen below Rogers Quarry than above it. Bioaccumulation studies indicated that selenium, arsenic, and possibly thallium were elevated in largemouth bass from the quarry in comparison to levels in reference fish. These fish also had severe deformities present in bony structures. Similar deformities in fish from the stream proper were noticed by the fish community studies in McCoy Branch below the quarry. A fish community was absent in McCoy Branch above the quarry, while the stream immediately below the quarry had an atypical community for a small stream with many species tolerant of degraded conditions. The benthic macroinvertebrate community assessment indicated the maximum impacts were above the quarry, with all sites showing more impact than companion reference sites. Some temporal recovery was noted in the benthic community above the quarry, which was not seen at downstream sites. Overall, McCoy Branch demonstrated moderate impacts on the biological communities, with perhaps some improvement since the cessation of fly ash disposal. If the system is beginning to recover, then the additional biomonitoring covered in this report should detect further improvement in the above areas.

## WATER QUALITY

The water quality data show that although some metal concentrations, levels of radioactivity, and organic constituents are still above reference values, the average values of these contaminants in surface water and groundwater samples in McCoy Branch watershed generally have been decreasing yearly since 1988. During this period, nitrate-nitrogen values have actually fallen below reference standards. The groundwater near Rogers Quarry has become less turbid as well. These improving conditions coincide with the elimination of Y-12 Plant discharge of fly ash into upper McCoy Branch.

Phase I and II surface water sampling yielded greater levels of typical coal ash metal contaminants, organics, and radioactivity near the point of discharge in the upstream portion of McCoy Branch, with the highest values at or near the FCAP. The leaching of these contaminants from the base of the FCAP produced the observed downstream gradient. The elevated contaminant levels in the sediments of McCoy Branch floodplain are believed to primarily exist due to the spilling over of the FCAP and subsequent rising water levels in the creek during heavy rains. Like the surface water samples, the groundwater samples exhibited higher levels of metals in McCoy Branch compared to reference samples from an unaffected stream. However, unlike the surface water samples, the downstream decline in concentrations of metals was not observed in groundwater samples. Radioactivity and the quantity of organic compounds detected in the groundwater were greatest near the FCAP.

## TOXICITY TESTING

The fathead minnow tests provided no strong evidence for acute or chronic toxicity of water at any of the tested sites. However, survival of fathead minnow larvae was <80% in

full-strength water from one or more sites in 4 of the 11 test periods that involved testing with fish. In each of the tests where minnow survival was suspect, minnow growth was greater than the control.

The results of the *C. dubia* tests also provided no strong evidence for toxicity. In 17 of the 26 site-test combinations that used full-strength McCoy Branch water, survival of *C. dubia* was 100% and in 5 of the 26 site-test combinations, it was 90%. This distribution of survival values is very similar to that obtained when using *C. dubia* to assess non-contaminated stream sites on the ORR. In 18 of the 26 site-test combinations involving full-strength water from McCoy Branch sites, *C. dubia* reproduction was  $\geq 20$  offspring per female, which is well above the EPA's criterion for control acceptability (15 offspring per female). In 11 of the 26 site-test combinations, *C. dubia* reproduction in full-strength McCoy Branch water exceeded reproduction in the controls.

The feeding rates of *Elimia* in water from MCK 1.6 and MCK 1.9 (upper McCoy Branch) were very similar to feeding rates of the snails in diluted mineral water controls for all three test periods. Water temperature strongly influenced the feeding rates of the snails, as would be expected based on simple physiological principles, but there was no significant interaction between the temperature and sediment treatments. The snail *in situ* tag-release-recapture study results indicated that *Pleurocera* and *Elimia* find it difficult to colonize McCoy Branch. Water quality factors such as pH, conductivity, alkalinity, hardness, and temperature probably can be ruled out as restricting factors in this regard. Conditions that can not yet be ruled out as restricting factors may include low but biologically significant concentrations of contaminants, either in the water or associated with the sediments or food; physical features of the sediment (e.g., limited suitable habitat on ash particles may be more corrosive than particles of rock origin during flood events); or more subtle aspects of water quality that are biologically important but which do not involve contaminants (e.g., cation ratios, such as Na:K, Ca:Mg, etc.).

The two earthworm tests showed that FCAP ash might be problematic for soil organisms. In the *in situ* test, earthworm survival in FCAP ash at the six sites was substantially lower than in peat moss. The low survival of the worms in the FCAP ash did not appear to be explained by pH, water, or organic matter content. In the laboratory test, ash from FCAP-1 site adversely affected earthworm survival. On the average, worms lost more body weight in all FCAP ash treatments than they did in any of the control- or reference-soil treatments. FCAP ash texture, or ash-associated contaminants, are factors that could account for earthworm mortality in the *in situ* and laboratory tests and/or weight-loss patterns noted in the laboratory test.

Water quality data from MCK 1.6 and MCK 1.9 were evaluated by ANOVA and by inspecting correlative relationships among conductivity, alkalinity, and hardness. The results of these analyses indicate that water quality conditions at MCK 1.6 and MCK 1.9 are similar to other stream sites on the ORR where impacts could be described as slight to moderate. These analyses provide a reasonable means for detecting major chemical perturbations (i.e., those that alter naturally established relationships among conductivity, alkalinity, and hardness), but may not be very sensitive to the presence of low concentrations of toxic contaminants. The water-quality conditions in McCoy Branch, based on consideration of relationships among conservative water-quality factors, were more similar to non-contaminated reference streams on the ORR than they were to conditions typical for streams such as EFPC.

## BIOACCUMULATION STUDIES

Concentrations of selenium and arsenic are elevated in largemouth bass (*Micropterus salmoides*) from Rogers Quarry relative to bass and sunfish from other sites in east Tennessee. Only arsenic was found to exceed conservatively based screening criteria; however virtually all biological materials from the quarry exceeded this criterion for arsenic. Cessation of inputs of fly ash to the system has not resulted in the expected decrease in arsenic and selenium concentrations in fish, suggesting that internal cycling of these elements within the quarry is maintaining elevated concentrations in the biota.

Elimination of slurried fly ash discharges to the quarry was followed by a steady increase in concentrations of mercury in the axial muscle of resident largemouth bass. Average mercury concentrations in bass (adjusted for covariance with fish weight) increased from 0.02  $\mu\text{g/g}$  to 0.17  $\mu\text{g/g}$  in 3 years. Aqueous selenium concentrations in the quarry decreased from 25  $\mu\text{g/L}$  to  $< 2 \mu\text{g/L}$  after elimination of fly ash discharges, but selenium concentrations in bass remained about three times background levels. Previous studies have shown selenium addition to be a viable means of ameliorating mercury contamination in fish in low alkalinity, low pH waters of northern Europe and Canada. These results suggest that selenium in the fly ash has effectively blocked the accumulation of methylmercury in harder, more alkaline waters during the period of ash discharge to the quarry. After ash discharge ceased, mercury accumulation increased as selenium levels declined.

## TERRESTRIAL BIOACCUMULATION STUDIES

Metals in the ash, selenium and arsenic in particular, are taken up and bioaccumulated in the vegetation on the FCAP. Additionally, chromium was found in greater concentrations in ground cover on the FCAP, compared to the Sluice and reference sites. Se and As concentrations found in deciduous foliage were significantly greater than that found at the sluice and reference sites. Selenium concentrations may cause possible adverse effects on herbivorous wildlife, including small mammals and white-tailed deer. The influx of selenium to soil by means of autumn leaf fall may also present some potential for toxicity to litter fauna.

Average concentrations of arsenic and selenium found in small mammals collected at the FCAP are significantly greater than those from the reference and Sluice sites. Contaminant concentrations and tolerance limits exceed levels which may pose a toxicological risk to the small mammals and their predators. Although cadmium, mercury, and chromium bioaccumulated in small mammals on Upper McCoy Branch sites, concentrations were relatively low. Because heavy metals, with the exception of thallium, were found in small mammals at the FCAP, these animals serve as a potential pathway for contaminant transfer to predators and other wide-ranging species.

## FISH COMMUNITY ASSESSMENT

Data on the fish populations in McCoy Branch and Rogers Quarry demonstrated that both the stream and the quarry have received stress from the coal ash operations. McCoy Branch was found to have more species of fish compared to the reference stream, Scarboro Creek, but was also found to be missing common fish species found in Scarboro Creek and



other area streams. The lack of these species may be due to past extermination in McCoy Branch and the prevention of recolonization from area streams because of the barrier posed by Melton Hill Reservoir.

Observed abnormalities such as deformed heads, eroded or missing fins were observed in fish from both McCoy Branch and Rogers Quarry but not in the reference streams. The number and severity of abnormalities in fish declined from fall 1990 through fall 1993 for both McCoy Branch and Rogers Quarry and may be the result of the decrease in fly ash entering the system.

Severe abnormalities in fish decreased over time and with an increase in distance from Rogers Quarry. Severe abnormalities decreased in fish from Rogers Quarry from 1991 through 1993. This decrease may be associated with a decrease in the amount of fly ash released to the quarry and associated components present in the water. During 1991, a greater percentage of severe abnormalities were observed in the centrarchids in Rogers Quarry as compared to the successive downstream sites of MCK 1.6 (1%) and McCoy Branch embayment (0).

A study on the introduction of a native fish species to McCoy Branch above Rogers Quarry was conducted as part of the monitoring for recovery in the watershed. Prior to 1992, upper McCoy Branch did not have an established fish community; without a fish population, any measurement of recovery of the stream following cessation of the fly ash disposal was limited to improved water quality and changes in benthic invertebrate communities. Therefore, as part of the biomonitoring of McCoy Branch, a fish introduction study was initiated for upper McCoy Branch. The study involved transplanting banded sculpins (*Cottus carolinae*) into upper McCoy Branch and following their population parameters (i.e., survival, recruitment). The introduced banded sculpins survived and were in good condition. However, there is no direct evidence that the introduced sculpins are reproducing in McCoy Branch. Therefore, whether McCoy Branch has sufficiently recovered from past coal fly-ash disposal practices to support a fish population has not yet been determined.

## BENTHIC MACROINVERTEBRATE COMMUNITY ASSESSMENT

An additional 4 years of data obtained on the benthic macroinvertebrate community in McCoy Branch since January 1990, provided evidence that a substantial and sustained recovery occurred in McCoy Branch through October 1993, particularly upstream of Rogers Quarry. Changes in the macroinvertebrate community strongly coincided with operational changes at the Y-12 Plant steam plant that initially included a substantial reduction in the amount of ash discharged to McCoy Branch, and was then soon followed by complete elimination of all fly ash discharges to the stream. Both total and EPT (a measure of sensitive species) richness increased substantially at MCK 1.9 within the first year after the changes began, and, following this, values for both parameters remained considerably higher. Full recovery probably has not occurred, although this may be clarified in the future with results from additional reference sites. However, continued impacts appear to be only slight to mild, and may be caused by unstable and/or altered habitat associated with such factors as the continued presence of fly ash.

## FUTURE STUDIES

Quarterly *C. dubia* toxicity tests (acute and chronic) will continue at MCK 1.60 and MCK 1.9 to monitor changes in water quality. Surveys of prior release sites will be made to determine whether survivors of the original snail release are reproducing. Based on this information, additional snail release studies may be initiated to determine whether snails can survive, grow, and reproduce in upper McCoy Branch. Annual monitoring of selenium, arsenic, and mercury in largemouth bass from Rogers Quarry will be continued in order to quantify the time scale of response of these elements to source reduction/elimination and the response of fish to these changes. Fish populations will be surveyed at MCK 1.6 and at an additional site in upper McCoy Branch twice a year to continue to monitor any recovery of the system. Benthic macroinvertebrate samples will continue to be collected on a quarterly basis. However, as done for this report, only samples collected during the spring and fall sampling periods will be processed unless data from the summer and winter seasons are needed to further quantify responses to changes in chemical parameters.

## 1. INTRODUCTION

The 1984 Hazardous and Solid Waste Amendments to the Resource Conservation and Recovery Act (RCRA) required assessment of all current and former solid waste management units. Such an assessment or RCRA Facility Investigation (RFI) was required of the Y-12 Plant for their Filled Coal Ash Pond (Murphy 1988). The filled coal ash pond (FCAP) was constructed in the 1950s on the upper portion of the McCoy Branch watershed. It received coal ash through a pipeline in a slurry from the coal-fired steam plant at Y-12 Plant. Because disposal of coal ash in the FCAP, and later in McCoy Branch and Rogers Quarry, was not consistent with the Tennessee Water Quality Act, several remediation steps (e.g., extending pipeline to Rogers Quarry) were implemented or planned for McCoy Branch to address disposal problems. The initial RFI plan examined the FCAP (Murphy 1988) and associated disposal concerns (e.g., heavy metals associated with fly ash disposal). An expanded RFI plan was written to also assess remediation in McCoy Branch and some downstream components of this waste management unit (e.g., Rogers Quarry) (Murphy and Loar 1988).

The McCoy Branch RFI plan included provisions for biological monitoring of the McCoy Branch watershed. As outlined in the first McCoy Branch biomonitoring report (Ryon et al. 1992), the objectives of the biological monitoring were to (1) document the degree of improvement in biological quality of McCoy Branch after completion of the pipeline and after all ash discharges to Rogers Quarry are terminated; (2) provide guidance on the need for additional remediation; and (3) evaluate the effectiveness of the remedial actions that are implemented. The data from the biological monitoring program will also be sufficient to determine whether the classified uses of McCoy Branch, as identified in the State of Tennessee Water Quality Management Plan for the Clinch River Basin (TDPH 1978), are being adequately protected and maintained (Murphy and Loar 1988). These objectives are still a primary mission of the McCoy Branch biomonitoring program; but, during the 1991-93 period, providing support for a risk assessment analysis of McCoy Branch became an important additional objective.

The overall strategy for the biological assessment remained the same as described in the initial report and was based on four considerations: (1) Because remediation efforts are phased temporally, the biological monitoring would also be implemented in stages with increased monitoring as the system recovered. (2) The size of the stream restricted the application of assessment techniques to certain areas. (3) The fauna of McCoy Branch appeared depauperate; therefore, some constraints on the parameters to be measured were necessary, and the need was recognized for a limited number of reference sites. (4) Because the time frame for reaching hydrologic equilibrium following remediation is unknown, the biological monitoring must consider changes in hydrologic regime and improvement in water quality (Murphy and Loar 1988). As more became known about McCoy Branch, the monitoring program was expanded to cover the three principal physical areas of the system: (1) the upper section of the watershed (above Rogers Quarry), (2) Rogers Quarry, and (3) the lower section (between the quarry and the Clinch River). The biological monitoring applied to McCoy Branch consisted of an integrated multitiered program with five primary tasks: (1) toxicity monitoring, (2) bioaccumulation assessments, (3) terrestrial assessment, (4) fish community assessments, and (5) benthic community assessments.

The results of these tasks are presented in this report. The results focus on the period from 1991 to 1993, but selected data from previous years are also presented.

## 2. DESCRIPTION OF THE WATERSHED AND CONTAMINANTS

(R. P. HOFFMEISTER)

### 2.1 WATERSHED CHARACTERISTICS

The McCoy Branch watershed is located south of the Y-12 Plant (Fig. 2.1) within the Department of Energy's (DOE) Oak Ridge Reservation (ORR). The ORR, which is located in the Valley and Ridge physiographic province, is characterized by southwest-northeast oriented, parallel ridges of sandstone, shale, and cherty dolomite, separated by valleys of less resistant limestone and shale (Murphy 1988). The McCoy Branch watershed drains Fanny Knob on the southern slope of Chestnut Ridge and supplies water to Melton Hill Reservoir. Land use in the watershed includes grass-covered slopes and fields, selected marshy pockets in the floodplain, agricultural buildings, roads, and heavily forested slopes.

The eastern branch of McCoy Branch has been extensively modified by coal ash disposal. (This report will deal only with the eastern branch.) In connection with the coal ash disposal, McCoy Branch (Fig. 2.2) has been segmented into four basic areas: (1) the headwater sections of the stream and a large (8 ha) filled coal ash pond (FCAP) created by a 19-m high earthen dam; (2) a section of free-flowing stream that originates from a spring at the back of the dam downstream into Rogers Quarry and that, after rain, is supplemented by flow over the top of the dam; (3) Rogers Quarry, a deep, steep-sided lake; and (4) a section of free-flowing stream reaching from Rogers Quarry into an embayment of Melton Hill Reservoir. The western branch of McCoy Branch also enters this embayment.

The headwaters of McCoy Branch consist of two intermittent streams with minimal discharge (Turner et al. 1986). A large pipe initially used to transfer coal ash slurry from Y-12 Plant to the FCAP is located between these two streams. In the past, the flow from these three sources combined in the ash pond, which contains the ash slurry from the early years of disposal. The channel in the pond was shallow and meandered over the surface of the ash to the earthen dam. During ash sluicing and high storm runoff, flows reached the dam and were transported downstream via an overflow spillway on the eastern side of the dam.

Water in McCoy Branch below the dam derives primarily from several large springs. The spring at the base of the dam is heavily vegetated and is fed by groundwater discharge. From the dam the stream flows approximately 0.9 km to Rogers Quarry. In this reach the stream is typically narrow (about 0.5 to 2.0 m in width) and fast-flowing and consists of a series of runs and riffles. Just above Rogers Quarry lies a marshy area with a large spring between McCoy Branch (to the east) and a municipal sludge farm to the west. McCoy Branch flows into this marshy area before being diverted to Rogers Quarry (Murphy and Loar 1988).

Rogers Quarry is a 4-ha lake located about midway in the eastern branch of McCoy Branch. The quarry was used as a source of stone in the 1940s and was abandoned when it began to fill with water in the 1950s (Bogle and Turner 1989). After leaving Rogers Quarry, the stream flows through Bethel Valley to the McCoy Branch Embayment on the Melton Hill Reservoir. The stream below the quarry is heavily vegetated and receives flow from several spring-fed tributaries.

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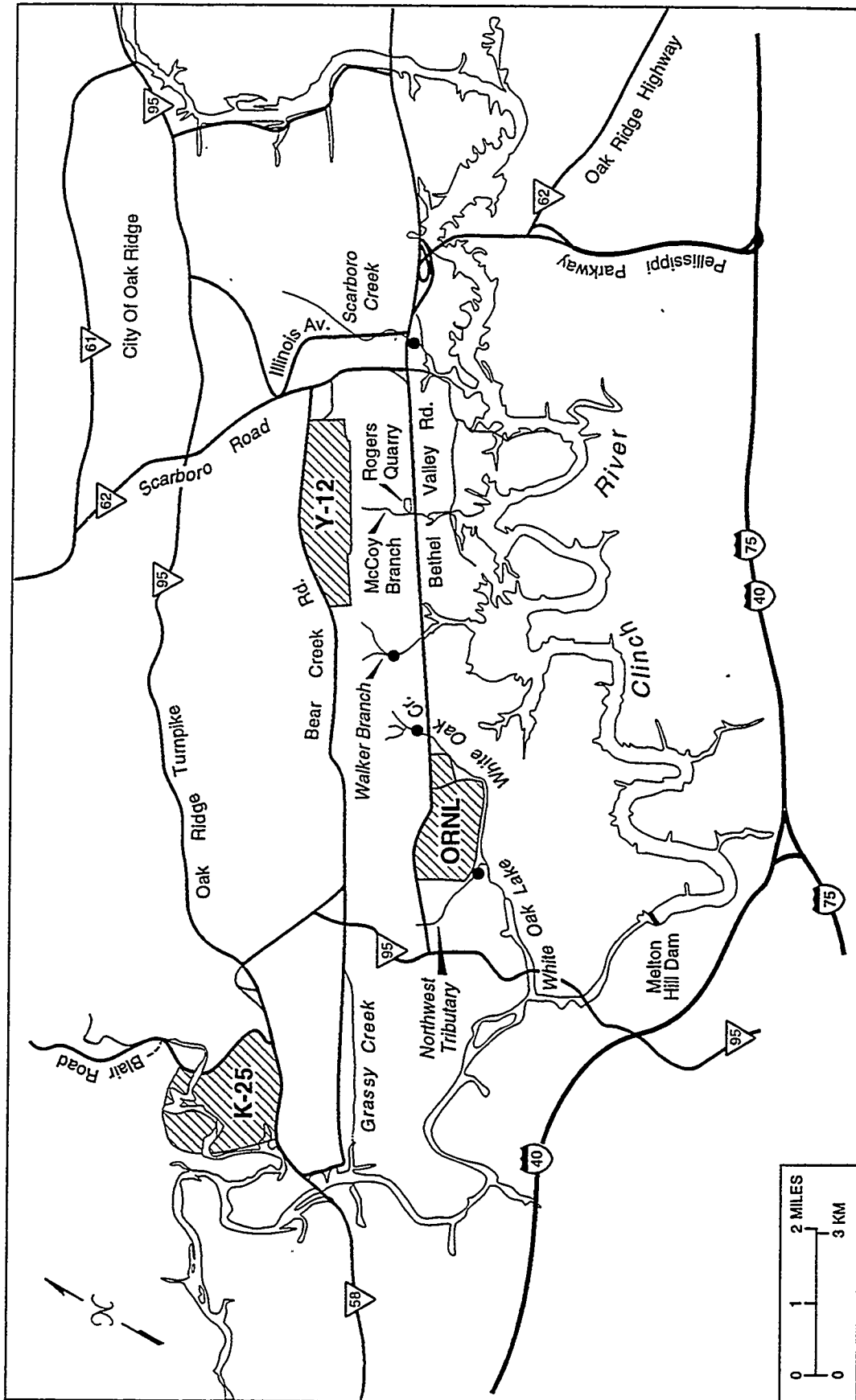


Fig. 2.1. Map of the McCoy Branch Watershed in relation to the Oak Ridge Reservation and reference streams.

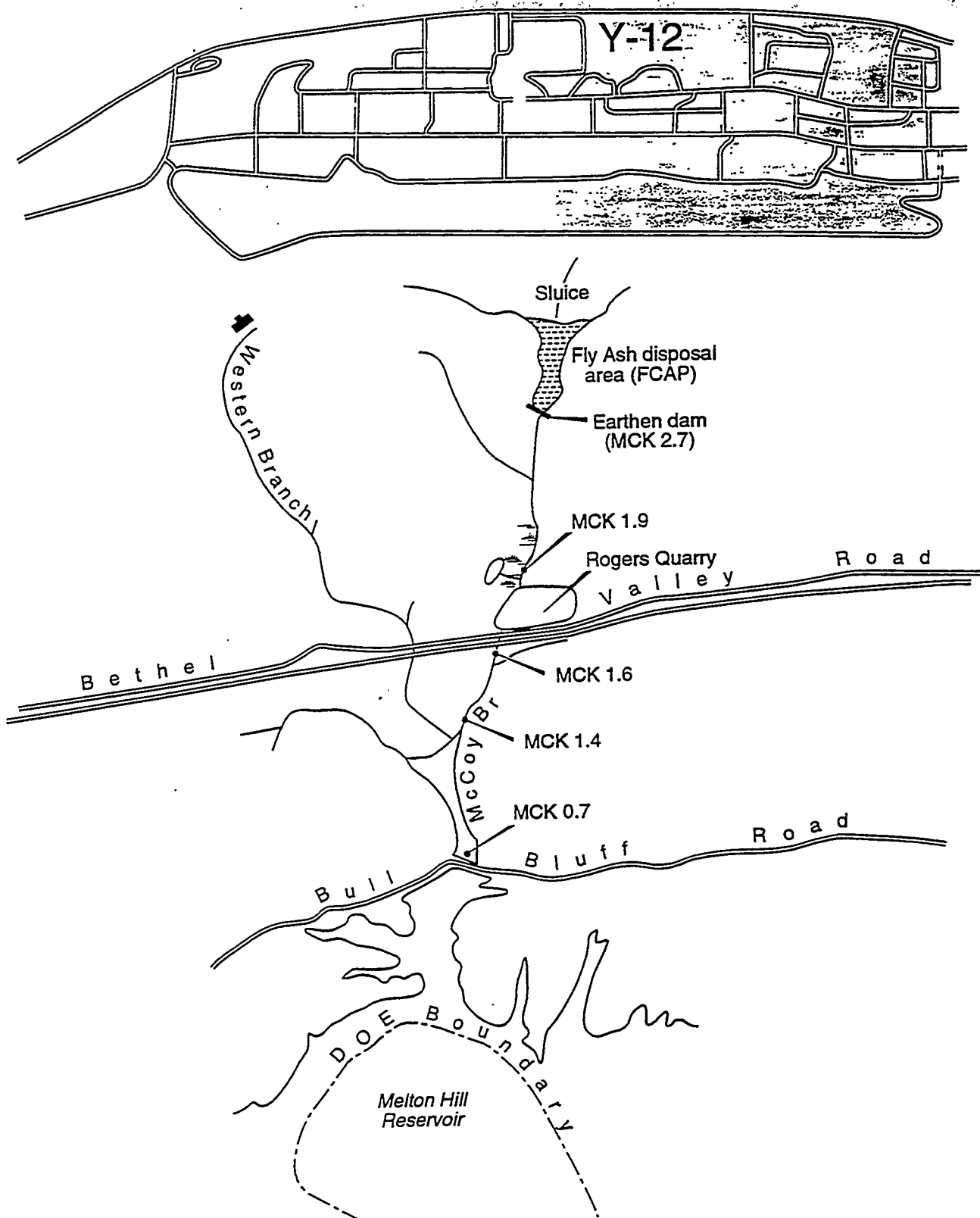


Fig. 2.2. Map of the McCoy Branch showing the primary features and monitoring sites.

McCoy Branch is geographically separated from the Y-12 Plant by Chestnut Ridge. The distance from the ash slurry pipe on top of the ridge to the discharge into Melton Hill Reservoir is approximately 1.5 km. The elevation at the ash slurry pipe at the top of the ridge is about 335 m; elevation at the point where McCoy Branch discharges into Melton Hill Reservoir is approximately 244 m. The watershed has a drainage area of about 148 ha above Rogers Quarry, 63 ha of which lie above the earthen dam located in the headwaters.

## 2.2 GEOHYDROLOGY

McCoy Branch is underlain by the Knox group, a highly crystalline dolomite interbedded with shale and mudstone beds, a few very thin sandstone beds, aphanitic limestone lenses, numerous calcite and quartz filled fractures and cavities, and chert beds and nodules (Murphy 1988). Ketelle and Huff (1984) found that water movement in the bedrock of the Knox Group on Chestnut Ridge is controlled by the location and orientation of these cavities. Primary orientation of cavity systems are controlled by the local bedding orientation and orientation of penetrative joints and fractures, which are widened by dissolution. The actual groundwater flow paths in the bedrock are expected to resemble trellis drainage patterns, flowing parallel to strike and diverted by shorter cross-strike channels to other strike-controlled zones or emanated in surface streams. Within both the soil and bedrock aquifers, flow is from the higher topographic areas toward the lower areas, with gradients indicating flow toward the nearest perennial surface water features (Ketelle and Huff 1984; Murphy 1988).

Currently, the major source of water for the upstream reach of McCoy Branch is spring flow and precipitation. Craig and Tschantz (1986) estimated the combined base flow of these springs to be in the range of 0.42–0.53 million L/d. For a more comprehensive characterization of the geology and hydrology of the FCAP, see Murphy (1988), Jones and Mishu (1986), and Turner et al. (1986).

## 2.3 CONTAMINANT DISCHARGES

Water quality of McCoy Branch has been affected primarily by coal ash discharge. The ash sluice water discharged into McCoy Branch was enriched with total suspended solids, sulfates, phosphorous, and various metals (Turner et al. 1986). Sluicing of the ash was intermittent, with seasonal variation in the amount of ash discharged from about 0.76 million L/d in summer to 3.8 million L/d in winter (Murphy 1988; Bogle and Turner 1989). Ash residues remain in the FCAP as well as the streambed and floodplain downstream of the dam.

### 2.3.1 Description and History of Discharges

Prior to May 1990, the major source of water for McCoy Branch was the coal-ash slurry pumped from the steam plant at the Y-12 Plant. The steam plant, built in 1954, has undergone several renovations and upgrades including conversion to a baghouse-type fly ash collection system in 1985 (Turner et al. 1986). The plant consists of four boiler units that used pulverized coal as the sole fuel source from 1954 to 1988. On December 30, 1988, two



of the four boilers were converted to enable them to use natural gas as the primary fuel source and coal as a secondary source (D. M. Harvey, Y-12 Steam Plant, personal communication to J. G. Smith, Environmental Sciences Division, Oak Ridge National Laboratory). On February 15, 1990, the remaining two boilers had the dual fuel capacity installed. The steam plant produces two types of ash when burning coal. Historically, fly ash (dry ash) generated at the steam plant was removed by a wet collection system, and bottom ash (ash collected from the bottom of the boiler in wet form) was removed by high pressure water jets. The ash was then sluiced (pumped) in series—first the fly ash and then the bottom ash—as a slurry through an 8-in.-diam. iron pipe over the crest of Chestnut Ridge (CDM 1994). Gravity flow carried the effluent to the FCAP, which provided sedimentation for the ash slurry before discharge into McCoy Branch (Turner et al. 1986; M. A. Kane, Y-12 Plant, personal communication to R. L. Hinzman, Environmental Sciences Division, Oak Ridge National Laboratory). Constructed in 1955, the pond was expected to hold a volume of approximately 129-acre-ft of ash slurry and 20 years of steam plant ash but was filled to within almost 1 m of the top of the dam by July 1967 (CDM 1994; Murphy 1988). Subsequently, flow from McCoy Branch was used to channel the slurry to Rogers Quarry.

Between 1967 and 1993, Rogers Quarry was used for the disposal of ash slurry from the Y-12 Plant. The quarry was also used between 1965 and 1981 for the disposal of weapons-related and classified items (Table 5 in McCauley 1986; Bogle and Turner 1989). For a complete description of Rogers Quarry see Bogle and Turner (1989). On November 13, 1989, ash slurry discharge to the upper reaches of McCoy Branch was terminated by extending the pipeline directly to Rogers Quarry, which has an estimated life expectancy for ash disposal of 65 to 115 years (D. M. Harvey, Y-12 Plant steam plant, personal communication to J. G. Smith, Environmental Sciences Division, Oak Ridge National Laboratory; Murphy 1988; M. A. Kane, Y-12 Plant, personal communication to R. L. Hinzman, Environmental Sciences Division, Oak Ridge National Laboratory).

Discharges into McCoy Branch and Rogers Quarry were in violation of the Tennessee Water Quality Act. Several actions were performed to comply with the appropriate regulations. The Y-12 Plant steam plant switched to a higher grade of washed coal, increasing efficiency and decreasing the total volume of coal utilized in 1986. The steam plant was converted in the winter of 1988 to use natural gas as the primary fuel type. The targeted yearly ratio of energy derived from gas compared to that from coal is approximately 5 to 1, with a higher percentage of coal being used in winter months. A dry vacuum system was installed in May 1990 to collect dry fly ash. Fly ash is now put in a landfill, and all sluice water discharge to Rogers Quarry has been terminated. By January 1994, a bottom ash dewatering system was installed and functional at the steam plant. The wet bottom ash is sent through the system and the ash residue is trucked to a landfill, while the wash down water is recycled in the system for future wash down use. The last wet bottom ash discharge to Rogers Quarry occurred in July 1993 (R. Ahl, Y-12 Plant steam plant, personal communication to R. P. Hoffmeister, Environmental Sciences Division, Oak Ridge National Laboratory).

### 2.3.2 Source Characterization

Since the Y-12 steam plant changed to a higher grade of washed coal in 1986, there has been a steady decline in the amount of coal burned (Fig. 2.3). In 1984, over 120,000 tons of coal were consumed by the plant. The consumption rate fell by nearly 15% by the end of 1986. The largest decline in the amount of coal burned occurred between 1988 and 1989

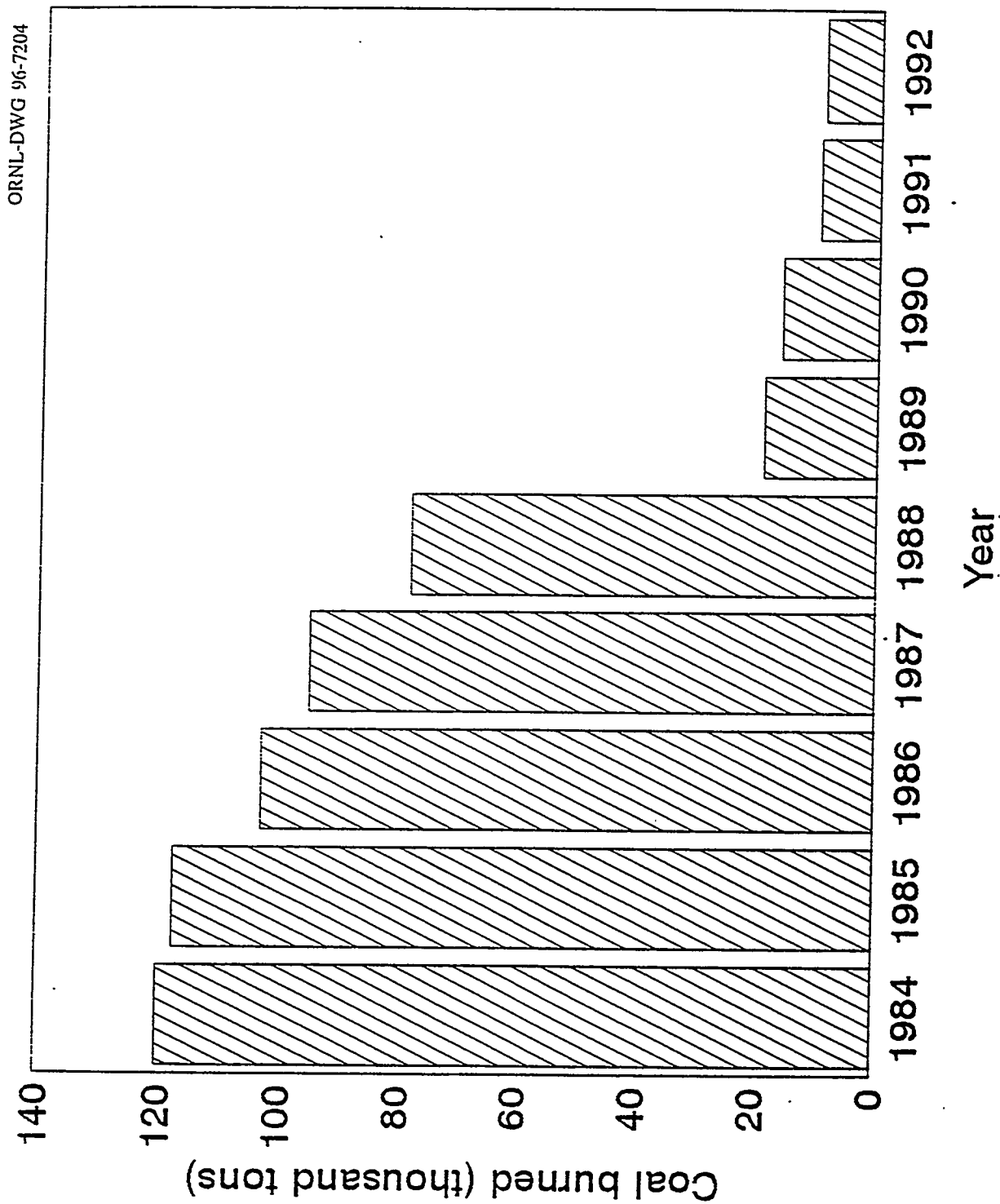


Fig. 2.3. Amount of coal burned (thousand tons) by Y-12 Steam Plant from 1984 to 1992. Source: D. M. Harvey, Y-12 Steam Plant, personal communication to J. G. Smith, Environmental Sciences Division, ORNL.

when the steam plant converted the primary fuel source to natural gas. The amount of coal burned decreased from 78,463 tons in 1988 to 18,882 tons in 1989; a reduction of 76%. By the end of 1993, coal consumption had decreased over 96% since 1984. The steam plant was converted back to using coal as the primary fuel source on January 17, 1994, and is using natural gas only in emergency situations (D. M. Harvey, Y-12 Plant steam plant, personal communication to R. P. Hoffmeister, Environmental Sciences Division, Oak Ridge National Laboratory). This conversion was made because coal is cheaper to burn than gas. The dry ash is processed at the plant's dry ash facility and the wet bottom ash is sent through the bottom ash dewatering system.

The chemical constituents of coal include all naturally occurring elements (El-Mogazi et al. 1988). The resulting fly ash from the combustion of coal contains nearly all of these elements in trace amounts. Typically, the elemental constituents found most in fly ash include Al, Ca, C, Fe, Mn, Mg, K, P, Si, Na, S, and Ti (El-Mogazi et al. 1988). Major trace elements in fly ash are arsenic, boron, cadmium, chromium, copper, lead, molybdenum, nickel, selenium, and zinc. Several mutagenic and carcinogenic polycyclic aromatic hydrocarbons (PAH) organic compounds have been detected in coal fly ash, but a thorough list has not been produced because of the complexity in isolating them (El-Mogazi et al. 1988).

## 2.4 SURFACE WATER

### 2.4.1 Surface Water Chemistry

A Remedial Investigation (RI) program consisting of two phases was conducted on McCoy Branch beginning in 1990 to characterize contaminant levels due to the coal ash sluicing by the Y-12 Plant. Phase I concentrated on sampling sediment soils, surface waters, and groundwaters of upper McCoy Branch. Sampling was expanded in Phase II to include more locations along the upper reaches of the stream to the FCAP and the sluice channel.

Phase II surface water sampling of Upper McCoy Branch involved additional sampling of the stream above Rogers Quarry near springs and in the FCAP. Table 3.10 in CDM (1994) includes those metals and one organic compound whose values exceed ambient water quality standards. All of the eight metals (Al, As, Be, Cu, Fe, Pb, Hg, and Zn) exceeding these standards had their highest values measured after a rain event. Rain events, especially heavy rains, increase leaching rates of contaminants contained in the sediments of Upper McCoy Branch and the FCAP. These samples were taken near a spring at the foot of the dam and represent leachate from the bottom of the FCAP. The data showed that six of the eight metals had higher concentrations in the FCAP than in a reference pond. Other metals such as Ba, Ca, Cr, Mg, Mn, K, Na, and V also had their highest levels in the FCAP. All of these trace metals were found at higher levels in the upper east end of the FCAP than the upper west end. Al, As, Ca, Mn, K, Na, and Z concentrations were higher in Upper McCoy Branch than in reference streams. The organic compound bis(2-Ethylhexyl)Phthalate was detected at the highest levels at a spring location nearly midway between the FCAP and Rogers Quarry. Levels of  $\text{Th}^{228}$ ,  $\text{Th}^{230}$ ,  $\text{U}^{234}$ , and  $\text{U}^{238}$  in the FCAP exceeded those in a reference pond. Gross alpha and gross beta existed near the spring at the base of the dam. Compared to the data in Phase I (Appendix A, Tables A-8 to A-13; Ryon et al. 1992) the data in Phase II also show the trend of decreasing contaminant concentrations downstream of the quarry. This was determined for arsenic in a detailed study for McCoy Branch by Ford and colleagues (1993). They found the highest levels of arsenic at the outfall of Rogers Quarry.

Surface water data collected from a National Pollutant Discharge Elimination System (NPDES) station downstream of Rogers Quarry at Outfall 302 showed a decrease in mean weekly levels of sulfate, selenium, arsenic, and iron since the Y-12 Plant steam plant converted from coal to natural gas as its primary fuel source in 1988 (Table 2.1). The sharp increase in the mean selenium value in 1993 was due to a change in the analytical detection limit for that parameter (K. Hanzelka, HSEA Division, Y-12 Plant, personal communication to R. P. Hoffmeister, Environmental Sciences Division, Oak Ridge National Laboratory). Water turbidity and water flow rates leaving the quarry have been decreasing since 1989. Since this time Rogers Quarry has acted more as a settling basin for the remaining contaminants upstream of the quarry.

**Table 2.1. Mean weekly values for selected surface water quality parameters from National Pollutant Discharge Elimination System station Outfall 302 for 1986 through 1993**

Parameter	1986	1987	1988	1989	1990	1991	1992	1993
Arsenic (mg/L)	0.21	0.26	0.2	0.07	0.05	0.04	0.05	0.04
Iron (mg/L)	0.56	0.13	0.1	0.14	0.11	0.08	0.06	0.06
Selenium (mg/L)	0.02	0.03	0.02	0.01	.004	.002	.002	.043
Sulfate (mg/L)	91.6	76.2	92.0	51	32	22	21	19
Turbidity (NTU)	4.1	2.7	3.0	3.48	2.25	1.60	2.61	0.67
Flow (L/s)	— <sup>a</sup>	—	33.7	46.9	43.8	39.0	14.9	12.8
Temperature (deg.C)	17.6	17.5	17	17.5	17.9	19.4	12.8	11.4
pH (units)	6.8/9.5 <sup>b</sup>	7.6/9.3	6/9	6.9/9.1	7.1/9.0	7.0/9.2	7.3/8.6	7.3/8.2

<sup>a</sup>Data not available.

<sup>b</sup>Minimum value/maximum value.

Sources: 1993 data: K. Hanzelka, Health, Safety, Environment, and Accountability Division; Y-12 Plant, personal communication. 1989 through 1992 data: Kornegay et al. Vol.2, 1990b, 1991b, 1992b, 1993b. 1987 and 1988 data: Rogers et al., Vol.2, 1988b, 1989b. 1986 data: Oakes et al. 1987, Vol.2.

The Y-12 Plant had a history of compliance problems with pH levels at Rogers Quarry. As Table 2.1 shows, pH levels above the NPDES upper limit of 8.5 existed from 1986 through 1992. Fig. 2.4 shows the level of pH noncompliance of surface water leaving the quarry since 1982. The percentage of noncompliances began to increase in 1984, and by 1987, the Y-12 Plant was in NPDES compliance only 62% of the time. The compliance percentage rose to over 80% in both 1988 and in 1990 but fell again to 62% in 1991.

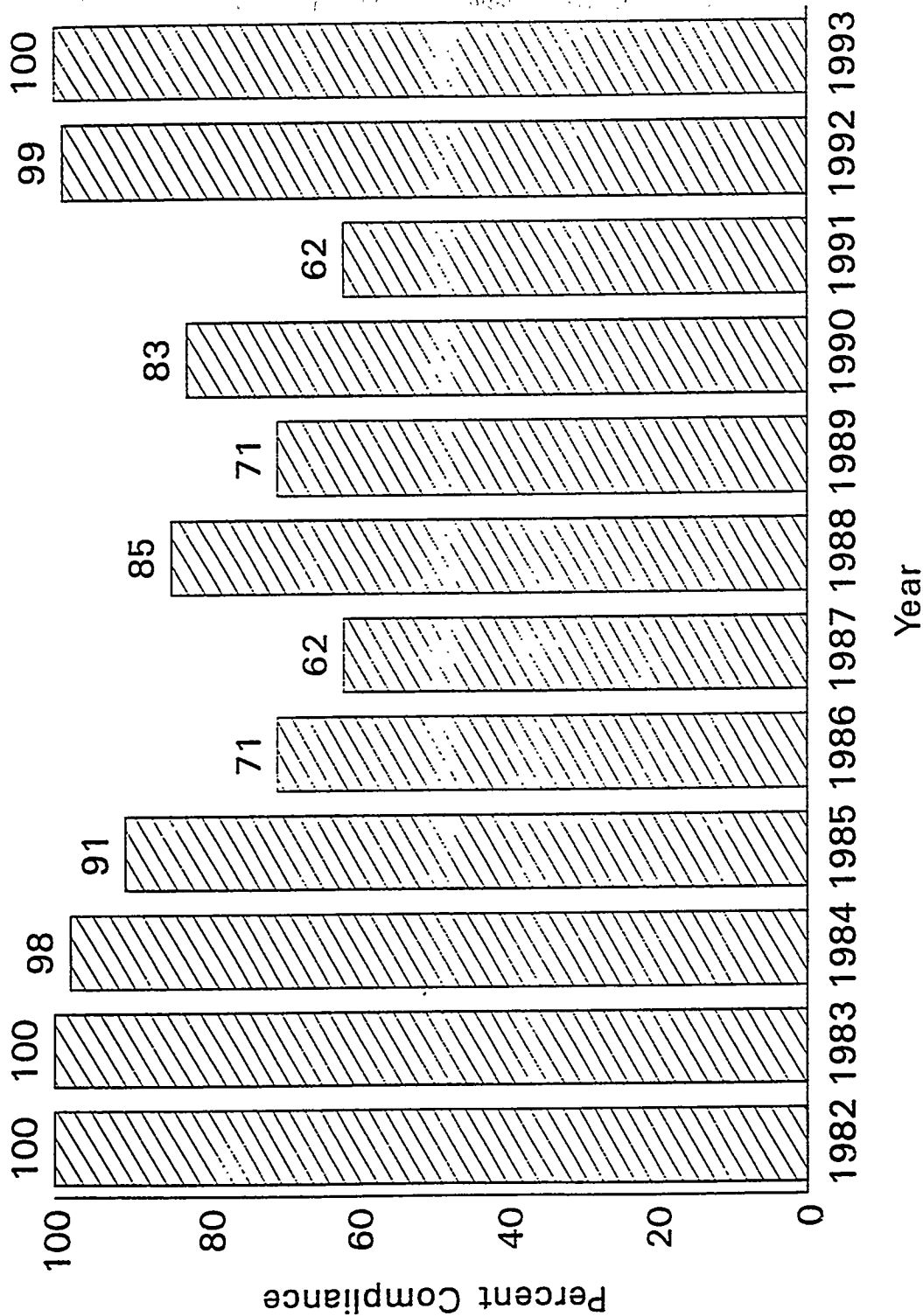


Fig. 2.4. Y-12 Plant NPDES pH compliance of surface water at Rogers Quarry, Outfall 302 from 1982 to 1993. Sources: 1993 data: K. Hanzelka, Health, Safety, Environment, and Accountability Division, Y-12 Plant, personal communication. 1989 through 1992 data: Kornegay et al. (1990a, 1991a, 1992a, 1993a). 1987 and 1988 data: Rogers et al. (1988a, 1989a). 1982 through 1986 data: Oakes et al. (1987).

In 1991, there were 19 pH related noncompliances at Rogers Quarry (Kornegay et al. 1993a). The elevated pH levels have been attributed to heavy algal growth in the quarry. The increased water clarity in the quarry resulting from the elimination of the Y-12 Plant steam plant discharges has allowed sunlight to penetrate deeper into the water column. The resulting increase in photosynthetic activity lowered the amounts of dissolved CO<sub>2</sub> and formed less carbonic acid, thus raising the pH. Although the elevated pH is a naturally occurring phenomenon, the Y-12 Plant was obligated to rectify the noncompliances. In March 1992, a subsurface discharge pipe was installed at the outlet of the quarry approximately 30 meters below the surface. This design released cooler water, enriched in CO<sub>2</sub>, which was sampled for pH compliance (B. Hummel, Site Shift Operations, Y-12 Plant, personal communication to R. P. Hoffmeister, Environmental Sciences Division, Oak Ridge National Laboratory). At the time of this report, the Y-12 Plant was running at 100% pH compliance.

#### 2.4.2 TEMPERATURE

Temperature data from 1991 to 1993 showed little variation between McCoy Branch upstream of Rogers Quarry and two reference locations in upper White Oak Creek and Scarboro Creek (Table 2.2 and Fig. 2.5). Until 1993, water temperatures in McCoy Branch below Rogers Quarry during the winter were similar to those above the quarry and in the reference streams. In 1993, however, the mean monthly temperatures in lower McCoy Branch were substantially cooler and it was not until August that the water temperature began to approximate those upstream and in Scarboro Creek. During this time, the mean monthly temperature in lower McCoy Branch was as much as 4.4°C cooler than in upper McCoy Branch. Mean monthly temperatures in the summer of 1991 in McCoy Branch below the quarry were higher than those in upper McCoy Branch and upper White Oak Creek by as much as 11.7°C. These findings are similar to those from 1990 (Ryon et al. 1992). Seasonal temperature differences may be attributed to thermal stratification and layering in Rogers Quarry. After March 1992, subsurface discharges from the quarry avoided extremely elevated summer temperatures. The lower temperatures in McCoy Branch below the quarry early in 1993 may be the result of very cold discharge water from the quarry during an unusually frigid winter.

#### 2.5 SEDIMENTS

In a study by Turner et al. (1986), coal ash sluice water discharged from the Y-12 Plant steam plant was reported to contain elevated amounts of Al, Ba, B, Ca, Fe, Mg, Na, As, Sr, K, total suspended solids, total phosphorous, sulfide, and sulfate when compared to background water concentrations (Appendix A, Tables A.3 to A.6, Ryon et al. 1992). Turner et al. (1986) also noted that the FCAP provided little or no treatment, did not remove suspended solids, and that the flow of McCoy Branch was so small it was incapable of providing appreciable dilution to the ash sluice water.

Ash deposits, which occurred when the creek overran its banks, vary greatly along the stream. In a preliminary study, Murphy (1988) found that immediately downstream of the ash pond dam, the deposits covered the floodplain through which McCoy Branch flows. Further downstream, the deposits were limited to no more than a couple of meters horizontally from the edge of the creek.

Table 2.2. Mean ( $\pm 1$  SD) monthly water temperature ( $^{\circ}\text{C}$ ) at two locations in McCoy Branch and in two reference streams (White Oak Creek and Scarboro Creek) from 1991 to 1993

Minimum and maximum values in parentheses

MONTH	1991			1992			1993		
	MCK 1.9 <sup>a</sup>	MCK 1.6 <sup>b</sup>	WCK 6.8 <sup>c</sup>	MCK 1.9 <sup>d</sup>	MCK 1.6 <sup>e</sup>	SCK 2.2	MCK 1.9	MCK 1.6	SCK 2.2
Jan.	11.2 $\pm$ 0.9 (8.8-12.6)	10.2 $\pm$ 0.5 (9.1-11.2)	10.3 $\pm$ 1.7 (1.1-17.9)	10.0 $\pm$ 1.5 (5.5-12.7)	9.7 $\pm$ 0.6 (7.5-11.1)	10.6 $\pm$ 1.2 (7.8-13.2)	10.7 $\pm$ 1.1 (6.7-12.9)	9.7 $\pm$ 0.3 (8.0-11.1)	11.1 $\pm$ 0.9 (8.0-13.5)
Feb.	10.8 $\pm$ 1.1 (7.4-13.2)	9.6 $\pm$ 0.6 (6.4-11.1)	10.0 $\pm$ 1.4 (6.8-12.8)	10.2 $\pm$ 1.7 (5.8-13.7)	9.5 $\pm$ 1.1 (6.4-11.8)	10.5 $\pm$ 1.2 (6.5-13.3)	9.4 $\pm$ 1.3 (5.6-13.2)	8.8 $\pm$ 0.4 (6.4-9.5)	10.0 $\pm$ 1.3 (6.3-13.3)
Mar.	12.1 $\pm$ 1.2 (9.6-15.9)	12.3 $\pm$ 1.8 (9.7-16.8)	11.5 $\pm$ 1.3 (8.9-15.1)	11.7 $\pm$ 1.4 (8.8-15.6)	9.5 $\pm$ 1.1 (8.7-14.1)	10.7 $\pm$ 0.8 (8.5-12.3)	10.7 $\pm$ 1.6 (5.6-15.1)	8.4 $\pm$ 0.3 (7.2-10.4)	11.0 $\pm$ 1.7 (5.6-15.6)
Apr.	13.4 $\pm$ 1.2 (10.8-16.8)	14.5 $\pm$ 1.2 (12.1-16.5)	13.3 $\pm$ 1.2 (10.2-16.7)	13.3 $\pm$ 2.1 (8.3-19.0)	9.7 $\pm$ 0.6 (8.6-13.0)	NA <sup>f</sup>	12.8 $\pm$ 1.4 (10.2-16.8)	8.8 $\pm$ 0.3 (8.3-11.8)	13.3 $\pm$ 1.5 (10.5-17.4)
May.	NA	19.7 $\pm$ 2.2 (15.8-24.2)	14.7 $\pm$ 1.1 (11.4-19.6)	14.1 $\pm$ 1.3 (11.4-17.2)	NA	NA	14.2 $\pm$ 1.1 (11.7-16.8)	9.8 $\pm$ 0.6 (8.9-14.9)	15.2 $\pm$ 1.4 (11.8-20.2)
Jun.	NA	25.3 $\pm$ 1.2 (22.5-28.1)	15.4 $\pm$ 0.8 (13.6-19.5)	15.2 $\pm$ 0.6 (13.1-16.8)	NA	NA	15.5 $\pm$ 0.8 (12.8-17.6)	11.3 $\pm$ 0.9 (9.7-15.9)	16.6 $\pm$ 1.2 (13.5-19.1)
July	NA	28.0 $\pm$ 0.7 (25.7-30.0)	16.3 $\pm$ 0.8 (15.1-19.7)	16.6 $\pm$ 0.6 (15.4-19.2)	NA	NA	16.9 $\pm$ 0.6 (15.7-21.7)	14.8 $\pm$ 1.2 (12.6-20.8)	18.3 $\pm$ 1.1 (15.7-21.1)
Aug.	16.9 $\pm$ 1.0 (15.0-21.1)	27.4 $\pm$ 1.2 (25.0-30.3)	17.0 $\pm$ 1.0 (14.7-19.4)	16.7 $\pm$ 0.7 (15.1-19.1)	18.1 $\pm$ 0.8 (16.2-20.2)	17.4 $\pm$ 1.0 (15.0-19.6)	17.2 $\pm$ 0.5 (15.9-18.6)	17.1 $\pm$ 2.3 (12.6-22.3)	18.2 $\pm$ 1.1 (15.4-21.0)
Sep.	16.5 $\pm$ 1.4 (12.7-20.1)	25.8 $\pm$ 2.0 (21.2-29.2)	16.5 $\pm$ 1.9 (11.4-20.4)	16.5 $\pm$ 0.8 (13.4-18.1)	19.2 $\pm$ 1.0 (17.5-21.6)	17.1 $\pm$ 1.1 (12.6-19.6)	16.2 $\pm$ 1.3 (11.9-19.5)	17.4 $\pm$ 2.2 (13.4-22.2)	17.0 $\pm$ 1.7 (11.8-22.3)
Oct.	14.3 $\pm$ 1.5 (11.1-18.0)	19.6 $\pm$ 1.7 (16.3-24.5)	13.7 $\pm$ 2.1 (9.3-18.8)	13.6 $\pm$ 1.0 (10.4-16.0)	16.5 $\pm$ 1.2 (13.7-19.8)	13.8 $\pm$ 1.5 (9.0-16.8)	13.1 $\pm$ 1.9 (8.1-16.9)	15.1 $\pm$ 1.6 (11.2-17.7)	13.8 $\pm$ 1.8 (9.5-18.0)
Nov.	11.2 $\pm$ 2.0 (7.8-15.5)	13.8 $\pm$ 1.6 (9.9-19.0)	10.3 $\pm$ 2.4 (5.3-15.4)	11.4 $\pm$ 1.7 (7.8-15.5)	12.4 $\pm$ 0.8 (10.8-16.0)	11.7 $\pm$ 1.8 (8.0-16.8)	9.2 $\pm$ 2.3 (4.6-14.4)	11.1 $\pm$ 0.8 (9.7-14.8)	11.2 $\pm$ 1.7 (8.2-15.3)
Dec.	11.4 $\pm$ 1.1 (9.1-14.6)	11.6 $\pm$ 1.1 (9.4-21.6)	11.7 $\pm$ 1.0 (9.7-13.9)	9.7 $\pm$ 1.4 (6.7-12.6)	10.8 $\pm$ 0.8 (7.1-12.6)	9.8 $\pm$ 1.3 (6.3-13.1)	9.2 $\pm$ 1.9 (4.6-12.6)	9.9 $\pm$ 0.3 (8.6-10.9)	10.2 $\pm$ 1.4 (6.4-13.3)

Note: Data were obtained with a Ryan TempMentor digital temperature recorder with values recorded hourly. MCK = McCoy Branch kilometer; WCK = White Oak Creek kilometer; SCK = Scarboro Creek kilometer.

<sup>a</sup>Disregarded values occurred in March and December. Abnormally low minimum and high maximum temperature values may have resulted from exposure of the temperature monitor to air during ice formation or flooding. In some cases, the first value recorded after deployment is invalid. All these values have been disregarded when calculating means.

<sup>b</sup>Disregarded values occurred in April, August, and December.

<sup>c</sup>Days of record for incomplete months were: April (29 d), August (29 d), and December (12 d).

<sup>d</sup>Disregarded values occurred in December.

<sup>e</sup>Disregarded values occurred in August.

<sup>f</sup>Data not available.

Sources: WCK 6.8 data: Hinzman and Loar 1992.

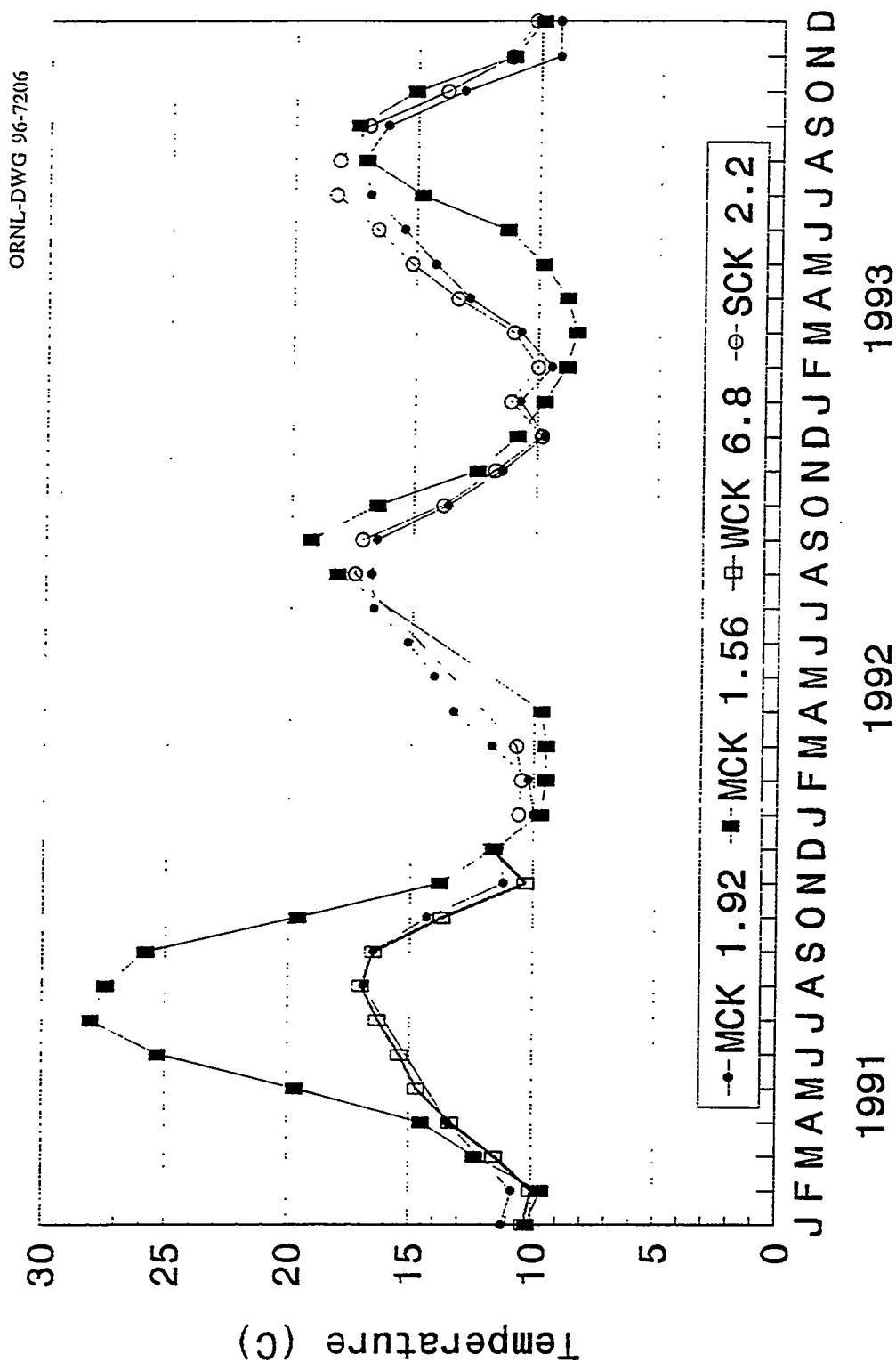


Fig 2.5. Mean monthly water temperatures (°C) in McCoy Branch above Rogers Quarry at McCoy Branch kilometer (MCK) 1.9, below Rogers Quarry (MCK 1.6), and in two reference streams, upper White Oak Creek at White Oak Creek kilometer (WCK) 6.8, and Scarboro Creek (SCK) 2.2 from 1991 to 1993. Data were obtained from Ryan TempMentor digital temperature recorders.



Results from the Phase I sampling of soils within the McCoy Branch floodplain for the RI project indicated that elevated levels of Ba, Ca, Cd, Fe, K, Mg, Mn, Na, and Ni exist in the sediments compared to those found in samples taken in 1974-75 (Appendix A, Tables A.15 to A.17, Ryon et al. 1992). Levels of radioactivity for each sampling location are given in Appendix A, Table A.18 (Ryon et al. 1992).

Table 2.3 shows the sediment sampling results just downstream of the FACP of Phase II of the RI. Reference samples were also taken at stream locations on Walker Branch and White Oak Creek (CDM 1994). The numerical results represent a composite of three aliquots taken at that specific site. High levels of those metals found in Phase I were also present in Phase II, except for Ca, Cd, and Mg. Aluminum, arsenic, and potassium were measured in high levels in Phase II. These trace metals are common constituents of coal ash. All of these metals in Phase II were detected at much higher concentrations in McCoy Branch than in the reference streams. Alpha and Beta levels of radioactivity were estimated values and were substantially higher than those found in Phase I. Levels of  $\text{Th}^{228}$  and  $\text{Th}^{232}$  were also much higher in McCoy Branch than in either of the reference streams. This data suggested that previous fly ash sluicing led to high contaminant concentrations in the sediments of upper McCoy Branch.

## 2.6 GROUNDWATER

Coal ashes typically exhibit leachability of ash constituents. Groundwater may be a receptor of contaminants from the coal ash disposal system and may act as a pathway for contamination migration near the Y-12 Plant (Murphy 1988). Filtered groundwater samples taken in 1990 near Upper McCoy Branch showed that concentration levels of 12 metals exceeded those at background sites north of the FCAP (Table 3.16, CDM 1994). These metals include Al, Ba, Ca, Co, Cu, Fe, Mg, Mn, K, Se, Na, and Zn. Although these metals existed in elevated amounts, most did not exceed TDEC maximum contaminant levels (CDM 1994). Aluminum, iron, and manganese were among those that did. For 8 of the 12 metals, the highest concentrations were located at the same sampling site (GW-672) approximately 600m downstream from the FCAP dam. Note that the concentrations generally did not decrease downstream as they did in the surface water samples (Table 3.16, CDM 1994). The complex geologic structure underlying McCoy Branch yielded different leaching capabilities for different elements. High values for nitrate, sulfate, and total organic carbon were present in the groundwater but were not detected in the background samples. Total uranium, gross alpha, and gross beta were also measured above background samples.

Phase II filtered groundwater data showed elevated levels of the same metals as in Phase I except for cobalt (Table 3.17, CDM 1994). All observed levels were above reference levels detected north of the FCAP. Barium, calcium, and sodium were detected at all sample locations but had much higher concentrations downstream of the FCAP. Manganese was detected at all locations except for one reference site, while potassium and zinc were detected at half of all sampling sites. Questions arose as to the appropriateness of using groundwater wells as background (Phase I) and reference (Phase II) sites due to their proximities to the FCAP (CDM 1994) and the Chestnut Ridge Security Pits. Also, the actual direction of groundwater flow along Chestnut Ridge is uncertain. Although higher metal concentrations were found in the groundwater, a stronger comparison showing the effects of past sluicing might be made if the background/reference points were at some other location.

Table 2.3. Levels of selected parameters in Phase II sediment sampling of upper McCoy Branch

Parameter <sup>a</sup>	Upper McCoy Branch SD-004-01 MCK 2.4 <sup>b</sup>	Reference streams			
		Walker Branch			White Oak Creek
		SD-005-01	SD-006-01	SD-007-01	
Aluminum	9,320.00 <sup>b</sup>	6,030.00	3,870.00	4,900.00	2,810.00
Arsenic	167.00 E <sup>c</sup>	7.60	4.40	13.40	3.50 E
Barium	309.00	218.00	118.00	68.00	87.80
Copper	19.10	14.80	2.00	2.70	ND <sup>d</sup>
Iron	21,900.00	11,600.00	19,400.00	20,900.00	20,300.00 E
Manganese	4,030.00	3,370.00	2,040.00	1,490.00	1,310.00 E
Nickel	30.70	11.50	5.90	9.20	ND
Potassium	1,750.00	785.00	ND	ND	ND
Sodium	385.00	ND	ND	ND	32.40
Alpha	18.00 E	NA <sup>e</sup>	NA	NA	NA
Beta	16.00 E	NA	NA	NA	NA
Thorium <sup>228</sup>	1.000	0.840	0.360	0.720	0.620
Thorium <sup>232</sup>	1.100	0.580	0.260	0.710	0.660

Note: MCK = McCoy Branch kilometer.

<sup>a</sup>Metal concentrations in mg/kg.

<sup>b</sup>Approximate stream location in kilometers; radioactivity and Thorium measurements in picocuries per gram.

<sup>c</sup>Estimated value.

<sup>d</sup>Not detected.

<sup>e</sup>Not analyzed.

Source: CDM, 1994

Gross alpha and gross beta activity were highest above the FCAP. Thorium<sup>232</sup> and Th<sup>232</sup> were found at only one reference site. Uranium<sup>234</sup> and U<sup>238</sup> had levels higher in Upper McCoy Branch than at the reference locations. The five organic compounds were observed at the highest levels in the reference wells above the FCAP.

Groundwater results from Rogers Quarry from 1988 to 1993 are given in Table 2.4. The average arsenic concentration above detection limit was twice as high as the reference value in 1988 and was detected in all eight samples. In 1992, arsenic was found in only one of the 16 samples and its concentration (0.061 mg/L) was slightly above the reference value (0.05 mg/L). In 1993, the mean value of those values above the detection limit for aluminum was five times higher than the reference value of 0.200 mg/L. Aluminum was detected in one third of the samples. Iron and manganese were detected in nearly every sample since 1988. The mean of iron levels above the detection limit declined yearly from 1.3 mg/L in 1988 to 0.46 mg/L in 1992, before rising in 1993 to 0.66 mg/L. All of these means were well above the reference value of 0.3 mg/L. The average of the manganese values above the detection limit fluctuated from year to year. The highest mean value was measured in 1989 at 0.15 mg/L while the lowest was 0.055 mg/L in 1991. As with iron, all mean values were above the reference value (0.05 mg/L). Sample analysis showed that nitrate-nitrogen concentrations have been decreasing since 1989. It has been detected in 25% of the samples ranging from average values above the detection limit of 37 mg/L in 1989 to less than half that in 1990 (16 mg/L). In 1992, levels fell to 5.5 mg/L and in 1993 to 4.0 mg/L.

Since 1989, turbidity in Rogers Quarry groundwater has been monitored. The results show that until 1992, the mean value above the detection limit had also been decreasing sharply. In 1992, the mean turbidity rose to 7.7 NTU and in 1993, it increased to 9.6 NTU. Note that in 1992 the regulatory reference value changed from 5 NTU to 1 NTU. All mean values were above the reference value.

## 2.7 DISCUSSION AND CONCLUSIONS

These data show that although some metal concentrations, levels of radioactivity, and organic constituents are still above reference values, the average values of these contaminants in surface water and groundwater samples in McCoy Branch watershed have generally been decreasing yearly since 1988. During this period, nitrate-nitrogen values have actually fallen below reference standards. The groundwater near Rogers Quarry has become less turbid as well. These improving conditions coincide with the elimination of Y-12 Plant discharge of fly ash into upper McCoy Branch.

Phase I and II surface water sampling yielded greater levels of typical coal ash metal contaminants, organics, and radioactivity near the point of discharge in the upstream portion of McCoy Branch, with the highest values at or near the FCAP. The leaching of these contaminants from the base of the FCAP produced the observed downstream gradient. The elevated contaminant levels in the sediments of McCoy Branch floodplain are believed to primarily exist due to the spilling over of the FCAP and subsequent rising water levels in the creek during heavy rains. Like the surface water samples, the groundwater samples exhibited higher levels of metals in McCoy Branch compared to reference samples from an unaffected stream. However, unlike the surface water samples, the downstream decline in concentrations of metals levels was not observed in groundwater samples. Radioactivity and the quantity of organic compounds detected in the groundwater were greatest near the FCAP.

Table 2.4. Levels of selected parameters in Rogers Quarry groundwater samples taken from 1988 to 1993

Parameter	1988	1989	1990	1991	1992	1993
<b>Arsenic (mg/L)</b>						
Number det./Number samp.	8/8	NA <sup>a</sup>	NA	NA	1/16	NA
average <sup>b</sup>	0.097	NA	NA	NA	0.061	NA
ref. value	0.05	NA	NA	NA	0.05	NA
<b>Aluminum (mg/L)</b>						
Number det./number samp.	NA	NA	NA	NA	10/16	5/16
average	NA	NA	NA	NA	0.26	0.96
ref. value	NA	NA	NA	NA	0.200	0.200
<b>Iron (mg/L)</b>						
Number det./number samp.	12/14	4/4	7/8	8/8	16/16	16/16
average	1.3	1.3	0.51	0.45	0.46	0.66
ref. value	0.3	0.3	0.30	0.3	0.3	0.300
<b>Manganese (mg/L)</b>						
Number det./number samp.	13/14	4/4	6/8	8/8	15/16	15/16
average	0.056	0.15	0.097	0.055	0.11	0.076
ref. value	0.05	0.05	0.050	0.05	0.05	0.050
<b>Nitrate-nitrogen (mg/L)</b>						
Number det./number samp.	NA	1/4	2/8	NA	4/16	4/16
average	NA	37	16	NA	5.5	4.0
ref. value	NA	10	10	NA	10	10
<b>Turbidity (NTU)</b>						
Number det./number samp.	NA	NA/4	NA/8	NA/8	16/16	16/16
average	NA	13	12	6.5	7.7	9.6
ref. value <sup>c</sup>	NA	5	5	5	1	1

<sup>a</sup>Not analyzed for.<sup>b</sup>Average of values above detection limit.<sup>c</sup>National Primary Drinking Water Regulatory Reference Value.

Sources: 1993 data: K. Hanzelka, Health, Safety, Environment, and Accountability Division, Y-12 Plant, personal communication. 1989 through 1992 data: Kornegay, et al., 1990b, 1991b, 1992b, 1993b. 1988 data: Rogers et al., 1989b.

### 3. AMBIENT TOXICITY MONITORING

(A. J. Stewart, L. F. Wicker, and J. M. Bricker)

#### 3.1 INTRODUCTION

Various tests were used to assess the toxicity of water samples from three sites in McCoy Branch and of ash samples from the FCAP. In one test both water and sediment toxicity were assessed. Tests used to routinely assess water quality included 7-d laboratory tests with fathead minnow (*Pimephales promelas*) larvae and with *Ceriodaphnia dubia* (a freshwater microcrustacean). These two tests commonly are used to estimate effluent and ambient water toxicity. On several occasions, a 4-d laboratory feeding-rate test with a pleurocerid (gill-breathing) snail (*Elimia clavaeformis*) was also used to assess water toxicity. During two of the feeding-rate tests, sediment collected from McCoy Branch kilometer (MCK) 1.6 and MCK 1.9 was tested with the snails, as well. Two types of 14-d tests with earthworms (*Eisenia foetida*) were used to assess the biological quality of ash from the FCAP. One of these was conducted *in situ*; the other involved quantifying responses of the worms to FCAP ash samples under laboratory conditions. The results of various other biological or toxicological tests of water in McCoy Branch (e.g., *in situ* snail-release study at MCK 1.9 and MCK 1.6; a life-cycle test with *C. dubia* using water from MCK 1.9 and 7-d fathead minnow larvae and *C. dubia* toxicity tests for January 1989 to April 19, 1990, at MCK 1.9) were given in another report (Ryon et al. 1992; Appendix B).

Information about the locations of the sites tested, the month and date when the tests were initiated, and the types of tests that were used to assess biological conditions, is summarized in Table 3.1. In each test used to assess water quality, measurements were also made of pH, conductivity, alkalinity, and hardness. In tests used to assess ash samples, measurements were made of ash pH and organic content (by loss of mass with ignition). The water quality data obtained during the tests above are also summarized in this chapter, as is information about the FCAP ash samples that were tested with *E. foetida*.

#### 3.2 MATERIALS AND METHODS

##### 3.2.1 Fathead Minnow Larvae Survival and Growth Tests

Seven-day static-renewal tests with fathead minnow larvae are commonly used to estimate the toxicity of effluents and receiving waters. This test has been standardized by the Environmental Protection Agency (EPA) (method 1000.0; Weber et al. 1989); it involves rearing newly hatched replicate groups of larval minnows in beakers containing control water and beakers containing water or effluent samples, under controlled temperature, lighting and food conditions in the laboratory. Water samples for the tests are collected and handled using chain-of-custody procedures in accordance with Standard Operating Procedures for the Environmental Science Division's Toxicology Laboratory Quality Assurance Program (Kszos et al. 1989). Generally, ten larvae are used in each replicate beaker, with four replicates per test. The minnows are inspected daily for mortality; dead fish, if any, are removed, and the old test solution is then siphoned off and replaced with fresh test solution. At the end of a

Table 3.1. Assessment of water quality from selected sites on McCoy Branch

Site tested	Date when test was initiated													
	July 30, 1990	Apr. 11, 1991	July 11, 1991	Oct. 24, 1991	Jan. 31, 1992	Apr. 2, 1992	July 9, 1992	Sept. 17, 1992	Jan. 14, 1993	Mar. 25, 1993	Apr. 19, 1993	July 29, 1993	Sept. 16, 1993	Feb. 1994
MCK 1.6	C, F	C	C, F	C, F	C, F	C, F	C, F	C	C, F, SF <sup>a</sup>	C, F	SF	C, F, SF <sup>b</sup>	C, F	—
MCK 1.9	C, F	C	C, F	C, F	C, F	C, F	C, F	C	C, F, SF <sup>a</sup>	C, F	SF	C, F, SF <sup>b</sup>	C, F	—
MCK 2.3	—	—	—	—	—	—	—	—	—	—	—	—	—	SR <sup>c</sup>
MCK 2.5	—	—	—	—	—	—	—	—	—	C, F	—	—	—	—
MCK 2.7	—	C	—	—	—	—	—	—	—	—	—	—	—	—
FCAP	—	—	—	—	—	—	EF <sup>e</sup>	—	—	—	—	BL <sup>d</sup>	—	—

Note: Codes for test types are as follows: C = *C. dubia* 7-d survival and reproduction test; F = fathead minnow larvae 7-d survival and growth test; SF = snail 4-d feeding-rate test; SR = *in situ* snail tag-release-recapture test; BF = *in situ* 14-d *Eisenia foetida* survival and growth test; EL = laboratory 14-d *E. foetida* survival and growth test. "—" = not tested. MCK = McCoy Branch kilometer.

<sup>a</sup>Snail feeding-rate test started on January 18, 1993.

<sup>b</sup>Snail feeding-rate test started on July 27, 1993.

<sup>c</sup>*In situ* test with earthworms started on July 1, 1993.

<sup>d</sup>Laboratory test with earthworms started on August 1, 1993.

<sup>e</sup>Study in progress at the time of report preparation.

7-d test period, survival and growth of the minnow larvae in the effluent or ambient water samples are compared to the survival and growth of the minnow larvae in the control water. Statistically significant reductions in minnow survival or growth in the effluent or ambient water samples are used as evidence for acute and chronic toxicity, respectively. In some ambient applications, low mean survival of fathead minnow larvae test can occur due to the presence of facultative pathogenic microorganisms; in these instances, the test may yield "false positives" for acute toxicity (Kszos and Stewart 1992). In most effluent applications, the fathead minnow test appears to perform reliably. Examples of the use of 7-d fathead minnow tests to assess ambient toxicity in streams can be found in Norberg-King and Mount (1986) and Stewart et al. (1990).

In each test period in which fathead minnow larvae were used to conduct water-quality assessments, various concentrations of water (e.g., 80%, 60%, 40%) were tested in addition to full-strength water from McCoy Branch. Dilutions, when used, were prepared using the same water as used in the control (i.e., diluted mineral water).

### 3.2.2 *Ceriodaphnia dubia* Survival and Reproduction Tests

Seven-day static-renewal tests with the freshwater microcrustacean *C. dubia* are also used to estimate toxicity of effluents and receiving waters. This test has been standardized for use in effluent testing by the EPA (method 1002.0; Weber et al. 1989). The test involves rearing neonate *C. dubia* (<24-h old) individually in small beakers containing either control water or effluent or ambient water under controlled temperature, lighting, and food conditions in a laboratory. These daphnids are typically parthenogenic (i.e., females can produce offspring in the absence of males) and achieve reproductive maturity in 2 to 4 days. In a typical 7-d test, *C. dubia* neonates used to start the test can produce three or more broods of new neonates. The contents of each beaker are inspected visually each day of the test; when new neonate *C. dubia* are present in a test beaker, they are counted and discarded; only adult daphnids are transferred individually to fresh test solution. *C. dubia* survival and reproduction in control water in a concurrently conducted test is compared to survival and reproduction of the daphnids in the test solutions to provide estimates of acute and chronic toxicity respectively.

*C. dubia* in general are more sensitive than fathead minnow larvae to many toxicants. Inherent differences in procedures for the fathead minnow larvae and *C. dubia* test methods exist (e.g., groups of ten fish per replicate, versus single daphnids per replicate; measurements of weight change for the fish, as estimates of growth, versus numbers of neonates produced per daphnid in the *C. dubia* test). In addition, the difference in number of replicates typically used in each test method (four for the fish, ten for the daphnids) also tends to increase the resolution of the *C. dubia* test, relative to the fathead minnow test. Reproduction of *C. dubia* and other daphnids is closely linked to food quality and quantity. Thus, mean reproduction of *C. dubia* in ambient water samples that contain bacteria, algae or detritus can exceed mean reproduction of *C. dubia* in the controls (cf. Kszos and Stewart 1992). Additionally, water quality factors such as conductivity, hardness, alkalinity, and pH also can affect survival or reproduction of *C. dubia*, or virtually any other type of bioassay. Thus, in ambient applications, test-to-test variation in *C. dubia* mean reproduction is often nearly as great as site-to-site variation in *C. dubia* reproduction (Stewart and Loar 1994). These findings show that the results of *C. dubia* "toxicity" tests must be considered carefully if they are to provide meaningful information about water quality in ambient applications.

Examples of the use of *C. dubia* tests to assess ambient water quality are provided by Burton et al. (1987), Nimmo et al. (1990), Stewart et al. (1990), Norberg-King et al. (1991), Kszos et al. (1992), and Stewart et al. (1994).

In each test period in which *C. dubia* were used to conduct water-quality assessments, various concentrations of water (e.g., 80%, 60%, 40%) were tested in addition to full-strength water. Dilutions, when used, were prepared using the same water as was used in the control (i.e., diluted mineral water).

### 3.2.3 Snail Feeding-Rate Tests

Pleurocerid (gill-breathing) snails are key grazers and influential competitors in many streams (Hawkins and Furnish 1987, Burris et al. 1990, Hill 1992, Stewart et al. 1993). These animals can occur in high densities, sometimes comprising more than 90% of the macroinvertebrate biomass (Hawkins and Furnish 1987, Elwood et al. 1981). Snails respond behaviorally to differences in water quality. When toxicants are present, for example, they may cease feeding, slough their tentacles or retract their foot, or actively attempt to avoid the conditions by moving downstream or crawling out of the water (Burris et al. 1990, Harry and Aldrich 1963, Arthur and Leonard 1970, Winger et al. 1984, Burris 1987). These findings suggest that quantifiable changes in snail activities could be used to describe water quality.

Laboratory tests that measured the feeding rates *Elimia clavaeformis*, a pleurocerid snail that is abundant in many streams on the Oak Ridge Reservation, were conducted to provide information about water quality in McCoy Branch both upstream and downstream of Rogers Quarry.

*E. clavaeformis* are common and abundant in most streams within the White Oak Creek (WOC) drainage basin, and are present in several other streams that drain the south slope of Chestnut Ridge (e.g., Walker Branch, Scarboro Creek, Ish Creek). Although *Elimia* do not presently occur in McCoy Branch, snail distribution patterns on the Oak Ridge Reservation suggest that these snails may have inhabited the stream in the past and, with continued improvements in water quality, may eventually recolonize the stream. Specimens used for the laboratory tests reported here were collected from minimally disturbed areas in upper WOC [near WOC kilometer (WCK) 6.8]. A 20% concentration of degassed mineral water (DMW) was used as a negative control. Grab samples of water were collected daily from McCoy Branch for use in the tests.

In the laboratory, the snails were held in 12-L aquaria supplied with a constant flow of dechlorinated tap water. They were fed lettuce *ad libitum* and acclimated stepwise from the temperature at collection to the testing temperature (25°C). The tests were conducted under controlled conditions (25°C and a 16:8 L:D photoperiod) using a static-renewal procedure similar to the fathead minnow larvae test procedure. Twelve snails, selected randomly, were placed in a 600-mL beaker and exposed to 250 mL of treatment water; four replicate beakers were used to assess each treatment. Three green leaf lettuce disks (total area of 3 cm<sup>2</sup>) were added to each beaker each day for food; the disks were weighted with two staples so that they would not float. Initial and final wet weights of the disks were recorded each day. The open top of each beaker was covered with mesh to prevent the snails from escaping.

Snail feeding-rate tests were conducted on three dates (Table 3.1). The tests differed slightly with respect to objectives and methodology. Relevant differences among the tests are described as follows:



- Test 1. Water was collected from MCK 1.9 (upstream of Rogers Quarry at the weir) and MCK 1.6 (downstream of Bethel Valley Road). Snails were exposed to full-strength water and water diluted by 50% with DMW. The tests were conducted at 25°C.
- Test 2. Water was collected from MCK 1.9 and MCK 1.6. Sediment was collected from MCK 2.5. Snails were exposed to full-strength water, and full-strength water plus sediment (1 g sediment/L) from each site. The tests were conducted at 25°C.
- Test 3. Water and associated sediment was collected from MCK 1.9 and MCK 1.6. Snails were exposed to full-strength water with ~1.5 g of sediment from the site. Each treatment was conducted at 15°C and 25°C.

In each test, feeding rate (expressed as mean number of grams of wet weight of lettuce eaten within each beaker per day) of the snails was used as the response variable. The data were analyzed using PROC GLM (SAS 1985). Whenever possible, the tests were conducted concurrently with toxicity tests performed with fathead minnow larvae (*P. promelas*) and *C. dubia* (cf. Table 3.1). The snail-feeding assay described above is not yet an EPA-approved procedure, but has been used successfully to assess water quality at the inlet and outfall of Lake Reality in upper East Fork Poplar Creek (Hinzman 1993) and to evaluate stormwater quality entering White Oak Creek (Hinzman et al. 1994).

### 3.2.4 Snail *In Situ* Tag-Release-Recapture Test

This study was designed to determine whether water, sediment and food quality conditions in McCoy Branch have improved enough to allow recolonization of the stream by pleurocerid snails (Bricker and Stewart 1994). The study involved collecting 700 *E. clavaeformis* and 700 *Pleurocera uncialis* from Scarboro Creek downstream of Bethel Valley Road, individually tagging and weighing each animal, then releasing all of them together at one site in McCoy Branch (MCK 2.4) on February 15, 1994. After the snails had been released, they were periodically monitored; individuals were recaptured and reweighed at 3 and 8 weeks after the initial release to provide estimates of growth (cf. Hill 1992, Hill et al. 1995). Recaptured individuals at both time points were also analyzed for ammonium release rates using a spectrophotometric method described by Verdouw et al. (1978), and individuals recovered after 8 weeks were analyzed for respiration rates by measuring the change in dissolved oxygen content using a micro-Winkler technique (Fox and Wingfield 1938). Snails recaptured after 3 weeks were re-released at MCK 2.4. Measurements were made of respiration and ammonium release rates of *Elimia* and *Pleurocera* in Scarboro Creek and Northwest Tributary, a tributary of White Oak Creek, to permit comparison of the physiological status of the two species in McCoy Branch with physiologically stable populations.

At the start of the experiment, additional (non-tagged) snails of both species were also placed in cages at various locations in both McCoy Branch and Scarboro Creek for 1 week, using methods similar to those described by Hinzman et al. (1994). The caged snails were exposed to stream water, but did not have direct access to sediment or periphyton. After 7 days, the caged snails in both streams were monitored for survival and analyzed for ammonium release.

### 3.2.5 Earthworm Tests

Earthworm tests are used for solid waste and soil toxicity assessments. These soil-dwelling invertebrates are ecologically significant because they modify texture and fertility of soil and are important as food to many small mammals and birds. Earthworms are exposed to contaminants in soils both externally (through direct contact with soil), and internally (through ingestion of soil particles). These animals also grow and reach reproductive age rapidly. Thus, they are potentially excellent indicators of a soil's environmental condition and may be useful in ecological risk assessments. An EPA-approved method for testing with earthworms is described by Greene et al. (1989). This method allows assessment of soils by quantifying effects of soils on earthworm survival and growth.

Two types of earthworm tests were used to assess ash in the FCAP. One, conducted in July 1992 (Table 3.1), involved *in situ* testing at six locations in the FCAP. The other, conducted in the laboratory during August 1993, was used to assess ash samples collected from three locations in the FCAP. The procedures for these two tests are described in the following section.

#### 3.2.5.1 *In situ* earthworm test

Six sites, running along a north-south transect, were selected for testing. It was hypothesized that there might be a north-south gradient in ash toxicity caused by differences in the weathering duration of the ash, with "newer" ash being more likely near the north end of the FCAP, and "older" ash predominating at the south end of the FCAP. The *in situ* tests were considered to be exploratory in nature; an experimental design that used randomly selected sites (and a broader range in reference sites) might have been more appropriate for assessing spatial heterogeneity in toxicity of FCAP ash. The locations of the six sites used in the *in situ* tests began with site 1, approximately 25 m north of the earthen dam. Sites 2 through 6 were separated by 23 m, 61 m, 35 m, 38 m, and 33 m, respectively, in a transect to the south of site 1. At each site, four treatments were evaluated: (1) non-altered site ash, (2) site ash augmented with fermented alfalfa (used as *Eisenia* food), (3) pH-adjusted peat moss, and (4) pH-adjusted peat moss augmented with fermented alfalfa. Each replicate consisted of a 0.5-gallon plastic bucket filled with test medium. The buckets were partially buried in the FCAP ash, deep enough so that their tops were nearly level with the ash surface. Six earthworms, pre-weighed as a batch from laboratory cultures were placed on top of the substrate in each bucket. The tops of the buckets were then covered with mesh to prevent the worms from escaping. Buckets containing FCAP ash were filled with ash from the hole dug to bury the buckets. The peat moss was obtained as a commercial product. Enough  $\text{CaCO}_3$  was added to the peat moss (about 1%, on a dry mass basis) before the test to neutralize the peat moss's acidity. The peat moss also was saturated with distilled water prior to starting the test. Because rain events occurred throughout the 14-d testing period, no water was added to any of the treatments during the test. The fermented alfalfa used as a food supplement in the food-augmented combinations was prepared using the method described by Greene et al. (1989).

After 14 days, the 24 buckets were removed from the FCAP and taken to the laboratory. The ash in each bucket was then hand sorted to harvest the surviving earthworms, and analyzed for pH, water content (by drying at 100°C), and organic matter content (by ashing at 500°C). Surviving earthworms were purged of their gut contents before being weighed.

The number of worms surviving in each bucket (expressed as a proportion; arcsine square-root transformed) was used as the response variable in this test. The transformed survival data were then analyzed using a random effects ANOVA for a nested design, with substrate type (FCAP soil versus peat moss) and food-amendment (yes or no) serving as independent variables. The experimental design was not appropriate for distinguishing differences in worm responses among sites; it was suitable for detecting differences in worm survival in fly-ash soil versus pH-adjusted peat moss, and differences in worm survival in the two ash types attributable to the presence of food. Growth data could not be analyzed for reasons discussed in the results section.

### 3.2.5.2 Laboratory earthworm test

This test also used *Eisenia foetida*, and generally followed the EPA-approved test procedure described by Greene et al. (1989). The test was conducted during August 1–14, 1993 (Table 3.1). Three samples of ash were tested; the samples were from FCAP-1, FCAP-2, and the sluice site. A reference site (SO 12-1; Fig 3.15 in CDM 1994) and an artificial soil (peat moss, sand, and clay; Greene et al. 1989) were tested concurrently, using the same methods. An additional treatment was introduced to the test by comparing autoclaved versus non-autoclaved ash from each site. The autoclaved samples were sterilized at 1.6 kg/cm<sup>2</sup> for 15 min to rid the ash of possible microbial pathogens. The experimental design described above involved testing ten types of materials (autoclaved versus non-autoclaved samples of artificial soil, a reference site soil, FCAP-1, FCAP-2, and the sluice ash).

After the samples had been collected and autoclaved, distilled water was added to each soil (or ash) type to achieve approximately a 50% moisture fraction (the consistency of a thoroughly wet, but not muddy soil). Each ash or soil type was tested by adding 400 g of hydrated soil (or ash) to each of four 1-pint canning jars. Ten earthworms (pre-weighed as a group) were then placed on top of the material in each jar. Lids (with a 1/16-inch hole to permit gas exchange) were screwed onto the jars, and each jar was placed in an environmental chamber. The temperature in the chamber was  $25 \pm 2^\circ\text{C}$ ; the light regime was 16 h light and 8 h dark. Fourteen days after the test had started, the jars were removed and the material in each jar was hand-sorted to remove all surviving worms. The worms in each replicate were rinsed with distilled water to remove adhering soil (or ash) particles before being weighed.

The number of worms surviving in each replicate was analyzed using a one-way ANOVA; the survivorship data were transformed (arcsine square root) prior to analysis. A multiple-comparison T-test (equivalent to Fischer's least significant difference test) was used to determine which differences, if any, were statistically significant (SAS 1985a).

Growth of the worms in each replicate was estimated by subtracting the mean final mass of the earthworms (grams mean individual fresh mass per jar) from the mean initial weight of the earthworms (grams mean individual fresh mass per replicate). One-way ANOVA was then used to determine the influence of autoclaving and soil (or ash) source on earthworm growth.

### 3.2.6 Soil (Ash) Invertebrate Survey

A preliminary survey of invertebrates living in the ash was conducted in July 1993. The purpose of this survey was to determine the abundance of earthworms present at the site prior

to performing an earthworm bioaccumulation study. Five samples, each consisting of 1 ft<sup>3</sup> of soil, were hand-sorted using a 2.36-mm sieve. The samples were excavated approximately 5 m apart along a transect with FCAP-1. The surface ash was primarily barren except for a few larval and adult arthropods. Specimens found in the samples included

- Sample 1: 0 macroinvertebrates
- Sample 2: 2 small beetle larvae (Coleoptera)
- Sample 3: 1 millipede (Diplopoda)
- Sample 4: 1 millipede
- Sample 5: 3 millipedes

Two additional samples (each 1 ft<sup>3</sup> soil) were collected from the sluice area and hand-sorted. Similar organisms, at comparable abundances, were found in these samples: sample number 1, 2 millipedes and 1 beetle larva; sample number 2, 3 millipedes. No earthworms were found in any of the samples of ash from the FCAP or the sluice sites. The specimens collected from the ash were not sufficiently numerous or massive for contaminant analysis. The apparent absence of earthworms in the ash suggests that this may not be a significant pathway for current contaminant exposure for birds (i.e., woodcocks, robins) or other vermivores.

### 3.2.7 Analyses of Physicochemical Water Quality Factors

Water samples for use in the fathead minnow tests, *C. dubia* tests, and snail-feeding tests described earlier (Sects. 3.2.1, 3.2.2, and 3.2.3) were collected as grab samples daily for 7 days in most of the test periods. The water temperature was measured at the time of sample collection, and portions of each sample were analyzed for pH, conductivity, alkalinity (EPA method 130.1), and hardness (EPA method 130.2) in the laboratory. The methods used for each of these analyses are described by Kszos et al. (1989). Because multiple samples were collected from each site during each test period, it was possible to analyze the results of the water quality measurements using two-way ANOVA, using test periods and sites as class variables. Response variables in this analysis included temperature, pH, conductivity, alkalinity, and hardness.

## 3.3 RESULTS

### 3.3.1 Fathead Minnow and *Ceriodaphnia* Tests

The results of the fathead minnow tests and the *C. dubia* tests are summarized in Table 3.2. In most cases tests with these two species were conducted concurrently, used the same water samples, and involved the same dilution series. In this table, test date refers to the date the test was initiated. In general, *C. dubia* has been shown to be more sensitive than fathead minnows to diverse contaminants (Constable and Orr 1994, Stewart et al. 1990). Thus, interpretation of the results of the ambient toxicity tests for the McCoy Branch sites in this section emphasizes the results the *C. dubia* tests, compared to the results of the fathead minnow tests.

During the 12 test periods, the distribution of survival values for *C. dubia* in full-strength water samples from the McCoy Branch sites was as follows: 100% survival in 17 of 26 cases, 90% in 5 of 26 cases, 70% in 3 of 26 cases, and 60% in one case (Table 3.2).

**Table 3.2. Results of fathead minnow and *Ceriodaphnia* tests of ambient water samples from various sites in McCoy Branch**

Test date	Water source	Conc. (%)	<i>Pimephales promelas</i>		<i>Ceriodaphnia dubia</i>	
			Survival (%) (mean $\pm$ SD)	Growth (mg/fish) (mean $\pm$ SD)	Survival (%)	Reproduction (mean $\pm$ SD)
July 30, 1990	Control	100	100.0 $\pm$ 0.0	0.53 $\pm$ 0.10	90	24.0 $\pm$ 3.4
	MCK 1.6	100	100.0 $\pm$ 0.0	0.64 $\pm$ 0.04	90	18.3 $\pm$ 10.8
		80	97.5 $\pm$ 5.0	0.69 $\pm$ 0.11	—	—
	MCK 1.9	100	100.0 $\pm$ 0.0	0.66 $\pm$ 0.09	90	11.1 $\pm$ 8.1
		80	—	—	100	11.5 $\pm$ 9.7
		60	—	—	100	21.0 $\pm$ 9.0
		40	—	—	70	15.6 $\pm$ 8.6
Apr. 11, 1991	Control	100	—	—	90	30.1 $\pm$ 7.7
	MCK 1.6	100	—	—	100	31.2 $\pm$ 6.4
		80	—	—	100	33.0 $\pm$ 6.2
		60	—	—	100	29.1 $\pm$ 6.4
		40	—	—	100	29.2 $\pm$ 5.9
	MCK 1.9	100	—	—	100	21.8 $\pm$ 2.8
		80	—	—	100	19.5 $\pm$ 3.5
		60	—	—	100	21.1 $\pm$ 2.1
		40	—	—	100	20.3 $\pm$ 0.8
	MCK 2.7	100	—	—	100	22.3 $\pm$ 3.5
		80	—	—	100	25.3 $\pm$ 4.7
		60	—	—	100	26.6 $\pm$ 4.6
		40	—	—	90	28.8 $\pm$ 6.0
July 11, 1991	Control	100	100.0 $\pm$ 0.0	0.73 $\pm$ 0.06	100	19.1 $\pm$ 6.0
	MCK 1.6	100	95.0 $\pm$ 5.8	0.72 $\pm$ 0.12	100	23.8 $\pm$ 3.9
		80	97.5 $\pm$ 5.0	0.76 $\pm$ 0.05	100	21.7 $\pm$ 6.2
		60	82.5 $\pm$ 22.2	0.70 $\pm$ 0.06	100	23.2 $\pm$ 6.4
		40	97.5 $\pm$ 5.0	0.68 $\pm$ 0.11	100	24.6 $\pm$ 5.3

Table 3.2 (continued)

Test date	Water source	Conc. (%)	<i>Pimephales promelas</i>		<i>Ceriodaphnia dubia</i>	
			Survival (%) (mean $\pm$ SD)	Growth (mg/fish) (mean $\pm$ SD)	Survival (%)	Reproduction (mean $\pm$ SD)
Oct. 24, 1991	MCK 1.9	100	95.0 $\pm$ 5.8	0.72 $\pm$ 0.12	100	19.7 $\pm$ 5.3
		80	97.5 $\pm$ 5.0	0.76 $\pm$ 0.05	100	23.6 $\pm$ 2.3
		60	97.5 $\pm$ 5.0	0.67 $\pm$ 0.11	70	20.0 $\pm$ 6.1
		40	100.0 $\pm$ 0.0	0.68 $\pm$ 0.07	90	19.3 $\pm$ 6.8
	Control	100	95.0 $\pm$ 5.8	0.37 $\pm$ 0.01	100	26.8 $\pm$ 3.3
		MCK 1.6	90.0 $\pm$ 14.1	0.50 $\pm$ 0.04	100	30.3 $\pm$ 4.0
			70.0 $\pm$ 21.6	0.53 $\pm$ 0.04	100	20.5 $\pm$ 8.2
			57.5 $\pm$ 20.6	0.67 $\pm$ 0.13	100	25.5 $\pm$ 3.6
			80.0 $\pm$ 33.7	0.52 $\pm$ 0.06	100	27.4 $\pm$ 3.7
	MCK 1.9	100	87.5 $\pm$ 12.6	0.55 $\pm$ 0.05	100	27.2 $\pm$ 2.9
		80	90.0 $\pm$ 8.2	0.50 $\pm$ 0.03	100	26.5 $\pm$ 2.5
		60	95.0 $\pm$ 5.8	0.52 $\pm$ 0.07	90	25.9 $\pm$ 2.5
		40	90.0 $\pm$ 8.2	0.51 $\pm$ 0.03	100	26.0 $\pm$ 3.7
Jan. 31, 1992	Control	100	90.0 $\pm$ 14.1	0.51 $\pm$ 0.06	70	26.9 $\pm$ 7.2
		MCK 1.6	87.5 $\pm$ 12.6	0.46 $\pm$ 0.12	100	30.4 $\pm$ 4.0
			100.0 $\pm$ 0.0	0.50 $\pm$ 0.04	90	32.6 $\pm$ 4.3
			87.5 $\pm$ 12.6	0.46 $\pm$ 0.05	80	29.7 $\pm$ 4.4
		40	—	—	100	27.0 $\pm$ 4.7
	MCK 1.9	100	75.0 $\pm$ 43.6	0.49 $\pm$ 0.11	70	23.7 $\pm$ 4.2
		80	62.5 $\pm$ 45.0	0.42 $\pm$ 0.10	80	24.0 $\pm$ 7.5
		60	65.0 $\pm$ 43.6	0.47 $\pm$ 0.03	60	27.6 $\pm$ 2.9
		40	—	—	80	26.3 $\pm$ 5.7
	Control	100	92.5 $\pm$ 5.0	0.48 $\pm$ 0.08	90	24.9 $\pm$ 2.6
		MCK 1.6	47.5 $\pm$ 18.9	0.60 $\pm$ 0.03	70	24.0 $\pm$ 8.6
			42.5 $\pm$ 32.0	0.54 $\pm$ 0.19	60	21.8 $\pm$ 5.2
			67.5 $\pm$ 34.0	0.60 $\pm$ 0.05	60	25.8 $\pm$ 4.8
		40	—	—	70	27.4 $\pm$ 2.3

Table 3.2 (continued)

Test date	Water source	Conc. (%)	<i>Pinephales promelas</i>		<i>Ceriodaphnia dubia</i>	
			Survival (%) (mean $\pm$ SD)	Growth (mg/fish) (mean $\pm$ SD)	Survival (%)	Reproduction (mean $\pm$ SD)
	MCK 1.9	100	82.5 $\pm$ 23.6	0.59 $\pm$ 0.09	60	21.8 $\pm$ 4.6
		80	85.0 $\pm$ 5.8	0.61 $\pm$ 0.08	40	23.3 $\pm$ 4.9
		60	67.5 $\pm$ 27.5	0.69 $\pm$ 0.12	80	23.8 $\pm$ 9.9
		40	—	—	40	20.8 $\pm$ 5.3
July 9, 1992	Control	100	95.0 $\pm$ 10.0	0.39 $\pm$ 0.02	90	29.7 $\pm$ 1.7
		100	95.0 $\pm$ 10.0	0.43 $\pm$ 0.03	100	22.0 $\pm$ 4.9
		80	80.0 $\pm$ 16.3	0.44 $\pm$ 0.06	100	24.6 $\pm$ 6.7
		60	87.5 $\pm$ 5.0	0.46 $\pm$ 0.06	90	18.9 $\pm$ 9.3
	MCK 1.6	40	92.5 $\pm$ 9.6	0.40 $\pm$ 0.04	100	22.8 $\pm$ 5.8
		100	82.5 $\pm$ 17.1	0.45 $\pm$ 0.06	100	21.0 $\pm$ 3.1
		80	87.5 $\pm$ 9.6	0.48 $\pm$ 0.04	100	24.0 $\pm$ 2.5
		60	65.0 $\pm$ 17.3	0.50 $\pm$ 0.06	100	26.5 $\pm$ 2.8
	MCK 1.9	40	65.0 $\pm$ 25.1	0.53 $\pm$ 0.10	100	23.9 $\pm$ 3.2
		100	—	—	100	23.7 $\pm$ 5.4
		100	—	—	100	21.8 $\pm$ 4.8
		75	—	—	100	24.6 $\pm$ 5.2
Sept. 17, 1992	MCK 1.6	50	—	—	100	25.3 $\pm$ 5.9
		100	—	—	100	25.8 $\pm$ 4.1
		75	—	—	90	24.0 $\pm$ 4.2
	MCK 1.9	50	—	—	90	22.9 $\pm$ 4.1
		100	87.5 $\pm$ 5.0	0.42 $\pm$ 0.07	100	27.9 $\pm$ 3.1
		100	92.5 $\pm$ 9.6	0.45 $\pm$ 0.06	90	27.3 $\pm$ 7.6
Jan. 14, 1993	MCK 1.6	75	90.0 $\pm$ 8.2	0.43 $\pm$ 0.07	100	28.4 $\pm$ 7.0
		50	85.0 $\pm$ 10.0	0.49 $\pm$ 0.03	100	27.8 $\pm$ 3.8
	MCK 1.9	100	92.5 $\pm$ 9.6	0.40 $\pm$ 0.03	100	27.5 $\pm$ 3.6
		75	87.5 $\pm$ 9.6	0.46 $\pm$ 0.06	90	28.7 $\pm$ 5.5
		50	92.5 $\pm$ 9.6	0.49 $\pm$ 0.05	80	29.3 $\pm$ 6.8

Table 3.2 (continued)

Test date	Water source	Conc. (%)	<i>Pimephales promelas</i>		<i>Ceriodaphnia dubia</i>	
			Survival (%) (mean $\pm$ SD)	Growth (mg/fish) (mean $\pm$ SD)	Survival (%)	Reproduction (mean $\pm$ SD)
Mar. 25, 1993	Control	100	92.5 $\pm$ 9.6	0.42 $\pm$ 0.04	100	7.0 $\pm$ 4.7
	MCK 1.6	100	77.5 $\pm$ 18.9	0.57 $\pm$ 0.10	100	7.4 $\pm$ 4.3
	MCK 1.93	100	75.0 $\pm$ 26.5	0.53 $\pm$ 0.08	90	12.9 $\pm$ 2.9
	MCK 2.5	100	55.0 $\pm$ 36.9	0.62 $\pm$ 0.18	100	14.5 $\pm$ 6.0
July 29, 1993	Control	100	95.0 $\pm$ 5.8	0.33 $\pm$ 0.09	90	28.0 $\pm$ 5.2
	MCK 1.6	100	80.0 $\pm$ 21.6	0.40 $\pm$ 0.07	100	22.9 $\pm$ 4.9
		80	85.0 $\pm$ 10.0	0.38 $\pm$ 0.03	100	21.8 $\pm$ 7.4
		60	75.0 $\pm$ 20.8	0.39 $\pm$ 0.06	100	21.5 $\pm$ 8.0
	MCK 1.9	100	82.5 $\pm$ 5.0	0.35 $\pm$ 0.02	100	22.8 $\pm$ 5.3
		80	80.0 $\pm$ 8.2	0.35 $\pm$ 0.09	80	25.4 $\pm$ 4.2
		60	80.0 $\pm$ 11.5	0.36 $\pm$ 0.08	90	21.3 $\pm$ 8.5
	Control	100	97.5 $\pm$ 5.0	0.54 $\pm$ 0.06	90	27.1 $\pm$ 5.8
Sept. 16, 1993	MCK 1.6	100	72.5 $\pm$ 17.1	0.58 $\pm$ 0.10	90	27.3 $\pm$ 5.8
		80	75.0 $\pm$ 23.8	0.65 $\pm$ 0.14	100	23.2 $\pm$ 6.5
		60	75.0 $\pm$ 26.5	0.65 $\pm$ 0.10	100	19.4 $\pm$ 5.3
	MCK 1.9	100	62.5 $\pm$ 17.1	0.68 $\pm$ 0.09	100	18.5 $\pm$ 6.8
		80	82.5 $\pm$ 15.0	0.59 $\pm$ 0.02	100	17.7 $\pm$ 6.8
		60	60.0 $\pm$ 31.6	0.73 $\pm$ 0.14	100	19.3 $\pm$ 8.7

Note: A dash (—) indicates not tested.

The distribution of *C. dubia* survival values in the McCoy Branch tests is very similar to the distribution of its survival values reported for control water and water from noncontaminated reference sites in headwater streams on the ORR (Loar 1991, Stewart 1996). Thus, overall survival patterns for *C. dubia* in tests of full-strength water samples from McCoy Branch do not provide evidence for acute toxicity.

During the 12 test periods, mean reproduction of *C. dubia* in the controls exceeded 20 offspring per female in 10 cases and was lower than 20 offspring per female in 2 cases. Mean reproduction of *C. dubia* in full-strength water from the McCoy Branch sites was greater than 20 offspring per female in 20 of 26 cases. All three McCoy Branch sites had low values for *C. dubia* reproduction during the test that started on March 25, 1993; reproduction in the control for these tests was also low. When the tests for March 25, 1993,



were excluded from consideration, mean reproduction of *C. dubia* was less than 20 offspring per female in 1 of 11 cases for controls, and less than 20 offspring per female in 4 of 23 cases involving full-strength water from McCoy Branch. In the 3 cases where *C. dubia* mean reproduction was less than 20 offspring per female in full-strength McCoy Branch water, one value was 19.7 offspring per female (July 11, 1991; MCK 1.9), two were greater than 18 offspring per female (18.3 for July 30, 1990, at MCK 1.6, and 18.5 for September 16, 1993, at MCK 1.9), and one was 11.1 (July 30, 1990; MCK 1.9). In three of these four cases, minnow survival was 100% and minnow growth in full strength water from McCoy Branch differed from minnow growth in the corresponding control by less than 10%; in one of the cases (September 16, 1993), minnow survival was low (62.5%) but minnow growth was high (0.68 mg/fish, vs 0.54 mg/fish in the control).

Minnow tests of McCoy Branch water from one or more sites were conducted during ten test periods (Table 3.2). Minnow survival and growth data were available for 21 cases involving full-strength water from McCoy Branch. In 3 of the 21 cases, mean survival of the minnows was less than 70%, a value that may be used as an arbitrary but reasonable criterion to suggest toxicity, given the EPA's 80% survival criterion for test acceptability (Weber et al. 1989). These "low survival" outcomes occurred in water from MCK 1.6 during the April 2, 1992, test (survival was  $47.5 \pm 18.9\%$ ), MCK 2.5 during the March 25, 1993, test ( $55.0 \pm 36.9\%$ ), and MCK 1.9 during the September 16, 1993, test (survival was  $62.5 \pm 17.1\%$ ). In all three "low survival" cases, minnow growth in the potentially toxic water was greater than in the corresponding control.

In various instances, minnow survival in one or more samples of diluted McCoy Branch water was lower than 70% (Table 3.2). In several of these instances, there was no consistent concentration-response pattern, such that greater mortality occurred in beakers that contained higher concentrations of McCoy Branch water. Examples of this situation can be found by inspecting the results of the minnow tests for MCK 1.9 for January 31, 1992, April 2, 1992, and July 9, 1992 (Table 3.2), and for MCK 1.6 for October 24, 1991, and April 2, 1992.

The results of the *C. dubia* and fathead minnow tests described above do not provide consistent, strong evidence for either acute or chronic toxicity to either fathead minnows or *C. dubia*.

### 3.3.2 Snail Feeding-Rate Tests

Water from any of the McCoy Branch sites, in any test period, did not affect the feeding rates of snails relative to feeding rates of snails in the controls (Table 3.3). In the first test, snails in full-strength water from MCK 1.9 and the 50% concentration of water from MCK 2.5 ate approximately twice as much as those in the controls (0.07 mg/day for controls, versus 0.17 mg/day for MCK 1.6 and MCK 1.9; Table 3.3). This difference was significant ( $p = 0.05$ ). In the second test, the mean feeding rate of snails in the controls was virtually identical to that in water either from MCK 1.6 or MCK 1.9 (0.17 mg/day). In this test, too, the addition of sediment to the water from the two McCoy Branch sites did not affect snail feeding rates (Table 3.3). In the third test, a temperature effect on feeding rate was very evident: at 25°C, snails ate 2.5 to 3.5 times more lettuce per day than they did at 15°C. Water temperature alone explained about 92.4% of variation in feeding rates; the site to temperature interaction was not significant ( $p = 0.829$ ).

Collectively, the results of the three feeding-rate tests suggest that water and sediment quality in McCoy Branch may not be factors that limit the recolonization of the stream by

Table 3.3. Results of 3-d snail (*Elimia clavaeformis*) feeding tests with water and sediment from McCoy Branch

Site/Treatment	Mean wet weight (g) eaten $\pm$ SE
<i>Test No. 1 Jan. 18, 1993</i>	
DMW <sup>a</sup>	0.07 $\pm$ 0.026
MCK 1.6-100%	0.14 $\pm$ 0.028
MCK 1.6-50%	0.16 $\pm$ 0.026
MCK 1.9-100%	0.17 $\pm$ 0.039
MCK 1.9-50%	0.13 $\pm$ 0.021
<i>Test No. 2 Apr. 19, 1993</i>	
DMW	0.17 $\pm$ 0.007
MCK 1.6	0.17 $\pm$ 0.007
MCK 1.6+sediment	0.17 $\pm$ 0.007
MCK 1.9	0.17 $\pm$ 0.011
MCK 1.9+sediment	0.18 $\pm$ 0.007
<i>Test No. 3 July 27, 1993</i>	
DMW-15°C	0.06 $\pm$ 0.004
DMW-25°C	0.15 $\pm$ 0.012
MCK 1.6+sediment-15°C	0.05 $\pm$ 0.005
MCK 1.6+sediment-25°C	0.14 $\pm$ 0.008
MCK 1.9+sediment-15°C	0.04 $\pm$ 0.005
MCK 1.93+sediment-25°C	0.14 $\pm$ 0.007

<sup>a</sup>DMW refers to diluted mineral water, which was used as a negative control and, in some of the McCoy Branch water toxicity tests, as a diluent.

*Elimia*. Longer-term tests that consider the possibility of effects of water or sediment on snail growth and/or reproduction would be needed to assess this possibility.

### 3.3.3 Snail *In Situ* Tag-Release-Recapture Test

One week after the test started, the caged snails in McCoy Branch and Scarboro Creek (a reference stream) were examined for survival and ammonium release. No mortality had occurred in any cage and the ammonium release rates of individual caged snails were similar

between streams. Ammonium release rates ( $\mu\text{g NH}_4^+/\text{g wet weight/hr}$ ), given as mean  $\pm$  SE (N), of *Elimia* and *Pleurocera* caged in McCoy Branch were  $1.80 \pm 0.12$  (45) and  $1.66 \pm 0.061$  (45), and those of *Elimia* and *Pleurocera* caged in Scarboro Creek were  $1.83 \pm 0.08$  (45) and  $1.42 \pm 0.08$  (45) respectively.

Three weeks after the tagged snails were released, the populations were monitored. Only 14% of the *Elimia* and 14% of the *Pleurocera* could be found, and over 90% of the animals found were downstream from the release site. The ammonium release rates of *Pleurocera* recovered from McCoy Branch at this time were greater than the ammonium release rates of *Pleurocera* in Scarboro Creek or Northwest Tributary; however, the ammonium release rates of *Elimia* were similar to those of *Elimia* in the two other streams (Fig. 3.1). Neither *Pleurocera* nor *Elimia* had measurable growth, based on changes in blotted wet weight; yet, fecal pellets were released into the sample vials by the recovered snails, indicating recent feeding. Many of the recovered snails, especially *Elimia*, showed evidence of physical damage attributed to high flows resulting from exceptionally wet weather during February and March; the damage included chipped and broken shells. After being weighed and analyzed for ammonium release, the tagged-recaptured snails were re-released at MCK 2.4.

Eight weeks after the initial release date, the two populations were monitored again. Only 10 *Elimia* and 17 *Pleurocera* could be found, despite extensive searches of submersed substrates upstream and downstream from the release site. All of these animals were found well downstream from the release point (one as far as 350 m). As seen with the first recovery, many of the snails had chipped and broken shells and no significant growth. However, loss of shell mass due to damage by the high flows may have prevented the detection of growth. Interestingly, the leading edge of the shell (i.e., near the operculum, where new shell material is deposited to permit shell growth) was visibly thin and very brittle for virtually all of the recaptured snails. This condition could indicate impairment of calcium-depositing physiological processes of snails involved with shell growth.

At the 8-week monitoring point, the ammonium release rates of *Elimia* and *Pleurocera* in McCoy Branch were extremely low compared to ammonium release rates of snails in Northwest Tributary at this time (Table 3.4). Mean respiration rates ( $\mu\text{g O}_2/\text{g wet weight/hr}$ ) of both species of snails recovered from McCoy Branch 8 weeks after initial release were about 20% lower than the respiration rates, given as mean  $\pm$  SE (N), of snails from Northwest Tributary [*Elimia*,  $42.5 \pm 3.4$  (10); *Pleurocera*,  $45.0 \pm 6.9$  (10)]; however, the variation about each mean rate was moderately large.

The low ammonium release rates, negligible growth rates, poor shell biomineralization, and gross movement patterns (downstream rather than upstream; see Burris et al. 1990) of *Elimia* and *Pleurocera* in McCoy Branch suggested that the snails were under physiological stress. As a logical outgrowth of the tag-release-recapture test, and with consideration given to information about the kinds of contaminants likely to be present in coal-ash, two hypotheses were formulated:

1. Physiological effects of stress in *Pleurocera*, as evidenced from measurements and observations during the tag-release-recapture test, can be attributed to exposure to coal-ash contaminated sediment.
2. Selenium and arsenic, contaminants concentrated in coal-ash, are responsible for the physiological effects on *Pleurocera*.

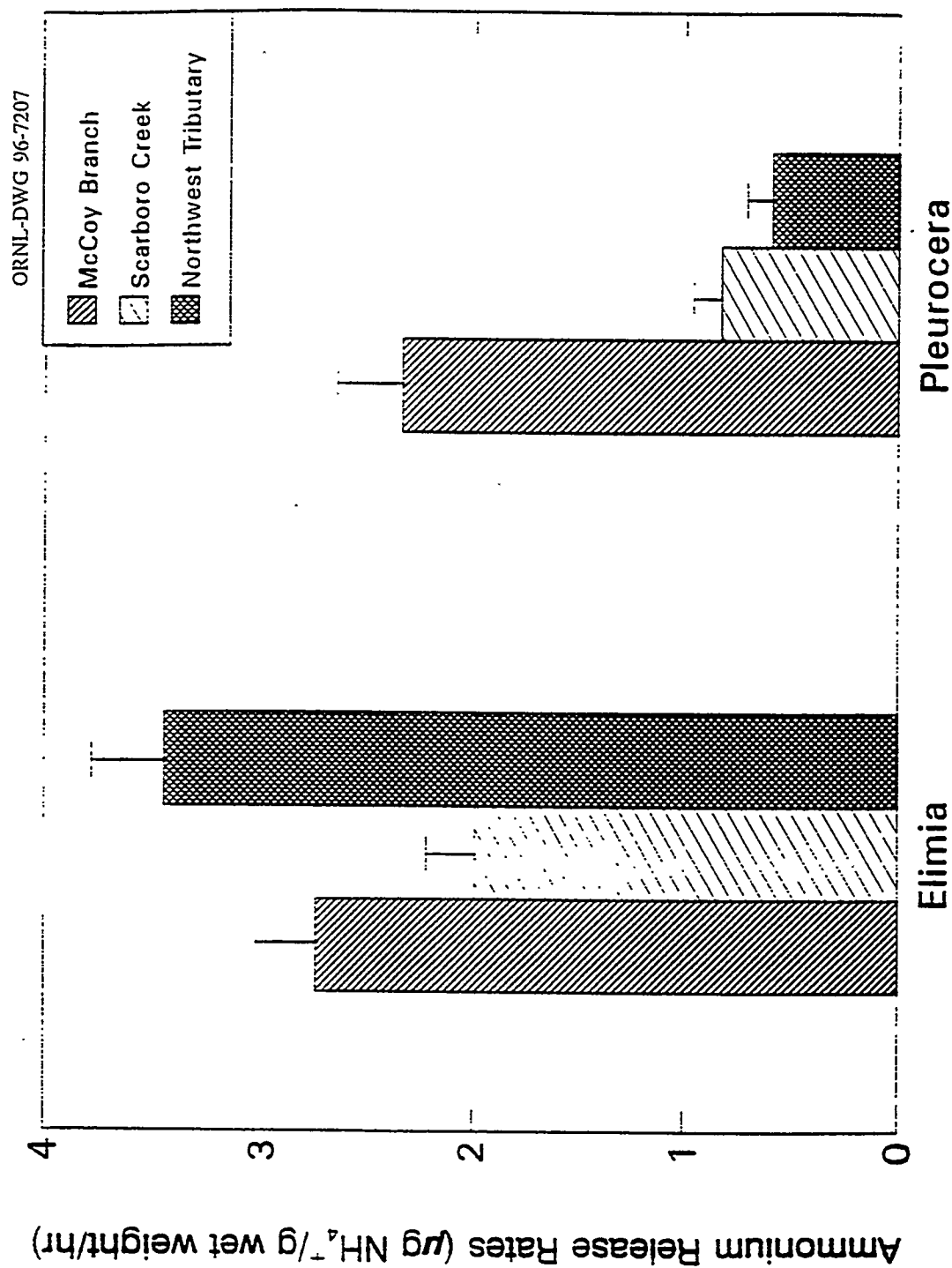


Fig. 3.1. Mean ammonium release rates of *Elimia clavaeformis* and *Pleurocera unciata unciata* in McCoy Branch (3 weeks after initial release), Scarborough Creek, and Northwest Tributary. Vertical lines are  $\pm 1$  standard error.

Table 3.4. Ammonium release rates, given as mean  $\pm$  SE (N), for *Elimia clavaeformis* and *Pleurocera uncialis uncialis* in McCoy Branch (8 weeks after initial release) and Northwest Tributary

Rates expressed as $\mu\text{g NH}_4^+/\text{g wet weight/hr}$		
Source stream for snails	Type of snail	
	<i>Elimia</i>	<i>Pleurocera</i>
McCoy Branch	0.10 $\pm$ 0.06 (10)	0.14 $\pm$ 0.04 (10)
Northwest Tributary	2.25 $\pm$ 0.30 (10)	2.83 $\pm$ 0.42 (10)

These two hypotheses will be tested by experiments conducted in the laboratory to exclude potentially confounding influences of stress due to scouring floods or other environmental factors. Although the hypotheses are expected to be germane to both species of snail, only *Pleurocera* will be used, because this snail tends to associate with sediments more strongly than *Elimia* (Neuhoff and Stewart 1994).

### 3.3.4 *In Situ* Test with Earthworms

Earthworm survival was higher in the peat moss soil than in the FCAP ash samples, and mean survival in the food-added treatments was greater than in the non-fed treatments of the same soil (or ash) type (Table 3.5). The ANOVA for nested designs using the transformed survival data revealed significant differences among survival means due to substrate type ( $p = 0.029$ ). The differences in means attributed to food were not statistically significant ( $p = 0.428$ ).

The measurements of soil (or ash) pH, moisture content and organic matter content showed that the FCAP ash was slightly acidic (pH 6.7-6.9) (Table 3.6). This ash had a water content of 47% to 48%, and an organic content of 30% to 34% (dry weight basis; Table 3.6). These conditions are within the ranges considered tolerable for earthworm survival (Lee 1985). The food-addition treatment slightly increased the pH in both the ash and the peat-moss soil (by 0.2 units, on average; Table 3.6). Survival in the FCAP soils was too low to allow accurate estimates of earthworm growth.

### 3.3.5 Laboratory Test with Earthworms

Results of the 14-d laboratory test with earthworms are summarized in Table 3.7. Mean survival of the worms in the reference and control (artificial) soil treatments was  $> 90\%$ , both for autoclaved and non-autoclaved treatments. Mean survival of worms among the three FCAP ash samples ranged from 32.5% (non-autoclaved ash from the Sycamore site (FCAP-1)) to 95% (autoclaved ash from the Sluice site and the Willow site (FCAP-2)). An ANOVA revealed statistically significant treatment-related differences in survival ( $F$  ratio = 4.55,  $p < 0.0008$ ). The multiple-level T-test showed that worm survival in ash from the FCAP-1 (both autoclaved and non-autoclaved) was significantly lower than either the control or reference soils ( $p < 0.05$ ) and that mean survival of the worms in ash from the FCAP-2 and Sluice site was not detectably different from their mean survival in the control or reference soils.

Table 3.5. Results of *in situ* test with earthworms

Soil type	Treatment	N	Survival fraction (mean $\pm$ SD)
peat moss	food added	6	0.611 $\pm$ 0.344
peat moss	no food	6	0.444 $\pm$ 0.251
fly ash	food added	6	0.056 $\pm$ 0.086
fly ash	no food	6	0.028 $\pm$ 0.068

Table 3.6. Results of soil and ash analyses for *in situ* test with earthworms

Soil type	Treatment	N	pH	Water content (%) (mean $\pm$ SD)	Organic content (% of dry matter) (mean $\pm$ SD)
peat moss	food added	6	4.8 $\pm$ 0.4	78 $\pm$ 2.2	96 $\pm$ 0.2
peat moss	no food	6	4.6 $\pm$ 0.4	78 $\pm$ 1.8	97 $\pm$ 0.8
fly ash	food added	6	6.8 $\pm$ 0.7	48 $\pm$ 6.2	34 $\pm$ 9.0
fly ash	no food	6	6.6 $\pm$ 0.9	47 $\pm$ 5.4	30 $\pm$ 7.8

(Table 3.7). ANOVA also was used to inspect the relationship between earthworm survival for autoclaved versus non-autoclaved FCAP ash. The results of this analysis indicated that apparent increases in worm survival in the autoclaved samples were not statistically significant ( $p = 0.152$ ). Thus, the mean effect of autoclaving on worm survival, for all sites compared simultaneously, was not great.

The results of the 14-d laboratory test described above suggest that ash at the FCAP-1 may have been toxic to earthworms. In the FCAP-1 ash, mean survival of the worms increased considerably (by 61%; from 32.5% to 52.5%; Table 3.7) with autoclaving. The increase in survival in the FCAP-1 sample caused by autoclaving does not eliminate the possibility that worm survival was affected by pollutants, because autoclaving can rid soils of volatile or semivolatile contaminants, in addition to killing potential pathogens or parasites. The results of this test suggest that pathogenic organisms do not account for the apparent toxicity of FCAP-1 ash.

On average, earthworms tended to lose more body weight in the FCAP ash treatments than they did in any of the control- or reference-soil treatments (Table 3.7). The greatest loss in body weight (about 50.5%) occurred in non-autoclaved ash from the FCAP-1 (Table 3.7). The losses in body weight among the soil (or ash) types, though, did not differ significantly when all treatments were considered simultaneously ( $p = 0.066$ ). In general, reproduction is considered to be more sensitive than growth to toxic chemicals. Soil toxicity tests that incorporate earthworm reproduction as a response variable might be more useful than tests that include only survival and growth as response variables.

Table 3.7. Results of 14-d laboratory tests with earthworms exposed to sediments from McCoy Branch watershed and reference areas

Soil or ash source	N	Treatment	Initial pH	Survival fraction (mean $\pm$ SD)	Multiple-level T-test <sup>a</sup>	Change in fresh mass (g; mean $\pm$ SD)
Reference	4	autoclaved	---	0.925 $\pm$ 0.050	a	0.0392 $\pm$ 0.0385
Reference	4	none	4.96	0.950 $\pm$ 0.058	a	-0.0032 $\pm$ 0.0355
Artificial	4	autoclaved	---	0.950 $\pm$ 0.100	a	-0.0088 $\pm$ 0.0648
Artificial	4	none	6.74	0.925 $\pm$ 0.050	a	-0.0410 $\pm$ 0.0522
Sluice	4	autoclaved	---	0.950 $\pm$ 0.100	a	-0.0558 $\pm$ 0.0311
Sluice	4	none	5.67	0.825 $\pm$ 0.150	ab	-0.0560 $\pm$ 0.0526
FCAP-1	4	autoclaved	---	0.525 $\pm$ 0.299	bc	-0.0818 $\pm$ 0.0373
FCAP-1	4	none	5.69	0.325 $\pm$ 0.330	c	-0.1820 $\pm$ 0.2027
FCAP-2	4	autoclaved	---	0.950 $\pm$ 0.058	a	-0.0778 $\pm$ 0.0862
FCAP-2	4	none	6.75	0.750 $\pm$ 0.379	ab	-0.0632 $\pm$ 0.0629

<sup>a</sup>Comparisons are for fraction of animals surviving.

### 3.3.6 Physicochemical Water Quality Factors

Measurements of pH, conductivity, alkalinity, and hardness were made on water from 2 or more sites in McCoy Branch on 12 of the 13 test periods shown in Table 3.1. Summary statistics for these parameters, by test, for MCK 1.6 and MCK 1.9, are shown in Tables 3.8 and 3.9.

The chemical data from MCK 1.6 and MCK 1.9 were extensive compared to sites MCK 2.5 and MCK 2.7 (the latter two sites had seven observations only; see Table 3.8 in Ryon et al. 1992). Data from MCK 1.6 and MCK 1.9 were analyzed statistically by SAS (PROC GLM) to reveal spatial and temporal contributions to differences in pH, conductivity, alkalinity, hardness, and the ratios of alkalinity to conductivity (A:C) and alkalinity to hardness (A:H). This analysis used site and test period (see Table 3.1) as class variables. Site, test, and the interaction between site and test explained 81.3%, 93.5%, 96.4%, and 77.3% of the variation in pH, conductivity, alkalinity, and hardness respectively (Table 3.10). Site, test, and the interaction between site and test explained 83.0% of the variation in A:C and 58.9% of the variation in A:H. Each of the three explanatory variables included in the model (test, site, and test to site) was highly significant ( $p < 0.0001$ ) for each water quality variable alone, and for both ratios. Thus, for data pooled temporally, the two sites were distinguishable statistically ( $p < 0.0001$ , with  $N = 84$ ) on the basis of any one of the four physicochemical parameters alone. The two sites could also be resolved statistically based on A:C and A:H, as well, but these two ratios were less characteristic of the sites than measurements of pH, alkalinity, hardness, or conductivity measurements alone. Based on the magnitude of the means computed by pooling data temporally, alkalinity (29.4%), hardness (18.1%), and conductivity (15.0%) differences between the two sites were more easily discerned than differences in pH (1.6%), A:C (11.7%), or A:H (8.7%) (Table 3.10).

Conductivity, alkalinity, and hardness are considered to be conservative properties of water, in that they are influenced more by fundamental geochemical processes and hydrological conditions than by biological processes (Stewart 1988). Inputs of wastewaters, or significant inputs of groundwater, or seepage water enriched in any constituent that affects pH, conductivity, or hardness, can alter the "chemical fingerprint" of stream water; the inputs are discernable by examining the relationships among conductivity, alkalinity, and hardness conditions. Based on measurements of conductivity, alkalinity, and hardness, water at MCK 1.6 and MCK 1.9 could be characterized as being similar to water in WOC between sites WCK 5.1 and WCK 4.4. The chemical conditions at MCK 1.6 and MCK 1.9 were much more typical of natural conditions than water at any site in East Fork Poplar Creek (EFPC).

## 3.4 DISCUSSION AND CONCLUSIONS

### 3.4.1 Fathead Minnow and *Ceriodaphnia* Tests

The fathead minnow tests provided no strong evidence for acute or chronic toxicity of water at any of the tested sites. Survival of fathead minnow larvae was  $<80\%$  in full-strength water from one or more sites in 4 of the 11 test periods that involved testing with fish. The site-date combinations in which minnow survival was suspect included: January 31, 1992 (MCK 1.9, 75% survival); April 2, 1992 (MCK 1.6, 47.5% survival); March 25, 1993, when survival at three sites was 55.0% to 77.5%; and September 16, 1993,



Table 3.8. Summary statistics of water quality factors for McCoy Branch kilometer (MCK) 1.6 for various toxicity test periods (see Table 3.1)

Values are 7-d means, standard deviation of the mean (in parentheses), and ranges for each 7-d test period

Factor	Analysis period													
	July 1990	Apr. 1991	July 1991	Oct. 1991	Jan. 1992	Apr. 1992	July 1992	Sept. 1992	Jan. 1993	Mar. 1993	July 1993	Sept. 1993		
pH (S.U.)	8.49 (0.09) 8.41-8.67	7.98 (0.15) 7.74-8.28	7.87 (0.05) 7.78-7.96	7.87 (0.14) 7.75-8.14	8.12 (0.05) 8.02-8.21	8.08 (0.08) 7.91-8.18	8.11 (0.08) 8.04-8.25	8.05 (0.04) 8.00-8.11	7.88 (0.09) 7.76-7.97	8.18 (0.09) 8.02-8.27	8.03 (0.05) 7.93-8.06	7.95 (0.06) 7.86-8.04		
Conductivity ( $\mu$ S)	232.9 (3.6) 227-237	241.4 (13.9) 225-260	212.6 (12.6) 192-229	235.3 (19.6) 193-259	251.5 (21.9) 225-276	249.0 (22.8) 218-277	271.6 (9.4) 258-281	288.5 (5.5) 283-296	278.6 (13.3) 255-291	265.4 (12.8) 243-277	278.3 (11.6) 253-286	284.9 (5.6) 273-290		
Alkalinity (mg/L)	82.9 (1.5) 81-84	95.4 (7.9) 86-113	92.1 (7.3) 82-101	105.1 (9.4) 94-118	110.1 (10.8) 98-125	107.1 (11.6) 94-119	123.7 (2.1) 120-126	125.5 (2.6) 123-130	123.3 (4.9) 120-134	118.0 (7.7) 106-130	125.6 (2.1) 123-129	132.3 (5.5) 128-144		
Hardness (mg/L)	124.6 (6.2) 112-130	126.0 (12.5) 100-140	114.1 (8.5) 100-128	128.7 (12.3) 114-154	129.1 (12.8) 112-152	129.4 (13.6) 110-154	150.9 (10.1) 140-166	157.7 (10.1) 146-172	149.1 (10.9) 134-164	169.4 (46.0) 134-268	156.0 (15.0) 136-180	150.3 (10.6) 140-168		

Note: See also Table 3.1, this document.

Table 3.9. Summary statistics of water quality factors for McCoy Branch kilometer (MCK) 1.9 for various toxicity test periods

Values are 7-d means, standard deviation of the mean (in parentheses), and ranges for each 7-d test period

Factor	Analysis period											
	July 1990	Apr. 1991	July 1991	Oct. 1991	Jan. 1992	Apr. 1992	July 1992	Sept. 1992	Jan. 1993	Mar. 1993	July 1993	Sept. 1993
pH (S.U.)	8.07 (0.07) 7.95-8.15	7.64 (0.10) 7.49-7.78	7.60 (0.07) 7.52-7.72	7.12 (0.14) 7.50-7.87	8.06 (0.05) 8.00-8.17	7.99 (0.05) 7.91-8.05	8.05 (0.16) 7.75-8.26	8.03 (0.07) 7.92-8.09	7.77 (0.11) 7.61-7.93	8.01 (0.14) 7.74-8.16	8.10 (0.06) 8.01-8.17	8.01 (0.06) 7.90-8.09
Conductivity ( $\mu$ S)	336.1 (18.1) 297-352	274.3 (22.8) 244-307	314.3 (7.7) 299-321	345.1 (5.1) 336-350	284.0 (6.4) 276-293	268.0 (13.0) 253-286	328.9 (16.9) 296-350	360.5 (5.5) 353-365	259.7 (21.4) 226-284	196.9 (21.9) 157-226	338.3 (3.7) 334-343	345.0 (3.1) 340-348
Alkalinity (mg/L)	165.0 (4.3) 158-171	120.4 (10.5) 104-134	158.3 (4.3) 151-163	175.1 (2.4) 170-177	141.9 (13.0) 131-170	129.4 (7.0) 121-141	169.7 (2.7) 167-174	179.2 (1.9) 177-182	117.4 (13.1) 102-138	84.0 (10.0) 72-101	171.3 (2.1) 169-174	179.1 (2.0) 177-183
Hardness (mg/L)	189.7 (4.4) 184-194	143.1 (13.6) 128-164	177.1 (7.6) 170-192	194.3 (5.2) 190-204	151.4 (8.2) 144-166	146.3 (8.2) 140-162	185.1 (9.6) 176-202	204.0 (13.5) 192-230	149.7 (18.4) 120-176	122.8 (21.5) 92-164	185.1 (11.5) 170-196	196.0 (9.9) 186-210

Note: See also Table 3.1, this document.

Table 3.10. Analysis of variance results for pH, conductivity, alkalinity, and hardness data from McCoy Branch kilometer (MCK) 1.6 and MCK 1.9

Means computed from 84 observations						
Site	pH	Conductivity	Alkalinity	Hardness	Alk:Cond	Alk:Hard
Mean at MCK 1.6	8.05	264.4	115.4	144.2	0.436	0.806
Mean at MCK 1.9	7.92	304.1	149.3	170.3	0.487	0.872
R <sup>2a</sup>	0.813	0.935	0.964	0.773	0.830	0.589
F-ratio for: test (df=11)	38.50	72.12	119.41	13.58	22.85	10.79
site (df=1)	83.12	506.57	1369.99	145.03	310.98	46.62
test to site (df=11)	10.78	71.60	105.52	17.72	12.73	3.77
Site difference (%) <sup>b</sup>	1.6	15.0	29.4	18.1	11.7	8.2

<sup>a</sup>R<sup>2</sup> is the proportion of variation explained collectively by site, test, and the interaction between site and test, using ANOVA (PROC GLM, type III sum of squares). Model and error degrees of freedom were 23 and 144, respectively, for each water quality factor.

<sup>b</sup>Computed, for each water quality factor, as the difference between means for the two sites, divided by the smaller mean, times 100.

when survival was 62.5% at MCK 1.9 and 72.5% at MCK 1.6 (Table 3.2). In each of the tests where minnow survival was suspect, minnow growth was greater than the control. In most cases, tests in which minnow survival was low also had higher-than-usual levels of among-replicate variation (Table 3.2). Other studies have found that low mean survival of fathead minnow larvae is fairly common in ambient toxicity testing situations; low mean survival, if accompanied by an unusually large variation in survival among replicates, may indicate the presence of pathogenic bacteria or fungi (Kszos and Stewart 1992). The unusually large variation in survival among replicates for the minnows in each of the suspect site-date combinations provides better evidence for pathogens than for chemical contaminants.

The results of the *C. dubia* tests also provided no strong evidence for toxicity. In 17 of the 26 site-test combinations that used full-strength McCoy Branch water, survival of *C. dubia* was 100%; in 5 of the 26 site-test combinations, it was 90%; in 3 cases, survival was 70%, and in 1 case, it was 60% (Table 3.2). This distribution of survival values is very similar to that obtained when using *C. dubia* to assess non-contaminated stream sites on the ORR (Loar 1991). In April, 1992, when two site-date combinations yielded *C. dubia* survival values that were  $\leq 70\%$ , there was no evidence of a dose-response relationship, even though multiple concentrations were tested. Additionally, the *C. dubia* that survived in the two full-strength water samples produced large numbers of offspring.

In 18 of the 26 site-test combinations involving full-strength water from McCoy Branch sites, *C. dubia* reproduction was  $\geq 20$  offspring/female, which is well above the EPA's criterion for control acceptability (15 offspring/female). In 11 of the 26 site-test combinations, *C. dubia* reproduction in full-strength McCoy Branch water exceeded reproduction in the controls. In the July 30, 1990, test period, mean reproduction of *C. dubia* in full-strength water from MCK 1.9 was low ( $< 12$  offspring/female), but no dose-response pattern in MCK 1.9 dilutions was evident (Table 3.2). During the March 1993 test, reproduction of the daphnids was low both in the controls (7.0 offspring/female) and in McCoy Branch water (MCK 1.6, MCK 1.9 and MCK 2.5). In this instance, the mean numbers of offspring produced by daphnids in water from the three McCoy Branch sites (7.4, 12.9, and 14.5 offspring/female respectively) was greater than that of the control (7.0 offspring/female) (Table 3.2). This response does not rule out the possibility that water from McCoy Branch was toxic during the March 1993 test, but the more parsimonious explanation is that the daphnids used in the test were stressed inadvertently either before or during the test.

#### 3.4.2 Snail Feeding-Rate Tests

The feeding rates of *Elimia* in water from MCK 1.6 and MCK 1.9 were very similar to feeding rates of the snails in diluted mineral water controls in all three test periods (Table 3.3). At 25°C, the mean feeding rates ranged from about 0.14 mg to 0.18 mg of wet weight of lettuce eaten per day (Table 3.3). In two of the tests, water from MCK 1.6 and MCK 1.9 was spiked with a high concentration of McCoy Branch sediment (1 g/L and 1.5 g/L) to determine whether sediment contaminants or physical nature of the sediments interfered with snail feeding. The feeding rates of the snails were not inhibited by the sediment in either of these tests (Table 3.3). Water temperature strongly influenced the feeding rates of the snails, as would be expected based on simple physiological principles, but there was no significant interaction between the temperature and sediment treatments. Thus, we conclude that the snail test results provide no evidence for acute toxicity either from the

water or sediments of MCK 1.6 or MCK 1.9. It is possible that longer exposures to these media would reveal snail behavioral differences that might be related to the presence of contaminants.

### 3.4.3 Snail *In Situ* Tag-Release-Recapture Test

The study results to date indicate that *Pleurocera* and *Elimia* find it difficult to colonize McCoy Branch. Water quality factors such as pH, conductivity, alkalinity, and temperature can probably be ruled out as restricting factors in this regard given the range of conditions in ORR streams presently inhabited by *Elimia* (see Hill 1992, Hill et al. 1995). Based on visual inspections of substrates, food quantity appears to be great enough to sustain either *Elimia* or *Pleurocera*. Conditions that cannot yet be ruled out as restricting factors may include low but biologically significant concentrations of contaminants, either in the water or associated with the sediments or food; physical features of the sediment (e.g., ash particles may be more corrosive than particles of rock origin during flood events); or more subtle aspects of water quality that are biologically important but which do not involve contaminants (e.g., cation ratios, such as Na:K, Ca:Mg, etc.). Several of these possibilities are being explored.

### 3.4.4 Laboratory and *In Situ* Earthworm Tests

The two earthworm tests showed that FCAP ash might be problematic for soil organisms. In the *in situ* test, earthworm survival in FCAP ash at the six sites was substantially lower than in peat moss (Table 3.6). The addition of food increased survival in both the ash and peat-moss soil, but survival even in food-augmented FCAP ash was still very low compared to non-amended peat moss. The low survival of the worms in the FCAP ash did not appear to be explained by pH or by water or organic matter content.

In the laboratory test, ash from the FCAP-1 site adversely affected earthworm survival. On the average, worms lost more body weight in all FCAP ash treatments than they did in any of the control- or reference-soil treatments (Table 3.7). This difference was nearly significant ( $p = 0.066$ ). On average, autoclaving the FCAP ash samples used in this test increased mean survival of the earthworms, but this effect was not statistically significant ( $p = 0.152$ ). Additionally, mean survival in autoclaved FCAP-1 site ash was only 52.5%. FCAP ash texture, or ash-associated contaminants, are factors that could account for earthworm mortality in the *in situ* and laboratory tests and/or weight-loss patterns noted in the laboratory test.

### 3.4.5 Physicochemical Water Quality Factors

Water quality data from MCK 1.6 and MCK 1.9 were evaluated by ANOVA and by inspecting correlative relationships among conductivity, alkalinity, and hardness. The results of these analyses indicate that water quality conditions at MCK 1.6 and MCK 1.9 are similar to other stream sites on the ORR where impacts could be described as slight to moderate. The analysis used provides a reasonable means for detecting major chemical perturbations (i.e., those that alter naturally established relationships among conductivity, alkalinity, and hardness), but may not be very sensitive to the presence of low concentrations of toxic

contaminants. The analyses of the water quality factors showed that the McCoy Branch sites tested for toxicity could be distinguished from one another statistically based on factors such as conductivity, alkalinity, and hardness. However, these analyses do not provide evidence for or against the presence of toxicants. Additionally, because no strong evidence for toxicity was detected, no relationships could be established between toxicity and measurements of pH, conductivity, alkalinity, or hardness. The water-quality conditions in McCoy Branch, based on consideration of relationships among conservative water-quality factors, were more similar to non-contaminated reference streams on the ORR than they were to conditions typical for polluted streams, such as EFPC.

### 3.5 FUTURE STUDIES

Water from two sites in McCoy Branch (MCK 1.6 and MCK 1.9) will be tested quarterly for acute and chronic toxicity to *C. dubia*. The samples will not be tested with fathead minnow larvae, for two reasons: *C. dubia* are more sensitive than fathead minnows to various toxic metals and organic chemicals, and, for ambient applications, the *C. dubia* test system is more robust and less prone to artifacts than the minnow test system (Kszos and Stewart 1995, Stewart 1996).

During August and October 1995, substrates in McCoy Branch upstream and downstream from the snail release site (MCK 2.4) will be surveyed for juvenile and adult *Pleurocera* and *Elimia*. The results of these surveys will be reported with other 1995 results in subsequent reports. If no snails of either genus can be found in the swamp, it is reasonable to suspect that sediment or water quality, rather than floods, limits invasion of McCoy Branch by pleurocerid snails. A simplified re-release plan (e.g., non-tagged *Pleurocera* and/or *Elimia*) will be developed to test the idea that poor water or sediment quality, not floods, accounts for the snail-free situation.

## 4. BIOACCUMULATION STUDIES

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

### 4.1 INTRODUCTION

Bioaccumulation monitoring conducted in Rogers Quarry in 1990 identified only two inorganic substances associated with fly ash leachate, arsenic and selenium, as bioaccumulation concerns (Ryon et al. 1992). No organic contaminants or other trace elements were found to be clearly elevated above background levels. Thallium was detected in bass from Rogers Quarry in 1990, but the measured concentrations were very close to the detection limit of the analytical procedure. Mercury was of interest in the 1990 sampling data, but concentrations of this element were extremely low in Rogers Quarry fish. Bioaccumulation monitoring subsequent to the initial 1990 screening focused on evaluating expected temporal changes in arsenic, selenium, and mercury in Rogers Quarry bass that resulted from the elimination of inputs of fly ash and leachate. Data on concentrations of other elements were obtained incidental to the analysis of arsenic and selenium.

### 4.2 MATERIALS AND METHODS

Largemouth bass (*Micropterus salmoides*) were collected by angling from Rogers Quarry in July 1991, 1992, and 1993 and from Lambert Quarry, a nearby quarry with no history of fly ash disposal, in 1991. The fish were placed on ice and returned to the laboratory, where they were weighed and measured. The bass were filleted and skinned, and a 5 g portion of the anterior dorsal portion of the fillet was removed for analysis for mercury, arsenic, selenium, and any other trace metals at the Oak Ridge National Laboratory Analytical Chemistry Division. Mercury was analyzed by cold vapor atomic absorption spectrophotometry after digestion in a mixture of perchloric and nitric acids (Feldman 1974; EPA 1991, method 245.5). Arsenic, beryllium, and uranium were analyzed by inductively coupled plasma mass spectrometry (ICP/MS) (EPA 1991, method 200.8), and antimony, cadmium, chromium, copper, lead, nickel, selenium, silver, thallium, and zinc by inductively coupled plasma optical emission spectrometry following digestion in nitric acid (EPA 1991, method 200.7). In 1993, all analyses except mercury were carried out by ICP/MS.

Statistical evaluation of the data was conducted using linear regression, analysis of variance, Levenes test for homogeneity of variances, and analysis of covariance (SAS 1985a). Statistical comparisons used 0.05 as the predetermined level of significance.

### 4.3 RESULTS AND DISCUSSION

#### 4.3.1 Metals in Fish

Mean concentrations of arsenic, selenium, and mercury in individual bass from Rogers Quarry and Lambert Quarry are summarized in Table 4.1, and data for individual fish are

**Table 4.1. Mean concentrations ( $\mu\text{g/g}$  wet wt,  $\pm$  SE) of arsenic, selenium, and mercury in axial muscle of largemouth bass from Rogers Quarry, Melton Hill Reservoir, and a reference site**

Expressed as micrograms per gram wet weight  $\pm$  SE

Site	Date	Arsenic	Selenium	Mercury	Mercury,adj <sup>a</sup>
Rogers Quarry	7/90	0.29 $\pm$ 0.02	3.0 $\pm$ 0.10	0.014 $\pm$ 0.002	0.020 $\pm$ 0.002
Rogers Quarry	7/91	0.26 $\pm$ 0.03	3.3 $\pm$ 0.07	0.054 $\pm$ 0.006	0.048 $\pm$ 0.003
Rogers Quarry	7/92	0.27 $\pm$ 0.01	2.2 $\pm$ 0.08	0.11 $\pm$ 0.009	0.084 $\pm$ 0.007
Rogers Quarry	7/93	0.22 $\pm$ 0.01	2.2 $\pm$ 0.13	0.19 $\pm$ 0.009	0.166 $\pm$ 0.011
Melton Hill Reservoir	7/90	0.14 $\pm$ 0.01	1.00 $\pm$ 0.29	0.073 $\pm$ 0.008	-
Lambert Quarry (reference)	7/91	<0.05	0.71 $\pm$ 0.10	0.93 $\pm$ 0.09	-

<sup>a</sup>Means adjusted by analysis of covariance,  $\ln(\text{Hg})$  vs  $\ln(\text{wgt})$ . Differences between adjusted means are statistically significant, (Analysis of covariance,  $p < 0.0001$ ).

Note: N = 8 samples/site.

presented in Table 4.2. Arsenic concentrations, found to be elevated in 1990, remained at the same level, about 0.2 to 0.3  $\mu\text{g/g}$  over the 1990-93 period. Selenium exhibited similar behavior, remaining in the 2-3  $\mu\text{g/g}$  range. Both elements were elevated several fold above background concentrations, as typified by Lambert Quarry bass. Thallium was not detected ( $<0.02$   $\mu\text{g/g}$ ) in bass from Rogers Quarry in 1991 and 1992; however, very low concentrations (0.045 and 0.021  $\mu\text{g/g}$ ) were detected in two of eight fish in 1993.

Concentrations of antimony, cadmium, chromium, copper, lead, nickel, silver, thallium, and uranium were below detection limits in 1991 and 1992, when inductively coupled plasma/atomic emission spectroscopy was used for analyses. Detection limits were improved by using inductively coupled plasma/mass spectrometry for analyses in 1993. Mean (micrograms per gram wet wt,  $\pm$  SE) metal concentrations measured in Rogers Quarry bass in 1993 by this method were: silver,  $<0.04$ ; beryllium,  $<0.003$ ; cadmium,  $<0.02$ ; chromium,  $<0.05$  (two detectable measurements, 0.07 and 0.08  $\mu\text{g/g}$ ); copper,  $0.08 \pm 0.03$ ; nickel,  $<0.05$ ; lead,  $<0.02$ ; antimony,  $<0.05$ , uranium,  $<0.003$ , and zinc,  $3.5 \pm 0.9$ .

The mean concentrations of metals were all below concentrations used by EPA (EPA 1990) and others (Hoffman et al. 1984, Travis et al. 1986) to screen for levels of contamination that pose no threat for human consumption, with the exception of arsenic. Arsenic was present at concentrations above screening criteria; however the screening concentration for arsenic is below the detection limit for arsenic in fish, and well above concentrations typical of most biological materials (Bowen 1979).



Table 4.2. Concentrations of arsenic, beryllium, selenium, thallium, uranium, zinc, and mercury in largemouth bass (*Micropterus salmoides*) from Rogers Quarry and Lambert Quarry, Anderson County, Tennessee, 1991-93

Expressed as micrograms per gram wet weight												
Site	Date	Tag	Sex	Wgt (g)	Lgth (cm)	As	Be	Se	Tl	U	Zn	Hg
RQ	7/15/91	1344	M	646	37	0.37	<0.003	3.7	0.02	0.003	4	0
		1332	F	639	35.9	0.29	<0.003	3.1	0.02	0.003	4	0
		1443	F	634	36.7	0.24	<0.003	3.5	0.02	0.003	4	0
		1531	F	735	36.5	0.29	<0.003	3.2	0.02	<0.003	3	0
		1338	F	607	36.4	0.29	<0.003	3.3	0.02	<0.003	6	0
		1442	F	1471	45.8	0.13	<0.003	3.1	<0.02	<0.003	5	0
		1445	M	623	36.7	0.33	<0.003	3.3	<0.02	<0.003	4	0
		1447	M	229	26.8	0.14	<0.003	3.1	0.02	0.003	4	0
LQ	8/30/91	5998	F	520	37.6	<0.05	<0.003	0.69	<0.02	<0.003	4	1
		5999	M	446	34.2	<0.05	<0.003	1.2	<0.02	<0.003	3	1
		5958	M	707	40.5	<0.05	<0.003	<0.4 9	<0.02	<0.003	4	1
		9429	F	478	36.4	<0.05	<0.003	0.86	<0.02	<0.003	4	1
	10/4/91	9443	M	495	36.2	<0.05	<0.003	0.77	<0.02	<0.003	4	1
		9444	M	503	39.5	<0.05	<0.003	0.58	<0.02	<0.003	4	1
		9445	M	419	33	<0.05	<0.003	0.66	<0.02	<0.003	4	1
		9446	M	383	35.2	<0.05	<0.003	0.67	<0.02	<0.003	4	1
RQ	7/20/92	3300	M	673	39.2	0.3	<0.003	2.36	<0.02	<0.003	0	0
		3301	M	673	39.3	0.28	<0.003	2.42	<0.02	<0.003	0	0
		3302	M	673	40.1	0.24	<0.003	2.58	<0.02	<0.003	0	0
		3303	M	673	36.4	0.33	<0.003	2.28	<0.02	<0.003	0	0
		3304	F	673	41.3	0.19	<0.003	1.9	<0.02	<0.003	0	0
		3305	F	673	41	0.24	<0.003	1.89	<0.02	<0.003	0	0
		3306	M	673	47.5	0.27	<0.003	2.24	<0.02	<0.003	0	0
		3309	M	673	39.5	0.27	<0.003	2.25	<0.02	<0.003	0	0

Table 4.2 (continued)

Site	Date	Tag	Sex	Wgt (g)	Lgth (cm)	As	Be	Se	Tl	U	Zn	Hg
RQ	7/28/93	4940	M	651	40	0.24	<0.003	2.3	<0.02	<0.003	3	0
		4941	M	588	39.7	0.24	<0.003	1.9	<0.02	<0.003	6	0
		4942	M	590	35.9	0.27	<0.003	1.9	<0.02	<0.003	5	0
		4943	F	706	39.5	0.28	<0.003	2.5	<0.02	<0.003	6	0
		4944	M	533	34.6	0.24	<0.003	2.1	0.048	<0.003	5	0
		4945	F	760	41.2	0.22	<0.003	1.7	<0.02	<0.003	5	0
		4946	F	796	39.9	0.19	<0.003	2	0.028	<0.003	4	0
		4947	F	751	41.6	0.2	<0.003	2	<0.02	<0.003	6	0

#### 4.3.2 Effect of Reduced Fly Ash Input on Concentrations of Selenium and Mercury in Fish

Selenium concentrations in largemouth bass from Rogers Quarry were clearly elevated, averaging over 2  $\mu\text{g/g}$  on each sampling (Table 4.1). Typical background or reference site concentrations of selenium in fish in this region appear to be well below 1  $\mu\text{g/g}$ . The mean concentration in bass from nearby Lambert Quarry was 0.7  $\mu\text{g/g}$  (Table 4.1). Analyses conducted by the Tennessee Valley Authority (TVA) on largemouth bass from reservoirs in the Tennessee River system averaged  $0.29 \pm 0.10$   $\mu\text{g/g}$  (Dycus 1989). In analyses conducted as part of the U.S. Fish and Wildlife Service National Contaminant Monitoring Program, the mean concentration of selenium in bass from sites in the United States east of the Mississippi River was  $0.40 \pm 0.10$  (Lowe et al. 1985).

Concentrations of selenium in fish from other sites impacted by fly ash discharges exceeded those observed in Rogers Quarry, sometimes by a wide margin. In Belews Lake, North Carolina, mean muscle concentrations of selenium in collections of various sunfish (*Lepomis*) species at different sites in the lake typically ranged from 10 to 20  $\mu\text{g/g}$  (Cumbie and Van Horn 1978). Sunfish in Martins Lake, Texas, averaged about 7  $\mu\text{g/g}$  Se in muscle (Sorenson et al. 1982). Bluegill from Hyco Reservoir, North Carolina, averaged 6–8  $\mu\text{g/g}$  selenium (Gillespie and Baumann 1986).

Selenium has remained elevated in Rogers Quarry bass (Table 4.1) despite the elimination of inputs to the system. Similar results were observed in Martins Lake, where selenium remained high in fish collected in July 1981 where ash pond effluent was discharged from September 1978 to May 1979 (Sorensen et al. 1982). Selenium bioaccumulation is dominated by food chain uptake (Graham et al. 1992; Turner and Swick 1983), and adverse toxicological and ecological effects in natural waters have generally been reported at aqueous concentrations far below those found to be toxic through direct aqueous exposure in laboratory tests (Hodson 1990). It appears that aqueous inputs of selenium to aquatic systems are to some extent stockpiled in the biological components of the systems, acting to buffer against rapid response of selenium in biota to a decrease in aqueous phase concentrations.

Since 1985, mercury concentrations have been measured in sunfish and other species, including largemouth bass, from numerous sites in east Tennessee as part of remedial

investigations and monitoring programs at DOE facilities in Oak Ridge. Mercury concentrations in largemouth bass collected from Rogers Quarry in 1990 (Table 4.1) were the lowest observed in any species of fish in those monitoring programs. Mercury concentrations in the largemouth bass from Rogers Quarry are among the lowest reported for this species in the United States.

Concentrations measured by TVA in bass from 11 other sites in the upper Tennessee Valley all equaled or exceeded  $0.2 \mu\text{g/g}$  (Dycus 1989). The lowest mean concentrations observed in an extensive survey (96 sites) of lakes and streams in Florida were  $0.04$  and  $0.07 \mu\text{g/g}$  in 2 highly eutrophic lakes (Hand and Friedman 1990). Concentrations at other locations typically exceeded  $0.2 \mu\text{g/g}$  in that study, with most exceeding  $0.5 \mu\text{g/g}$ . Only one of 39 composite largemouth bass samples analyzed in the National Contaminant Biomonitoring Program, 1978–1981, contained  $0.02 \mu\text{g/g}$  or less mercury (Lowe et al. 1985).

Mean mercury concentrations measured in largemouth bass from Rogers Quarry increased continually in each year since 1990 (Table 4.1). Mean concentrations of mercury (log<sub>e</sub> transformed, analysis of covariance) were statistically different among all four collections (1990 < 1991 < 1992 < 1993). Mercury concentrations in bass increased by a factor of eight between 1990 and 1993, reaching a level typical of largemouth bass from nearby Melton Hill reservoir (Table 4.1) but below that typical of other TVA reservoirs (Dycus 1989). Fish collected from nearby Lambert Quarry contained much higher concentrations of mercury (Table 4.1). Although there is no history of mercury disposal in that quarry, the very high levels of mercury in fish, coupled with the proximity to industrial sites that used large quantities of mercury in the past, raise suspicions that these levels are not typical background concentrations for bass from quarries in this region. Likewise, the mercury disposal history of Rogers Quarry is unknown and the eventual steady state concentration of mercury in bass there is uncertain. The gradual increase in mercury content in bass followed the abrupt decrease in waterborne selenium concentrations that occurred in 1989.

Several researchers have speculated that additional selenium in solution would have an inhibitory effect on mercury bioaccumulation (Rudd et al. 1980; Rudd et al. 1983; Turner and Rudd 1983; Turner and Swick 1983). Bjornberg et al. (1988) postulated that this was due to the extremely low solubility of mercuric selenide in water. The presence of selenide acts to reduce the activity of  $\text{Hg}^{+2}$  in solution, reducing the availability of the key precursor to methylmercury formation. Others found that the accumulation of methylmercury from food was inhibited when selenium was accumulated along with mercury in the target species (Rudd et al. 1980; Rudd et al. 1983; Turner and Rudd 1983; Turner and Swick 1983). These researchers found that aqueous exposure concentrations of  $1\text{--}10 \mu\text{g/L}$  selenite were effective at elevating selenium concentrations in food organisms enough to reduce mercury accumulation.

Anderson and Smith (1977) found that mercury concentrations in fish from a cooling water lake at a coal-fired power plant were substantially lower than those in fish from similar lakes in the region, despite evidence of mercury inputs to the lake from plant operation. Ash from the plant was disposed of in diked settling ponds near the lake, drainage from which may have added selenium to lake waters. Several lakes in Sweden, in which mercury concentrations in fish were excessive, were treated with additions of  $0.5$  to  $5 \mu\text{g/L}$  Se as sodium selenite (Bjornberg 1987; Paulsson and Lundbergh 1991). Substantial decreases in the concentrations of mercury were observed in fish from treated lakes. Large increases in selenium concentrations in fish were associated with these treatments, with perch increasing from  $0.3 \mu\text{g/g}$  before treatment to  $5\text{--}7 \mu\text{g/g}$  afterwards. The increase in mercury concentrations in Rogers Quarry bass occurs at selenium concentrations in fish somewhat below those found to inhibit mercury accumulation in Sweden. However, selenium concentrations in the bass were within roughly a factor of two of those observed in fish in the Swedish studies, ranging from  $2$  to  $3 \mu\text{g/g}$  over the 1990–92 period. These

results suggest that selenium accumulated in the tissues of fish is not very effective at inhibiting the accumulation of methylmercury, but rather that continuous exposure to additional selenium in food or water is needed to retard bioaccumulation of mercury.

#### 4.4 CONCLUSIONS

Concentrations of selenium and arsenic are elevated in largemouth bass from Rogers Quarry relative to bass and sunfish from other sites in east Tennessee. Only arsenic exceeds conservatively based screening criteria; however virtually all biological materials exceed this criterion for arsenic. Cessation of inputs of fly ash to the system has not resulted in the expected decrease in arsenic and selenium concentrations in fish, suggesting that internal cycling of these elements within the quarry is maintaining elevated concentrations in the biota.

Elimination of slurried fly ash discharges to the quarry was followed by a steady increase in concentrations of mercury in the axial muscle of resident largemouth bass (*Micropterus salmoides*). Average mercury concentrations in bass (adjusted for covariance with fish weight) increased from 0.02  $\mu\text{g/g}$  to 0.17  $\mu\text{g/g}$  in 3 years. Aqueous selenium concentrations in the quarry decreased from 25  $\mu\text{g/L}$  to < 2  $\mu\text{g/L}$  after elimination of fly ash discharges, but selenium concentrations in bass remained about three times background levels. Previous studies have shown selenium addition to be a viable means of ameliorating mercury contamination in fish in low alkalinity, low pH waters of northern Europe and Canada. These results suggest that selenium may also be effective at blocking the accumulation of methylmercury in harder, more alkaline waters.

#### 4.5 FUTURE STUDIES

Annual monitoring of selenium, arsenic, and mercury in bass from Rogers Quarry will be continued in order to quantify the time scale of response of these elements to source reduction/elimination.

## 5. TERRESTRIAL BIOACCUMULATION STUDIES

(L. A. Baron and C. T. Garten)

### 5.1 INTRODUCTION

Ash typically contains nutrient elements for plant and animal survival and growth; however, toxic elements may also be present (Furr et al. 1976). Vegetation and small mammals (e.g., mice, shrews, voles) living on the FCAP are subjected to elevated concentrations of metals (Al, As, Ba, Be, Ca, Cd, Co, Cr, Cu, Fe, Hg, K, Mg, Na, Ni, Pb, Se, Tl, V, and Zn) found in the fly ash.

Although contaminants may not reach phytotoxic levels, heavy metals (As and Se) accumulated in plant tissue may exist at potentially toxic levels for herbivorous wildlife such as white-tailed deer and small mammals (Adriano et al. 1980; Furr et al. 1977). Many studies have shown that Se is readily absorbed and accumulated by plants (Arthur et al. 1992a, 1992b; El-Mogazi et al. 1988; Adriano et al. 1980; Scanlon and Duggan 1979; and Furr et al. 1977). Ideally, plants should contain 0.05 to 0.3 ppm (dry weight) of Se to provide sufficient dietary intake for animal metabolism. Higher concentrations (3 to 40 ppm) of Se may result in chronic toxicity to foraging animals (Ohlendorf 1989).

Small mammals may be exposed to contaminants through consumption of vegetation, surface water, and incidental exposure to ash while foraging or burrowing (Suter 1993). Small mammals are known to bioaccumulate a variety of heavy metals from contaminated environments (Talmage and Walton 1991). Talmage and Walton (1991) found a general relationship between concentrations of contaminants in soil or food and concentrations in tissues of several species, especially for nonessential heavy metals such as Cd, Pb, and Hg. Heavy metals may reach potentially toxic concentrations to the small mammals or their wide-ranging predators (i.e., birds of prey, snakes, foxes).

The primary objectives of these studies were to determine the degree of bioaccumulation of heavy metals (As, Cd, Cr, Hg, Pb, Se, and Tl) in vegetation and small mammals in the McCoy Branch watershed above Rogers Quarry and to provide information indicating the potential for transfer of contaminants through the food chain and possible risks to higher-level consumers.

### 5.2 MATERIALS AND METHODS

#### 5.2.1 Vegetation

Plants were sampled at two distinct vegetative zones on the FCAP (FCAP-1: Sycamore area and FCAP-2: Willow area) (Fig. 2.2) to determine concentrations of As, Cd, Cr, Pb, Se, and Tl. The plants were sampled in mid-September 1992, and included: red maple (*Acer rubrum*), boxelder (*Acer negundo*), eastern cottonwood (*Populus deltoides*), eastern red cedar (*Juniperus virginiana*), sycamore (*Platanus occidentalis*), willow (*Salix* spp.), a grass (*Eulalia viminea*), and Japanese honeysuckle (*Lonicera japonica*). The sluice site, located on a hillside north of the FCAP in the vicinity of the former effluent channel from the Y-12 Plant, was

sampled at the same time. Plants sampled at the sluice included: red maple, sycamore, eastern red cedar, and *Eulalia*. Individual trees were sampling units, and only leaves were collected from each tree. Grasses (*Eulalia* spp.) were sampled by compositing both leaves and stems at each sampling point. Honeysuckle was sampled in a manner similar to grasses. The samples were not washed and were frozen during storage. Washed samples would not be representative of the plant material consumed by herbivores or litter detritivores. Controls were selected from archived, dry foliage samples collected in August or September (1988 or 1991) from Walker Branch watershed (Fig. 2.1). The control samples included leaves from dogwood (*Cornus florida*), sourwood (*Oxydendrum arboreum*), American beech (*Fagus grandifolia*), sweet gum (*Liquidambar styraciflua*), eastern red cedar, sycamore, boxelder, red maple, and *Eulalia*.

Plant samples were freeze-dried and ground to a fine, homogenous powder using a Tecator sample mill. Samples were then submitted to Roy F. Weston, Inc., for metals analysis. The samples were prepared for analysis as follows: (1) a 200 mg portion was weighed into a Teflon acid digestion bomb (Parr Model 4782), (2) concentrated nitric acid (2.5 mL) was added to the sample, which was irradiated in a microwave oven for 90 seconds at 350 watts, (3) the resultant solution was diluted with deionized water to give a final volume of 100 mL. Total metals analysis (As, Cd, Cr, Pb, Se, and Tl) was performed by Graphite Furnace Atomic Absorption (GFAA). Mercury was analyzed by Cold Vapor Atomic Absorption (CVAA). Quality was assured for these analyses by the use of standard procedures, calibration of instruments, and maintenance of laboratory control charts for all instruments. Statistical comparisons were made using analysis of variance (SAS 1988).

### 5.2.2 Small Mammals

Small mammals were trapped from FCAP-1, FCAP-2, and the Sluice locations (Fig. 2.2). Small mammals were also trapped from Walker Branch watershed, a reference site (Fig. 2.1). Seventy-five Sherman live traps were placed on an 11 by 7 trap grid, approximately 3 m apart, at both FCAP-1 and FCAP-2 (only 5 traps in 11th row). Forty pit-fall traps, spaced approximately 5 m apart, were also installed at FCAP-1. Pit-fall traps were only used at FCAP-1 to improve the success of capturing shrews. These traps did not enhance trapping success and, therefore, were not employed at other locations.

One hundred Sherman live traps were placed on a 10 by 10 trap grid approximately 3 m apart at both the sluice and Walker Branch sites. The traps were baited with a mixture of peanut butter and oats and checked daily. Trapped animals were sacrificed for analysis. Small mammals were identified to species according to Burt and Grossenheider (1976) and body measurements (weight, body length, and tail length) were recorded. Whole-body samples were then washed with Prell® shampoo to remove external contamination and thoroughly rinsed with deionized water. Samples were then stored at  $-20^{\circ}\text{C}$  in a locked freezer until submission for laboratory analysis. One laboratory rinsate sample per ten small mammal samples was also analyzed to determine if the processing procedure (i.e. use of Prell® shampoo) was a source of contamination.

Samples were shipped overnight in a cooler containing dry ice to Southwest Research Institute (SWRI) for analysis. Staff at SWRI homogenized each whole-body sample using a stainless steel blender. Three 2-g aliquots of homogenate were completely digested with boiling nitric and perchloric acid until total dissolution. The digested samples were analyzed

for As, Cd, Cr, Pb, Se, and Tl by GFAA; Hg analyses were performed by CVAA. Metal concentrations were reported on both a wet-weight and dry-weight corrected basis.

### 5.2.3 Statistical Analyses

Heavy metal concentrations, standard deviations, and the coefficient of variation (CV) were calculated from the 3 aliquots analyzed for each animal. The CV is the standard deviation expressed as a fraction of the mean, and is used to compare variation among aliquots independent of the magnitude of their means. The CV values were used to evaluate the degree of homogeneity achieved by the grinding procedure.

For risk assessment purposes, upper tolerance limits (to be defined later) were determined for all small rodents collected in the upper McCoy Branch watershed. Animals used to calculate tolerance limits and total rodent means in upper McCoy Branch watershed include: white-footed mice (*Peromyscus leucopus*), harvest mice (*Reithrodontomys* sp.), and a pine vole (*Microtus pinetorum*) collected from FCAP-1, FCAP-2, and Sluice areas. Insectivores, such as the short-tailed shrew (*Blarina brevicauda*), were excluded from these calculations for various reasons. These reasons include (1) shrews tended to have much higher metal concentrations than other genera; (2) shrews have different dietary habits; (3) shrews comprise a small percentage of the diet of most mammalian predators (Greenburg et al. 1988; Story et al. 1982); and (4) shrews may have increased exposure due to a fossorial lifestyle. Tolerance limits were not calculated for rodents collected at Walker Branch due to extremely low sample size.

Tolerance limits are used to indicate the limits within which a proportion (P) of a population can be expected to be found. Knowing the population mean (X) and standard deviation (S), it is possible to determine a constant K such that we can assert, with 95% confidence, that the proportion of the population contained between  $X - KS$  and  $X + KS$  is at least P (Hines and Montgomery 1980). With 95% confidence, the conclusion could be reached that a proportion of the population (e.g., 99%) would be less than the upper tolerance limit. The tolerance limits were calculated using the log-transformed means of metal concentrations, which were found to more nearly satisfy the assumptions for normality and homogeneity of variance required for conventional parametric statistical analyses. Thus, there is a 95% certainty that 99% of the heavy metal distribution is less than the upper limit. However, there are limitations for the use of tolerance limits. Small sample sizes in conjunction with large variability of heavy metal concentrations may result in unrealistic tolerance limits.

Analysis of variance procedures (PROC GLM; SAS 1988) were performed to determine if metal contamination differed among sites or species (*Peromyscus* vs *Reithrodontomys*). Because *Peromyscus* was the most commonly trapped small mammal from the FCAP, Sluice, and Walker Branch sites, a comparison of differences of heavy metal concentrations between sites was conducted using the log-transformed means for this species.

## 5.3 RESULTS

### 5.3.1 Vegetation

Heavy metal concentrations for individual plant species collected at Walker Branch watershed (Control), FCAP-1, FCAP-2, and the sluice sites are presented in Table A.1 (Appendix A). Average concentrations of metals in deciduous trees, eastern red cedar, and groundcover collected from all sites are summarized in Table 5-1.

**Thallium** - Concentrations of thallium in plants from the United States do not normally exceed  $0.5 \mu\text{g/g}$  ash (Gough et al. 1979), or  $\approx 7 \mu\text{g/g}$  dry mass (based on an ash weight:dry weight ratio of 0.075 for leaves). Thallium was not present above the detection limit of  $1 \mu\text{g/g}$  dry mass in any of the 70 plant samples analyzed (Table A.1).

**Lead** - Lead was not above the detection limit of  $1 \mu\text{g/g}$  dry mass in any control samples or in samples from the sluice site (Table A.1). Two vegetation samples from FCAP-1 had lead concentrations between 1 and  $2 \mu\text{g/g}$  dry mass, but 95 % of the vegetation samples from the FCAP were less than the detection limit.

**Arsenic** - Arsenic in control samples was not above the detection limit ( $1.00 \mu\text{g As/g}$  dry mass). Slightly elevated mean As concentrations were found in deciduous tree leaves ( $1.53$  to  $1.83 \mu\text{g/g}$ ) at the FCAP-1 and FCAP-2. These mean As concentrations were significantly different from controls ( $p < 0.01$ ).

**Cadmium** - Cadmium was detected in 1 of 19 control samples, and the concentration ( $0.30 \mu\text{g/g}$ ) was slightly above the detection limit ( $0.25 \mu\text{g Cd/g}$  dry mass). Cadmium was also detected in 1 of 12 samples collected at the sluice site (the concentration was  $0.60 \mu\text{g/g}$ ; all other samples were at or below the detection limit). Under natural conditions, Cd concentrations in plants are low (Shacklette 1972), and concentrations exceeding  $3 \mu\text{g Cd/g}$  in plant tissues are associated with reduced growth (Gough et al. 1979). Cadmium concentrations at the FCAP were not outside of the range expected for vegetation from natural environments (Connor and Shacklette 1975).

**Chromium** - Chromium was not above the detection limit of  $3 \mu\text{g/g}$  dry mass in plant samples from the control or sluice locations. With the exception of a few samples, Cr was not found above the analytical detection limit in leaves from trees sampled on the FCAP. However, all samples of ground cover (honeysuckle and grasses) at the FCAP had detectable levels of Cr. The average concentration was  $10.07 \mu\text{g/g}$ . Based on plants growing in serpentine soils, toxicity symptoms can occur at foliar concentrations exceeding  $18 \mu\text{g Cr/g}$  dry mass (Gough et al. 1979).

**Mercury** - Mercury concentrations in plants from the FCAP and sluice sites did not exceed those found in deciduous tree leaves from the control site. In most instances, Hg concentrations in plants from the FCAP were at or below the limit of detection ( $0.05 \mu\text{g Hg/g}$  dry matter). Mercury concentrations in tree leaves from the control site were actually greater than those at FCAP-1 or the sluice site ( $p < 0.001$ ). The reported range of natural Hg concentrations in plants from Missouri is  $<0.025$  to  $0.050 \mu\text{g/g}$  dry matter (Connor and Shacklette 1975).

**Selenium** - Selenium concentrations in leaves from deciduous trees at FCAP-1 and FCAP-2 were significantly greater than those found in trees from the control and sluice sites ( $p < 0.001$ ). The highest Se levels were found in leaves from deciduous trees at FCAP-2 (mean =  $29.1 \mu\text{g/g}$ , Table 5.1). Ground covers and eastern red cedar at the FCAP exhibited Se concentrations that were less than those found in leaves from deciduous trees ( $p < 0.05$ ), but were higher than reference site concentrations. Sensitivity to Se toxicity is so variable



Table 5.1. Mean ( $\pm$  SD) concentrations of metals detected in leaves from deciduous trees, eastern red cedars, and ground covers from a control area (Walker Branch watershed), FCAP-1, FCAP-2, and the sluice area

Less than values represent contaminant levels below the detection limit

Vegetation type	Location	n	Concentration ( $\mu\text{g/g}$ dry matter)					
			As	Cd	Cr	Pb	Hg	Se
Deciduous trees <sup>a</sup>	Control	14	1.01 $\pm$ 0.05 <sup>b</sup>	0.25 $\pm$ 0.01	< 3.00	< 1.00	0.11 $\pm$ 0.05 <sup>b</sup>	< 1.00 <sup>b</sup>
	FCAP-1	18	1.53 $\pm$ 0.76 <sup>b</sup>	0.62 $\pm$ 0.66 <sup>b</sup>	3.04 $\pm$ 0.19	< 1.00	0.06 $\pm$ 0.02 <sup>c</sup>	14.8 $\pm$ 12.7 <sup>c</sup>
	FCAP-2	12	1.83 $\pm$ 0.58 <sup>b</sup>	1.74 $\pm$ 0.96 <sup>c</sup>	< 3.00	1.07 $\pm$ 0.23	< 0.05 <sup>c</sup>	29.1 $\pm$ 24.5 <sup>d</sup>
	Sluice	6	< 1.00 <sup>b</sup>	0.31 $\pm$ 0.14 <sup>b</sup>	< 3.00	< 1.00	0.07 $\pm$ 0.01 <sup>c</sup>	1.55 $\pm$ 0.87 <sup>b</sup>
Eastern red cedar	Control	2	< 1.00	< 0.25	< 3.00	< 1.00	< 0.05	< 1.00
	FCAP-1	3	< 1.00	< 0.25	< 3.00	< 1.00	< 0.05	2.43 $\pm$ 0.42
	Sluice	3	< 1.00	< 0.25	< 3.00	< 1.00	0.06 $\pm$ 0.01	< 1.00
Ground covers	Control	3	1.07 $\pm$ 0.12	< 0.25	< 3.00	< 1.00	< 0.05	< 1.00
	FCAP-1	6	1.07 $\pm$ 0.12	< 0.25	10.07 $\pm$ 5.44	1.02 $\pm$ 0.04	< 0.05	2.12 $\pm$ 0.76
	Sluice	3	< 1.00	< 0.25	< 3.00	< 1.00	< 0.05	1.37 $\pm$ 0.21

<sup>a</sup>Deciduous trees included the following: red maple, sourwood, dogwood, American beech, sweet gum, sycamore, boxelder, cottonwood, and willows. Ground covers were limited to a grass (*Eutalia*) and Japanese honeysuckle.

<sup>b,c,d</sup>For deciduous trees, values in the same column with different superscripts are significantly different ( $p < 0.05$ ).

among plants that general toxicity levels for plants cannot be established (Gough et al. 1979). Vegetation growing in seleniferous areas of Canada contain from 3 to 4000  $\mu\text{g Se/g dry matter}$  (Gough et al. 1979).

### 5.3.2 Small Mammals

A total of 24 small mammals were trapped from the FCAP, Sluice, and Walker Branch locations in 2,980 trap-nights (Table 5.2). No animals were caught in the pitfall traps at FCAP-1. Tolerance limits for rodents at upper McCoy Branch watershed ranged from 0.165  $\mu\text{g/g}$  for cadmium to 26.98  $\mu\text{g/g}$  for selenium (Table 5.3). Mean concentrations of metals (wet-weight), derived from individual animal means are also summarized in Table 5.3. Mercury (sluice and Walker Branch) and Cr (Walker Branch) concentrations found in *Blarina* exceed maximum levels for all other small mammal species collected (Table 5.4).

Wet and dry-weight corrected raw data and summary statistics for whole body metal concentrations from small mammals from the FCAP, sluice, and Walker Branch locations are provided in Tables A.2-A.5. Most wet weight metal concentrations were qualified as estimated during the validation process (Table A.2).

Variation among the three aliquots from each animal is due to analytical variability (including homogenization). The EPA specifies that the relative percent difference (RPD) values for duplicate samples in a solid matrix should be less than 35% if the reported value is at least five times the contract required detection limit (CRDL) or minimum detection limit (MDL) (Engels 1992). A CV value of 25% is equivalent to an RPD of 35%. Distribution percentiles were calculated using all 24 small mammal samples for those analytes where there were no values below the detection limit:

Se: 99% of the CV values were < 20.1%

Pb: 75% of the CV values were < 30.5%

Hg: 95% of the CV values were < 24.2%

Cr: 75% of the CV values were < 23.1%

Except for Pb, CV distributions for analytes indicate that analytical variability was within acceptable limits, thus suggesting that the homogenization process was adequate.

**Thallium** - Thallium was only detected in two small mammals collected from FCAP-1 (1 *Peromyscus*) and FCAP-2 (1 *Reithrodontomys*). One aliquot for each animal had a detectable thallium value (mean = 0.243  $\mu\text{g/g}$  and 0.202  $\mu\text{g/g}$  respectively; Table A.2). Thallium was not detected in any animals from Walker Branch.

**Lead** - Lead was detected in small mammals ( $n = 24$ ; 72 aliquot samples) from all locations. Concentrations ranged from 0.094  $\mu\text{g Pb/g}$  to 10.27  $\mu\text{g Pb/g}$  (Table A.3). Lead concentrations found in rodents at upper McCoy Branch were slightly greater than those found in *Peromyscus* at Walker Branch ( $1.030 \pm 2.408 \mu\text{g/g}$  and  $0.334 \pm 0.228 \mu\text{g/g}$ , respectively; Table 5.3). Although lead concentrations in rodents collected from upper McCoy Branch locations were relatively low (< 0.82  $\mu\text{g Pb/g}$  for 15 of 17 animals), one animal (a *Reithrodontomys* from FCAP-2) had an average concentration of 10.27  $\mu\text{g Pb/g}$ . When excluding the outlier, the tolerance limit (20.10  $\mu\text{g/g}$ ; Table 5.3) would be substantially lower.

**Arsenic** - Arsenic was detected in 37 of 54 and 3 of 18 small mammal aliquots (3 aliquots per small mammal) collected from upper McCoy Branch and Walker Branch locations, respectively (Table A.2). The mean arsenic concentration measured in *Peromyscus* at the FCAP is significantly higher ( $p < 0.05$ ) than that found in *Peromyscus* at the Sluice or Walker Branch locations (mean =  $0.171 \pm 0.121 \mu\text{g As/g}$  (FCAP),  $0.051 \pm 0.001 \mu\text{g As/g}$  (Sluice), and  $0.054 \pm 0.007 \mu\text{g As/g}$  (Walker Branch), respectively; Table 5.3).

Table 5.2. Species (number of animals trapped), collection dates, number of trap nights, and number of animals collected per trap-night at FCAP-1, FCAP-2, Sluice, and Walker Branch locations

Site	FCAP-1	FCAP-2	Sluice	Walker Branch
Species	White-footed mice (7) ( <i>Peromyscus leucopus</i> )	White-footed mice (1)  Eastern harvest mice (3) ( <i>Reithrodontomys humulus</i> )  Fulvous harvest mice (1) ( <i>Reithrodontomys fulvescens</i> )	White-footed mice (4)  Short-tailed shrew (1) ( <i>Blarina brevicauda</i> )  Pine vole (1) ( <i>Microtus pinetorum</i> )	White-footed mice (4)  Short-tailed shrew (2)
Dates collected	June 23-29, 1993	August 5-18, 1993	September 9-14, 1993	September 29 - October 13, 1993
Trap-nights	525	675	480	1300
Animals/ Trap-nights	0.0133	0.0074	0.0125	0.0046

Table 5.3. Mean concentrations ( $\pm$  SD, range in parentheses) ( $\mu\text{g/g}$ ) of metals derived from individual animal means for each genus and all rodents collected at upper McCoy Branch, FCAP, Sluice, and Walker Branch areas

Analyte	Upper McCoy Branch		FCAP			Sluice		Walker Branch	
	Upper McCoy Branch rodents <sup>a</sup>	Tolerance limit <sup>b</sup>	<i>Peromyscus</i> (n=8)	<i>Reithrodontomys</i> (n=4)	<i>Peromyscus</i> (n=4)	<i>Microtus</i> (n=1)	<i>Peromyscus</i> (n=4)		
As	0.131 $\pm$ 0.098 (0.050 - 0.434)	0.924	0.171 $\pm$ 0.121 <sup>c</sup> (0.081 - 0.434)	0.139 $\pm$ 0.064 <sup>cd</sup> (0.060 - 0.216)	0.051 $\pm$ 0.001 <sup>d</sup> (0.050 - 0.053)	0.093 $\pm$ 0.013	0.054 $\pm$ 0.007 <sup>d</sup> (<0.05 - 0.064)		
Cd	0.022 $\pm$ 0.016 (0.007 - 0.065)	0.174	0.021 $\pm$ 0.014 <sup>c</sup> (0.008 - 0.047)	0.028 $\pm$ 0.026 <sup>c</sup> (<0.010 - 0.065)	0.018 $\pm$ 0.014 <sup>c</sup> (0.007 - 0.038)	0.022 $\pm$ 0.003	0.022 $\pm$ 0.007 <sup>c</sup> (0.013 - 0.027)		
Cr	1.256 $\pm$ 0.354 (0.987 - 2.253)	2.706	1.389 $\pm$ 0.477 <sup>c</sup> (0.987 - 2.253)	1.116 $\pm$ 0.095 <sup>c</sup> (1.004 - 1.227)	1.083 $\pm$ 0.038 <sup>c</sup> (1.035 - 1.117)	1.457 $\pm$ 0.129	1.587 $\pm$ 1.498 <sup>c</sup> (0.679 - 3.823)		
Pb	1.030 $\pm$ 2.408 (0.094 - 10.267)	20.102	0.520 $\pm$ 0.469 <sup>c</sup> (0.139 - 1.453)	2.662 $\pm$ 5.070 <sup>c</sup> (0.094 - 10.267)	0.558 $\pm$ 0.234 <sup>c</sup> (0.271 - 0.821)	0.476 $\pm$ 0.054	0.334 $\pm$ 0.228 <sup>c</sup> (0.124 - 0.648)		
Hg	0.037 $\pm$ 0.025 (0.011 - 0.105)	0.268	0.033 $\pm$ 0.014 <sup>c</sup> (0.016 - 0.056)	0.051 $\pm$ 0.045 <sup>c</sup> (0.012 - 0.106)	0.038 $\pm$ 0.015 <sup>c</sup> (0.025 - 0.053)	0.011 $\pm$ 0.002	0.027 $\pm$ 0.007 <sup>c</sup> (0.018 - 0.034)		
Se	1.831 $\pm$ 1.481 (0.329 - 5.640)	26.980	2.563 $\pm$ 1.580 <sup>c</sup> (0.446 - 5.64)	2.007 $\pm$ 1.222 <sup>c</sup> (.644 - 3.387)	0.506 $\pm$ 0.184 <sup>d</sup> (0.329 - 0.764)	0.569 $\pm$ 0.064	0.261 $\pm$ 0.012 <sup>d</sup> (0.248 - 0.277)		
Tl	0.203 $\pm$ 0.010 (<.200 - 0.243)		0.205 $\pm$ 0.015 <sup>c</sup> (< 0.200 - 0.243)	0.200 $\pm$ 0.001 <sup>c</sup> (<0.200 - 0.202)	<0.200 <sup>c</sup>	<0.200	< .200		

<sup>a</sup>Upper McCoy Branch Rodents: Mean concentration found in all rodents collected within Chestnut Ridge Operable Unit 2.

<sup>b</sup>Tolerance Limits calculated for maximum contaminant concentration that may be found in 99% of the small mammal population with a confidence level of 95%. Tolerance limit calculated only for rodents at upper McCoy Branch.

<sup>c</sup>Means with like superscripts do not differ significantly ( $p > 0.05$ ) for each Analyte.

<sup>d</sup>Tolerance limits could not be calculated for non-detectable values.

Table 5.4. Mean concentrations ( $\mu\text{g/g}$ ) of metals detected in *Blarina brevicauda* at Walker Branch watershed<sup>a</sup> (n=2) and the Sluice site<sup>b</sup> (n=1)

Analyte	Walker Branch	Sluice
As	0.059 $\pm$ 0.012	0.177 $\pm$ 0.016
Cd	0.066 $\pm$ 0.013	0.073 $\pm$ 0.014
Cr	3.647 $\pm$ 2.522	0.797 $\pm$ 0.139
Hg	0.337 $\pm$ 0.145	0.481 $\pm$ 0.121
Pb	1.533 $\pm$ 0.316	0.677 $\pm$ 0.108
Se	1.195 $\pm$ 0.342	2.330 $\pm$ 0.176
Tl	< 0.200	< 0.200

<sup>a</sup>Mean metal concentrations in two individual shrews collected at Walker Branch watershed are calculated using three replicates per individual.

<sup>b</sup>Mean metal concentrations in single individual shrew collected at the Sluice Site was calculated using three replicates.

**Cadmium** - Cadmium was detected in 36 of 54 and 17 of 18 small mammal aliquots (3 aliquots per small mammal) collected from upper McCoy Branch and Walker Branch locations, respectively (Table A.2). Mean cadmium concentrations in rodents from upper McCoy Branch sites ( $0.022 \pm 0.016 \mu\text{g Cd/g}$ ) was not significantly different ( $p = 0.749$ ) from levels in rodents collected at Walker Branch ( $0.022 \pm 0.007 \mu\text{g Cd/g}$ ) (Table 5.3). Cadmium concentrations in small mammals found at upper McCoy Branch and Walker Branch are below reported concentrations found at other reference sites (0.1 to  $1.4 \mu\text{g/g}$ ; Talmage and Walton 1991).

**Chromium** - Chromium was detected in small mammals collected (n= 24; 72 aliquot samples) at all locations having mean concentrations ranging from  $0.679 \mu\text{g Cr/g}$  to  $5.4 \mu\text{g Cr/g}$  (Table A.3). Levels of chromium in rodents at upper McCoy Branch locations were not significantly different ( $p = 0.687$ ) from concentrations found in rodents at Walker Branch (mean =  $1.256 \pm 0.354 \mu\text{g/g}$  and  $1.587 \pm 1.498 \mu\text{g/g}$ , respectively). Acute and chronic adverse effects of chromium to mammals are caused mainly by Cr VI compounds (Eisler 1986). In most soils Cr is primarily present as precipitated  $\text{Cr}^{+3}$  and is not bioavailable, with little evidence of Cr biomagnifying through food chains in its inorganic form (Eisler 1986). Since these analyses resulted in total chromium values, the amount of Cr VI present in the soil and biota was not determined.

**Mercury** - Mercury was detected in small mammals collected (n=24; 72 aliquot samples) at all locations. Mean concentrations ranged from  $0.012 \mu\text{g Hg/g}$  to  $0.481 \mu\text{g Hg/g}$ . Mercury concentrations found in rodents from upper McCoy Branch sites were higher than concentrations found in rodents from Walker Branch. However, this difference was not statistically significant ( $p = 0.573$ ). Mercury concentrations found in *Blarina* (Table 5.4), at both sluice ( $0.481 \pm 0.121 \mu\text{g Hg/g}$ ) and Walker Branch ( $0.337 \pm 0.145 \mu\text{g Hg/g}$ ) were substantially higher (approximately 13 times greater for *Blarina* at the sluice site) than concentrations found in rodents ( $0.037 \pm 0.025 \mu\text{g Hg/g}$ ) at upper McCoy Branch.

**Selenium** - Selenium was detected in small mammals collected (n=24; 72 aliquot samples) at all locations with mean concentrations ranging from  $0.248 \mu\text{g Se/g}$  to  $5.64 \mu\text{g}$

Se/g. The mean selenium concentration found in rodents (*Peromyscus* and *Reithrodontomys*) at the FCAP was significantly higher ( $p < 0.05$ ) than concentrations found in *Peromyscus* at the sluice and Walker Branch locations (Table 5.3).

## 5.4 DISCUSSION

### 5.4.1 Vegetation

Chromium and selenium were the metals found in highest concentrations in plants from sites at the FCAP. Following root uptake, there is little translocation of Cr to aboveground plant parts (Parr and Taylor 1980). Therefore, elevated concentrations of Cr in ground covers on the FCAP are probably due to the resuspension of fine dust to leaf surfaces (leaves were not washed prior to analysis in this study). Previous studies have shown no increase in Cr and Pb uptake by woody plants growing on fly ash, which indicates that these elements are fixed in fly ash and are not present in bioavailable forms (Scanlon and Duggan 1979).

The highest Se concentration observed for tree leaves from the FCAP was 97  $\mu\text{g/g}$  dry mass. For comparison, severe toxicity to plants (more than 80% yield reduction) in bush beans and alfalfa is associated with Se concentrations of 900 to 1000  $\mu\text{g/g}$  dry leaf mass (Wallace et al. 1980; Wan et al. 1988). However, for many agricultural plants the level of tolerance to Se is  $< 50 \mu\text{g/g}$  dry mass (Wan et al. 1988). Alfalfa yields can be reduced by 10% when Se concentrations in plant tissues exceed 25 to 30  $\mu\text{g/g}$  dry mass (Soltanpour and Workman 1980). Comparable reductions in yield have been observed in bush beans when foliar levels reach 25  $\mu\text{g Se/g}$  dry mass (Wallace et al. 1980). Based on measured concentrations and reported toxicity levels in agricultural plants, there is some potential for Se toxicity in trees growing on the FCAP. However, due to the widely varying resistance to Se toxicity in plants (Gough et al. 1979) and because no distinction has been made between internal and external contamination in this first survey, the importance of potential Se toxicity to plants at the FCAP is not known. This possibility needs to be evaluated in follow-up studies of Se toxicity to tree seedlings. It is unlikely that the Se concentrations measured in tree leaves can be attributed to surface contamination by resuspended dust because leaves were usually sampled at a height of more than 1 m from ground level.

Concentrations of Se in plants on the FCAP are high enough to warrant further study of possible toxicity effects on small mammals and soil invertebrates. The influx of Se to soil by means of autumn leaf fall may present some potential for toxicity to litter fauna. Complexation with organic matter, adsorption of selenium to Fe and Al oxides and oxyhydroxides, and high sulfate concentrations in soil all tend to reduce the root uptake of Se (Logan et al. 1987). Wild plants (white sweet clover) growing in fly ash have been found to accumulate over 200 ppm of Se (Gutenmann et al. 1976). Selenium accumulates in protein-containing tissues of plants, like seeds, because of biochemical and metabolic similarities to sulfur (Adriano 1986). No information is currently available on the levels of Se in seeds from the FCAP. However, studies suggest that metal concentrations in seeds and fruits are equal to or less than those in foliage (Scanlon and Duggan 1979; Arthur et al. 1992a and b; Energy Systems 1994). Concentrations of Se in seeds comparable to those measured in tree leaves ( $\approx 20 \mu\text{g/g}$ ) could be of concern from the standpoint of toxicity to small mammals. As a point of reference, Se concentrations in feeds  $> 5 \mu\text{g/g}$  dry mass are considered potentially toxic to animals (Gough et al. 1979; Adriano 1986).

The ecological risk assessment performed for Chestnut Ridge OU 2 RI/FS indicated that growth and possibly reproduction of plants at FCAP, sluice, and upper McCoy Branch sites may be depressed by  $\geq 20\%$  based on results of media analyses. Ag, Al, Sb, As, Ba, Cd, Cr, Co, Fe, Hg, Mn, Ni, Se, V, and Zn were all found in ash, soil, and shallow groundwater at concentrations high enough to cause reduced growth. FCAP ash tended to have higher concentrations of all contaminants than the sluice or UMB ashes. Al, As, Se, and V represented the highest risks to plants (Energy Systems 1994).

#### 5.4.2 Small Mammals

Cadmium, chromium, lead, mercury, and thallium concentrations in small mammals collected from upper McCoy Branch were similar to concentrations in animals collected from the reference site. However, one small mammal living on FCAP-2 had an average lead concentration of  $10.27 \mu\text{g/g}$  ( $28.107 \text{ ppm}$  dry weight); the tolerance limit for the population was  $20.1 \mu\text{g Pb/g}$ . Previous studies have shown that comparable lead body burdens did not appear to adversely affect or impact populations of western harvest mice ( $3.1\text{--}10.8 \mu\text{g/g}$  dry weight), white-footed mice ( $17 \mu\text{g/g}$  dry weight), and deer mice ( $> 50 \mu\text{g/g}$  fresh weight) (Getz et al. 1977; Beyer et al. 1985b; Blus et al. 1987). However, exposure to elevated levels of lead can adversely affect survival, growth, reproduction, development, behavior, learning and metabolism. Specifically, lead inhibits blood delta aminolevulinic acid dehydratase (ALAD) activity in mice at concentrations of  $\geq 0.05 \mu\text{g Pb/g}$  body weight (Eisler 1988a). Cellular alterations were also associated with renal lead levels of  $8.1 \mu\text{g Pb/g}$  in small mammals (Goyer et al. 1970). Additionally, survival was reduced by acute oral doses as low as  $5 \mu\text{g/g}$  body weight in rats (Eisler 1988a; Schroeder et al. 1965).

Arsenic and selenium were found in significantly higher ( $p < 0.05$ ) concentrations in *Peromyscus* at the FCAP compared to *Peromyscus* collected at reference and sluice locations. *Riethodontomys* sp. at FCAP-2 also had significantly higher metal concentrations ( $p < 0.05$ ) as compared to *Peromyscus* at the reference site. The mean concentration ( $1.83 \pm 1.48 \mu\text{g/g}$ ) and upper tolerance limit for Se ( $26.98 \mu\text{g/g}$ ) in rodents at upper McCoy Branch exceed documented toxicological values for small mammals and have potential repercussions for predators (e.g., red fox, red-tailed hawk). In mammals, chronic Se poisoning is induced by diets containing 1 to 44 ppm Se (Harr 1978). Symptoms include liver cirrhosis, lameness, loss of hair, emaciation, reduced conception and increased fetal resorption (Lo and Sandi 1980). Fishbein (1977) reported minimum toxic concentrations of Se for a lifetime exposure in rats of  $0.35 \mu\text{g Se/g}$ ; this concentration in the diet caused changes in liver chemistry. Longevity and histology of the heart, kidney, and spleen were also modified when rats were exposed to a  $0.75 \mu\text{g Se/g}$  diet. Selenium can cause congenital malformations in mice, rats, swine and cattle (Harr 1978). Selenium is also embryotoxic and teratogenic to birds (mallards) (Hoffman and Heinz 1988).

Arsenic was also found in concentrations (upper McCoy Branch rodent mean =  $0.131 \pm 0.098 \mu\text{g/g}$ ; tolerance limit =  $1.087 \mu\text{g/g}$ ) which may cause adverse effects in rodents and their predators. Arsenic is a carcinogen and teratogen. Other effects include reduced growth, hearing/sight loss, liver/kidney damage, and death (Eisler 1988b). Wildlife mortality and malformations have been observed for chronic doses of  $1\text{--}10 \mu\text{g/g}$  body weight and dietary concentrations of  $5\text{--}50 \text{ ppm}$  (Eisler 1988b). Blakely et al. (1980) showed immuno-suppressive effects from a dose of  $0.10 \mu\text{g As/g}$  from mice drinking water containing  $0.5 \text{ mg As/L}$  (sodium arsenite) for 3 weeks. NRCC (1978) has shown that a maternal dose of  $5 \mu\text{g As}^{+5}/\text{g}$

BW (arsenate) in hamsters (*Cricetus* sp) and 10  $\mu\text{g}$   $\text{As}^{+5}/\text{g}$  in mice (*Mus* spp.) caused some fetal mortality and fetal malformations (mice only). Mice that were exposed to 5 ppm sodium arsenite in drinking water for three generations showed a reduction in litter size (Schroeder and Mitchener 1971). A dose of 0.38  $\mu\text{g}/\text{g}$  over a lifetime was sufficient to cause a slight decrease in the median lifespan of laboratory mice (Schroeder and Balassa 1967).

#### 5.4.3 Species and Site Specific Differences

Statistical analyses (PROC GLM/Tukeys; SAS) showed that Se and As concentrations in *Peromyscus* at FCAP-1 were higher ( $p < 0.05$ ) than concentrations in *Peromyscus* at the sluice and Walker Branch sites. Metal concentrations in *Peromyscus* at the sluice and Walker Branch sites were similar ( $p > 0.05$ ). As and Se concentrations found in *Reithrodontomys* were similar to that found in *Peromyscus* at the FCAP; however, As was not statistically different from *Peromyscus* at the sluice or Walker Branch sites. Therefore, it appears that mice in the FCAP area are bioaccumulating higher levels of Se than at other areas on upper McCoy Branch or the reference site.

A non-statistical comparison was made between *Blarina brevicauda* and rodents collected. Heavy metal concentrations, with the exception of As, in *Blarina* (sluice and Walker Branch sites) (Table 5.4) exceeded those in most *Peromyscus*. Average mercury concentrations in shrews were at least 9 times greater than average concentrations found in other species (Table 5.3; Table 5.4). Shrews have the potential to ingest larger concentrations of metals in their diet. Shrews are voracious feeders, ingesting an equivalent of their body weight each day. The shrew diet consists of earthworms, insects, millipedes, snails, slugs, and, occasionally, other small mammals (Talmage and Walton 1991; Burt and Grossenheider 1976). *Peromyscus* are opportunistic feeders, primarily herbivorous, whose diet consists of seeds, insects, fruit and green vegetation (Brown 1964). Boggess (1977) found higher metal body burden concentrations (e.g. lead) in insectivorous rodents relative to herbivores. Heavy metals present in the soil may also contribute to increased exposure in *Blarina* due to their burrowing lifestyle. Mean Se and Hg concentrations in *Blarina* collected at Walker Branch watershed and the sluice area exceed toxicological levels known to cause adverse effects in other organisms (Energy Systems 1994).

### 5.5 CONCLUSIONS

Metals in the ash, Se and As in particular, are taken up and bioaccumulated in the vegetation on the FCAP. Additionally, Cr was found in greater concentrations in groundcover at the FCAP compared to the sluice and reference sites. Se and As concentrations found in deciduous foliage at the FCAP were significantly greater than those found at the sluice and reference sites. Se concentrations may cause adverse effects on herbivorous wildlife, including small mammals and white-tailed deer (see ecological risk assessment; Energy Systems 1994). The influx of Se to soil by means of autumn leaf fall may also present some potential for toxicity to litter fauna.

Average concentrations of As and Se found in small mammals collected at the FCAP are significantly greater than those from the reference and sluice sites. Contaminant concentrations and tolerance limits exceed levels that may pose a toxicological risk to small mammals and their predators (see ecological risk assessment; Energy Systems 1994).



Although Cd, Hg, and Cr bioaccumulated in small mammals on upper McCoy Branch sites, concentrations were relatively low. Because heavy metals, with the exception of Tl, were found in small mammals at the FCAP, these animals serve as a potential pathway for contaminant transfer to predators and other wide-ranging species.

## 5.6 FUTURE STUDIES

Future studies performed on the terrestrial component of upper McCoy Branch will depend on the remedial alternatives selected as a result of the feasibility study. Studies will include monitoring activities to determine effectiveness of the designated remedial action. The likely remedial alternative includes surface water controls, dam stabilization, and environmental improvements. Environmental improvements involve wetland development and enhancement at the toe of the dam, placement of salt-licks for deer consumption surrounding the FCAP, and nutrient (sulfur and phosphorus) and organic matter addition to the ash on the FCAP and Sluice Channel area. The addition of nutrients and organic matter to the area will decrease Se and As uptake in vegetation and increase rate of vegetation growth. This addition will also increase the production of a leaf litter layer, thus forming a new soil protective layer for growth of new vegetation. Future studies would include heavy metal bioaccumulation in vegetation, monitoring natural succession of vegetation and overall quality and size of wetlands.

## 6. FISH POPULATION AND COMMUNITY ASSESSMENTS

(*E. M. Schilling and B. A. Carrico*)

### 6.1 INTRODUCTION

Fish population and community studies can be used to assess the ecological effects of changes in water quality and habitat. These studies offer several advantages over other indicators of environmental quality (see Karr et al. 1986; Karr 1987) and are especially relevant to assessing the biotic integrity of McCoy 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). Also, statements about the condition of the fish community are better understood by the general public (Karr 1981).

The fish populations and communities in the McCoy Branch watershed were evaluated in three separate studies. As a continuation of earlier sampling (Ryon et al. 1992), the fish community in lower McCoy Branch below Rogers Quarry was evaluated through semiannual quantitative samples. This area of the watershed is greatly influenced by proximity to Melton Hill Reservoir downstream and possibly by Rogers Quarry upstream. In conjunction with sampling conducted by the Fish Bioaccumulation Task (Sect. 4), limited studies were conducted on the fish populations of Rogers Quarry. These studies were primarily age determinations and assessments of abnormalities in a few selected species. A third section of the watershed, the stream below the FCAP and above Rogers Quarry, was studied by experimentally introducing a fish species, banded sculpin (*Cottus carolinae*), into the stream. This section of McCoy Branch had previously failed to support a resident fish community and banded sculpin were introduced to provide a mechanism to evaluate the recovery of this section of the stream.

### 6.2 LOWER McCOY BRANCH (*E. M. Schilling*)

The initial objectives of the instream fish monitoring task in lower McCoy Branch were (1) to characterize spatial and temporal patterns in the distribution and abundance of fishes in this section of McCoy Branch and (2) to document any effects on fish community structure and function resulting from implementation of remedial actions as discussed in the McCoy Branch RFI (Murphy and Loar 1988).

#### 6.2.1 Materials and Methods

Quantitative sampling of the fish population was conducted in MCK 1.6 by electrofishing biannually from fall 1990 through 1993. Similar quantitative samples were made at two

nearby reference sites [Grassy Creek kilometer (GCK) 2.4 and Scarboro Creek kilometer (SCK) 2.2] for comparisons of population parameters. Stream conditions of Grassy Creek declined; therefore, Scarboro Creek was added as a new reference site beginning in spring 1991. Grassy Creek was sampled in fall 1990 through fall 1992 and Scarboro Creek was sampled spring 1991 through fall 1993. The resulting data were used to (1) determine species composition, (2) estimate population size (numbers and biomass per unit area), and (3) determine length-frequencies of selected species. The lengths of the sampling reaches ranged from 44 to 49 m at McCoy Branch and from 36 to 60 m at the reference sites (Table 6.1). The sampling lengths in McCoy Branch and Grassy Creek were similar to those in 1989 and spring 1990 (Ryon et al. 1992).

#### 6.2.1.1 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 volts 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 volts) based on the specific conductance of the water. The 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 assist in the collection of stunned fish.

After 0.64-cm-mesh seines were placed across the upper and lower boundaries of the fish sampling reach 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 from each pass were collected and stored separately in perforated buckets downstream of the sampling reach during further sampling. When possible, aerators were used on buckets during processing to reduce mortality.

Following electrofishing, fish were anesthetized with MS-222 (tricaine methane-sulfonate), 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) 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, disposition (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 percent

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

Site <sup>a</sup> (km)	Date	Length (m)	Mean width (m)	Mean depth (m)	Surface area (m <sup>2</sup> )	Pool: riffle ratio
MCK 1.6	09/24/90	48	0.6	8.5	29	0.5
MCK 1.6	05/02/91	45	0.7	14.2	31	0.4
MCK 1.6	11/15/91	49	0.9	7.8	44	0.3
MCK 1.6	04/10/92	49	1.0	11.2	49	0.5
MCK 1.6	12/03/92	45	0.9	10.7	41	0.8
MCK 1.6	05/18/93	46	0.9	6.7	41	0.6
MCK 1.6	11/11/93	44	0.8	8.9	35	0.6
GCK 2.4	10/25/90	36	1.5	8.5	54	P <sup>b</sup>
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

<sup>a</sup>MCK = McCoy Branch kilometer; GCK = Grassy Creek kilometer; SCK = Scarboro Creek kilometer.

<sup>b</sup>P = Stream was a continuous pool; no riffle present.

cloud cover. Following completion of fish sampling, the length, width, and depth of the sampling reach were measured at each site.

#### 6.2.1.2 Population data analysis

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 per size class for each species. To calculate density and biomass per unit area, total numbers and biomass were divided by the water surface area ( $m^2$ ) 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).

#### 6.2.1.3 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 or if any unusual mortality had affected a size class (Anderson and Gutreuter 1983).

### 6.2.2 Results and Discussion

#### 6.2.2.1 Species richness and composition

A total of 23 species were collected in seven quantitative surveys of McCoy Branch from fall 1990 through fall 1993 (Table 6.2). In comparison, a total of six species were found at Grassy Creek in five surveys from fall 1990 through fall 1992; and a total of 16 species were found in six surveys of Scarboro Creek from spring 1991 through fall 1993. Species richness ranged from 8 to 15 species for the individual 7 surveys conducted at McCoy Branch, from 3 to 5 species for the individual 5 surveys conducted at Grassy Creek, and from 7 to 13 species for the 6 individual surveys conducted at Scarboro Creek (Table 6.3). Species richness ranged from 9 to 11 species for three earlier surveys conducted from May 1989 to May 1990 in McCoy Branch (Ryon et al. 1992).

The variability in species number and composition in McCoy Branch and Scarboro Creek is related to the proximity of these sites to Melton Hill Reservoir. Grassy Creek is located approximately twice as far from an embayment as either McCoy Branch or Scarboro Creek. The closeness of the reservoir allows species to be found in McCoy Branch and Scarboro Creek that are atypical for streams of that size. In comparison with Scarboro Creek and other streams in the area, several species expected to occur in McCoy Branch are lacking or have been found during only one sampling period and do not represent stable populations. Species that have been found during one sampling period include: central stoneroller (*Camptostoma anomalum*), striped shiner (*Luxilus chrysocephalus*), creek chub (*Semotilus atromaculatus*), white sucker (*Catostomus commersoni*), and redbreast sunfish (*Lepomis auritus*). The central stoneroller, creek chub, and redbreast sunfish have been collected six, four, and four times respectively out of the six sampling periods in Scarboro Creek. The absence of the northern hog sucker (*Hypentelium nigricans*) and banded sculpin at MCK 1.6 may reflect past extermination by coal ash discharges and the prevention of recolonization due to the presence of Melton Hill Reservoir. These species are typically found in streams and are not associated with lakes or reservoirs. Melton Hill Reservoir and its lotic waters may act as a barrier to migration of these fish species to McCoy Branch from nearby streams. The northern hog sucker and the banded sculpin were collected at SCK 2.2 in two and six, respectively, of the six sampling periods. The fish community in McCoy Branch contains species such as

Table 6.2. Fish species composition in McCoy Branch, Grassy Creek, and Scarboro Creek, fall 1990 through fall 1993

Species	Sites <sup>a</sup>		
	MCK 1.6 (7) <sup>b</sup>	GCK 2.4 (5)	SCK 2.2 (6)
<b>Clupeidae</b>			
Threadfin shad ( <i>Dorsoma petenense</i> )	1 <sup>c</sup>	-	-
<b>Cyprinidae</b>			
Central stoneroller ( <i>Campostoma anomalum</i> )	1	-	6
Spotfin shiner ( <i>Cyprinella spiloptera</i> )	3	-	3
Striped shiner ( <i>Luxilus chrysocephalus</i> )	1	1	-
Bluntnose minnow ( <i>Pimephales notatus</i> )	7	-	4
Fathead minnow ( <i>Pimephales promelas</i> )	2	-	1
Blacknose dace ( <i>Rhinichthys atratulus</i> )	3	5	6
Creek chub ( <i>Semotilus atromaculatus</i> )	1	5	4
<b>Catostomidae</b>			
White sucker ( <i>Catostomus commersoni</i> )	1	4	-
Northern hog sucker ( <i>Hypentelium nigricans</i> )	-	-	2
<b>Ictaluridae</b>			
Yellow bullhead ( <i>Ameiurus natalis</i> )	5	-	4
<b>Poeciliidae</b>			
Western mosquitofish ( <i>Gambusia affinis</i> )	1	-	1
<b>Atherinidae</b>			
Brook silverside ( <i>Labidesthes sicculus</i> )	1	-	-

Table 6.2 (continued)

Species	Sites <sup>a</sup>		
	MCK 1.6 (7) <sup>b</sup>	GCK 2.4 (5)	SCK 2.2 (6)
Cottidae			
Banded sculpin ( <i>Cottus caroliniae</i> )	-	2	6
Centrarchidae			
Redbreast sunfish ( <i>Lepomis auritus</i> )	1	-	4
Green sunfish ( <i>Lepomis cyanellus</i> )	7	2	6
Warmouth ( <i>Lepomis gulosus</i> )	2	-	-
Bluegill ( <i>Lepomis macrochirus</i> )	7	-	4
Redear sunfish ( <i>Lepomis microlophus</i> )	2	-	-
Hybrid sunfish	-	-	1
Spotted bass ( <i>Micropterus punctulatus</i> )	4	-	-
Largemouth bass ( <i>Micropterus salmoides</i> )	2	-	2
Percidae			
Greenside darter ( <i>Etheostoma blennioides</i> )	1	-	-
Blueside darter ( <i>Etheostoma jessiae</i> )	1	-	-
Snubnose darter ( <i>Etheostoma simoterum</i> )	4	-	-
Logperch ( <i>Percina caprodes</i> )	2	-	4
Number of species (N)	23	6	16

<sup>a</sup>MCK = McCoy Branch kilometer 1.6; GCK = Grassy Creek kilometer 2.4; SCK = Scarboro Creek kilometer 2.2.

<sup>b</sup>Numbers represent the number of sampling periods for each site.

<sup>c</sup>Numbers represent the number of sampling periods (N=1) that a given species was collected at the site and a '-' indicates that the species was not collected.

**Table 6.3. Total fish density (individuals/m<sup>2</sup>), total biomass (g/m<sup>2</sup>), and species richness in McCoy Branch, and in two reference streams, Grassy Creek and Scarboro Creek, for fall 1990-93, and spring 1991-93<sup>a</sup>**

Sampling periods	Sites <sup>b</sup>		
	MCK 1.6	GCK 2.4	SCK 2.2
Fall 1990			
Total density	1.64	3.41	NS <sup>c</sup>
Total biomass	11.35	5.80	
Species richness	8	3	
Spring 1991			
Total density	12.30	0.41	2.15
Total biomass	111.75	1.84	11.41
Species richness	15	4	7
Fall 1991			
Total density	1.03	4.35	6.19
Total biomass	7.97	5.63	26.55
Species richness	7	4	13
Spring 1992			
Total density	2.22	1.51	2.75
Total biomass	13.96	2.31	25.60
Species richness	7	3	11
Fall 1992			
Total density	3.87	4.37	4.91
Total biomass	9.80	4.00	32.21
Species richness	9	5	9
Spring 1993			
Total density	4.07	NS	4.04
Total biomass	43.58		42.23
Species richness	7		7
Fall 1993			
Total density	1.12	NS	7.56
Total biomass	2.93		48.33
Species richness	7		11

<sup>a</sup>The months sampled in the spring and fall were: September 1990 for McCoy Branch; October 1990 for Grassy Creek; May 1991 for McCoy Branch and Scarboro Creek; March 1991 for Grassy Creek; November 1991 for McCoy Branch, Grassy Creek, and Scarboro Creek; April 1992 for McCoy Branch, Grassy Creek, and Scarboro Creek; December 1992 for McCoy Branch, Grassy Creek, and Scarboro Creek; May 1993 for McCoy Branch, Grassy Creek, and Scarboro Creek; October 1993 for Grassy Creek and Scarboro Creek; and November 1993 for McCoy Branch.

<sup>b</sup>MCK=McCoy Branch kilometer; GCK=Grassy Creek kilometer; SCK=Scarboro Creek kilometer.

<sup>c</sup>NS= Not sampled



threadfin shad (*Dorsoma petenense*) and brook silverside (*Labidesthes sicculus*) that move into and out of the stream from the reservoir and have been collected only during one sampling period.

The logperch (*Percina caprodes*) has been found at both MCK 1.6 and SCK 2.2. This species also can migrate from the embayments into the streams. No other darter species have been found at SCK 2.2. In contrast, the snubnose darter (*Etheostoma simoterum*) was found at MCK 1.6 in four of the seven sampling periods. Two other darter species, greenside darter (*E. blennioides*) and blueside darter (*E. jessiae*), were each represented by a single specimen collected at MCK 1.6. These collections of greenside and blueside darters do not represent stable populations, but do indicate that the species is present downstream of the site or that they have immigrated from the embayment.

#### 6.2.2.2 Density and biomass

Population surveys of McCoy 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 Appendix B, Tables B-1 through B-7.

Densities at MCK 1.6 for the seven sampling periods ranged from 1.03 to 12.03 fish/m<sup>2</sup> (Table 6.3). The density of 12.03 fish/m<sup>2</sup> in spring 1991 included 6.13 fish/m<sup>2</sup> of bluegill (*Lepomis macrochirus*) and 3.19 fish/m<sup>2</sup> of green sunfish (*L. cyanellus*). Later samples did not include as many bluegill and green sunfish. These high densities may be affected by immigration from Melton Hill Reservoir and/or Rogers Quarry. Densities at GCK 2.4 and SCK 2.2 ranged from 0.41 to 4.37 fish/m<sup>2</sup> and 2.15 to 7.56 fish/m<sup>2</sup> respectively. The community at MCK 1.6 was dominated by bluntnose minnow (*Pimephales notatus*), bluegill, and green sunfish. During two of the sampling periods, yellow bullhead (*Ameiurus natalis*) and spotfin shiner (*Cyprinella spiloptera*) were also dominant species. The dominant species at GCK 2.4, blacknose dace (*Rhinichthys atratulus*), creek chub, and white sucker, were present in low numbers in McCoy Branch in three of the seven surveys. Scarborough Creek was dominated by blacknose dace, banded sculpin, and central stoneroller. Central stonerollers were present in low numbers in McCoy Branch during only one sampling period and banded sculpin was absent altogether. Bluntnose minnow and green sunfish were also dominant species at SCK 2.2 in terms of density in three and two, respectively, of the six surveys.

Fish biomass at MCK 1.6 ranged from 2.93 to 111.75 g/m<sup>2</sup> from fall 1990 through fall 1993 (Table 6.3). Spring 1991 biomass (111.75 g/m<sup>2</sup>) was very high compared to other sampling periods and to the reference streams and was represented primarily by bluegill and green sunfish. Fish biomass at the reference sites ranged from 1.84 to 5.63 g/m<sup>2</sup> at GCK 2.4 and 11.41 to 48.33 g/m<sup>2</sup> at SCK 2.2. In general, the dominant species in terms of biomass paralleled the trends observed in population densities.

#### 6.2.2.3 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,

bluntnose minnow, bluegill, and green sunfish, collected in McCoy Branch for the sampling periods of fall 1990 through fall 1993 (Appendix C, Figs C-1 through C-9). Bluegill were found in small numbers in fall 1990 and did not have the expected length-frequency of a normal population (Fig. C-1). Bluegill demonstrated the expected length-frequency distribution in spring 1991. A greater number of bluegill were found in spring 1991 as compared to the following sampling periods. The bluegill population did not exhibit the expected length-frequency distribution from fall 1991 through fall 1993 (Figs. C-1 through C-3). The bluegill population size at MCK 1.6 has declined since spring 1989. One reason for the decline may be associated with physical changes at the site. A small pool which was located in approximately the middle of the sampling site as gradually silted in over the last three years and thus provides much less habitat for bluegill.

Green sunfish did not exhibit the expected length-frequency distribution of a normal population during the sampling periods (Figs. C-4 through C-6). The population of green sunfish had a large number of intermediate size fish. Generally, the smaller size classes were lacking, indicating a lack of reproduction at the site and possible immigration to the site. Fall data for all years demonstrates a small population of green sunfish compared to spring data and supports the premise of immigration and emigration of the fish at this site.

The bluntnose minnow had a much narrower size range (Figs. C-7 through C-9). The size classes of bluntnose minnow were similar to those in spring 1989 through spring 1990 (Ryon et al. 1992). The bluntnose minnow population did not indicate much variation in the distribution of size classes in fall 1990 or fall 1991. The population in spring 1991 was dominated by a few large individuals. Spring and fall 1992 samples were represented by a greater number of intermediate size individuals. Density increased steadily from spring 1991 through fall 1992. The bluntnose minnow population in spring and fall 1993 was represented by a fewer number of individuals in the intermediate size classes. Density declined from fall 1992 through fall 1993. From the variations in distribution of size classes from 1990-93, it appears that there is both immigration and emigration of the bluntnose minnow populations at MCK 1.6 from Melton Hill Reservoir.

#### 6.2.2.4 Observed abnormalities

As part of the normal processing procedure for population estimates, observations of abnormalities or attached parasites are made of the fish specimens. Usually these abnormalities are limited to less than 1-5% of the population in a minimally stressed system; a highly stressed system would have 5% or more abnormalities (Karr et al. 1986). Such abnormalities may include spinal deformities, bone deformities, open skin lesions, fin rot or erosion, or distended eyes ("popeye").

Abnormalities were found in fish at McCoy Branch in the fall 1990 and spring 1991. In the fall 1990 sample, one green sunfish (which represented 8% of the green sunfish) had a deformed caudal fin. In the spring 1991 sample, 45 out of 190 bluegill (24%) had popeye, 1 redbreast sunfish out of 3 had a deformed fin, and 1 redear sunfish out of 2 had a deformed head. No abnormalities were noted for the fall 1991 through fall 1993 sampling periods. Extremely eroded fins were not present and the occurrence of deformed fins or heads was not as great as in 1989 and spring 1990 samples (Ryon et al. 1992). The decrease in the occurrence of abnormalities in fall 1990 and spring 1991 followed by the lack of abnormalities in fall 1991 through 1993 may be related to the decrease in the amount of fly ash released to Rogers Quarry. The reference streams did not have any specimens with deformities during the sampling periods.

### 6.2.3 Conclusions

Data on the fish populations in lower McCoy Branch demonstrated that the stream has received stress from the coal ash operations. McCoy Branch was found to have more species of fish compared to the reference stream, Scarboro Creek, but was also found to be missing common fish species such as northern hog sucker and banded sculpin found in Scarboro Creek and other area streams. The lack of these species may be due to past extermination and the prevention of recolonization due to the presence of Melton Hill Reservoir.

Abnormalities such as deformed heads, eroded or missing fins were observed in fish from McCoy Branch but not in the reference streams. These abnormalities in fish have declined from fall 1990 through fall 1993 for McCoy Branch, and this decline may be the result of a decrease in fly ash entering the system.

## 6.3 ROGERS QUARRY (*E. M. Schilling*)

### 6.3.1 Materials and Methods

In conjunction with the Bioaccumulation Studies (Section 4.), largemouth bass (*Micropterus salmoides*) and bluegill were collected from Rogers Quarry and the adjacent McCoy Branch embayment in July and May 1991, respectively, by angling and electrofishing. Largemouth bass were also collected from Rogers Quarry in July 1992 and 1993 by angling.

The collected fish were identified to species, measured for total length (cm) and weight (g), checked for obvious abnormalities, and had scales removed for age determination. Scale samples were taken from an area above the lateral line and slightly anterior to the insertion of the dorsal fin. Scales were placed in coin envelopes. Data recorded on the envelope included date, species, locality, length and weight of the fish, and any abnormalities of the fish. Impressions of the scales were made using a Wildco scale press and acetate slides. Enlarged images of the scales were projected on a screen using a Bruning 4020 microfiche reader with a 15-mm lens (48X). Where possible, at least ten scales from each fish were mounted and compared. If scales produced poor images due to size, the actual scales were placed between two slides for examination. The best representative scale was used for actual measurements of annuli. Scales identified as regenerated (lacinucleate) and those that were damaged or highly irregular in shape were not read. In some cases, no age data were obtained for a fish because all scale samples were unsuitable.

Age data and length at age were not calculated for fish from Rogers Quarry from 1993 because only a small portion of the total fish could be aged accurately. This small subset of fish could have produced an inaccurate representation of the population. Scales from the larger largemouth bass were generally eroded and could not be aged accurately. Many of the scales collected from the smaller largemouth bass taken from Rogers Quarry in 1993 appeared to have checks (false annuli) which made aging difficult, and may have introduced errors in the data analysis; therefore, these data were not used in the study.

### 6.3.2 Results

Abnormalities in fish collected from McCoy Branch embayment in May 1991 consisted of skin abrasions on two of 29 largemouth bass and one of 119 bluegill. The length of largemouth bass ranged from 9.5–54.4 cm, and age ranged from 1<sup>+</sup>– 4<sup>+</sup> years. The length of bluegill ranged from 4.8–17.2 cm, and age ranged from 1<sup>+</sup>– 5<sup>+</sup> years.

In contrast, the deformities in the fish collected from Rogers Quarry included deformed heads, ossified heads, caudal deformities, deformed lateral line, deformed and missing fins. Twenty-four of seventy-two (33%) largemouth bass collected in July 1992 had one or more of the above deformities. Two of five bluegill collected had deformed heads. The length of largemouth bass ranged from 21.4–46.0 cm, and age ranged from 2<sup>+</sup>– 4<sup>+</sup> years. The length of bluegill ranged from 10.9–19.7 cm, and age ranged from 2<sup>+</sup>– 4<sup>+</sup> years.

From the collection of Rogers Quarry in July 1992, 27 out of 74 (36%) of the largemouth bass exhibited one or more of the same type of abnormalities as was observed in 1991. Largemouth bass ranged from 30.7–47.4 cm. Age ranged from 2<sup>+</sup>– 4<sup>+</sup> years.

Largemouth bass lengths collected from Rogers Quarry in July 1993 did not exhibit the severe deformities that were seen in 1991 and 1992. Two of the 40 largemouth bass had much less severe abnormalities. One fish had skin abrasions and the other had fin erosion. This decrease in abnormalities may be associated with the decrease in fly ash released to Rogers Quarry.

Generally, the fish that exhibited the severe abnormalities such as deformed heads and missing fins also exhibited aberrations on the scales. Scales were often regenerated, eroded on the edges, and had abnormal layering of the circuli making it impossible to age some fish.

Differences in the ages of fish from McCoy Branch embayment and Rogers Quarry may be related to the time of collection. Fish collected in May from McCoy Branch embayment may not have laid down that year's annulus, whereas fish collected in July from Rogers Quarry had probably laid down annuli indicating the beginning of that year's growth.

### 6.3.3 Conclusions

Severe abnormalities in fish decreased over time and with an increase in distance from Rogers Quarry. Severe abnormalities decreased in fish from Rogers Quarry from 1991 through 1993. This decrease may be associated with a decrease in the amount of fly ash released to the quarry. During 1991, a greater percentage of severe abnormalities were observed in the Centrarchids in Rogers Quarry as compared to the successive downstream sites of MCK 1.6 (1%) and McCoy Branch embayment (0).

## 6.4 UPPER MCCOY BRANCH (*B. A. Carrico*)

### 6.4.1 Introduction

A study on the introduction of a native fish species to McCoy Branch above Rogers Quarry was conducted as part of the monitoring for recovery in the watershed. Prior to 1992, upper McCoy Branch did not have an established fish community; during sampling from 1989 to 1992, only four fish were taken. The quarry and inflow structures act functionally as barriers to fish movement into the upper section from the downstream fish

communities in lower McCoy Branch and Melton Hill Reservoir. Previous fly ash disposal techniques appear to have eliminated any resident fish populations in upper McCoy Branch and the quarry barrier has prevented substantial natural colonization. Without a fish population, any measurement of recovery of the stream following cessation of the fly ash disposal was limited to improved water quality and changes in benthic invertebrate communities. Therefore, as part of the biomonitoring of McCoy Branch, a fish introduction study was initiated for upper McCoy Branch.

The study involved transplanting banded sculpins (*Cottus carolinae*), a common member of fish communities found in headwater stream of that region, into upper McCoy Branch and following their population parameters. Banded sculpins were obtained from a reference stream, marked with subcutaneous acrylic paint injections, and then introduced into the McCoy Branch study area. Prior to the introduction, baseline water quality and habitat measurements were made and two study sites were established. Following introduction, population parameters (density, length frequency, and recruitment), and general condition indices (survival, distribution, and condition factor) were monitored at the study sites and at the banded sculpin source stream, Hansard Mill Branch. Selected life history traits were also included in the study, which could augment the evaluation of the colonization of McCoy Branch.

#### 6.4.2 Site Description

Two reaches approximately 50 m in length were designated on upper McCoy Branch as banded sculpin introduction sites. The upstream site at McCoy Branch kilometer, MCK 2.1, has a slightly sinuous configuration, with moderately shallow riffles, separated by medium-sized pools. The average pool riffle ratio was 0.7. Substrate at this site is moderately embedded, and is composed of mainly cobble, with some leaf litter, woody debris, and root wads. This site has a dense riparian cover. The downstream site MCK 2.0 has a slightly sinuous configuration, with moderately deep runs separated by narrow shallow riffles. The mean pool-riffle ratio was 0.5. Substrate at this site is moderately embedded, and is composed mainly of small cobbles, gravel, and root wads. The riparian canopy was slightly less than that at MCK 2.1. Approximately 100 m of stream separates the MCK 2.0 and MCK 2.1 study reaches. Below the sites, the stream is routed through a metal weir into a pipe that empties onto a rock face and flows down into the quarry.

Hansard Mill Branch was chosen as a source of sculpins for introduction into McCoy Branch because it (1) had a large existing population of banded sculpins and (2) it was similar to McCoy Branch with respect to drainage area and stream habitat characteristics. Also, the study sections of Hansard Mill Branch remain relatively unimpacted despite their close proximity to the road and a good amount of rural development found in the watershed. Hansard Mill Branch is in Knox County approximately 32 km Northeast of McCoy Branch and drains the same ridge as McCoy Branch. This stream flows approximately 2.2-km from its source of five springs until it empties into Bull Run Creek.

The two study sites chosen in Hansard Mill Branch were similar to the study areas in McCoy Branch. The upstream reference site on Hansard Mill Branch kilometer (HMK) 1.23, is slightly sinuous and is composed of moderately-wide, shallow riffles bordered by medium-sized pools. The average pool-riffle ratio was 0.3. At this site the substrate is slightly to moderately embedded, and is composed mainly of small gravel and cobbles, with some woody debris. The mean riparian cover at HMK 1.2 was more open than at the two McCoy Branch

sites. This site is approximately 800 m down from the stream's source, five springs of low to moderate discharge. The downstream site, HMK 0.4, is slightly sinuous and is composed of long, moderately-deep riffles bordered by medium-sized pools. The pool-rifle ratio at this site is 0.4. In this area, the substrate is moderately embedded and composed of bedrock outcrops, large flat cobbles, and coarse gravel. The riparian cover at HMK 0.4 is the most sparse of any of the sites. This site is approximately 1.05 km downstream from the upper reference site and about 410 m upstream from the stream's confluence with Bull Run Creek.

### 6.4.3 Materials and Methods

#### 6.4.3.1 Fish introduction

As noted earlier, banded sculpins to be introduced into McCoy Branch were obtained from Hansard Mill Branch. A Smith-Root model 15-A electrofisher was used to collect the sculpins on August 15, 1992 (110 sculpins), on August 28, 1992 (75 sculpins), and on October 2, 1992 (52 sculpins). These fish were brought to the laboratory and kept in a flow-through tank to allow them to recover from the electrofishing process. After recovery, the sculpins were anesthetized with MS-222, tricaine methanesulfonate, and injected with Liquitex non-toxic acrylic polymer emulsions as per Lotrich and Meredith (1974). The fish were anesthetized to allow for easier handling during the injection process. A 3-cc syringe with a number 23 needle was used to mark the fish. Water was added to the paint in an approximate ratio of 2-cc of water to 5-g paint to thin the paint for injection. The sculpins were injected in the loose subcutaneous area on the mid-ventral portion of the caudal peduncle. Two colors were used to separate the fish to be placed at the two McCoy Branch sites. Fish to be introduced at MCK 2.0 were injected with Liquitex Brilliant Orange (Cadmium Orange Hue), and the fish to be introduced at MCK 2.1 were injected with Liquitex Vivid Lime Green. By using different marks for each group of sculpins, movements of the fish could be detected within McCoy Branch. When used to mark fish in this manner (Lotrich and Meredith 1974), Liquitex paints are reported to have a 4 to 16 month range of durability, depending on color. In this study, fish paint markings have been detected up to 33 months after injection. Mortality rates associated with tagging were relatively low, approximately 8.5%. In Barrett and Grossman (1988), mortality of subcutaneously injected mottled sculpins (*Cottus bairdi*) obtained by electrofishing ranged from 0 to 15% at 30 days after treatment. In Lotrich and Meredith (1974), a 4% mortality rate was reported for a two week period following injection. The fish collected from Hansard Mill Branch for introduction, were held until introduction in a 620 L rectangular fiberglass tank with a continuous flow of dechlorinated tap-water. Water depth was set at 10 cm, and a pump was placed in the water at the head of the tank to create a riffle effect. The fish were fed every other day with frozen brine shrimp and red worms. Of the 237 banded sculpins brought back to the lab, 13 fish were lost due to electroshocking and handling stresses, 19 fish died after the injection procedure, 25 were kept in the lab for observation, and the rest were introduced into McCoy Branch. On October 3, 1992, the marked sculpins were stocked into the two McCoy Branch sites; 90 fish were released at each site. The banded sculpins introduced into McCoy Branch ranged in size from 3.5 to 11.1 cm TL.

A second group of sculpins was introduced into McCoy Branch in August 1993. The fish for this introduction were captured from just above and below the HMK 1.2 site. Twenty three fish were brought to the laboratory, allowed to recover from electrofishing stress in a

flow-through tank, and injected with Liquitex acrylic paint as described above. Different colors were used to mark this group of fish to separate it from the 1992 introductions. The fish stocked in the downstream section were marked with Liquitex Brilliant Blue (Phthalocyanine Blue), and the fish stocked in the upstream section were marked with Liquitex Medium Magenta (Quinarcidone Magenta). Mortality associated with this round of injections was low, only 1 dead in 23 fish injected (4.3%). Four fish perished before injections were done due to electroshocking and handling stresses. A different method was employed to disperse the fish into the stream than in the first introduction to reduce the high density situations created in the first round of introductions. Half of the fish were spread out evenly over the entire lower 200-m of the McCoy Branch which encompassed MCK 2.0 and the other half over the upper 200-m section encompassing MCK 2.1. On August 2, 1993, marked sculpins ranging in size from 5.0 to 11.6 cm TL were introduced into both sections of stream, nine fish in each section.

#### 6.4.3.2 Fish community

Fish community samples were made at the sites on a quarterly basis by blocking off 50-m segments of stream with 5-mm mesh nylon nets. These sections were sampled with a Smith-Root model 15-A electrofisher. The electrofisher was set at 400-600 V DC, with frequencies of 60-90 pulses per second. The cathode and anode probes were fitted with circular rings, and the anode probe ring was covered with 5-mm nylon mesh. This allowed the electrofisher operator to assist in netting stunned fish. Sampling was done by making three electrofishing passes in an upstream direction covering all habitat types and taking all stunned fish. At Hansard Mill Branch, fish species other than banded sculpins were tallied only, no lengths or weights were taken. All fish captured during the three sampling passes were: anesthetized with MS-222; identified to species; measured to the nearest mm TL; and weighed to the nearest 0.1 g. The fish were then returned to the same section of stream after recovering from the anesthetic.

During the course of the study, possible undersampling of the population became a concern in the McCoy Branch samples. The number of sculpins recovered during 3-pass estimates was lower than expected for McCoy Branch. Therefore, a new method of sampling was employed at both streams in an attempt to limit undersampling bias. More electrofishing passes were performed and in some instances stream sampling reaches were lengthened. At least four passes were made on each section; the number of sampling passes were continued at each reach until no more fish were recovered. This method is a modification of Riley and Fausch (1992). Numbers of fish captured and fish densities increased using the more stringent sampling methods.

Population sizes were estimated using data from multiple pass replacement method (Carle and Strub 1978). Fish densities were calculated using a FORTRAN program utilizing a weighted likelihood method (Railsback et al. 1989). The density measurements provided from this program will provide a clearer picture of the actual population numbers of sculpins in the two streams. The sculpin densities were calculated using data from the quarterly population surveys. To assess the condition of the fish after introduction into McCoy Branch, condition factors (Hile 1936) were calculated from the fish community and recruitment sample data. The formula for condition factor is:

$$\text{Condition factor (K)} = [\text{weight(g)} / \text{length(cm)}^3]100$$

This factor was used to assess the general condition of the fish and aid comparisons between McCoy Branch and Hansard Mill branch fish populations. The condition factors for the sculpin populations at both streams were compared using a one-way ANOVA procedure and Tukeys standardized range test (SAS 1985a,b).

#### 6.4.4 Results

The sculpin population in McCoy Branch seems to have stabilized after an initial dropoff (Table 6.4). While the introduced sculpins are the only fish species inhabiting McCoy Branch, there are several fish species at the Hansard Mill Branch sites. There were two to three fish species, including banded sculpin, blacknose dace, and creek chub found in samples of the HMK 1.2 site. At the HMK 0.4 site, five to six fish species, including banded sculpin, blacknose dace, creek chub, stoneroller (*Campostoma anomalum*), striped shiner (*Luxilus*

Table 6.4. Banded sculpin population survey results in upper McCoy Branch for 1993

Survey date	Number of sculpins captured <sup>a</sup>	Marked sculpins recovered	Area sampled, m <sup>2</sup>
Jan 6-7 1993	55	18 orange 37 green	240
April 9-16 1993	27	17 orange 10 green	246
June 7-11 1993	38	22 orange 15 green 1 NM <sup>b</sup>	540
October 19 1993	25	9 orange 7 green 1 magenta 4 blue 4 NM	270

<sup>a</sup>Introduced banded sculpins collected from McCoy Branch population surveys of MCK 2.1 and MCK 2.0.

<sup>b</sup>NM = no discernable mark.

*chrysocephalus*), northern hogsucker (*Hypentelium nigricans*), stripetail darter (*Etheostoma kennikotti*), and snubnose darter (*Etheostoma simoterum*), were collected in 1993 (Table 6.5).

The average fish density in upper McCoy Branch during 1993 was approximately 0.10 sculpins/m<sup>2</sup> of stream. This figure, while much lower than the one for Hansard Mill Branch, is comparable with those of other geographically related streams. Sculpin population densities at McCoy Branch have remained stable to slightly increasing during the study period. The average densities are somewhat lower than those found in Hansard Mill Branch. Densities for Hansard Mill Branch sculpins averaged 0.65 fish/m<sup>2</sup> in 1993.

Condition factors for sculpins remained constant in 1993 at McCoy Branch, an indication that the fish were surviving, feeding, and in generally good condition (Table 6.6). Condition



Table 6.5. Fish community composition and occurrence at sampling sites in the reference stream, Hansard Mill Branch for quarterly sampling in 1993

Fish species	HMK 1.2		HMK 0.4	
	Occurrence <sup>a</sup>	Percentage <sup>b</sup>	Occurrence <sup>a</sup>	Percentage <sup>b</sup>
Banded sculpin	4/4	61.9	2/2	11.6
Central stoneroller			2/2	42.9
Striped shiner			1/2	2.5
Blacknose dace	4/4	37.9	2/2	39.6
Creek chub	1/4	0.2		
Northern hogsucker			1	0.4
Stripetail darter			2	1.0
Snubnose darter			1	2.0

<sup>a</sup>Occurrence lists the number of sampling periods a species was captured out of the total number of samples made.

<sup>b</sup>Mean percentage for all sampling periods.

Table 6.6. Mean condition factors for banded sculpins in McCoy Branch and Hansard Mill Branch

Site	Year	Condition Factor	N
McCoy Branch	1993	1.207	107
Hansard Mill Br	1993	1.232	237

factors for Hansard Mill Branch fish showed a slight decrease for that same period. The ANOVA and Tukeys tests showed that there was not a statistically significant difference (at an alpha level of 0.05) in condition factor between the sculpin populations in two streams during 1993.

#### 6.4.5 Discussion

In August 1992, banded sculpins were transplanted from Hansard Mill Branch to McCoy Branch to determine whether McCoy Branch could support a population of fish. The fish have been monitored over the last two years to document their ability to survive and to colonize this system successfully. Observations of the physical characteristics in McCoy Branch have shown that suitable habitat is available, there is a sufficient food base, and the water quality is not a detriment to the health of the sculpins.

It is evident from the population sampling data that the transplanted banded sculpins are surviving, and even thriving in McCoy Branch. Condition factors of banded sculpins in

McCoy Branch are comparable to the reference stream, Hansard Mill Branch. The fish population in McCoy Branch may be higher than shown by the estimates. In the earlier surveys on this stream, only three passes were done. Later surveys with four or more passes indicated underestimates in the earlier samples. Sampling McCoy Branch for sculpins proved to be difficult even with four or more passes; there are places such as deeply undercut banks and sections where the stream goes underground creating areas where the fish can avoid capture by electrofishing. All of the previously mentioned factors, along with the cryptic coloration and lack of a swim bladder (which causes the fish to sink when stunned), could lead to an underestimate of the sculpin population in McCoy Branch.

Young of the year sculpins (size class 4 to 6 cm TL), did not appear in the 1993 surveys until the fall of the year. In Small (1975), maximum growth rates for first year banded sculpins ranged from 5.0 to 6.2 cm TL. It is possible that the juvenile fish recovered in 1993 were spawned in McCoy Branch. However, lack of direct spawning evidence creates uncertainty on whether they were bred in the McCoy system and found refuge in a spring or underground reach, or they gained access to the system from the outside. The latter is not considered a reasonable source, because potential donor sites for sculpins to upper McCoy Branch have not yet been located. Extensive sampling of all the spring outlets in the upper McCoy system has not yielded any sculpins. Sculpins have not been found in Rogers Quarry, the lower section of McCoy Branch, or Melton Hill Reservoir. There are no nearby streams that could serve as a source of sculpins for McCoy Branch.

Population sampling at McCoy Branch has failed to produce any post-larval sculpins (0.5–3.0 cm TL). Small sculpins have been taken regularly from Hansard Mill Branch and from BMAP fish population surveys on the ORR. To date, no sculpins less than 3.9 cm TL have been collected from McCoy Branch. This fact coupled with the lack of spawning evidence (gravid females, nests, and eggs or larval sculpins) causes uncertainty whether the introduced sculpins are reproducing in McCoy Branch. It is possible that the immature fish are escaping presently used sampling techniques by accessing hidden springs, deeply undercut banks, or sections of McCoy Branch where the stream flows underground. The fish may then re-enter the stream at a later date when they are larger and more susceptible to electrofishing techniques. Documenting reproductive activities is made more difficult by the fact that these fish may not utilize a nest as do other species of sculpins. Williams and Robbins (1970) reported that banded sculpins do not localize at a nest as do species in the *bairdi* group. Although the introduced sculpins are surviving and are in good condition, more study is needed to fully evaluate banded sculpin colonization of McCoy Branch.

#### 6.4.6 Conclusions

The banded sculpins introduced into upper McCoy Branch are surviving and in good condition. However, there is no direct evidence that the introduced sculpins are reproducing in McCoy Branch. Therefore, whether McCoy Branch had sufficiently recovered from past coal fly-ash disposal practices to support a fish population has not yet been determined. Banded sculpins are a K-selected species and it may require more time for the introduced sculpins to fully utilize their reproductive potential in the new ecosystem. Because there is little information on their spawning activities, a more detailed life history study will be required to allow for the verification of future sculpin reproduction in upper McCoy Branch.

## 6.5 FUTURE STUDIES

Routine, quantitative monitoring of fish density, biomass, richness, and population size structure will be conducted twice annually (spring and fall) at MCK 1.6. Following completion of the banded sculpin introduction study, a determination will be made whether a fish population site above Rogers Quarry will be added to the routine monitoring of McCoy Branch.

The fish in Rogers Quarry may be sampled in conjunction with the bioaccumulation sampling as done previously. These fish will be examined for length, weight, obvious anomalies, and sampled for scales. An expansion of the sampling effort in Rogers Quarry may include electrofishing in the shallow and margin habitats to provide some data on community composition. Any such effort would be conducted only as a qualitative sampling.

The banded sculpin introduction study will continue through 1995. Future efforts to be conducted within this study include: stream habitat analysis, benthic macroinvertebrate food availability analysis, and description of banded sculpin feeding habits and reproductive activities.

## 7. BENTHIC MACROINVERTEBRATE COMMUNITY ASSESSMENT

(J. G. Smith)

### 7.1 INTRODUCTION

Results of benthic macroinvertebrate studies from the first year of the McCoy Branch RFI (April 1989 through January 1990) indicated that McCoy Branch was impacted (Tolbert and Smith 1992). In April and July 1989, total density, total taxonomic richness, and the number of the pollution intolerant Ephemeroptera, Plecoptera, and Trichoptera taxa (or EPT richness) were substantially lower at MCK 1.9 than at reference site WCK 6.8. These metrics were also suppressed at the downstream-most site (MCK 1.4) compared with the reference site. However, marked increases in these metrics by January 1990 may have been indicative of the beginning of recovery possibly associated with changes in operations and discharges at the Y-12 steam plant.

Studies of the benthic macroinvertebrate community of McCoy Branch continued after January 1990 with the primary objective of evaluating the effectiveness of remedial actions occurring within the McCoy Branch watershed. Other objectives included (1) assisting in the evaluation of the ecological condition of the stream and (2) providing guidance on the need for additional remediation. This report summarizes results of this continued effort and includes additional data from the April and October sampling periods of 1990 through 1993. Where appropriate, this summary also incorporates the results obtained during the first year of the RFI. The samples collected during the winter and summer sampling periods after January 1990 were stored and will be processed only if further resolution of the data is needed for future reports.

### 7.2 MATERIALS AND METHODS

The sampling regime followed for the benthic macroinvertebrate task during the first year of the McCoy Branch RFI (Tolbert and Smith 1992) continued unmodified in subsequent years. This included the collection of quantitative benthos samples at quarterly intervals from two sites in McCoy Branch [McCoy Branch kilometer 1.9 (MCK 1.9) and MCK 1.4] and one relatively unimpacted reference site in White Oak Creek (WCK 6.8) (Figs. 2.1 and 2.2). During each collection period, three random benthic macroinvertebrate samples were collected from a riffle at each site with a Surber stream bottom sampler (0.09 m<sup>2</sup>; 363- $\mu$ m-mesh net). Samples were placed into pre-labeled glass jars and preserved in 80% ethanol. The ethanol was replaced with fresh ethanol within one week.

Supplemental data on water quality and stream characteristics were obtained 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), and visual determination of relative current velocity (very slow, slow, moderate, or fast) and substrate type based on a modified Wentworth particle size scale (Loar et al. 1985) were recorded for each sample. All chemical and physical measurements were made in accordance with established quality assurance

procedures (Smith 1992). These collected data were used only if needed for observational support.

In the laboratory the contents of each sample were placed into a U. S. Standard No. 60-mesh (250- $\mu$ m-mesh) sieve and rinsed with tap water to remove the ethanol. 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 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). Parametric statistical analyses performed included analysis of variance (ANOVA) models on three responses (or metrics): density (total number of organisms/0.1 m<sup>2</sup>), taxonomic richness (total number of taxa/sample), and total combined richness of the Ephemeroptera, Plecoptera, and Trichoptera (total number of EPT taxa/sample or EPT richness). Before performing the ANOVAs, the values for each metric were transformed [i.e.,  $\log_{10}(X+1)$  for density values, and 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)]. Two approaches were used to assess the data for temporal changes attributable to remedial actions. For each approach, a  $p < 0.05$  was considered statistically significant.

The first approach was a nested ANOVA to look for site differences (fixed effect) among the five 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. In this analysis, similar changes (i.e.,  $p > 0.05$ ) among the sites between seasons and across years indicated that no unusual changes could be statistically detected at the McCoy Branch sites.

For the second approach, a simple linear regression was used to estimate site-specific linear trends for each metric over the 5-year study period on season-specific data. The slopes of the regression lines were then compared by season for parallelism (or linear trends) with an ANOVA. This analysis assumes that the variability around the regression lines for each site is the same. Violation of this assumption reduces the ability to detect significant trends. Thus, with extensive variation, a significant effect ( $p < 0.05$ ) would show that temporal changes were different, but the absence of a difference could either mean that a difference did not truly exist or that, because of the excessive variability, the test could not detect a difference. Because differences did exist among the sites in the extent of variation, the analysis was repeated for all pairwise comparisons of sites to minimize the potential confounding effect of a third site.

## 7.3 RESULTS

### 7.3.1 Taxonomic Composition

A checklist of the benthic macroinvertebrates collected from each site in McCoy Branch and from WCK 6.8 is presented in Appendix D, Table D.1. The collection of macroinvertebrates during only 2 seasons per year over a 5-year period does not allow the compilation of a complete list of taxa from each site. 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 communities. Most major groups of macroinvertebrates common in streams (e.g., Hynes 1970) were represented by one or several taxa during the 5-year study period including the Oligochaeta (aquatic worms), Amphipoda (sideswimmers/scuds), Decapoda (crayfish), Hydracarina (mites), Gastropoda (snails), Bivalvia (mussels/clams), Ephemeroptera (mayflies), Plecoptera (stoneflies), Trichoptera (caddisflies), Odonata (dragonflies/damselflies), Megaloptera (dobsonflies/fishflies), Coleoptera (beetles) and Diptera (true flies). Many of these taxa were rare or not collected at all three sites [e.g., Hirudinea, Isopoda, Amphipoda, Decapoda, most Odonata, some Ephemeroptera (e.g., *Epeorus*, *Habrophlebiodes*), some Plecoptera (e.g., *Alloperla*, *Peltoperla*), and some Trichoptera (e.g., *Oecetis*, *Pycnopsyche*, *Dolophilodes*)], while others were common at all sites [e.g., Oligochaeta, Nematoda, *Baetis* (Ephemeroptera), *Cheumatopsyche* (Trichoptera), *Stenelmis* (Coleoptera), Chironomidae, and several other Diptera]. The most notable difference between WCK 6.8 and the two McCoy Branch sites was that WCK 6.8 consistently had several coexisting taxa within each of the major taxonomic groups. In particular, in all 5 years of the study, several taxa within the Ephemeroptera, Plecoptera, and Trichoptera coexisted at WCK 6.8. At the McCoy Branch sites, the number of taxa within these groups was less during the first year, but in the last 4 years the number of taxa within each of 3 groups increased.

### 7.3.2 Abundance

#### 7.3.2.1 Density

Total community density changed considerably through time at all three sites (Fig. 7.1). The most difference between the highest and lowest recorded mean values over the 5-year study period was exhibited by MCK 1.9. Furthermore, densities were lowest at this site during all but one sampling period (i.e., October 1990). At MCK 1.9, the highest mean density recorded during the study was 1831.7 organisms/0.1 m<sup>2</sup> in October 1990. The lowest mean density was 76.4 organisms/0.1 m<sup>2</sup> in April 1989, a difference of 24x between these two sampling periods. Density changed least at WCK 6.8, where it ranged from a low of 318.6 organisms/0.1 m<sup>2</sup> in October 1989 to a high of 1155.7 organisms/0.1 m<sup>2</sup> in October 1992, a difference of only 3.6x. Density values for WCK 6.8 exceeded those of the other two sites in only three of the ten sampling periods. Density values were highest at MCK 1.4 on six of the ten sampling dates, and the highest density observed during the study was at this site. The 7x difference that existed between the highest and lowest mean values during the study at this site was much less than for MCK 1.9 but still almost twice that for WCK 6.8.

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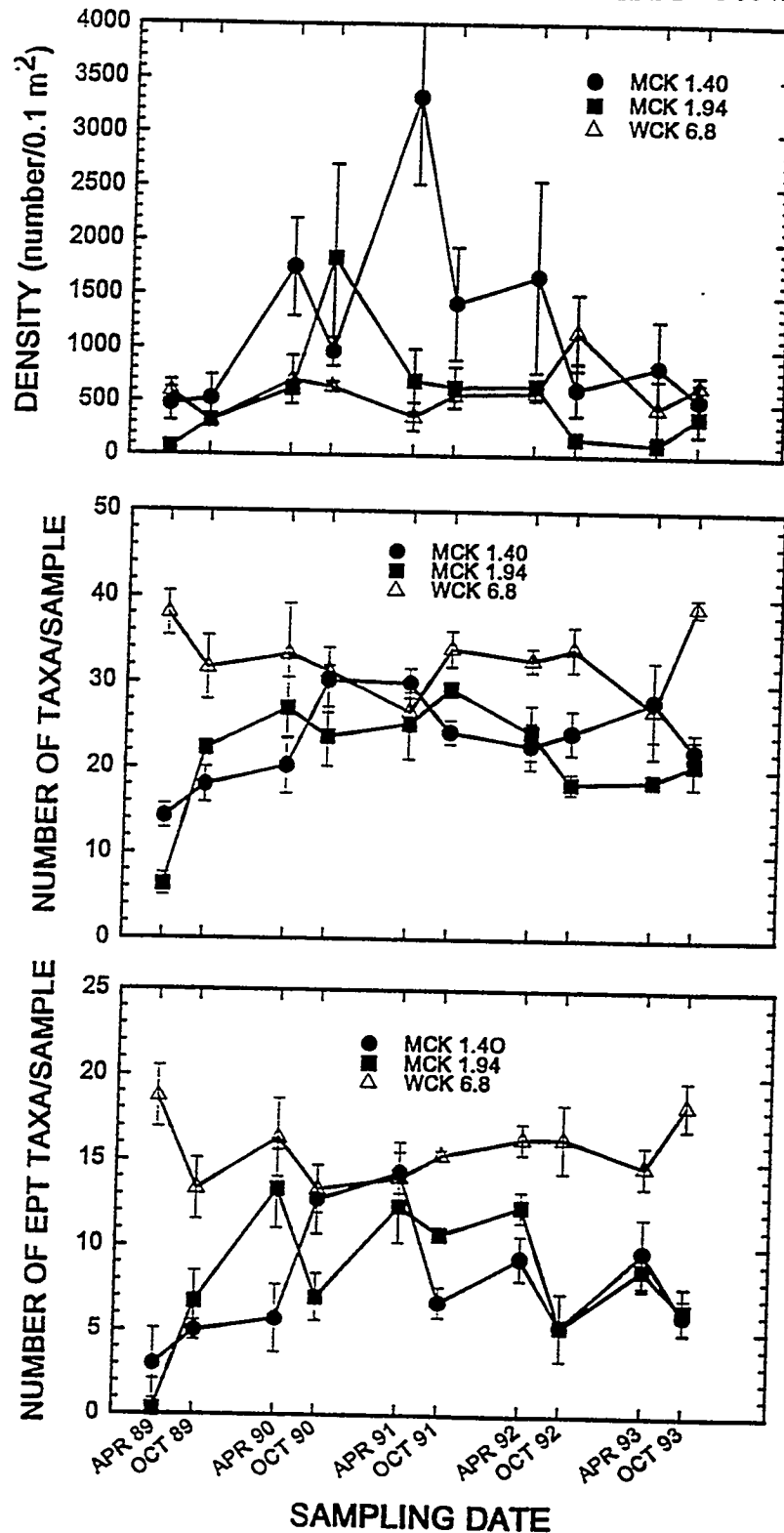


Fig. 7.1. Mean total density, mean total richness, and mean richness of the Ephemeroptera, Plecoptera, and Trichoptera (EPT richness) of the benthic macroinvertebrate communities in McCoy Branch and White Oak Creek, April 1989–October 1993. Vertical bars represent  $\pm 1$  SE. MCK = McCoy Branch kilometer; WCK = White Oak Creek kilometer.

Statistical analysis indicated that the patterns of change in density from the April to the October sampling periods were not the same between at least two sites during at least one year (Appendix E, Table E.1). Thus, during the study, two or all of the sites exhibited a different pattern of change from the April to October sampling periods.

The ANOVA of the regression slopes generated from the spring data were unable to detect any difference under the limitations of the test with the available data (Appendix F, Fig. F.1; Table E.1). If differences existed during the spring sampling period, their detection was obscured by variability. During the fall sampling periods for all years, only a difference between WCK 6.8 and MCK 1.9 was detected. By excluding data from 1989, a significant difference was detected from the three-site comparison. Pairwise comparisons detected differences between WCK 6.8 and each McCoy Branch study, while a difference between the slopes for MCK 1.9 and MCK 1.4 was not detected. This provided strong evidence that during the last 4 years of the study changes in density were similar at the McCoy Branch sites, but changes at these sites differed from those of the reference site.

### 7.3.2.2 Relative abundance

The macroinvertebrate communities at each site were generally dominated numerically by the Chironomidae, Diptera (excluding the Chironomidae), Coleoptera, Ephemeroptera, Plecoptera, and/or Trichoptera (Fig. 7.2). Each of these groups often accounted for over 10% of the total density. Except for the Gastropoda (primarily *Elimia*) at WCK 6.8, the proportion of the remaining major taxonomic groups rarely exceeded 5%.

The Chironomidae accounted for at least 15% of the total density at all sites during all sampling periods (Fig. 7.2). At MCK 1.4 and MCK 1.9 they accounted for > 50% of the total density during five and three of the ten sampling periods, respectively, most of which were April sampling periods. Particularly notable at MCK 1.9 was the large percentage of Chironomidae in the April 1989 sampling period, when they accounted for approximately 93% of the total density. After this time, however, their contribution to total density was much lower. In contrast to MCK 1.4 and MCK 1.9, the chironomids typically comprised < 30% of the total abundance at WCK 6.8. Seasonally, the percentage of chironomids at MCK 1.9 and MCK 1.4 was generally highest during the April sampling periods. At WCK 6.8, in contrast, there were no consistent temporal trends in the Chironomids.

Considered together, the Ephemeroptera, Plecoptera, and Trichoptera (or EPT) taxa consistently accounted for a high proportion of the community at WCK 6.8 (Fig. 7.2). At this site, the EPT taxa accounted for > 30% of the total density during nine of the ten sampling periods and > 50% in half of the sampling periods. At MCK 1.9, EPT taxa were virtually absent in April 1989, but then appeared to have "rebounded" in successive sampling periods when their relative abundances were generally comparable to those at WCK 6.8. At MCK 1.4, the relative abundance of the EPT taxa exceeded 10% in only five of ten sampling periods; four of these five sampling periods were during October. Seasonally, the proportion of the EPT taxa was generally much higher in the fall sampling periods at MCK 1.4, while at the other two sites the proportion of the EPT taxa tended to be highest during the spring sampling periods.

The Coleoptera comprised over 10% of the total density at MCK 1.4 during all but two sampling periods (Fig. 7.2). At each of the other two sites, the Coleoptera accounted for more than 10% of the total density during only four sampling periods. Seasonally, the proportion of the Coleoptera tended to be greatest during the October sampling periods at all three sites.



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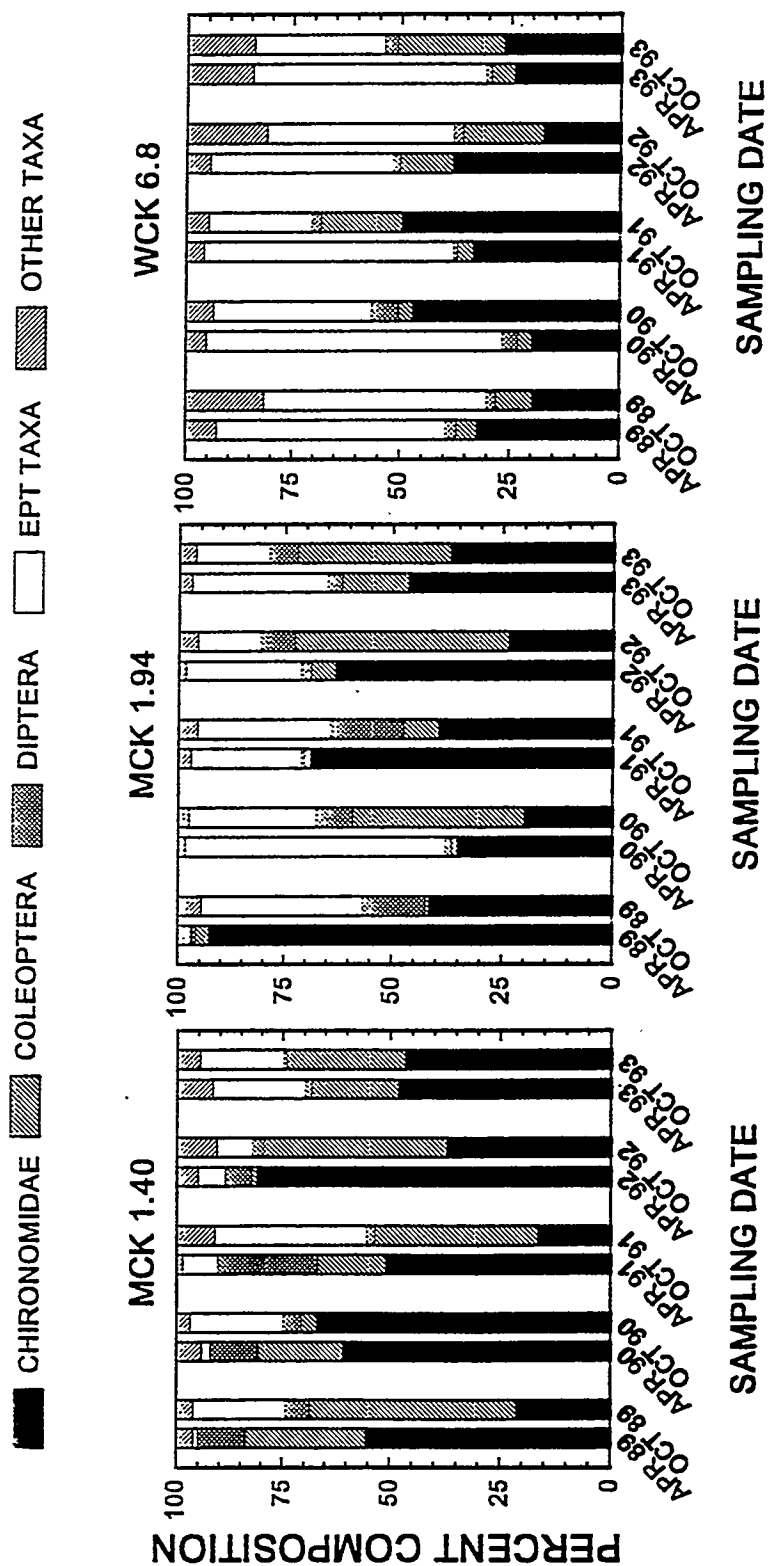


Fig. 7.2. Mean percent composition (percentage of total density) of selected benthic macroinvertebrate taxonomic groups in McCoy Branch and White Oak Creek, April 1989–October 1993. MCK = McCoy Branch kilometer; WCK = White Oak Creek kilometer.

Although the relative abundances of the Diptera (excluding the Chironomidae) were generally lower than those of the other numerically dominant groups, they still accounted for more than 5% of the total densities at both McCoy Branch sites during half of the ten sampling periods (Fig. 7.2). At WCK 6.8, the proportion of the Diptera exceeded 5% during only one sampling period and 2.5% during only three sampling periods. Seasonal patterns of the Diptera were similar at WCK 6.8 and MCK 1.9, where their proportions were usually highest during the October sampling periods. At MCK 1.4 the proportions of the Diptera were generally highest during the April sampling periods.

### 7.3.3 Taxonomic Richness

#### 7.3.3.1 Total richness

Total richness (i.e., total number of taxa/sample) was higher at WCK 6.8 than at either site on McCoy Branch during eight of ten sampling periods (Fig. 7.1). However, except for the April 1989 sampling period, the differences among sites during each sampling period did not exceed 2x. During the April 1989 sampling period, total richness at WCK 6.8 was 6x and 2.7x higher than at MCK 1.9 and MCK 1.4, respectively; richness at MCK 1.9 and MCK 1.4 differed by a factor of only 2.3x during this sampling period. The least difference among the three sites was observed in April 1991, when total richness differed by no more than a factor of 1.2x.

Total richness, like density, changed considerably through time at each site, although the magnitude of change was less (Fig. 7.1). The largest amount of change during the study occurred at MCK 1.9, where the difference between the lowest (April 1989) and highest (October 1991) values was 4.6x. As at MCK 1.9, the lowest value observed for total richness at MCK 1.4 was in April 1989, but the difference in richness between this sampling period and the sampling period having the highest richness value (October 1990) was only 2.1x. Total richness changed even less at WCK 6.8 over the 5-year period. At this site, the difference between the highest (April 1991) and lowest (October 1993) values observed was only 1.5x.

As observed for density, a statistically significant difference existed in seasonal changes among the three sites in total richness during one or more years, whether with or without data from year one (Table E.1, Figs F.1 and F.2). The slopes of the regression lines for the three sites over the study period during the spring were significantly different. Pairwise comparisons showed that changes in richness at both McCoy Branch sites differed from the change at WCK 6.8, but a difference between MCK 1.4 and MCK 1.9 was not detectable. When data for April 1989 were excluded, no statistical differences between sites were detected. No site differences in change could be detected with fall data when all three sites were examined together. However, with pairwise comparisons a significant difference between WCK 6.8 and MCK 1.9 was detected. When data from 1989 were excluded, significant differences were exhibited with an overall site analysis and with each pairwise comparison. As with the spring data, this suggested that after October 1989, taxonomic richness at the McCoy Branch sites became more similar to taxonomic richness at WCK 6.8. The fact that a difference between MCK 1.4 and WCK 6.8 was not detected for the fall sampling periods until after data for 1989 were excluded may indicate that change at MCK 1.4 was slower than at MCK 1.9. However, it is also possible that this inability to detect a difference was caused by data variability.

### 7.3.3.2 EPT richness

EPT richness, like total richness, was consistently higher at WCK 6.8 than at either McCoy Branch site over the 5-year study period (Fig. 7.1). Only in April 1991 was EPT richness higher at a McCoy Branch site (MCK 1.4) than at WCK 6.8. This sampling period was also the period during which the least difference was observed among the three sites (i.e.,  $< 1.2x$ ). The greatest difference among sites was observed during the April 1989 sampling period when mean EPT richness at WCK 6.8 was 56.7x and 9.1x higher than at MCK 1.9 and MCK 1.4, respectively.

Changes through time in EPT richness (combined total number of Ephemeroptera, Plecoptera, and Trichoptera taxa/sample) were much greater than those exhibited by total richness, particularly at MCK 1.9 (Fig. 7.1). At MCK 1.9, a difference of about 44x existed between the lowest value, which was observed in April 1989, and the highest value, which was observed in April 1990. After the April 1989 sampling period, however, the greatest difference between the highest and lowest mean values never exceeded 2.6x. The largest change between two sampling periods occurred between April and October 1989, when EPT richness increased by about 20.3x. However, after this period, the largest change between two sampling periods did not exceed 2.3x. As observed for density and total richness, the difference between the lowest (April 1989) and highest (April 1991) mean values at MCK 1.4 (4.8x) over the 5-year study period was less than at MCK 1.9 but greater than at WCK 6.8 (1.4x). The greatest change between sampling periods at MCK 1.4 was also less than at MCK 1.9; a change of approximately 2.2x was observed from April to October 1990 and from April to October 1991.

As exhibited by density and total richness, a statistically significant interaction occurred between site and sampling period (or season) within year whether with or without data from 1989, indicating that during two or more years, the seasonal changes exhibited were not the same between two or more of the sites (Table E.1). Comparisons of regression slopes gave similar results as those for total richness (Fig. F.1; Table E.1). The slopes of the regression lines for the spring sampling periods were significantly different, indicating that overall trends of two or all of the sites during the study were different. When data from 1989 were excluded, significant differences in linear trends among the sites were not detected. This may suggest that EPT richness was more stable during the last four years of the study although plots of the data (Figs. F.1 and F.2) show that values for MCK 1.4 continued to increase through 1991. During the fall sampling periods, trends at the two McCoy Branch sites differed from that at WCK 6.8. This significant difference persisted even after excluding data for 1989. This implied that different linear trends continued after the 1990 fall sampling period.

## 7.4 DISCUSSION

From April 1989 through January 1990, changes were observed in the benthic macroinvertebrate community of McCoy Branch suggestive of some recovery (Tolbert and Smith 1990). However, because no historical data for McCoy Branch were available from which the significance of these changes could be judged, it was not possible to make any definitive conclusions. The results from an additional 4 years of study strongly supported the previous conclusion that recovery was occurring during the first year of study. The rate and extent of recovery in McCoy Branch appeared to depend on location and time of year. Total

and EPT richness increased sharply at MCK 1.9 between April 1989 and April 1990. After the April 1990 sampling period, values for these metrics leveled off or declined only slightly across the remaining April sampling period but stayed well above the values recorded in April 1989. During the October sampling periods, neither total nor EPT richness values showed any changes at MCK 1.9 that persisted, while values at WCK 6.8 increased. Even so, the values observed at MCK 1.9 during all October sampling periods stayed well above those observed in April and July of 1989 (Fig. 7.1 this study; Tolbert and Smith 1992). These trends suggested that the normal April community existing at this site probably started changing after the April 1989 collection. Sometime after April 1990, changes ceased or slowed and a new community indicative of recovery stabilized. Although values for total and EPT richness remained less than at WCK 6.8, the magnitude of the differences was much less. The community structure and composition existing during the October sampling periods appeared stable by the time the first collection was made in this period in 1989. Without data from October sampling periods prior to 1989, it cannot be stated definitively that the community existing during the month of October from 1989 through 1993 was indicative of any recovery. It is possible that the perturbations impacting the community in the month of April were having no effects on the community existing in October.

Temporal trends for MCK 1.4 were not as straightforward as those for MCK 1.9. During the April sampling periods, total and EPT richness values increased sharply at MCK 1.4 in 1990 and then again in 1991 before declining slightly and stabilizing above those values observed in 1989 and 1990. Unlike MCK 1.9, where the community that existed during the October sampling periods appeared relatively stable during the study, total and EPT richness increased sharply in 1990 before apparently stabilizing in the last 3 years of the study. These results suggest that recovery occurred, but that it was behind and/or delayed relative to recovery at MCK 1.9. However, for MCK 1.9, without data prior to 1989 and with data from only five sampling periods for each quarter sampled, this conclusion remains speculative.

Even though the results obtained since 1989 suggest some recovery in McCoy Branch, the extent of recovery is not clear, nor is it clear whether some impacts continue. The results obtained from April 1989 through January 1990 indicated that McCoy Branch was adversely impacted, particularly at MCK 1.9 (this report and Tolbert and Smith 1992). This was particularly evident from the low relative abundances of taxa such as the Ephemeroptera and Trichoptera (this report and Tolbert and Smith 1992), taxa which are generally indicative of good water quality (e.g., Lenat 1988). Following the first year of study, the relative abundances and richness of these taxa increased considerably, particularly at MCK 1.9. However, the relative abundances of these taxa appeared to be persistently lower at the McCoy Branch sites than at WCK 6.8, and except during a few sampling periods, total and EPT richness appeared definitely suppressed at the McCoy Branch sites compared to WCK 6.8. However, EPT richness at WCK 6.8 has been found to be consistently higher than at some other reference stream sites draining south Chestnut Ridge (Smith 1993). Therefore, the differences remaining between McCoy Branch and WCK 6.8 may be natural and not disturbance-related. If the existing differences were impact-related, however, these results would suggest that they were slight to mild.

The changes observed in the macroinvertebrate community of McCoy Branch strongly coincide with operational changes at the Y-12 Plant steam plant (Section 2). After natural gas ignition and dual fuel capability were installed on burners 3 and 4 at the steam plant by the end of 1988, a 4.2-fold decrease in use of coal occurred, which would also translate to a similar reduction in ash sluiced to McCoy Branch. By November 1989, all ash from the

steam plant was sluiced directly to Rogers Quarry. The greatest change in the community at MCK 1.9 appeared to occur from April 1989 to October 1989, with some additional change by April 1990 (this study and Tolbert and Smith 1992). Thus, the reduction of sluiced-ash, followed soon by its total elimination, appears to have allowed significant recovery to occur in the macroinvertebrate community of upper McCoy Branch. The change at MCK 1.4 appeared to be less and possibly delayed relative to MCK 1.9, although this was not clear. However, the community at MCK 1.4 did appear to exhibit some recovery after the amount of ash discharged to McCoy Branch declined. Since the initial characterization of McCoy Branch indicated that MCK 1.4 was less impacted than MCK 1.9 (Tolbert and Smith 1992), these results also indicate that Rogers Quarry probably served as a "buffer" to the full effects of the ash discharges.

One additional change known to have occurred in the McCoy Branch watershed since April 1989 that could have affected MCK 1.4 appears to have had no notable effect on the invertebrates at this site. On March 4, 1992, release of water from Rogers Quarry was changed from a surface release to a release that was 10 m deep. Other than seasonal changes in all parameters that were also observed at the other two sites, no sustained trends were evident in the invertebrate community that would suggest that any significant changes occurred in response to the change.

As concluded by Tolbert and Smith (1992), it is not clear if the impacts observed were due to the direct or indirect action of fly ash on habitat (e.g., abrasion or loss of habitat by filling interstitial spaces) or to the possible toxic effects of metals (e.g., arsenic and selenium) leaching from the fly ash. Ephemeropterans are typically very sensitive to the presence of metals and are therefore either greatly reduced or eliminated in their presence (e.g., Hynes 1974; Wiederholm 1984; Clements 1994). Following the reduction and subsequent end to releases of fly ash into upper McCoy Branch, the relative abundance and number of Ephemeroptera taxa increased only slightly (Fig. 7.3). Thus, the presence of toxic concentrations of metals (e.g., selenium and arsenic) may still be a factor. However, the fact that overall recovery of the community occurred suggests that most metals probably leached rapidly from the fly ash, and that further recovery from any associated toxic effects will occur, but it will probably be relatively minor.

Since the macroinvertebrate community of McCoy Branch appears to have exhibited some recovery, if further recovery is hindered, the most likely cause(s) would be continued loss of habitat (e.g., reduction of interstitial spaces between substrate particles from fly ash or silt/sediment) or erosional effects associated with the continued presence of fly ash (e.g., abrasion) in the stream and flood plain. As the fly ash is flushed naturally from the floodplain and stream bottom, further changes may occur in the macroinvertebrate community as habitat improves.

## 7.5 RECOMMENDATIONS

Alterations to habitat within the stream and the riparian zone should be avoided since these activities would hinder (through habitat destruction) further recovery. If toxic quantities of metals exist, they will eventually be leached from the fly ash that remains. Similarly, any adverse effects to habitat associated with the remaining fly ash will decline as the fly ash is flushed naturally from the watershed.

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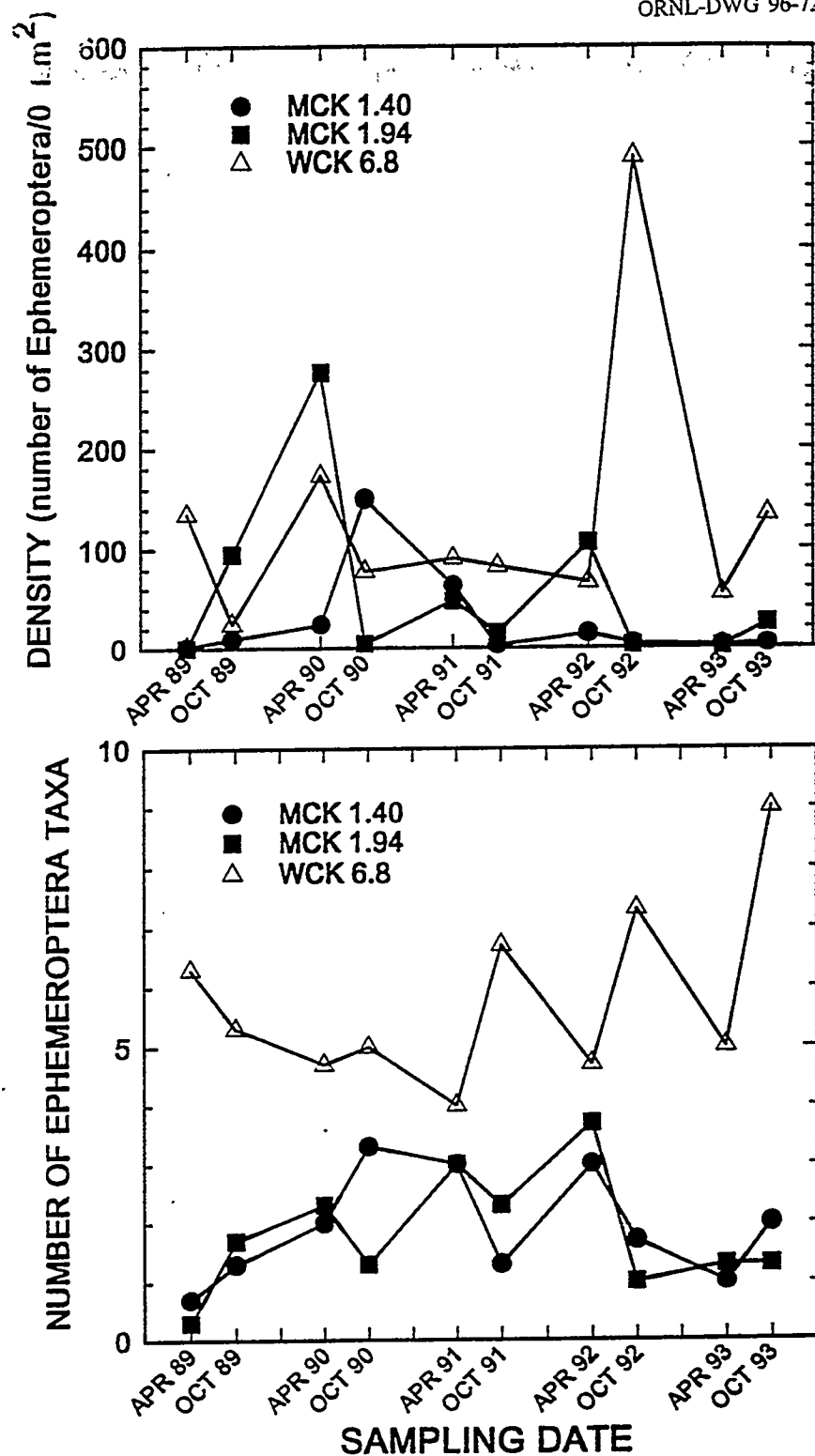


Fig. 7.3. Mean density and mean number of taxa of the Ephemeroptera in McCoy Branch and White Oak Creek, April 1989–October 1993. MCK = McCoy Branch kilometer; WCK = White Oak Creek kilometer.

## 7.6 SUMMARY AND CONCLUSIONS

An additional 4 years of data obtained on the benthic macroinvertebrate community in McCoy Branch since January 1990 provided strong evidence that substantial recovery began in McCoy Branch upstream of Rogers Quarry sometime after April 1989. Results to date also suggest that some recovery has probably occurred downstream of Rogers Quarry. Changes in the macroinvertebrate community strongly coincided with operational changes at the Y-12 Plant steam plant that initially included a substantial reduction in the amount of ash discharged to McCoy Branch, followed by complete elimination of all fly ash discharges to the stream. Total and EPT richness increased substantially at MCK 1.9 within the first year after the changes began, and then higher values persisted throughout the remainder of the study. The extent of recovery should be clarified as results from additional reference sites become available. If impacts are still occurring, the results to date would suggest that they were slight to mild and may be caused by unstable and/or altered habitat associated with such factors as the continued presence of fly ash.

## 7.7 FUTURE STUDIES

Benthic macroinvertebrate samples will continue to be collected on a quarterly basis. However, as done for this report, only samples collected during the spring and fall sampling periods will be processed unless data from the summer and winter seasons are needed. This provides a data base that coincides with collections of fishes (Section 6), and it provides a minimal data set needed to detect ecological change. In the future, data from additional reference sites will be incorporated into the analyses. This will provide a better data base from which natural temporal changes vs those associated with changes from remedial actions or additional perturbations can be more accurately interpreted, and it will also allow a more accurate assessment of the ecological conditions of McCoy Branch.

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**Appendix A**

**CONCENTRATIONS OF METALS IN PLANTS AND SMALL  
MAMMALS FOR MCCOY BRANCH WATERSHED**



Table A.1. Concentrations of metals in plants from the Filled Coal Ash Ponds (FCAP-1 and FCAP-2), Sluice, and control areas (Walker Branch Watershed)

Location	Sample ID	Date Sampled	Sample Leaves	As µg/g	Cd µg/g	Cr µg/g	Pb µg/g	Hg µg/g	Se µg/g	U µg/g
Control	55	22-Aug-91	Red maple	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.06	1.00	1.00 UJ
Control	56	23-Aug-91	Red maple	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.05	1.00	1.00 UJ
Control	59	08-Sep-88	Red maple	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.09	1.00	1.00 UJ
Control	62	08-Sep-88	Sourwood	1.20 UJ	0.25 UJ	3.00	1.00 UJ	0.12	1.00	1.00
Control	63	08-Sep-88	Dogwood	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.14	1.00	1.00
Control	64	08-Sep-88	Dogwood	1.00 UJ	0.25 J	3.00	1.00 UJ	0.21	1.00	1.00
Control	65	08-Sep-88	American beech	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.18	1.00	1.00
Control	66	08-Sep-88	American beech	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.13	1.00	1.00
Control	67	08-Sep-88	Sweet gum	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.11	1.00	1.00
Control	68	02-Apr-91	Eastern redcedar	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.05	1.00	1.00 UJ
Control	69	08-Sep-88	Sweet gum	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.08	1.00	1.00
Control	70	02-Apr-91	Eastern redcedar	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.05	1.00	1.00 UJ
Control	71	22-Aug-91	Grass	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.05	1.00	1.00
Control	72	22-Aug-91	Grass	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.05	1.00	1.00 UJ
Control	73	22-Aug-91	Grass	1.20 UJ	0.25 UJ	3.00	1.00 UJ	0.05	1.00	1.00 UJ
Control	74	23-Aug-91	Sycamore	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.05	1.00	1.00 UJ
Control	75	22-Aug-91	Boxelder	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.06	1.00	1.00 UJ
Control	76	08-Sep-88	Red maple	1.00 UJ	0.30 J	3.00 UJ	1.00	0.16	1.00	1.00 UJ
Control	77	08-Sep-88	Sourwood	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.06	1.00	1.00
FCAP-1	1	14-Sep-92	Red maple	3.80	0.25 UJ	3.00 UJ	1.00 UJ	0.12	8.50 J	1.00
FCAP-1	2	14-Sep-92	Red maple	2.80	0.25 UJ	3.00 UJ	1.00 UJ	0.10	5.80 J	1.00
FCAP-1	3	14-Sep-92	Cottonwood	1.00	2.10 J	3.00	1.00 UJ	0.05	25.90 J	1.00
FCAP-1	4	14-Sep-92	Sycamore	1.60 J	0.25 UJ	3.00	1.00 UJ	0.05	9.70 J	1.00
FCAP-1	5	14-Sep-92	Eastern redcedar	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.06	2.30 J	1.00
FCAP-1	6	14-Sep-92	Willow	1.00 J	2.00 J	3.00	1.00 UJ	0.05	20.50 J	1.00
FCAP-1	7	14-Sep-92	Sycamore	1.80	0.25 J	3.00	1.00 UJ	0.05	5.30 J	1.00 UJ
FCAP-1	8	14-Sep-92	Eastern redcedar	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.05	2.10 J	1.00 UJ
FCAP-1	12	14-Sep-92	Grass	1.30	0.25 UJ	20.20	1.00 UJ	0.05	1.80 J	1.00

Table A.1 (continued)

Location	Sample ID	Date Sampled	Sample Leaves	As µg/g	Cd µg/g	Cr µg/g	Pb µg/g	Hg µg/g	Se µg/g	Tl µg/g
FCAP-1	13	14-Sep-92	Sycamore	1.50	0.25 UJ	3.00	1.00 UJ	0.05	2.80 J	1.00
FCAP-1	14	14-Sep-92	Red maple	2.20	0.25 UJ	3.00	1.00 UJ	0.10	9.30 J	1.00
FCAP-1	15	14-Sep-92	Grass	1.00	0.25 UJ	7.50	1.10 J	0.05	1.70 J	1.00
FCAP-1	16	14-Sep-92	Honeysuckle	1.00	0.25 UJ	12.30	1.00 UJ	0.05	2.10 J	1.00
FCAP-1	17	14-Sep-92	Sycamore	1.00 J	0.25 UJ	3.00 UJ	1.00 UJ	0.05	7.50 J	1.00
FCAP-1	18	14-Sep-92	Boxelder	1.20 J	0.25 UJ	3.00 UJ	1.00 UJ	0.05	6.00 J	1.00 UJ
FCAP-1	19	14-Sep-92	Boxelder	1.50 J	0.25 UJ	3.80	1.00 J	0.06	4.70 J	1.00 UJ
FCAP-1	20	14-Sep-92	Grass	1.10 J	0.25 UJ	6.00	1.00 J	0.05	1.10 J	1.00 UJ
FCAP-1	21	14-Sep-92	Sycamore	1.90 J	0.25 UJ	3.00 UJ	1.00 UJ	0.05	7.80 J	1.00 UJ
FCAP-1	22	14-Sep-92	Boxelder	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.09	12.60 J	1.00 UJ
FCAP-2	23	15-Sep-92	Weeping Willow	2.10 J	2.40 J	3.00	1.80 J	0.05	31.20 J	1.00 UJ
FCAP-2	24	15-Sep-92	Cottonwood	1.90 J	1.80 J	3.00 UJ	1.00 UJ	0.05	97.00 J	1.00 UJ
FCAP-2	26	15-Sep-92	Sycamore	2.10 J	0.25 J	3.00	1.00 UJ	0.05	9.60 J	1.00 UJ
FCAP-2	27	15-Sep-92	Willow	1.80 J	2.40 J	3.00	1.00 UJ	0.05	36.50 J	1.00 UJ
FCAP-2	34	15-Sep-92	Sycamore	2.20 J	0.25 UJ	3.00	1.00 UJ	0.05	18.10 J	1.00 UJ
FCAP-1	35	14-Sep-92	Honeysuckle	1.00 UJ	0.25 UJ	7.80	1.00 UJ	0.05	2.90 J	1.00 UJ
FCAP-2	37	15-Sep-92	Sycamore	2.10 J	0.25 UJ	3.00	1.00 UJ	0.05	12.00 J	1.00 UJ
FCAP-2	39	15-Sep-92	Weeping Willow	3.10 J	1.40 J	3.00	1.00 UJ	0.05	20.30 J	1.00 UJ
FCAP-2	40	15-Sep-92	Cottonwood	1.00 UJ	2.20 J	3.00	1.00 UJ	0.05	1.00 UJ	1.00 UJ
FCAP-2	42	15-Sep-92	Willow	1.70 J	2.60 J	3.00	1.00 UJ	0.05	25.20 J	1.00
FCAP-2	43	15-Sep-92	Cottonwood	1.50 J	2.50 J	3.00	1.00 UJ	0.05	22.30 J	1.00 UJ
FCAP-1	52	14-Sep-92	Sycamore	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.06	22.50	1.00 UJ
FCAP-1	45	14-Sep-92	Cottonwood	1.10 J	0.45 J	3.00	1.00 UJ	0.05	39.40 J	1.00
FCAP-2	46	15-Sep-92	Weeping Willow	1.40 J	2.40 J	3.00	1.00 UJ	0.05	32.50	1.00 UJ
FCAP-1	48	14-Sep-92	Willow	1.10 J	0.60 J	3.00	1.00 UJ	0.05	15.70	1.00 UJ
FCAP-1	49	14-Sep-92	Willow	1.00 UJ	1.80 UJ	3.00	1.00 UJ	0.05	13.30	1.00 UJ
FCAP-1	51	14-Sep-92	Honeysuckle	1.00 UJ	0.25 UJ	6.60	1.00 UJ	0.05	3.10	1.00 UJ
FCAP-2	54	15-Sep-92	Willow	1.00 UJ	2.40 J	3.00	1.00 UJ	0.05	43.00	1.00 UJ



Table A.1 (continued)

Location	Sample ID	Date Sampled	Sample Leaves	As µg/g	Cd µg/g	Cr µg/g	Pb µg/g	Hg µg/g	Se µg/g	Tl µg/g
FCAP-1	55	14-Sep-92	Cottonwood	1.00 UJ	1.20 J	3.00	1.00 UJ	0.05	49.50	1.00 UJ
FCAP-1	56	14-Sep-92	Eastern reedcedar	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.05	2.90	1.00 UJ
SLUICE	3	15-Sep-92	Sycamore	1.00	0.25 UJ	3.00	1.00 UJ	0.08	1.00 U	1.00
SLUICE	10	15-Sep-92	Sycamore	1.00	0.25 UJ	3.00	1.00 UJ	0.07	1.00 U	1.00
SLUICE	15	15-Sep-92	Grass	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.05	1.60 J	1.00 UJ
SLUICE	27	15-Sep-92	Red maple	1.00 UJ	0.60 J	3.00	1.00 UJ	0.06	3.10 J	1.00 UJ
SLUICE	29	15-Sep-92	Grass	1.00 UJ	0.25 UJ	3.00 UJ	1.00 UJ	0.05	1.30 J	10.00 UJ
SLUICE	30	15-Sep-92	Eastern reedcedar	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.06	1.00 U	1.00 UJ
SLUICE	32	15-Sep-92	Eastern reedcedar	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.05	1.00 U	1.00 UJ
SLUICE	36	15-Sep-92	Grass	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.05	1.20 J	1.00 UJ
SLUICE	38	15-Sep-92	Eastern reedcedar	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.07	1.00 U	1.00 UJ
SLUICE	41	15-Sep-92	Red maple	1.00	0.25 UJ	3.00	1.00 UJ	0.08	1.00 U	1.00
SLUICE	47	15-Sep-92	Red maple	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.08	1.10	1.00 UJ
SLUICE	43	15-Sep-92	Sycamore	1.00 UJ	0.25 UJ	3.00	1.00 UJ	0.07	2.10	1.00 UJ

J = Estimated UJ = Less than detection limit

UJ = Estimated not-detect

Table A.2. Concentrations of metals in small mammals from the Filled Coal Ash Ponds (FCAP-1 and FCAP-2), Sluice, and reference site (Walker Branch Watershed)

Location	Species	Sample	Cd mg/kg	Se mg/kg	Pb mg/kg	As mg/kg	Tl mg/kg	Hg mg/kg	Cr mg/kg
FCAP-1	Peromyscus	02-001	0.020	2.05 J	0.568	0.132 UJ	0.200 UJ	0.017	1.34 J
FCAP-1	Peromyscus	02-001	0.111	2.02 J	0.920	0.0500	0.200 UJ	0.021	1.10 J
FCAP-1	Peromyscus	02-001	0.010 UJ	1.81 J	0.345	0.070 UJ	0.200 UJ	0.024	0.835 J
FCAP-1	Peromyscus	68-003	0.011	6.55 J	0.298	0.632	0.200 UJ	0.046	1.30 J
FCAP-1	Peromyscus	68-003	0.010 UJ	5.22 J	0.375	0.355	0.200 UJ	0.043	0.93 J
FCAP-1	Peromyscus	68-003	0.010 UJ	5.11 J	0.340	0.315	0.200 UJ	0.031	0.922 J
FCAP-1	Peromyscus	17-004	0.027 J	1.65 J	0.308	0.290 J	0.200 UJ	0.027	0.970 J
FCAP-1	Peromyscus	17-004	0.010 UJ	1.76 J	0.132	0.168	0.200 UJ	0.03	1.47 J
FCAP-1	Peromyscus	17-004	0.018	1.76 J	0.145	0.175	0.200 UJ	0.025	1.06 J
FCAP-1	Peromyscus	71-005	0.028	4.16 J	0.158	0.175 UJ	0.200 UJ	0.046	1.90 J
FCAP-1	Peromyscus	71-005	0.031	3.85 J	0.242	0.100 UJ	0.328 UJ	0.051	0.988 J
FCAP-1	Peromyscus	71-005	0.029	3.80 J	0.455	0.090 UJ	0.200 UJ	0.052	1.20 J
FCAP-1	Peromyscus	33-006	0.010 UJ	0.488 J	0.140	0.160 UJ	0.200 UJ	0.026	0.842 J
FCAP-1	Peromyscus	33-006	0.010 UJ	0.472 J	0.170	0.105 UJ	0.200 UJ	0.025	0.998 J
FCAP-1	Peromyscus	33-006	0.010 UJ	0.388 J	0.205	0.053 UJ	0.200 UJ	0.024	1.12 J
FCAP-1	Peromyscus	17-007	0.039 J	2.88 J	1.54	0.310	0.200 UJ	0.059	2.30 J
FCAP-1	Peromyscus	17-007	0.036	2.43 J	1.50	0.228	0.200 UJ	0.063	1.79 J
FCAP-1	Peromyscus	17-007	0.028	2.44 J	1.32	0.158	0.200 UJ	0.045	1.95
FCAP-1	Peromyscus	03-008	0.010 UJ	1.82 J	0.142	0.155 UJ	0.200 UJ	0.016	2.75 J
FCAP-1	Peromyscus	03-008	0.010 UJ	1.79 J	0.152	0.088 UJ	0.200 UJ	0.016	2.30 J
FCAP-1	Peromyscus	03-008	0.010 UJ	1.83 J	0.122	0.050 UJ	0.200 UJ	0.017	1.71 J
FCAP-2	Reithrodontomys	38-009	0.026 J	2.91 J	0.110	0.210 UJ	0.200 UJ	0.118	0.975 J
FCAP-2	Reithrodontomys	38-009	0.030	2.46 J	0.170	0.105 UJ	0.200 UJ	0.097	1.11 J
FCAP-2	Reithrodontomys	38-009	0.021	2.42 J	0.132	0.095 UJ	0.200 UJ	0.102	1.16 J
FCAP-2	Reithrodontomys	09-010	0.077 J	3.46 J	12.20	0.240 UJ	0.200 UJ	0.072	1.59 J
FCAP-2	Reithrodontomys	09-010	0.052	3.14 J	8.60	0.090 UJ	0.205 J	0.074	1.01 J
FCAP-2	Reithrodontomys	09-010	0.066	3.56 J	10.00	0.103 UJ	0.200 UJ	0.066	1.08 J

Table A.2 (continued)

Location	Species	Sample	Cd mg/kg	Se mg/kg	Pb mg/kg	As mg/kg	Tl mg/kg	Hg mg/kg	Cr mg/kg
FCAP-2	Peromyscus	11-011	orig	2.32 J	1.06	0.143 U	0.200 U	0.027	1.26 J
FCAP-2	Peromyscus	11-011	dup	2.44 J	1.5	0.050 U	0.200 U	0.024	0.632 J
FCAP-2	Peromyscus	11-011	trip	2.40 J	0.34	0.050 U	0.200 U	0.035	1.66 J
FCAP-2	Reithrodontomys	03-012	orig	1.63 J	0.2	0.278	0.200 U	0.013	0.965 J
FCAP-2	Reithrodontomys	03-012	dup	1.31 J	0.135	0.148	0.200 U	0.012	1.12 J
FCAP-2	Reithrodontomys	03-012	trip	1.26 J	0.118	0.222	0.200 U	0.012	0.928 J
FCAP-2	Reithrodontomys	33-013	orig	0.57 J	0.075	0.080 U	0.200 U	0.016	0.67 J
FCAP-2	Reithrodontomys	33-013	dup	0.63 J	0.102	0.050 U	0.200 U	0.016	1.47 J
FCAP-2	Reithrodontomys	33-013	trip	0.728 J	0.105	0.050 U	0.200 U	0.016	1.31 J
SLUICE	Microtus	12-014	orig	0.502	0.53	0.108 J	0.200 U	0.011 J	1.55
SLUICE	Microtus	12-014	dup	0.630	0.422	0.083 U	0.200 U	0.012	1.31
SLUICE	Microtus	12-014	trip	0.575	0.475	0.088 J	0.200 U	0.009 J	1.51
SLUICE	Peromyscus	29-015	orig	0.446	0.938	0.050 U	0.200 U	0.026 J	1.052
SLUICE	Peromyscus	29-015	dup	0.462	0.765	0.050 U	0.200 U	0.026 J	1.08
SLUICE	Peromyscus	29-015	trip	0.455	0.76	0.050 U	0.200 U	0.022 J	1.22
SLUICE	Peromyscus	79-016	orig	0.453	0.61	0.050 U	0.200 U	0.038 J	1.10
SLUICE	Peromyscus	79-016	dup	0.438	0.542	0.050 U	0.200 U	0.046 J	1.10
SLUICE	Peromyscus	79-016	trip	0.535	0.79	0.050 U	0.200 U	0.061 J	1.01
SLUICE	Peromyscus	60-017	orig	0.818	0.26 J	0.050 U	0.200 U	0.053 J	1.06
SLUICE	Peromyscus	60-017	dup	0.652	0.282	0.050 U	0.200 U	0.053 J	1.12
SLUICE	Peromyscus	60-017	trip	0.742	0.27	0.050 U	0.200 U	0.053 J	0.925
SLUICE	Blarina	28-018	orig	2.26	0.798 J	0.185 J	0.200 U	0.501 J	0.898
SLUICE	Blarina	28-018	dup	2.20	0.64	0.140 J	0.200 U	0.59 J	0.638 J
SLUICE	Blarina	28-018	trip	2.53	0.592	0.205 J	0.200 U	0.351 J	0.855
SLUICE	Peromyscus	93-019	orig	0.332	0.59	0.058 J	0.200 U	0.02 J	1.02
SLUICE	Peromyscus	93-019	dup	0.308	0.432	0.050 U	0.200 U	0.027 J	1.16
SLUICE	Peromyscus	93-019	trip	0.348	0.455	0.050 U	0.200 U	0.03 J	1.15

Table A.2 (continued)

Location	Species	Sample	Cd mg/kg	Se mg/kg	Pb mg/kg	As mg/kg	Ti mg/kg	Hg mg/kg	Cr mg/kg
Walker Branch	Blarina	77-021	orig	0.975 J	1.330	0.050 UJ	0.200 UJ	0.258	2.060 J
Walker Branch	Blarina	77-021	dup	0.963 J	1.020	0.050 UJ	0.200 UJ	0.234	1.710 J
Walker Branch	Blarina	77-021	trip	0.922 J	1.580	0.050 UJ	0.200 U	0.212	1.820 J
Walker Branch	Peromyscus	24-022	orig	0.272	0.410	0.050 UJ	0.200 U	0.034	1.170 J
Walker Branch	Peromyscus	24-022	dup	0.280 J	0.320	0.050 UJ	0.200 UJ	0.035	0.982 J
Walker Branch	Peromyscus	24-022	trip	0.278	0.300 U	0.050 UJ	0.200 U	0.034	0.925 J
Walker Branch	Peromyscus	70-023	orig	0.295	0.262 U	0.050 UJ	0.200 U	0.027	0.858 J
Walker Branch	Peromyscus	70-023	dup	0.202	0.202 U	0.050 UJ	0.200 U	0.024	0.815 J
Walker Branch	Peromyscus	70-023	trip	0.292 J	0.198 U	0.050 UJ	0.200 U	0.023	0.788 J
Walker Branch	Peromyscus	48-024	orig	0.255	0.165 U	0.050 UJ	0.200 U	0.017	0.755 J
Walker Branch	Peromyscus	48-024	dup	0.248	0.122 U	0.050 UJ	0.200 U	0.018	0.768 J
Walker Branch	Peromyscus	48-024	trip	0.265	0.085 U	0.050 UJ	0.200 U	0.020	0.515 J
Walker Branch	Blarina	88-025	orig	1.470 J	2.880	0.050 UJ	0.200 U	0.363	6.060 J
Walker Branch	Blarina	88-025	dup	1.500 J	1.330	0.050 UJ	0.200 UJ	0.406	6.270 J
Walker Branch	Blarina	88-025	trip	1.340 J	1.060	0.102 UJ	0.200 UJ	0.550	3.960 J
Walker Branch	Peromyscus	17-026	orig	0.282 J	0.602	0.065 UJ	0.200 UJ	0.026	4.530 J
Walker Branch	Peromyscus	17-026	dup	0.228 J	0.710	0.078 UJ	0.200 U	0.030	3.460 J
Walker Branch	Peromyscus	17-026	trip	0.235	0.632	0.050 U	0.200 UJ	0.031	3.480 J
LAD	rinsate	00-002	orig	0.0002 UJ	0.001 UJ	0.002 UJ	0.0004 UJ	0.0001 UR	0.02 UR
LAD	rinsate	RJ	orig	0.0002 U	0.001 UJ	0.001 U	0.002 U	0.0001 U	0.02 U
LAD	rinsate	00-027	orig	0.0004 UJ	0.002 UJ	0.001 UR	0.008 UJ	0.0001 UJ	0.005

U = less than detection limit (or sample qualified due to potential blank contamination)

J = Estimated  
R = Unusable

Table A.2 (continued)

Detection	Limits		
Cd:	0.010 mg/kg	Tl:	0.200 mg/kg
Se:	0.050 mg/kg	Hg:	0.005 mg/kg
Pb:	0.050 mg/kg	Cr:	0.125mg/kg
As:	0.050 mg/kg		

Table A.3. Average concentrations of metals in small mammals from the Filled Coal Ash Ponds (FCAP-1 and FCAP-2), Sluice, and reference site (Walker Branch Watershed)

Units expressed as wet weight

Location	Species	Sample	Cd mg/kg	Sc mg/kg	Pb mg/kg	As mg/kg	Pt mg/kg	Hg mg/kg	Cr mg/kg
FCAP-1	Peromyscus	02-001	Mean	1.973	0.611	0.084	0.200	0.021	1.092
FCAP-1	Peromyscus	02-001	Std. Dev	0.108	0.290	0.043	0.000	0.004	0.253
FCAP-1	Peromyscus	02-001	CV	0.055	0.475	0.509	0.000	0.170	0.231
FCAP-1	Peromyscus	68-003	Mean	5.64	0.338	0.434	0.200	0.040	1.051
FCAP-1	Peromyscus	68-003	Std. Dev	0.798	0.039	0.173	0.000	0.008	0.216
FCAP-1	Peromyscus	68-003	CV	0.141	0.114	0.398	0.000	0.198	0.206
FCAP-1	Peromyscus	17-004	Mean	1.723	0.195	0.211	0.200	0.027	1.167
FCAP-1	Peromyscus	17-004	Std. Dev	0.064	0.098	0.069	0.000	0.003	0.266
FCAP-1	Peromyscus	17-004	CV	0.037	0.503	0.325	0.000	0.092	0.228
FCAP-1	Peromyscus	71-005	Mean	3.937	0.285	0.122	0.243	0.049	1.363
FCAP-1	Peromyscus	71-005	Std. Dev	0.195	0.153	0.047	0.074	0.003	0.477
FCAP-1	Peromyscus	71-005	CV	0.052	0.050	0.382	0.305	0.065	0.350
FCAP-1	Peromyscus	33-006	Mean	0.446	0.172	0.106	0.200	0.025	0.987
FCAP-1	Peromyscus	33-006	Std. Dev	0.052	0.033	0.054	0.000	0.001	0.139
FCAP-1	Peromyscus	33-006	CV	0.116	0.190	0.505	0.000	0.040	0.141
FCAP-1	Peromyscus	17-007	Mean	2.583	1.453	0.232	0.200	0.056	2.013
FCAP-1	Peromyscus	17-007	Std. Dev	0.257	0.117	0.076	0.000	0.009	0.261
FCAP-1	Peromyscus	17-007	CV	0.099	0.081	0.33	0.000	0.170	0.130
FCAP-1	Peromyscus	03-008	Mean	1.813	0.139	0.098	0.200	0.016	2.253
FCAP-1	Peromyscus	03-008	Std. Dev	0.021	0.015	0.053	0.000	0.001	0.522
FCAP-1	Peromyscus	03-008	CV	0.011	0.110	0.544	0.000	0.035	0.231
FCAP-2	R. th. odontomys	38-009	Mean	2.597	0.137	0.137	0.200	0.105	1.082
FCAP-2	R. th. odontomys	38-009	Std. Dev	0.272	0.030	0.064	0.000	0.011	0.096
FCAP-2	R. th. odontomys	38-009	CV	0.105	0.221	0.466	0.000	0.104	0.088
FCAP-2	R. th. odontomys	09-010	Mean	3.387	10.267	0.144	0.202	0.071	1.227
FCAP-2	R. th. odontomys	09-010	Std. Dev	0.219	1.815	0.083	0.003	0.004	0.317
FCAP-2	R. th. odontomys	09-010	CV	0.193	0.065	0.576	0.014	0.059	0.258

Table A.3 (continued)

Location	Species	Sample	Cd mg/kg	Se mg/kg	Pb mg/kg	As mg/kg	Hg mg/kg	Cr mg/kg
FCAP-2	Peromyscus	11-011	Mean	2.387	0.967	0.081	0.029	1.184
FCAP-2	Peromyscus	11-011	Std. Dev	0.061	0.586	0.054	0.006	0.518
FCAP-2	Peromyscus	11-011	CV	0.026	0.606	0.663	0.000	0.438
FCAP-2	Reithrodontomys	03-012	Mean	1.400	0.151	0.216	0.012	1.004
FCAP-2	Reithrodontomys	03-012	Std. Dev	0.201	0.043	0.065	0.001	0.102
FCAP-2	Reithrodontomys	03-012	CV	0.143	0.287	0.302	0.000	0.102
FCAP-2	Reithrodontomys	33-013	Mean	0.644	0.094	0.060	0.016	1.150
FCAP-2	Reithrodontomys	33-013	Std. Dev	0.079	0.016	0.017	0.000	0.423
FCAP-2	Reithrodontomys	33-013	CV	0.123	0.176	0.289	0.000	0.368
SLUICE	Microtus	12-014	Mean	0.569	0.476	0.093	0.011	1.457
SLUICE	Microtus	12-014	Std. Dev	0.064	0.054	0.013	0.000	0.129
SLUICE	Microtus	12-014	CV	0.113	0.114	0.142	0.000	0.088
SLUICE	Peromyscus	29-015	Mean	0.455	0.821	0.050	0.025	1.117
SLUICE	Peromyscus	29-015	Std. Dev	0.007	0.101	0.000	0.002	0.091
SLUICE	Peromyscus	29-015	CV	0.015	0.123	0.000	0.094	0.081
SLUICE	Peromyscus	79-016	Mean	0.474	0.647	0.050	0.048	1.07
SLUICE	Peromyscus	79-016	Std. Dev	0.053	0.128	0.000	0.012	0.052
SLUICE	Peromyscus	79-016	CV	0.112	0.198	0.000	0.242	0.049
SLUICE	Peromyscus	60-017	Mean	0.764	0.271	0.050	0.053	1.035
SLUICE	Peromyscus	60-017	Std. Dev	0.065	0.011	0.000	0.000	0.100
SLUICE	Peromyscus	60-017	CV	0.085	0.011	0.000	0.000	0.096
SLUICE	Blarina	28-018	Mean	2.33	0.677	0.177	0.481	0.797
SLUICE	Blarina	28-018	Std. Dev	0.176	0.108	0.033	0.121	0.139
SLUICE	Blarina	28-018	CV	0.075	0.159	0.188	0.251	0.175
SLUICE	Peromyscus	93-019	Mean	0.329	0.492	0.053	0.025	1.110
SLUICE	Peromyscus	93-019	Std. Dev	0.020	0.085	0.005	0.005	0.078
SLUICE	Peromyscus	93-019	CV	0.061	0.173	0.000	0.199	0.070

Table A.3 (continued)

Location	Species	Sample	Cd mg/kg	Se mg/kg	Pb mg/kg	As mg/kg	Pi mg/kg	Hg mg/kg	Cr mg/kg
Walker Branch	Blarina	77-021	0.057	0.952	1.310	0.050	0.200	0.235	1.863
Walker Branch	Blarina	77-021	0.004	0.027	0.281	0.000	0.000	0.023	0.179
Walker Branch	Blarina	77-021	0.066	0.029	0.214	0.000	0.000	0.098	0.096
Walker Branch	Peromyscus	24-022	0.026	0.277	0.343	0.050	0.200	0.034	1.026
Walker Branch	Peromyscus	24-022	0.001	0.004	0.059	0.000	0.000	0.001	0.128
Walker Branch	Peromyscus	24-022	0.040	0.015	0.171	0.000	0.000	0.026	0.125
Walker Branch	Peromyscus	70-023	0.013	0.263	0.221	0.050	0.200	0.025	0.820
Walker Branch	Peromyscus	70-023	0.002	0.053	0.036	0.000	0.000	0.002	0.035
Walker Branch	Peromyscus	70-023	0.175	0.201	0.162	0.000	0.000	0.080	0.043
Walker Branch	Peromyscus	48-024	0.027	0.256	0.124	0.050	0.200	0.018	0.679
Walker Branch	Peromyscus	48-024	0.001	0.009	0.040	0.000	0.000	0.001	0.142
Walker Branch	Peromyscus	48-024	0.033	0.033	0.323	0.000	0.000	0.074	0.210
Walker Branch	Blarina	88-025	0.075	1.437	1.757	0.067	0.200	0.440	5.430
Walker Branch	Blarina	88-025	0.027	0.085	0.982	0.030	0.000	0.098	1.277
Walker Branch	Blarina	88-025	0.360	0.059	0.559	0.446	0.000	0.223	0.235
Walker Branch	Peromyscus	17-026	0.021	0.248	0.648	0.064	0.200	0.029	3.823
Walker Branch	Peromyscus	17-026	0.001	0.029	0.056	0.014	0.000	0.003	0.612
Walker Branch	Peromyscus	17-026	0.038	0.118	0.086	0.215	0.000	0.101	0.160



Table A.4. Concentrations (dry weight corrected) of metals in small mammals from Filled Coal Ash Ponds (FCAP-1 and FCAP-2), and the sluice

Location	Species	Sample	"	Cd mg/kg	Se mg/kg	Pb mg/kg	As mg/kg	Tl mg/kg	Hg mg/kg	Cr mg/kg	Solids %
FCAP-1	Peromyscus	02-001	orig	0.072	7.383	2.341	0.477	<0.719	0.062	4.811	0.278
FCAP-1	Peromyscus	02-001	dup	0.040	7.248	3.309	<0.180	<0.719	0.074	3.966	0.278
FCAP-1	Peromyscus	02-001	trip	0.399	6.664	1.241	0.252	<0.719	0.088	3.004	0.278
FCAP-1	Peromyscus	68-003	orig	<0.035	23.081	1.048	2.227	<0.704	0.163	4.577	0.284
FCAP-1	Peromyscus	68-003	dup	<0.035	18.398	1.320	1.250	<0.704	0.151	3.275	0.284
FCAP-1	Peromyscus	68-003	trip	<0.035	18.099	1.197	1.109	<0.704	0.108	3.248	0.284
FCAP-1	Peromyscus	17-004	orig	0.093	5.668	1.053	0.993	<0.685	0.092	3.322	0.292
FCAP-1	Peromyscus	17-004	dup	<0.034	6.036	0.454	0.574	<0.685	0.103	5.026	0.292
FCAP-1	Peromyscus	17-004	trip	0.060	6.010	0.497	0.599	<0.685	0.085	3.639	0.292
FCAP-1	Peromyscus	71-005	orig	0.083	12.621	0.477	0.530	<0.606	0.140	5.773	0.330
FCAP-1	Peromyscus	71-005	dup	0.093	11.674	0.735	0.303	0.992	0.153	2.992	0.330
FCAP-1	Peromyscus	71-005	trip	0.088	11.500	1.379	0.273	<0.606	0.156	3.652	0.330
FCAP-1	Peromyscus	33-006	orig	<0.033	1.598	0.459	0.525	<0.656	0.086	2.762	0.305
FCAP-1	Peromyscus	33-006	dup	<0.033	1.516	0.557	0.344	<0.656	0.082	3.270	0.305
FCAP-1	Peromyscus	33-006	trip	<0.033	1.270	0.672	0.172	<0.656	0.079	3.672	0.305
FCAP-1	Peromyscus	17-007	orig	0.134	9.846	5.291	1.062	<0.685	0.200	7.868	0.292
FCAP-1	Peromyscus	17-007	dup	0.124	8.313	5.120	0.779	<0.685	0.216	6.130	0.292
FCAP-1	Peromyscus	17-007	trip	0.095	8.339	4.521	0.539	<0.685	0.155	6.678	0.292
FCAP-1	Peromyscus	03-008	orig	<0.031	5.615	0.438	0.477	<0.615	0.050	8.469	0.325
FCAP-1	Peromyscus	03-008	dup	<0.031	5.500	0.469	0.269	<0.615	0.049	7.062	0.325
FCAP-1	Peromyscus	03-008	trip	<0.031	5.623	0.377	<0.154	<0.615	0.051	5.262	0.325
FCAP-2	Reithrodontomys	38-009	orig	0.101	11.367	0.430	0.820	<0.781	0.459	3.809	0.256
FCAP-2	Reithrodontomys	38-009	dup	0.116	9.619	0.664	0.410	<0.781	0.377	4.336	0.256
FCAP-2	Reithrodontomys	38-009	trip	0.081	9.463	0.518	0.371	<0.781	0.396	4.521	0.256
FCAP-2	Reithrodontomys	09-010	orig	0.243	10.899	29.314	0.757	<0.631	0.227	5.008	0.317
FCAP-2	Reithrodontomys	09-010	dup	0.164	9.921	23.462	0.284	0.647	0.233	3.194	0.317
FCAP-2	Reithrodontomys	09-010	trip	0.209	11.222	31.546	0.323	<0.631	0.207	3.415	0.317
FCAP-2	Reithrodontomys	33-013	orig	<0.026	2.011	0.265	0.282	<0.529	0.044	2.363	0.378
FCAP-2	Reithrodontomys	33-013	dup	<0.026	2.240	0.362	<0.132	<0.529	0.042	5.185	0.378
FCAP-2	Reithrodontomys	33-013	trip	<0.026	2.566	0.370	0.150	<0.529	0.041	4.630	0.378
FCAP-2	Peromyscus	11-011	orig	<0.035	8.207	3.746	0.504	<0.707	0.096	4.470	0.283
FCAP-2	Peromyscus	11-011	dup	<0.035	8.604	5.318	<0.177	<0.707	0.084	2.235	0.283
FCAP-2	Peromyscus	11-011	trip	<0.035	8.481	1.201	<0.177	<0.707	0.122	5.848	0.283

Table A.4 (continued)

Location	Species	Sample	Cd mg/kg	Se mg/kg	Pb mg/kg	As mg/kg	Tl mg/kg	Hg mg/kg	Cr mg/kg	Solids %
FCAP-2	R:ilth:odontomys	03-012	<0.034	5.612	0.690	0.957	<0.690	0.045	3.328	0.290
FCAP-2	R:ilth:odontomys	03-012	<0.034	4.526	0.466	0.509	<0.690	0.041	3.862	0.290
FCAP-2	R:ilth:odontomys	03-012	<0.034	4.328	0.405	0.767	<0.690	0.042	3.198	0.290
SLUICE	P:ronyscus	29-015	0.068	1.695	3.551	<0.189	<0.758	0.098	3.987	0.264
SLUICE	P:ronyscus	29-015	0.043	1.752	2.898	<0.189	<0.758	0.099	4.072	0.264
SLUICE	P:ronyscus	29-015	0.062	1.723	2.879	<0.189	<0.758	0.085	4.621	0.264
SLUICE	Micratus	12-014	0.066	1.757	1.853	0.376	<0.699	0.040	5.420	0.286
SLUICE	Micratus	12-014	0.084	2.203	1.477	0.288	<0.699	0.043	4.580	0.286
SLUICE	Micratus	12-014	0.085	2.010	1.661	0.306	<0.699	0.031	5.271	0.286
SLUICE	Peromyscus	79-016	0.084	1.466	1.987	<0.163	<0.651	0.124	3.575	0.307
SLUICE	Peromyscus	79-016	0.107	1.425	1.767	0.252	<0.651	0.151	3.591	0.307
SLUICE	Peromyscus	79-016	0.175	1.743	2.573	<0.163	<0.651	0.197	3.298	0.307
SLUICE	Peromyscus	60-017	<0.043	3.539	1.126	<0.216	<0.866	0.229	4.600	0.231
SLUICE	Peromyscus	60-017	<0.043	2.998	1.223	<0.216	<0.866	0.229	4.838	0.231
SLUICE	Peromyscus	60-017	<0.043	3.387	1.169	<0.216	<0.866	0.229	4.004	0.231
SLUICE	Blarina	28-018	0.205	6.969	2.454	0.569	<0.615	1.542	2.762	0.325
SLUICE	Blarina	28-018	0.193	6.762	1.962	0.431	<0.615	1.815	1.969	0.325
SLUICE	Blarina	28-018	0.275	7.792	1.823	0.631	<0.615	1.080	2.631	0.325
SLUICE	Peromyscus	93-019	0.040	1.346	2.389	<0.202	<0.810	0.082	4.140	0.247
SLUICE	Peromyscus	93-019	0.046	1.245	1.751	<0.202	<0.810	0.102	4.717	0.247
SLUICE	Peromyscus	93-019	0.066	1.407	1.842	<0.202	<0.810	0.121	4.656	0.247
REF	Peromyscus	17-026	0.085	1.170	2.498	0.270	<0.830	0.107	18.797	0.241
REF	Peromyscus	17-026	0.091	0.916	2.916	0.322	<0.830	0.126	14.357	0.241
REF	Peromyscus	17-026	0.086	0.975	2.622	<0.207	<0.830	0.130	14.44	0.241
REF	Peromyscus	24-022	0.110	1.115	1.680	<0.205	<0.820	0.138	4.795	0.244
REF	P:ro:myscus	24-022	0.102	1.148	1.312	<0.205	<0.820	0.145	4.205	0.244
REF	Peromyscus	24-022	0.104	1.139	1.230	<0.205	<0.820	0.141	3.910	0.244
REF	P:rc:myscus	48-024	0.101	0.977	0.632	<0.192	<0.766	0.067	2.893	0.261
REF	Peromyscus	48-024	0.102	0.950	0.467	<0.192	<0.766	0.067	2.943	0.261
REF	P:ro:myscus	48-024	0.108	1.015	0.326	<0.192	<0.766	0.076	1.973	0.261
REF	P:ro:myscus	70-023	0.047	1.024	0.910	<0.174	<0.694	0.094	2.979	0.288
REF	P:ro:myscus	70-023	<0.035	0.701	0.701	<0.174	<0.694	0.085	2.830	0.288
REF	P:ro:myscus	70-023	0.048	1.014	0.688	<0.174	<0.694	0.080	2.736	0.288

Table A.4 (continued)

Location	Species	Sample	Cd mg/kg	Se mg/kg	Pb mg/kg	As mg/kg	Tl mg/kg	Hg mg/kg	Cr mg/kg	Solids %
REF	Blarina	77-021	0.236	3.765	5.135	<0.193	<0.772	0.996	7.954	0.259
REF	Blarina	77-021	0.207	3.707	3.938	<0.193	<0.772	0.904	6.602	0.259
REF	Blarina	77-021	0.220	3.560	6.100	<0.193	<0.772	0.819	7.027	0.259
REF	Blarina	88-025	0.233	4.868	9.536	<0.166	<0.662	1.202	20.066	0.302
REF	Blarina	88-025	0.344	4.967	4.404	<0.166	<0.662	1.344	20.762	0.302
REF	Blarina	88-025	0.167	4.437	3.510	0.338	<0.662	1.821	13.113	0.302

Table A.5. Average concentrations (dry weight corrected) of metals in small mammals from the Filled Coal Ash Ponds (FCAP-1 and FCAP-2), and the sluice

Location	Species	Sample		Cd mg/kg	Se mg/kg	Pb mg/kg	As mg/kg	Tl mg/kg	Hg mg/kg	Cr mg/kg
FCAP-1	Peromyscus	02-001	mean	0.17	7.098	2.197	0.303	<0.719	0.074	3.927
FCAP-1	Peromyscus	02-001	Std. Dev.	0.162	0.382	0.043	0.154	0.000	0.013	0.904
FCAP-1	Peromyscus	02-001	C.V.	1.166	0.054	0.475	0.511	0.000	0.174	0.230
FCAP-1	Peromyscus	68-003	mean	<0.035	19.859	1.188	1.529	<0.704	0.141	3.700
FCAP-1	Peromyscus	68-003	Std. Dev.	0.000	2.794	0.136	0.609	0.000	0.029	0.760
FCAP-1	Peromyscus	68-003	C.V.	0.000	0.141	0.115	0.398	0.000	0.206	0.205
FCAP-1	Peromyscus	17-004	mean	0.062	5.905	0.668	0.722	<0.685	0.093	3.996
FCAP-1	Peromyscus	17-004	Std. Dev.	0.030	0.205	0.334	0.235	0.000	0.009	0.906
FCAP-1	Peromyscus	17-004	C.V.	0.000	0.035	0.500	0.326	0.000	0.097	0.227
FCAP-1	Peromyscus	71-005	mean	0.088	11.932	0.864	0.369	0.735	0.150	4.139
FCAP-1	Peromyscus	71-005	Std. Dev.	0.005	0.603	0.165	0.141	0.223	0.009	1.453
FCAP-1	Peromyscus	71-005	C.V.	0.057	0.051	0.538	0.381	0.303	0.057	0.351
FCAP-1	Peromyscus	33-006	mean	<0.033	1.460	0.563	0.347	<0.656	0.082	3.230
FCAP-1	Peromyscus	33-006	Std. Dev.	0.000	0.171	0.107	0.177	0.000	0.004	0.456
FCAP-1	Peromyscus	33-006	C.V.	0.000	0.117	0.190	0.509	0.000	0.043	0.141
FCAP-1	Peromyscus	17-007	mean	0.118	8.833	4.977	0.793	<0.685	0.190	6.892
FCAP-1	Peromyscus	17-007	Std. Dev.	0.020	0.878	0.404	0.262	0.000	0.032	0.889
FCAP-1	Peromyscus	17-007	C.V.	0.172	0.099	0.081	0.330	0.000	0.166	0.129
FCAP-1	Peromyscus	03-008	mean	<0.031	5.579	0.428	0.300	<0.615	0.050	6.931
FCAP-1	Peromyscus	03-008	Std. Dev.	0.000	0.069	0.047	0.164	0.000	0.001	1.608
FCAP-1	Peromyscus	03-008	C.V.	0.000	0.012	0.110	0.545	0.000	0.025	0.232
FCAP-2	Reithrodontomys	38-009	mean	0.099	10.150	0.537	0.534	<0.781	0.411	4.222
FCAP-2	Reithrodontomys	38-009	Std. Dev.	0.018	1.057	0.118	0.249	0.000	0.043	0.370
FCAP-2	Reithrodontomys	38-009	C.V.	0.177	0.104	0.220	0.466	0.000	0.104	0.088
FCAP-2	Reithrodontomys	09-010	mean	0.205	10.681	28.107	0.455	0.636	0.222	3.872
FCAP-2	Reithrodontomys	09-010	Std. Dev.	0.040	0.678	4.175	0.263	0.009	0.014	0.990
FCAP-2	Reithrodontomys	09-010	C.V.	0.193	0.063	0.149	0.577	0.015	0.063	0.256
FCAP-2	Reithrodontomys	33-013	mean	<0.026	2.272	0.332	0.188	<0.529	0.042	4.059
FCAP-2	Reithrodontomys	33-013	Std. Dev.	0.000	0.279	0.959	0.082	0.000	0.002	1.495
FCAP-2	Reithrodontomys	33-013	C.V.	0.000	0.123	0.177	0.436	0.000	0.036	0.368
FCAP-2	Peromyscus	11-011	mean	<0.035	8.431	3.422	0.286	<0.707	0.101	4.184
FCAP-2	Peromyscus	11-011	Std. Dev.	0.000	0.203	2.077	0.189	0.000	0.020	1.823
FCAP-2	Peromyscus	11-011	C.V.	0.000	0.024	0.607	0.660	0.000	0.193	0.436

Table A.5 (continued)

Location	Species	Sample	Cd mg/kg	Se mg/kg	Pb mg/kg	As mg/kg	Hg mg/kg	Cr mg/kg
FCAP-2	Rattus norvegicus	03-012	mean	4.822	0.520	0.744	<0.690	3.463
FCAP-2	Rattus norvegicus	03-012	Std. Dev.	0.692	0.150	0.225	0.000	0.000
FCAP-2	Rattus norvegicus	03-012	CV	0.143	0.288	0.302	0.000	0.102
SLUICE	Peromyscus	29-015	mean	1.723	3.109	<0.189	<0.758	4.227
SLUICE	Peromyscus	29-015	Std. Dev.	0.028	0.383	0.000	0.000	0.344
SLUICE	Peromyscus	29-015	CV	0.016	0.123	0.000	0.000	0.081
SLUICE	Microtus	12-014	mean	1.990	1.564	0.323	<0.699	5.090
SLUICE	Microtus	12-014	Std. Dev.	0.224	0.188	0.047	0.000	0.448
SLUICE	Microtus	12-014	CV	0.112	0.113	0.144	0.000	0.088
SLUICE	Peromyscus	79-016	mean	1.545	2.109	0.193	<0.651	3.488
SLUICE	Peromyscus	79-016	Std. Dev.	0.173	0.417	0.052	0.000	0.165
SLUICE	Peromyscus	79-016	CV	0.112	0.198	0.267	0.000	0.047
SLUICE	Peromyscus	60-017	mean	3.308	1.172	<0.216	<0.866	4.481
SLUICE	Peromyscus	60-017	Std. Dev.	0.279	0.019	0.000	0.000	0.429
SLUICE	Peromyscus	60-017	CV	0.084	0.042	0.000	0.000	0.096
SLUICE	Blarina	28-018	mean	7.174	2.079	0.544	<0.615	2.454
SLUICE	Blarina	28-018	Std. Dev.	0.545	0.332	0.102	0.000	0.425
SLUICE	Blarina	28-018	CV	0.076	0.159	0.188	0.000	0.173
SLUICE	Peromyscus	93-019	mean	1.333	1.994	<0.202	<0.810	4.504
SLUICE	Peromyscus	93-019	Std. Dev.	0.082	0.345	0.000	0.000	0.317
SLUICE	Peromyscus	93-019	CV	0.061	0.173	0.000	0.000	0.070
REF	Peromyscus	17-026	mean	1.030	2.689	0.266	<0.830	15.865
REF	Peromyscus	17-026	Std. Dev.	0.122	0.331	0.058	0.000	2.540
REF	Peromyscus	17-026	CV	0.118	0.086	0.216	0.000	0.160
REF	Peromyscus	24-022	mean	1.134	1.407	<0.205	<0.820	4.303
REF	Peromyscus	24-022	Std. Dev.	0.004	0.24	0.000	0.000	0.451
REF	Peromyscus	24-022	CV	0.395	0.17	0.000	0.000	0.105
REF	Peromyscus	48-024	mean	0.981	0.475	<0.192	<0.766	2.603
REF	Peromyscus	48-024	Std. Dev.	0.004	0.153	0.000	0.000	0.546
REF	Peromyscus	48-024	CV	0.037	0.322	0.000	0.000	0.210
REF	Peromyscus	70-023	mean	0.913	0.766	<0.174	<0.694	2.848
REF	Peromyscus	70-023	Std. Dev.	0.007	0.125	0.000	0.000	0.123
REF	Peromyscus	70-023	CV	0.167	0.163	0.000	0.000	0.043

Table A.5 (continued)

Location	Species	Sample	Cd mg/kg	Sc mg/kg	Pb mg/kg	As mg/kg	Tl mg/kg	Hg mg/kg	Cr mg/kg
REF	Blarina	77-021	mean	3.677	5.058	<0.193	<0.772	0.906	7.194
REF	Blarina	77-021	Std.Dev.	0.106	1.083	0.000	0.000	0.089	0.691
REF	Blarina	77-021	CV	0.029	0.214	0.000	0.000	0.098	0.096
REF	Blarina	88-025	mean	4.757	5.817	0.223	<0.662	1.456	17.98
REF	Blarina	88-025	Std.Dev.	0.282	3.252	0.099	0.000	0.324	4.230
REF	Blarina	88-025	CV	0.059	0.559	0.445	0.000	0.223	0.235

Table A.6. Dry weight corrected mean concentrations  $\pm$  SD (range in parentheses) of metals derived from individual animal means for each genus and all rodents collected at Chestnut Ridge OU2 (FCAP and Sluice areas) and Walker Branch

Expressed as milligrams per kilogram									
	Chestnut Ridge OU2	FCAP		Sluice			Walker Branch		
ANALYTE	CR OU2 Rodents*	Peromyscus (n=8)	Reithrodontomys (n=4)	Peromyscus (n=4)	Microtus (n=1)	Blarina (n=1)	Peromyscus (n=4)	Blarina (n=2)	
As	0.452 ± 0.342 (0.188 - 1.529)	.581 ± 0.432 (0.286 - 1.529)	0.480 ± 0.230 (0.188 - 0.744)	0.200 ± 0.012 (0.189 - 0.216)	0.323 ± 0.047	0.544 ± 0.102	0.209 ± 0.040 (0.174 - 0.266)	0.208 ± 0.021 (0.193 - 0.223)	
Cd	0.076 ± 0.052 (0.026 - 0.205)	0.072 ± 0.051 (0.031 - 0.170)	0.091 ± 0.083 (0.026 - 0.205)	0.068 ± 0.036 (0.043 - 0.122)	0.078 ± 0.011	0.224 ± 0.044	0.085 ± 0.029 (0.043 - 0.105)	0.235 ± 0.019 (0.221 - 0.248)	
Cr	4.377 ± 1.049 (3.235 - 6.931)	4.625 ± 1.442 (3.235 - 6.931)	3.904 ± 0.327 (3.463 - 4.222)	4.175 ± 0.475 (3.488 - 4.504)	5.090 ± 0.448	2.454 ± 0.425	6.405 ± 6.351 (2.603 - 15.865)	12.587 ± 7.627 (7.194 - 17.980)	
Pb	3.168 ± 6.551 (0.332 - 28.107)	1.788 ± 1.640 (0.428 - 4.977)	7.374 ± 13.822 (0.332 - 28.107)	2.096 ± 0.794 (1.172 - 3.109)	1.664 ± 0.188	2.079 ± 0.332	1.334 ± 0.983 (0.475 - 2.689)	5.437 ± 0.537 (5.058 - 5.817)	
Hg	0.131 ± 0.094 (0.038 - 0.411)	0.110 ± 0.046 (0.050 - 0.190)	0.180 ± 0.176 (0.042 - 0.411)	0.146 ± 0.062 (0.094 - 0.229)	0.038 ± 0.006	1.479 ± 0.372	0.105 ± 0.032 (0.070 - 0.141)	1.181 ± 0.388 (0.906 - 1.456)	
Se	6.289 ± 4.996 (1.333 - 19.859)	8.637 ± 5.445 (1.461 - 19.859)	6.981 ± 4.105 (2.272 - 10.681)	1.977 ± 0.901 (1.333 - 3.308)	1.990 ± 0.224	7.174 ± 0.545	1.015 ± 0.093 (0.913 - 1.134)	4.217 ± 0.764 (3.677 - 4.757)	
Tl	.702 ± 0.078 (<.529 - 0.866)	0.688 ± 0.038 (< 0.615 - 0.735)	0.659 ± 0.105 (<0.529 - 0.781)	<0.771 ± 0.091 (<0.651 - <0.866)	<0.699	<0.615	<0.778 ± 0.062 (<0.694 - <0.830)	<0.717 ± 0.078 (<0.662 - <0.772)	

\*CR OU2 Rodents: Mean concentration found in all rodents collected within Chestnut Ridge OU2.

**Appendix B**

**FISH DENSITY AND BIOMASS AT MCCOY BRANCH AND  
REFERENCE SITES, FALL 1990 TO FALL 1993**





**Table B.1. Fish densities and biomass in McCoy Branch, and in two reference streams, Grassy Creek and Scarboro Creek, for fall 1990**

Density expressed as individuals per square meter; biomass (in parenthesis) expressed as grams per square meter

Species	MCK 1.6	Sites <sup>a</sup>	
		GCK 2.4	SCK 2.2
Cyprinidae			NS <sup>b</sup>
Bluntnose minnow	0.52 (1.00)	-	
Blacknose dace	-	2.54 (2.11)	
Creek chub	-	0.83 (3.25)	
Ictaluridae			
Yellow bullhead	0.41 (6.66)	-	
Cottidae			
Banded sculpin	-	0.04 (0.44)	
Atherinidae			
Brook silverside	0.03 (0.04)	-	
Centrarchidae			
Green sunfish	0.41 (2.77)	-	
Bluegill	0.14 (0.36)	-	
Spotted bass	0.03 (0.21)	-	
Largemouth bass	0.03 (0.20)	-	
Percidae			
Snubnose darter	0.07 (0.11)	-	
Number of species (N)	8	3	
Total density	1.64	3.41	
Total biomass	(11.35)	(5.80)	

<sup>a</sup>MCK = McCoy Branch kilometer; GCK = Grassy Creek kilometer; SCK = Scarboro Creek kilometer.

<sup>b</sup>NS = Not sampled.

Table B.2. Fish densities and biomass (in parenthesis) in McCoy Branch, and in two reference streams, Grassy Creek and Scarboro Creek for spring 1991

Species	MCK 1.6	Sites <sup>a</sup> GCK 2.4	SCK 2.2
Clupeidae			
Threadfin shad	0.13 <sup>b</sup> (0.74) <sup>c</sup>	-	-
Cyprinidae			
Central stoneroller	-	-	0.16 (2.00)
Striped shiner	0.03 (0.23)	-	-
Bluntnose minnow	0.23 (0.99)	-	-
Fathead minnow	0.03 (0.09)	-	-
Blacknose dace	0.07 (0.08)	0.20 (0.19)	0.52 (1.92)
Creek chub	-	0.18 (1.10)	0.04 (0.08)
Catostomidae			
White sucker	0.03 (0.24)	0.01 (0.36)	-
Northern hog sucker	-	-	0.02 (0.69)
Ictaluridae			
Yellow bullhead	0.10 (1.28)	-	-
Cottidae			
Banded sculpin	-	0.02 (0.19)	1.25 (5.68)
Centrarchidae			
Redbreast sunfish	0.10 (5.77)	-	-
Green sunfish	3.19 (28.81)	-	0.12 (0.66)
Warmouth	0.10 (1.11)	-	-
Bluegill	6.13 (65.03)	-	-
Redear sunfish	0.07 (0.82)	-	-

Table B.2 (continued)

Species	MCK 1.6	Sites <sup>a</sup> GCK 2.4	SCK 2.2
Percidae			
Blueside darter	0.03 (0.06)	-	-
Snubnose darter	0.71 (1.31)	-	-
Logperch	0.32 (5.19)	-	0.04 (0.38)
Number of species (N)	15	4	7
Total density	12.30	0.41	2.15
Total biomass	(111.75)	(1.84)	(11.41)

<sup>a</sup>MCK = McCoy Branch kilometer; GCK = Grassy Creek kilometer; SCK = Scarboro Creek kilometer.

<sup>b</sup>Fish density expressed as number of individuals per square meter.

<sup>c</sup>Biomass expressed as grams per square meter.

Table B.3. Fish densities and biomass (in parenthesis) in McCoy Branch, and in two reference streams, Grassy Creek and Scarboro Creek for fall 1991

Species	MCK 1.6	Sites <sup>a</sup> GCK 2.4	SCK 2.2
<b>Cyprinidae</b>			
Central stoneroller	-	-	1.53 <sup>b</sup> (8.65) <sup>c</sup>
Spotfin shiner	0.05 (0.13)	-	0.69 (1.70)
Bluntnose minnow	0.50 (1.22)	-	0.71 (0.89)
Blacknose dace	-	3.24 (2.15)	0.61 (2.59)
Creek chub	-	0.72 (2.54)	0.06 (0.41)
<b>Catostomidae</b>			
White sucker	-	0.37 (0.77)	-
<b>Ictaluridae</b>			
Yellow bullhead	0.34 (3.87)	-	-
<b>Poeciliidae</b>			
Western mosquitofish	-	-	0.01 (0.01)
<b>Cottidae</b>			
Banded sculpin	-	-	1.58 (5.98)
<b>Centrarchidae</b>			
Redbreast sunfish	-	-	0.17 (0.85)
Green sunfish	0.14 (1.35)	0.02 (0.17)	0.46 (3.91)
Warmouth	0.05 (0.53)	-	-
Bluegill	0.23 (0.84)	-	0.29 (1.00)

Table B.3 (continued)

Species	MCK 1.6	Sites <sup>a</sup>	
		GCK 2.4	SCK 2.2
Hybrid sunfish	-	-	0.01 (0.10)
Spotted bass	0.02 (0.03)	-	-
Largemouth bass	-	-	0.01 (0.06)
Percidae			
Logperch	-	-	0.06 (0.40)
Number of species (N)	7	4	13
Total density	1.03	4.35	6.19
Total biomass	(7.97)	(5.63)	(26.55)

<sup>a</sup>MCK = McCoy Branch kilometer; GCK = Grassy Creek kilometer; SCK = Scarboro Creek kilometer.

<sup>b</sup>Fish density expressed as number of individuals per square meter.

<sup>c</sup>Biomass expressed as grams per square meter.

Table B.4. Fish densities and biomass (in parenthesis) in McCoy Branch, and in two reference streams, Grassy Creek and Scarboro Creek for spring 1992

Species	MCK 1.6	Sites <sup>a</sup> GCK 2.4	SCK 2.2
Cyprinidae			
Central stoneroller	-	-	0.87 <sup>b</sup> (10.00) <sup>c</sup>
Bluntnose minnow	1.08 (2.64)	-	0.04 (0.10)
Blacknose dace	-	1.01 (0.94)	0.37 (1.21)
Creek chub	-	0.39 (1.08)	0.01 (0.10)
Catostomidae			
White sucker	-	0.11 (0.29)	-
Northern hog sucker	-	-	0.07 (4.96)
Ictaluridae			
Yellow bullhead	0.14 (2.00)	-	0.01 (0.18)
Cottidae			
Banded sculpin	-	-	1.00 (5.36)
Centrarchidae			
Redbreast sunfish	-	-	0.03 (0.19)
Green sunfish	0.84 (8.75)	-	0.30 (3.10)
Bluegill	0.06 (0.23)	-	0.04 (0.25)
Redear sunfish	0.02 (0.06)	-	-
Percidae			
Snubnose darter	0.06 (0.15)	-	-

Table B.4 (continued)

Species	MCK 1.6	Sites <sup>a</sup>	
		GCK 2.4	SCK 2.2
Logperch	0.02 (0.13)	-	0.01 (0.15)
Number of species (N)	7	3	11
Total density	2.22	1.51	2.75
Total biomass	(13.96)	(2.31)	(25.60)

<sup>a</sup>MCK = McCoy Branch kilometer; GCK = Grassy Creek kilometer; SCK = Scarboro Creek kilometer.

<sup>b</sup>Fish density expressed as number of individuals per square meter.

<sup>c</sup>Biomass expressed as grams per square meter.



Table B.5. Fish densities and biomass (in parenthesis) in McCoy Branch, and in two reference streams, Grassy Creek and Scarboro Creek for fall 1992

Species	Sites <sup>a</sup>		
	MCK 1.6	GCK 2.4	SCK 2.2
<b>Cyprinidae</b>			
Central stoneroller	0.07 <sup>b</sup> (0.39) <sup>c</sup>	-	2.12 (17.40)
Spotfin shiner	0.90 (1.80)	-	0.18 (0.44)
Bluntnose minnow	1.88 (3.49)	-	0.61 (1.05)
Fathead minnow	0.10 (0.20)	-	-
Blacknose dace	0.07 (0.46)	3.85 (1.87)	0.30 (1.81)
Creek chub	-	0.42 (1.33)	-
Striped shiner	-	0.02 (0.25)	-
<b>Catostomidae</b>			
White sucker	-	0.06 (0.48)	-
<b>Ictaluridae</b>			
Yellow bullhead	-	-	0.05 (0.87)
<b>Poeciliidae</b>			
Western mosquitofish	0.07 (0.06)	-	-
<b>Cottidae</b>			
Banded sculpin	-	-	1.31 (6.13)
<b>Centrarchidae</b>			
Redbreast sunfish	-	-	0.01 (0.07)
Green sunfish	0.49 (1.66)	0.02 (0.07)	0.30 (4.30)

Table B.5 (continued)

Species	MCK 1.6	Sites <sup>a</sup>	
		GCK 2.4	SCK 2.2
Bluegill	0.27 (1.67)	-	0.03 (0.14)
Spotted bass	0.02 (0.07)	-	-
Number of species (N)	9	5	9
Total density	3.87	4.37	4.91
Total biomass	(9.80)	(4.00)	(32.21)

<sup>a</sup>MCK = McCoy Branch kilometer; GCK = Grassy Creek kilometer; SCK = Scarboro Creek kilometer.

<sup>b</sup>Fish density expressed as number of individuals per square meter.

<sup>c</sup>Biomass expressed as grams per square meter.

Table B.6. Fish densities and biomass (in parenthesis) in McCoy Branch, and in two reference streams, Grassy Creek and Scarboro Creek for spring 1993

Species	MCK 1.6	Sites <sup>a</sup> GCK 2.4	SCK 2.2
Cyprinidae		NS <sup>b</sup>	
Central stoneroller	-	-	2.49 <sup>c</sup> (31.76) <sup>d</sup>
Bluntnose minnow	0.59 (1.44)	-	-
Blacknose dace	-	-	0.47 (2.30)
Creek chub	-	-	0.03 (0.32)
Ictaluridae			
Yellow bullhead	0.02 (0.88)	-	0.01 (0.96)
Cottidae			
Banded sculpin	-	-	0.75 (3.94)
Centrarchidae			
Green sunfish	3.05 (38.10)	-	0.16 (1.69)
Bluegill	0.27 (2.03)	-	-
Largemouth bass	0.02 (0.84)	-	-
Percidae			
Greenside darter	0.02 (0.08)	-	-
Snubnose darter	0.10 (0.21)	-	-
Logperch	-	-	0.13 (1.26)

Table B.6 (continued)

Species	MCK 1.6	Sites <sup>a</sup>	
		GCK 2.4	SCK 2.2
Number of species (N)	7		7
Total density	4.07		4.04
Total biomass	(43.58)		(42.23)

<sup>a</sup>MCK = McCoy Branch kilometer; GCK = Grassy Creek kilometer; SCK = Scarboro Creek kilometer.

<sup>b</sup>NS = Not sampled.

<sup>c</sup>Fish density expressed as number of individuals per square meter.

<sup>d</sup>Biomass expressed as grams per square meter.

Table B.7. Fish densities and biomass (in parenthesis) in McCoy Branch, and in two reference streams, Grassy Creek and Scarboro Creek for fall 1993

Species	MCK 1.6	Sites <sup>a</sup> GCK 2.4	SCK 2.2
Cyprinidae		NS <sup>b</sup>	
Central stoneroller	-	-	3.46 <sup>c</sup> (33.95) <sup>d</sup>
Spotfin shiner	0.26 (0.71)	-	0.10 (0.21)
Bluntnose minnow	0.34 (0.38)	-	1.88 (3.28)
Fathead minnow	-	-	0.02 (0.03)
Blacknose dace	0.06 (0.19)	-	0.38 (2.69)
Creek chub	0.03 (0.03)	-	-
Ictaluridae			
Yellow bullhead	-	-	0.02 (0.11)
Cottidae			
Banded sculpin	-	-	0.68 (2.41)
Centrarchidae			
Redbreast sunfish	-	-	0.03 (0.28)
Green sunfish	0.23 (0.97)	-	0.28 (3.06)
Bluegill	0.17 (0.61)	-	0.69 (2.27)
Spotted bass	0.03 (0.04)	-	-
Largemouth bass	-	-	0.02 (0.04)
Number of species	7		11
Total density	1.12		7.56
Total biomass	(2.93)		(48.33)

<sup>a</sup>MCK = McCoy Branch kilometer; GCK = Grassy Creek kilometer; SCK = Scarboro Creek kilometer.

<sup>b</sup>NS = Not sampled.

<sup>c</sup>Fish density expressed as number of individuals per square meter.

<sup>d</sup>Biomass expressed as grams per square meter.

## **Appendix C**

### **LENGTH-FREQUENCY HISTOGRAMS FOR BLUEGILL, GREEN SUNFISH, AND BLUNTNOSSE MINNOW, FALL 1990-FALL 1993**



ORNL-DWG 96-7211

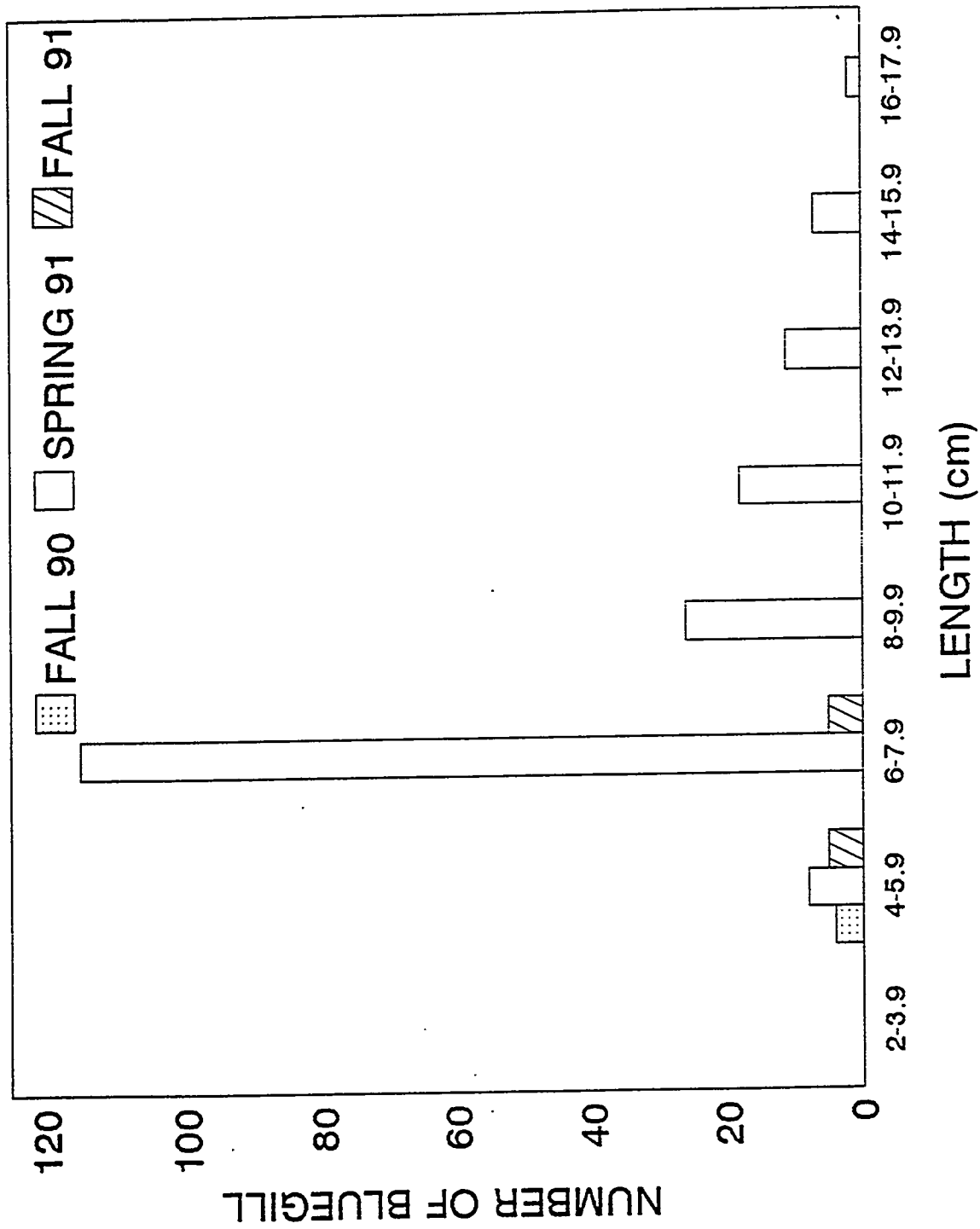


Fig. C.1. Length-frequency histogram for bluegill (*Lepomis macrochirus*) in McCoy Branch (MCK 1.56), fall 1990 through fall 1991.



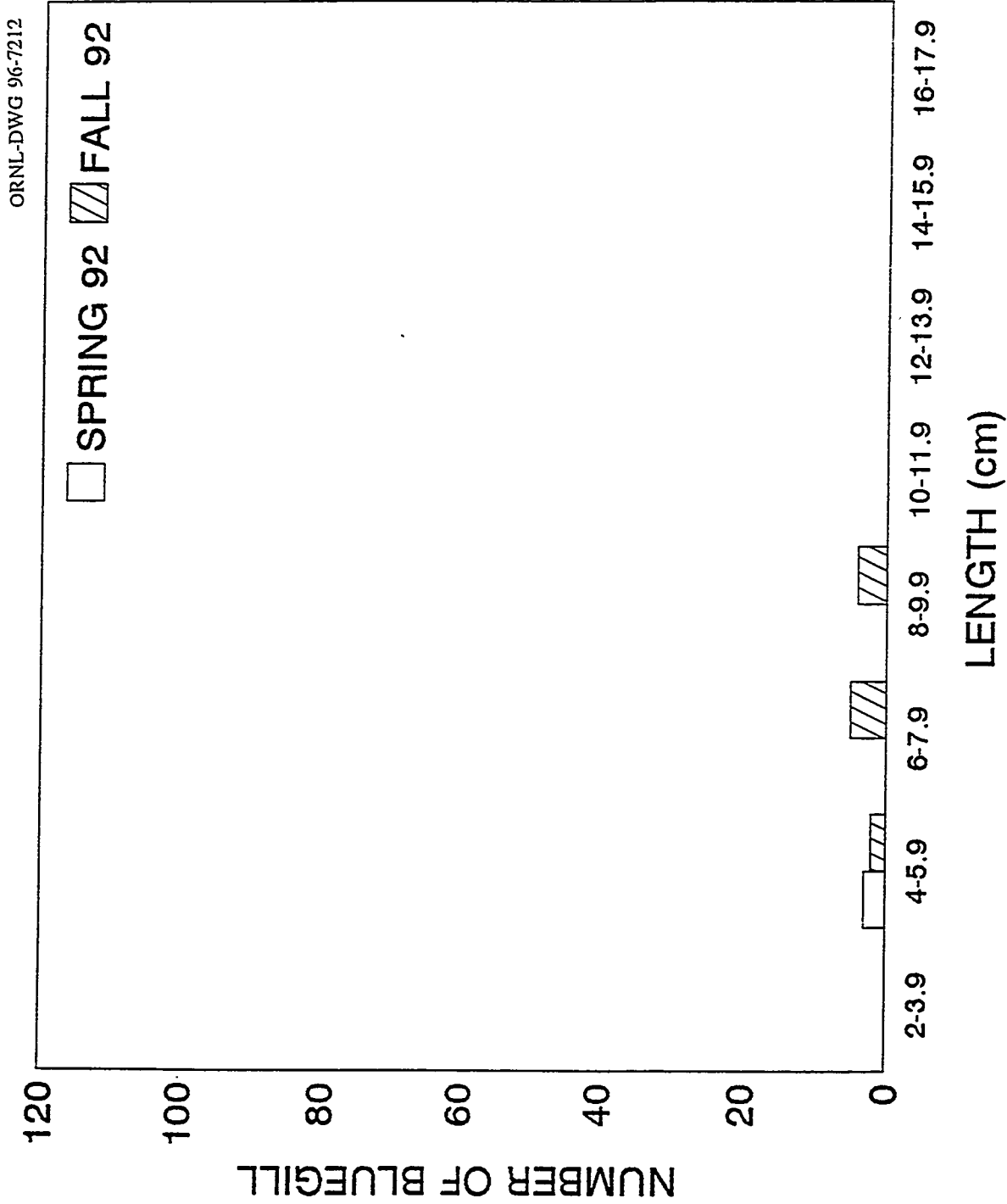


Fig. C.2. Length-frequency histogram for bluegill (*Lepomis macrochirus*) in McCoy Branch (MCK 1.56), 1992.

ORNLDWG 96-7213

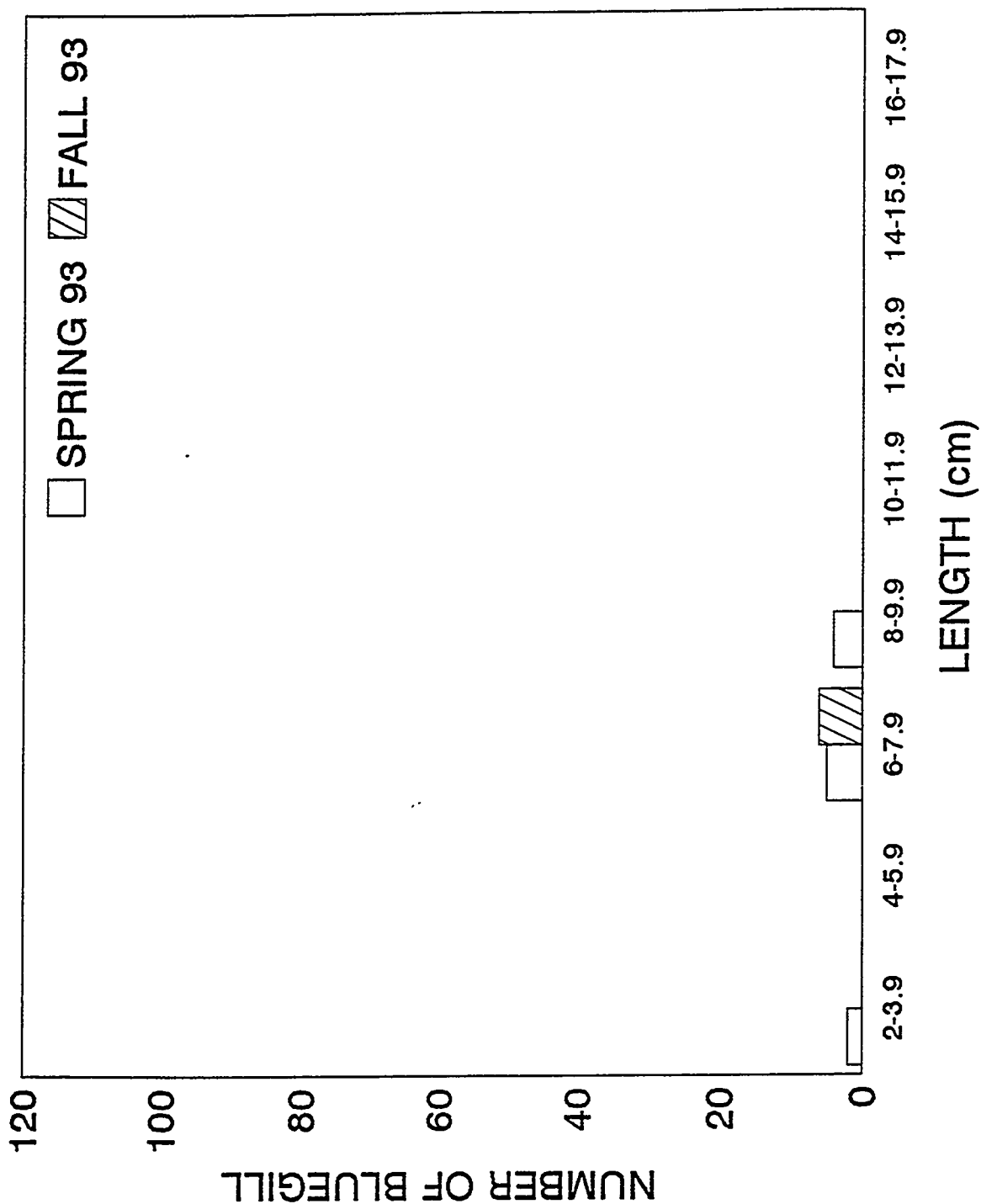


Fig. C.3. Length-frequency histogram for bluegill (*Lepomis macrochirus*) in McCoy Branch (MCK 1.56), 1993.

ORNLDWG 96-7214

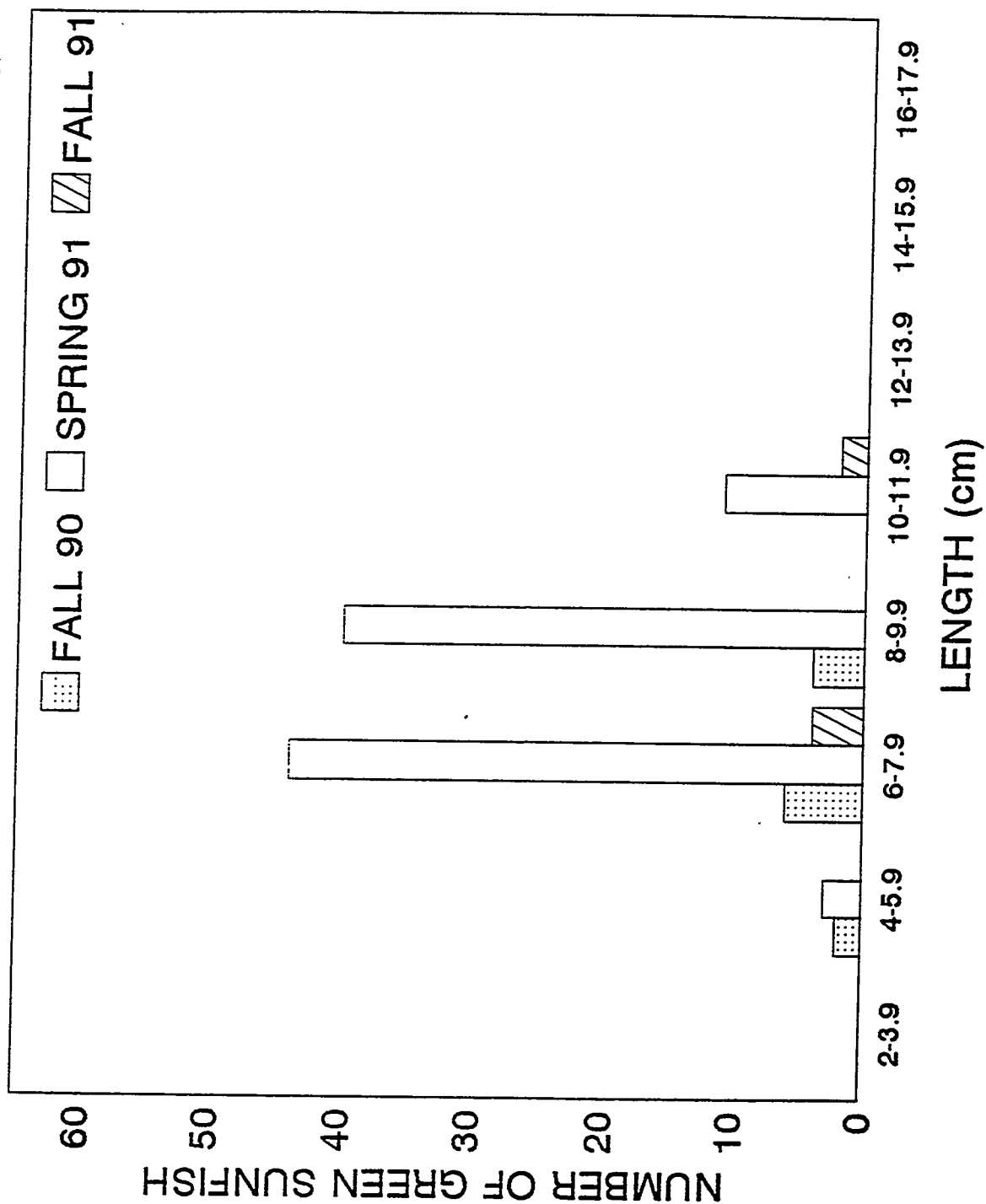


Fig. C.4. Length-frequency histogram for green sunfish (*Lepomis cyanellus*) in McCoy Branch (MCK 1.56), fall 1990 through fall 1991.

ORNLDWG 96-7215

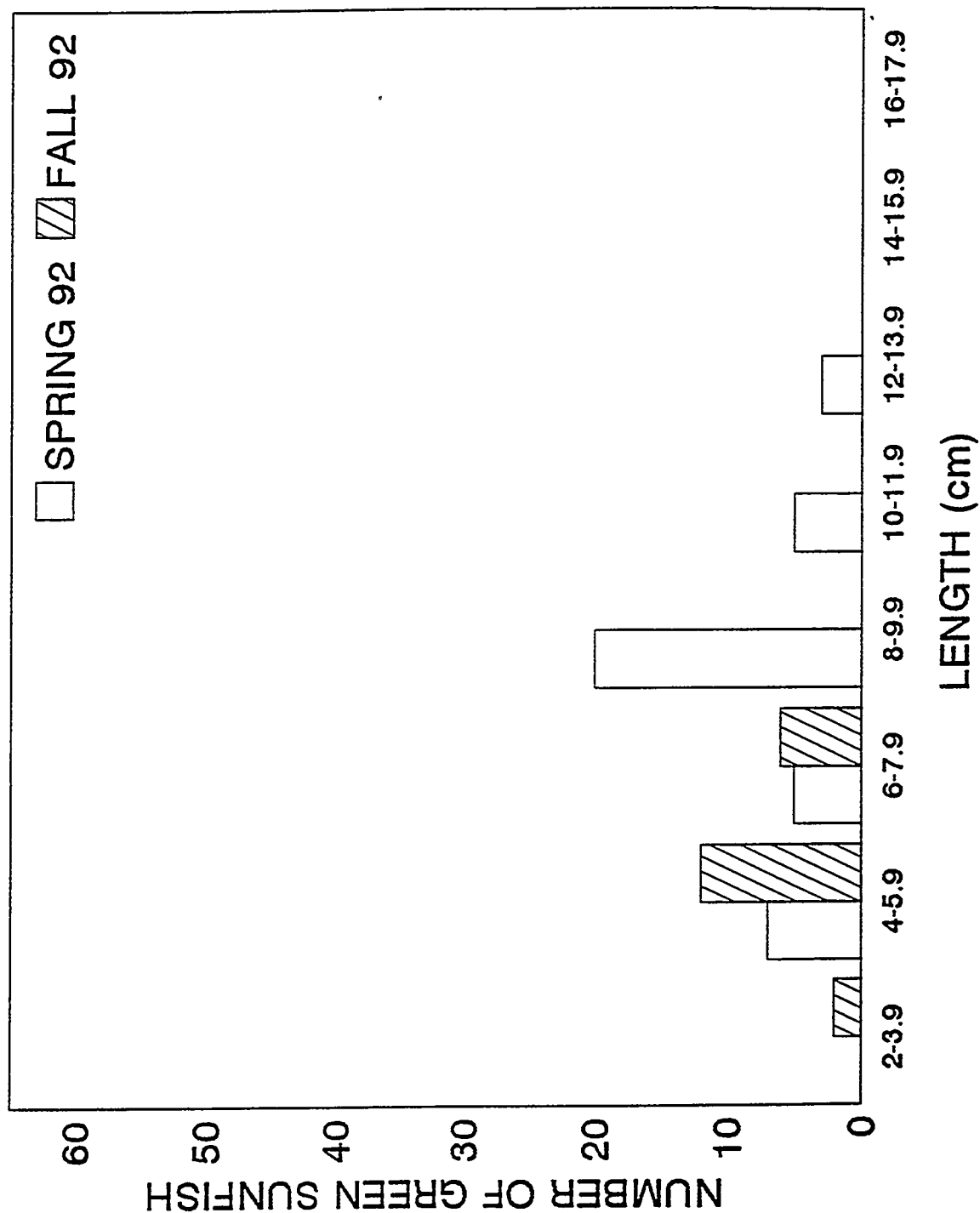


Fig. C.5. Length-frequency histogram for green sunfish (*Lepomis cyanellus*) in McCoy Branch (MCK 1.56), 1992.

ORNLDWG 96-7216

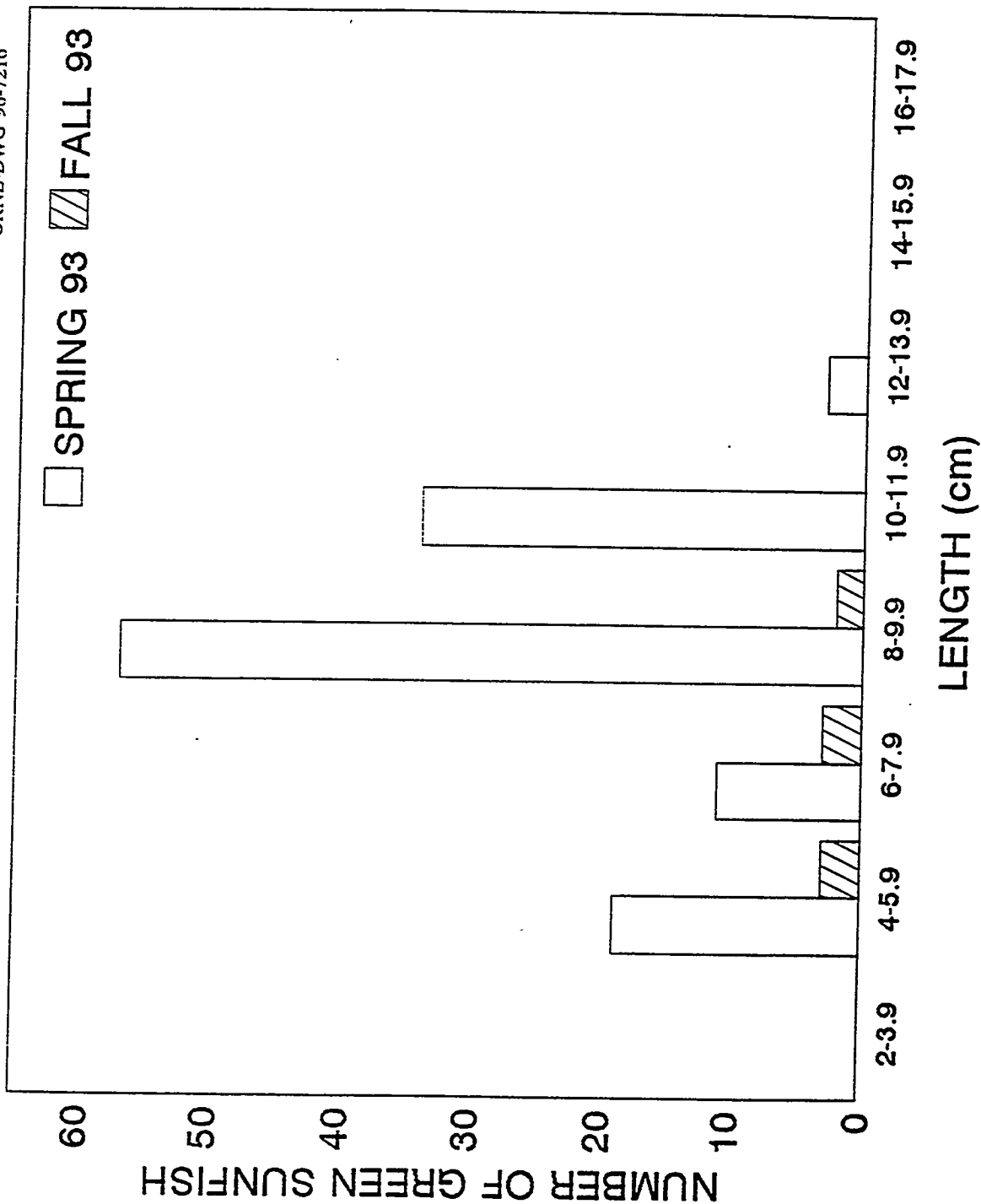


Fig. C.6. Length-frequency histogram for green sunfish (*Lepomis cyanellus*) in McCoy Branch (MCK 1.56), 1993.

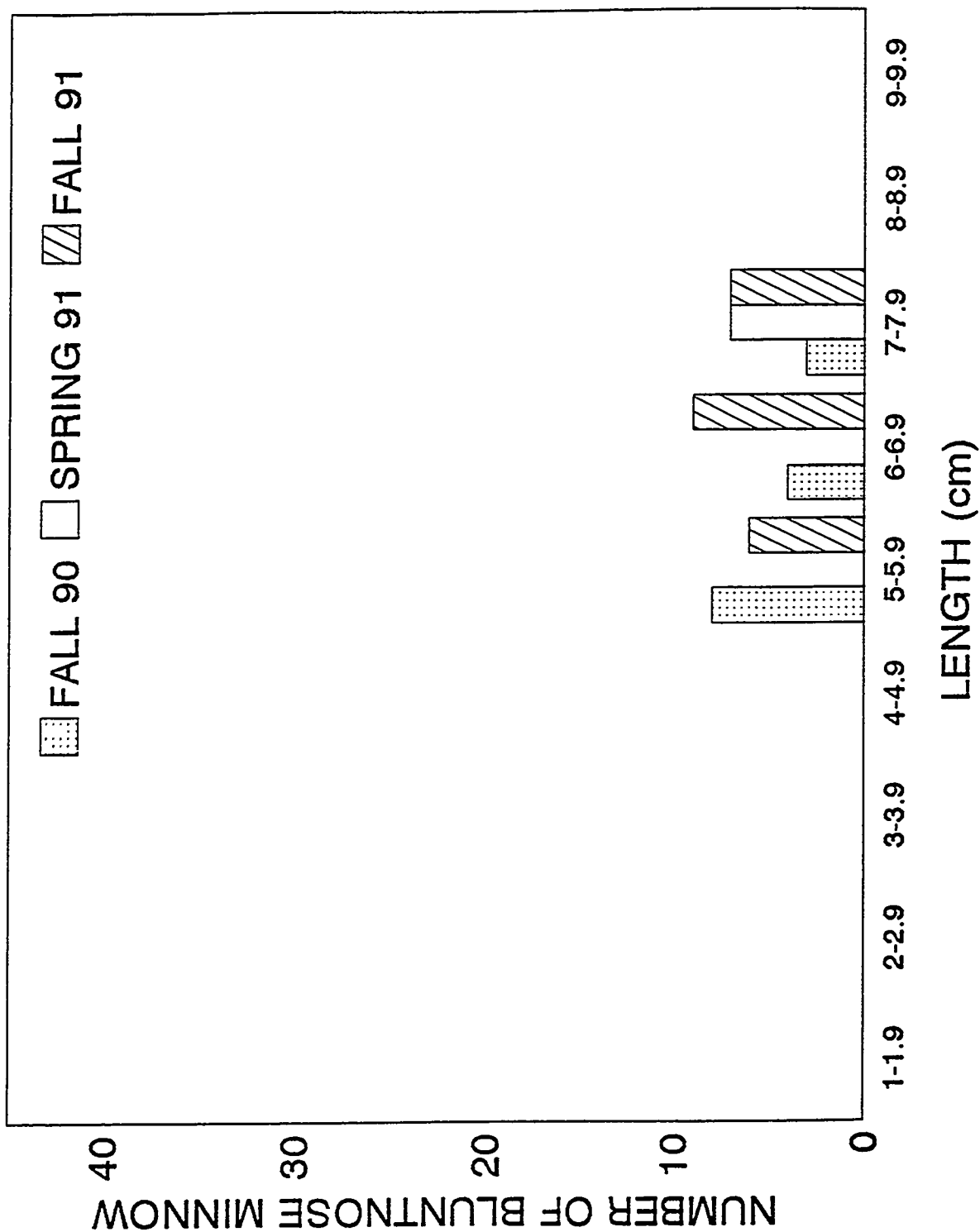


Fig. C.7. Length-frequency histogram for bluntnose minnow (*Pimephales notatus*) in McCoy Branch (MCK 1.56), fall 1990 through fall 1991.

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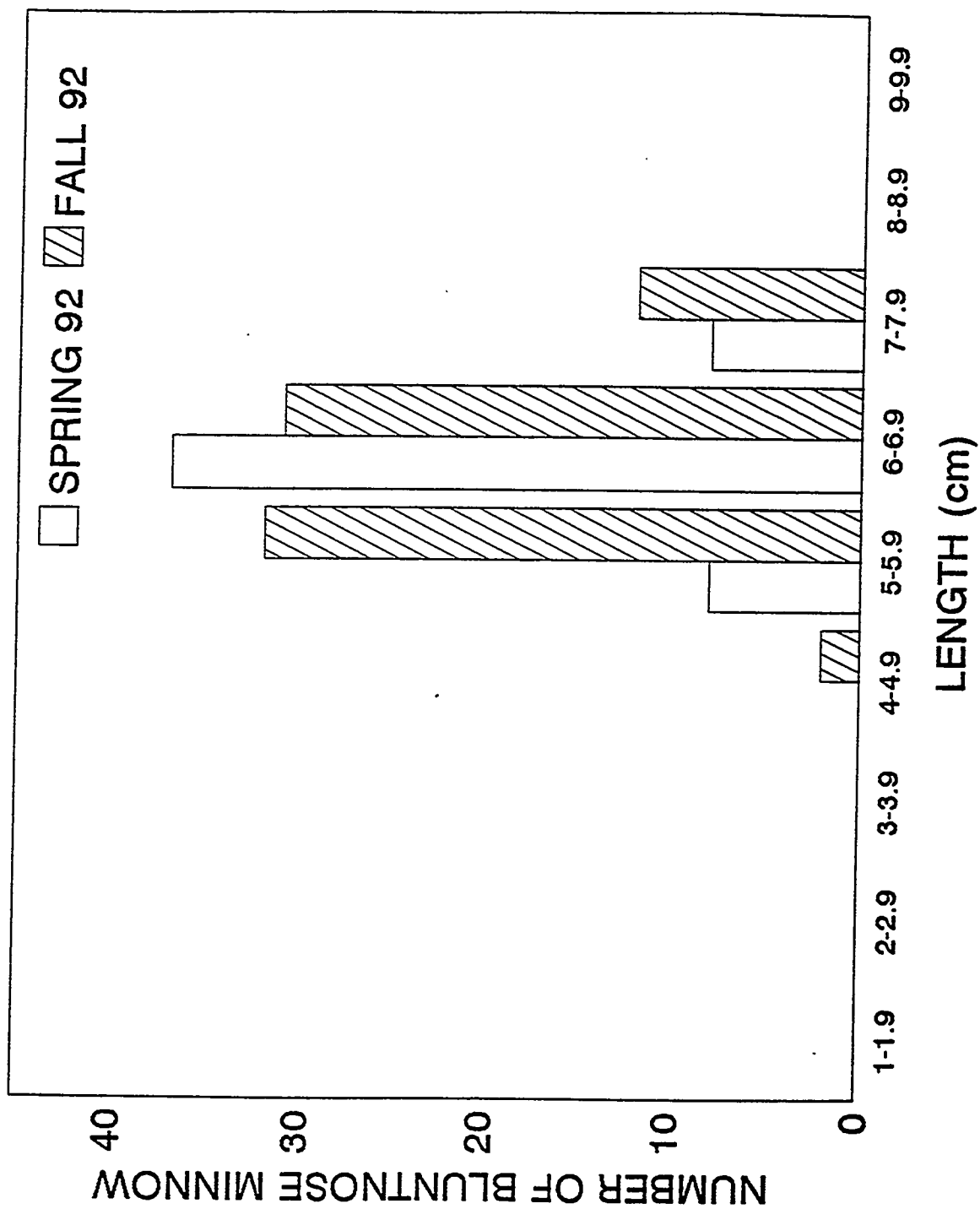


Fig. C.8. Length-frequency histogram for bluntnose minnow (*Pimephales notatus*) in McCoy Branch (MCK 1.56), 1992.

ORNLDWG 96-7219

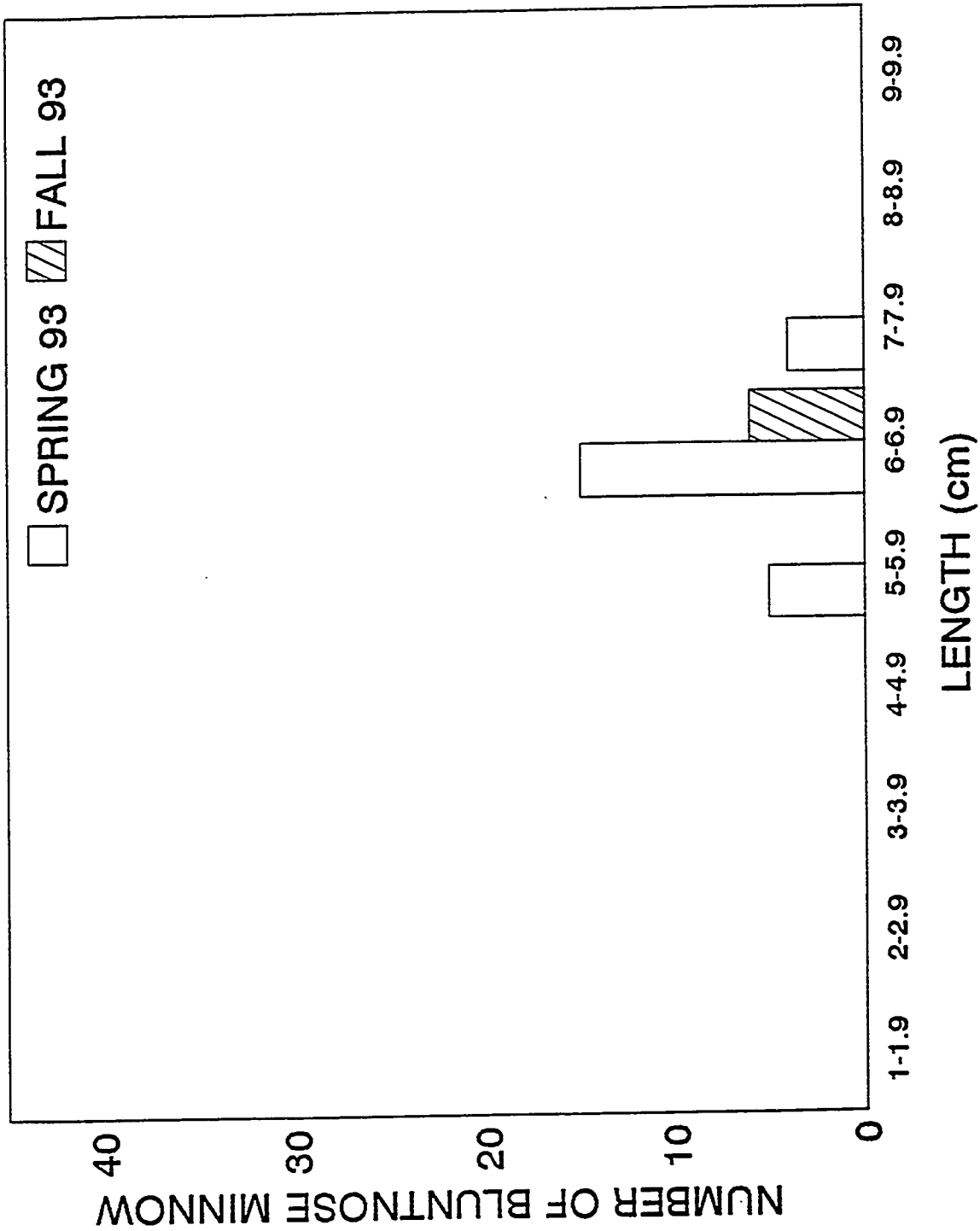


Fig. C.9. Length-frequency histogram for bluntnose minnow (*Pinephales notatus*) in McCoy Branch (MCK 1.56), 1993.



**Appendix D**

**CHECKLIST OF BENTHIC MACROINVERTEBRATE TAXA FROM  
MCCOY BRANCH AND WHITE OAK CREEK,  
APRIL 1989-OCTOBER 1993**



Table D.1. Checklist of benthic macroinvertebrates collected from McCoy Branch and White Oak Creek, April 1989–October 1993

Taxon	Site <sup>a,b</sup>		
	MCK 1.4	MCK 1.9	WCK 6.8
Coelenterata			
Hydridae			
<i>Hydra</i>	-	-	1
Turbellaria			
Tricladida			
Planariidae	1,2,3,5	1,2,3,4,5	1,2,3,4,5
Nematoda	1,2,3,4,5	2,3,4	1,2,3,4,5
Annelida			
Hirudinea	-	4	-
Hirudinea?	3,5	-	-
Oligochaeta	1,2,3,4,5	1,2,3,4,5	1,2,3,4,5
<i>Branchiura</i>			
<i>sowerbyi</i>	3,4	-	-
Crustacea			
Isopoda			
Asellidae			
<i>Lirceus</i>	3	-	-
Amphipoda			
Gammaridae			
<i>Crangonyx</i>	4	5	1,3,5
<i>Gammarus</i>	3	2	5
Decapoda			
Cambaridae	4,5	4	5
<i>Cambarus</i>	5	3	3,4
Hydracarina	4,5	1,3,4,5	1,3,4,5
Insecta			
Ephemeroptera			
Baetidae	-	1	-
<i>Baetis</i>	1,2,3,4,5	1,2,3,4,5	1,2,3,4,5
<i>Pseudocloeon</i>	1,3,4,5	3,4,5	1,2,3,4,5
<i>Pseudocloeon?</i>	-	-	5
Baetidae?	-	-	1

Table D.1 (continued)

Taxon	Site <sup>a,b</sup>		
	MCK 1.4	MCK 1.9	WCK 6.8
Ephemeroptera (continued)			
Caenidae			
<i>Caenis</i>	-	2	-
Ephemerellidae	-	-	1,2,3,4,5
<i>Ephemerella</i>	5	2,4	1,2,3,4,5
<i>Eurylophella</i>	1,2,3,4,5	2,3,4,5	1,2,3,4,5
Ephemeridae			
<i>Ephemerella</i>	-	-	1,3,4,5
Heptageniidae	-	-	1,3,4,5
<i>Epeorus</i>	-	3,4	2
<i>Stenacron</i>	-	-	1,4,5
<i>Stenonema</i>	-	-	1,2,3,4,5
Leptophlebiidae	2,4	1,3	1
<i>Habrophlebiodes</i>	2,3,4,5	3,4	1,2,3,4,5
<i>Paraleptophlebia</i>	2,3,4,5	2	1,3,4,5
Oligoneuriidae			
<i>Isonychia</i>	-	-	1,2,3,5
Odonata	3	-	-
Anisoptera			
Aeshnidae			
<i>Boyeria vinosa</i>	3,4,5	2	-
Cordulegastridae			
<i>Cordulegaster</i>	2,4,5	3,4,5	5
Gomphidae	-	-	1,4,5
<i>Stylogomphus</i>			
<i>albistylus</i>	-	2,3	1,2,3,4,5
<i>Stylopomphus?</i>	-	-	1,4
Gomphidae?	-	-	4
Libellulidae?	1	-	-
Zygoptera			
Calopterygidae			
<i>Calopteryx</i>	2,3,4,5	1,2,3,4,5	-
<i>Calopteryx?</i>	-	1	-
Coenagrionidae	4	-	-
<i>Argia</i>	5	-	-
<i>Enallagma</i>	2	-	-

Table D.1 (continued)

Taxon	Site <sup>a,b</sup>		
	MCK 1.4	MCK 1.9	WCK 6.8
Plecoptera	1	-	-
Capniidae			
<i>Allocapnia</i>	2,3,5	2,3	2,3,4,5
Capniidae/Leuctridae	-	-	1
Choloroperlidae	2	-	1,2,3,4,5
<i>Alloperla</i>	-	2	5
<i>Haploperla</i>	-	5	1,2,3,4,5
<i>Sweltsa</i>	-	-	1,2,3,4,5
Leuctridae			
<i>Leuctra</i>	2,3,5	2,3,4,5	1,2,3,4,5
Nemouridae			
<i>Amphinemura</i>	2,3,4,5	2,3,4,5	1,2,3,4,5
Peltoperlidae	-	-	3
<i>Peltoperla</i>	-	5	-
<i>Tallaperla</i>	2	1,3,4	2,3,4,5
<i>Viehoperla?</i>	-	-	1
Perlidae	3,4	-	1,3,4,5
<i>Acroneuria</i>	-	-	3
<i>Eccoptura xanthenes</i>	-	3	1,2,3,4,5
<i>Perlesta</i>	3,4,5	2,3	2,3,5
Perlodidae	-	1,4	2,3,5
<i>Isoperla</i>	1,2,3,4	2,3,5	2,3,5
Perlidae/Perlodidae	2,4,5	-	2,3,4,5
Megaloptera			
Corydalidae			
<i>Chauliodes</i>	-	5	-
<i>Nigronia fasciatus</i>	5	3,4,5	1,2,3,4,5
<i>Nigronia serricornis</i>	-	-	1,2,3
<i>Nigronia</i>	-	-	1
Sialidae			
<i>Sialis</i>	5	-	1,4,5
Trichoptera	3	3	4
Glossosomatidae			
<i>Agapetus</i>	3,5	2,3,4,5	1,2,3,4,5
<i>Glossosoma</i>	1,2	3,4	2,3,4,5
Goeridae			
<i>Goera</i>	5	5	1,2,3,4

Table D.1 (continued)

Taxon	Site <sup>a,b</sup>		
	MCK 1.4	MCK 1.9	WCK 6.8
Trichoptera (continued)			
Hydropsychidae	1,3	-	1
<i>Cheumatopsyche</i>	1,2,3,4,5	1,2,3,4,5	1,2,3,4,5
<i>Diplectrona modesta</i>	2,3,4,5	2,3,4,5	1,2,3,4,5
<i>Hydropsyche</i>	1,2,3,4,5	1,2,3,4,5	2,3,5
Hydroptilidae			
<i>Hydroptila</i>	1,2,3,4,5	2,3,4	1,2
<i>Hydroptila?</i>	-	2	-
<i>Ochrotrichia</i>	2,3,4,5	-	2
Lepidostomatidae			
<i>Lepidostoma</i>	-	2,3,4,5	1,2,4,5
Leptoceridae	3	-	-
<i>Oecetis</i>	3	2,3	-
Limnephilidae			
<i>Neophylax</i>	1,3,5	2,3,4,5	1,2,3,4,5
<i>Pycnopsyche</i>	2,5	2,3	2
<i>Pycnopsyche?</i>	2,5	-	-
Molannidae			
<i>Molanna</i>	2	1,2,3	4
Odontoceridae			
<i>Psilotreta</i>	-	-	1,2,4,5
Philopotamidae			
<i>Chimarra</i>	1,2,3,4,5	1,2,3,5	1
<i>Dolophilodes</i>			
<i>distinctus</i>	-	2	1,2
<i>Wormaldia</i>	-	1	2,3,4
Phryganeidae			
<i>Ptilostomis</i>	2	-	-
Polycentropodidae			
<i>Polycentropus</i>	2	2	1,2,3,4,5
Psychomyiidae			
<i>Lype diversa</i>	2	1,3,4,5	-
Rhyacophilidae			
<i>Rhyacophila</i>	5	3,4,5	1,2,3,4,5

Table D.1 (continued)

Taxon	Site <sup>a,b</sup>		
	MCK 1.4	MCK 1.9	WCK 6.8
Coleoptera			
Elmidae	1	-	-
<i>Dubiraphia</i>	2,4,5	3	5
<i>Microcylloepus</i>			
<i>pusillus</i>	1,2,3,4,5	1,2,3,4	-
<i>Optioservus</i>	1,2,3,4,5	2,3,4,5	1,2,3,4,5
<i>Stenelmis</i>	1,2,3,4,5	1,2,3,4,5	1,2,3,4,5
Eubriidae			
<i>Ectopria</i>	3,4	2	1,2,3,4,5
Psephenidae			
<i>Psephenus herricki</i>	1,3,4,5	3,4	1,2,3,4,5
Ptilodactylidae			
<i>Ancyrtarsus bicolor</i>	2	3,5	1,2,3,4,5
Diptera -	-	1	
Ceratopogonidae	1,2,3,5	1,2,3,4,5	1,2,3,4,5
<i>Atrichopogon</i>	-	-	4
Chironomidae	1,2,3,4,5	1,2,3,4,5	1,2,3,4,5
Chironominae	4	1	-
Chironomini	1,2,3,4,5	1,2,3,4,5	1,2,3,4,5
Tanytarsini	1,2,3,4,5	1,2,3,4,5	1,2,3,4,5
Diamesinae	-	1	-
Orthoclaadiinae	1,2,3,4,5	1,2,3,4,5	1,2,3,4,5
Tanypodinae	1,2,3,4,5	1,2,3,4,5	1,2,3,4,5
Dixidae			
<i>Dixa</i>	2,4	1,2,3,4	1,2,3,4,5
Empididae			
<i>Chelifera</i>	3,4,5	5	-
<i>Clinocera</i>	2	3	2
<i>Hemerodromia</i>	1,2,3,4,5	1,2,3,4,5	1,2,3,4,5
<i>Hemerodromia?</i>	-	-	1
Psychodidae			
<i>Pericoma</i>	-	-	1
Schiomyzidae	-	1	-
Simuliidae			
<i>Simulium</i>	1,2,3,4,5	1,2,3,4,5	1,2,3,4,5

Table D.1 (continued)

Taxon	Site <sup>a,b</sup>		
	MCK 1.4	MCK 1.9	WCK 6.8
Diptera (continued)			
Stratiomyidae	3	-	-
<i>Caloparyphus</i>	4	-	-
<i>Myxosargus</i>	1,2,3,5	-	-
<i>Odontomyia</i>	1	5	-
Tabanidae			
<i>Chrysops</i>	2	3	-
<i>Tabanus</i>	2	-	5
Tipulidae	-	1	-
<i>Antocha</i>	-	4,5	1,2,3,4,5
<i>Dicranota</i>	-	4	-
<i>Helius</i>	4	-	-
<i>Limnophila</i>	2	-	2
<i>Polymera</i>	-	-	1
<i>Pseudolimnophila</i>	-	1,3,4	1,3
<i>Pseudolimnophila?</i>	-	3	-
<i>Tipula abdominalis</i>	5	-	-
<i>Tipula</i>	2,5	1,2,3,4,5	1,2,4
<i>Tipula?</i>	-	2	-
Mollusca			
Gastropoda	-	1,2	-
Ancylidae			
<i>Ferrissia</i>	5	5	-
Lymnaeidae	2	3	-
<i>Pseudosuccinea</i>			
<i>columella</i>	2,4	1,2	-
<i>Pseudosuccinea?</i>	5	-	-
Lymnaeidae?	1,5	4	-
Physidae			
<i>Physella</i>	1,2,4,5	1,2,3,4	-
Planorbidae	1,2	2	-
<i>Gyraulus</i>	1	-	-
Pleuroceridae			
<i>Elimia</i>	5	-	1,2,3,4,5
<i>Elimia?</i>	-	-	4



Table D.1 (continued)

Taxon	Site <sup>a,b</sup>		
	MCK 1.4	MCK 1.9	WCK 6.8
Bivalvia			
Corbiculidae			
<i>Corbicula fluminea</i>	1,3,4,5	1,2,3,4	-
Sphaeriidae	4	4	1,2,3,4
<i>Pisidium</i>	-	-	1
<i>Sphaerium</i>	1,3	-	1

<sup>a</sup>MCK = McCoy Branch kilometer; WCK = White Oak Creek kilometer.

<sup>b</sup>The numbers associated with each taxon and site indicate the year(s) that the taxon was collected at least once, with 1=1989, 2=1990, 3=1991, 4=1992, and 5=1993. A blank indicates that a lower level of classification (i.e., 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.

**Appendix E**

**F-VALUES AND *P*-VALUES FOR NESTED ANOVAS AND  
REGRESSION ANALYSIS**



Table E.1. Statistics for nested ANOVAs<sup>a</sup> and ANOVA t-test for equal regression slopes across sites<sup>b</sup> using the response variables density, total richness, and EPT<sup>c</sup> richness, McCoy Branch, April 1989–October 1993

Model/Effect <sup>d</sup>	Variable					
	Density		Total Richness		EPT Richness	
	F-Value	p-Value	F-Value	p-Value	F-Value	p-Value
<b>Nested ANOVA</b>						
<b>All Years</b>						
Site <sub>(2,10)</sub>	5.57	0.0236	9.42	0.0050	11.27	0.0027
Year <sub>(4,5)</sub>	7.30	0.0256	2.99	0.1302	2.56	0.1649
Season(Year) <sub>(5,10)</sub>	0.50	0.7716	0.52	0.7590	0.82	0.5608
Site*Year <sub>(8,10)</sub>	0.96	0.5140	0.87	0.5714	0.74	0.6603
Site*Season(Year) <sub>(10,60)</sub>	3.30	0.0018	4.69	0.0001	6.07	0.0001
<b>Excluding 1989</b>						
Site <sub>(2,8)</sub>	5.26	0.0347	6.96	0.0177	8.21	0.0115
Year <sub>(3,4)</sub>	5.57	0.0653	1.73	0.2978	0.41	0.7565
Season(Year) <sub>(4,8)</sub>	0.60	0.6711	0.24	0.9076	0.73	0.5947
Site*Year <sub>(6,8)</sub>	1.33	0.3450	0.53	0.7719	0.30	0.9214
Site*Season(Year) <sub>(8,48)</sub>	2.54	0.0214	2.57	0.0202	5.22	0.0001
<b>Regression</b>						
<b>All Years - Spring<sup>e</sup></b>						
All Sites <sub>(2,39)</sub>	0.62	0.5429	4.61	0.0160	4.00	0.0263
MCK 1.4 vs WCK 6.8 <sub>(1,26)</sub>	0.55	0.4650	10.64	0.0031	8.19	0.0082
MCK 1.9 vs WCK 6.8 <sub>(1,26)</sub>	1.26	0.2723	6.04	0.0210	7.02	0.0135
MCK 1.4 vs MCK 1.9 <sub>(1,26)</sub>	0.19	0.6688	0.01	0.9383	0.10	0.7510
<b>All Years - Fall<sup>e</sup></b>						
All Sites <sub>(2,39)</sub>	3.13	0.0548	1.93	0.1588	2.56	0.0898
MCK 1.4 vs WCK 6.8 <sub>(1,26)</sub>	2.98	0.0962	0.82	0.3723	4.42	0.0454
MCK 1.9 vs WCK 6.8 <sub>(1,26)</sub>	5.86	0.0228	4.79	0.0377	4.38	0.0462
MCK 1.4 vs MCK 1.9 <sub>(1,26)</sub>	1.02	0.3228	0.93	0.3441	0.08	0.7753
<b>Excluding 1989 - Spring<sup>e</sup></b>						
All Sites <sub>(2,30)</sub>	1.03	0.3683	1.83	0.1772	1.61	0.2168
MCK 1.4 vs WCK 6.8 <sub>(1,20)</sub>	0.81	0.3798	1.35	0.2598	0.79	0.3840
MCK 1.9 vs WCK 6.8 <sub>(1,20)</sub>	2.53	0.1276	0.44	0.5144	1.18	0.2912
MCK 1.4 vs MCK 1.9 <sub>(1,20)</sub>	0.20	0.6610	3.91	0.0619	2.52	0.1280
<b>Excluding 1989 - Fall<sup>e</sup></b>						
All Sites <sub>(2,30)</sub>	5.07	0.0127	4.68	0.0171	7.09	0.0030
MCK 1.4 vs WCK 6.8 <sub>(1,20)</sub>	4.50	0.0466	11.61	0.0028	17.49	0.0005
MCK 1.9 vs WCK 6.8 <sub>(1,20)</sub>	9.06	0.0069	6.36	0.0203	5.14	0.0346
MCK 1.4 vs MCK 1.9 <sub>(1,20)</sub>	1.91	0.1822	0.02	0.8943	1.91	0.1821

<sup>a</sup>ANOVA = Analysis of variance.

<sup>b</sup>MCK = McCoy Branch kilometer; WCK = White Oak Creek kilometer

<sup>c</sup>EPT = Ephemeroptera, Plecoptera, and Trichoptera.

<sup>d</sup>Values in parentheses are the degrees of freedom (numerator, denominator).

<sup>e</sup>Spring = April sampling periods; Fall = October sampling periods.

**Appendix F**

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



ORNL-DWG 96-7220

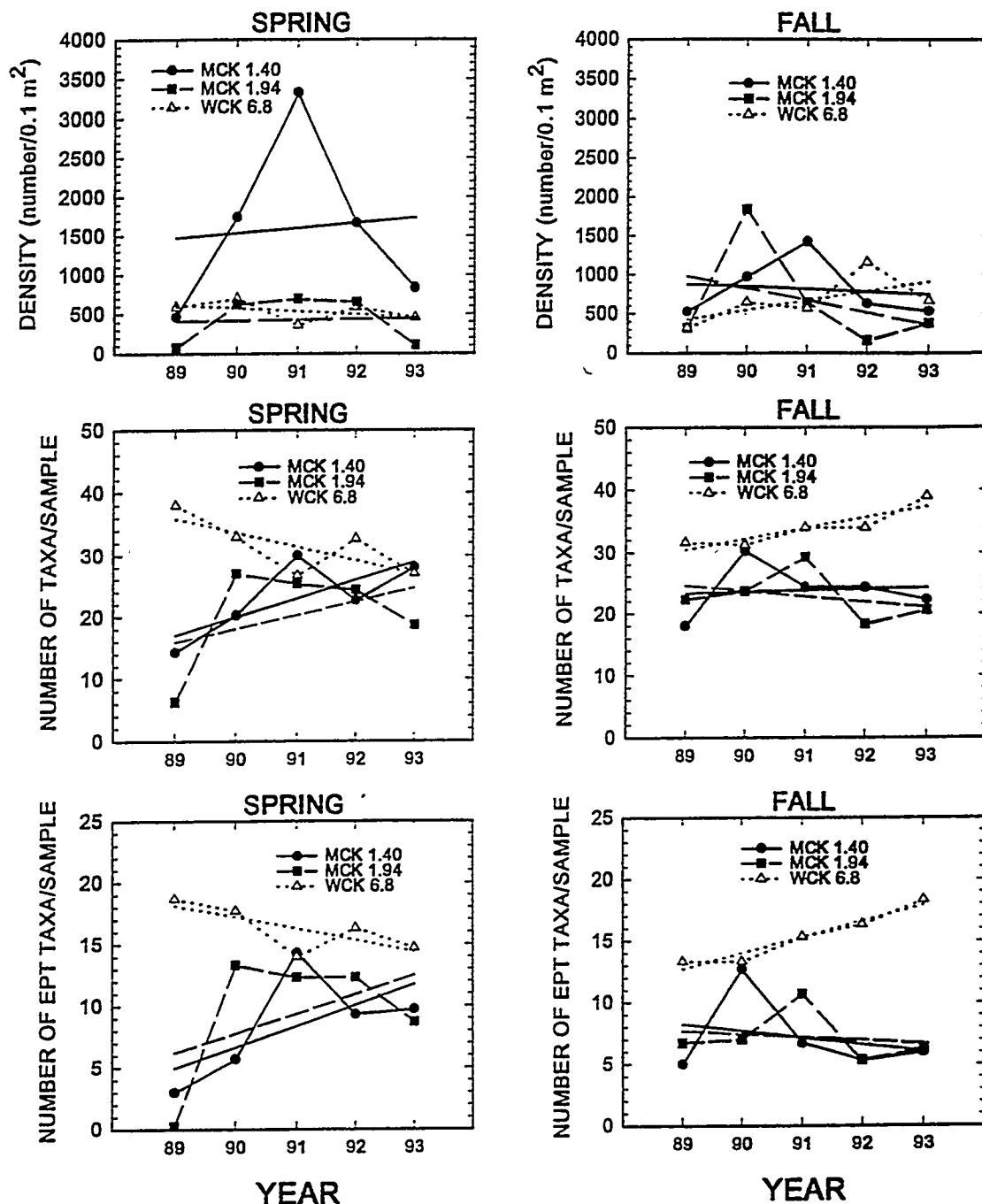


Fig. F.1. Seasonal regression lines for total density, total richness, and richness of the Ephemeroptera, Plecoptera, and Trichoptera (EPT richness) of the benthic macroinvertebrate communities in McCoy Branch and White Oak Creek, April 1989–October 1993. Estimates of error (i.e., standard error) are available in Fig. 7.1. MCK = McCoy Branch kilometer; WCK = White Oak Creek kilometer. Spring = April sampling periods; Fall = October sampling periods.

ORNL-DWG 96-7221

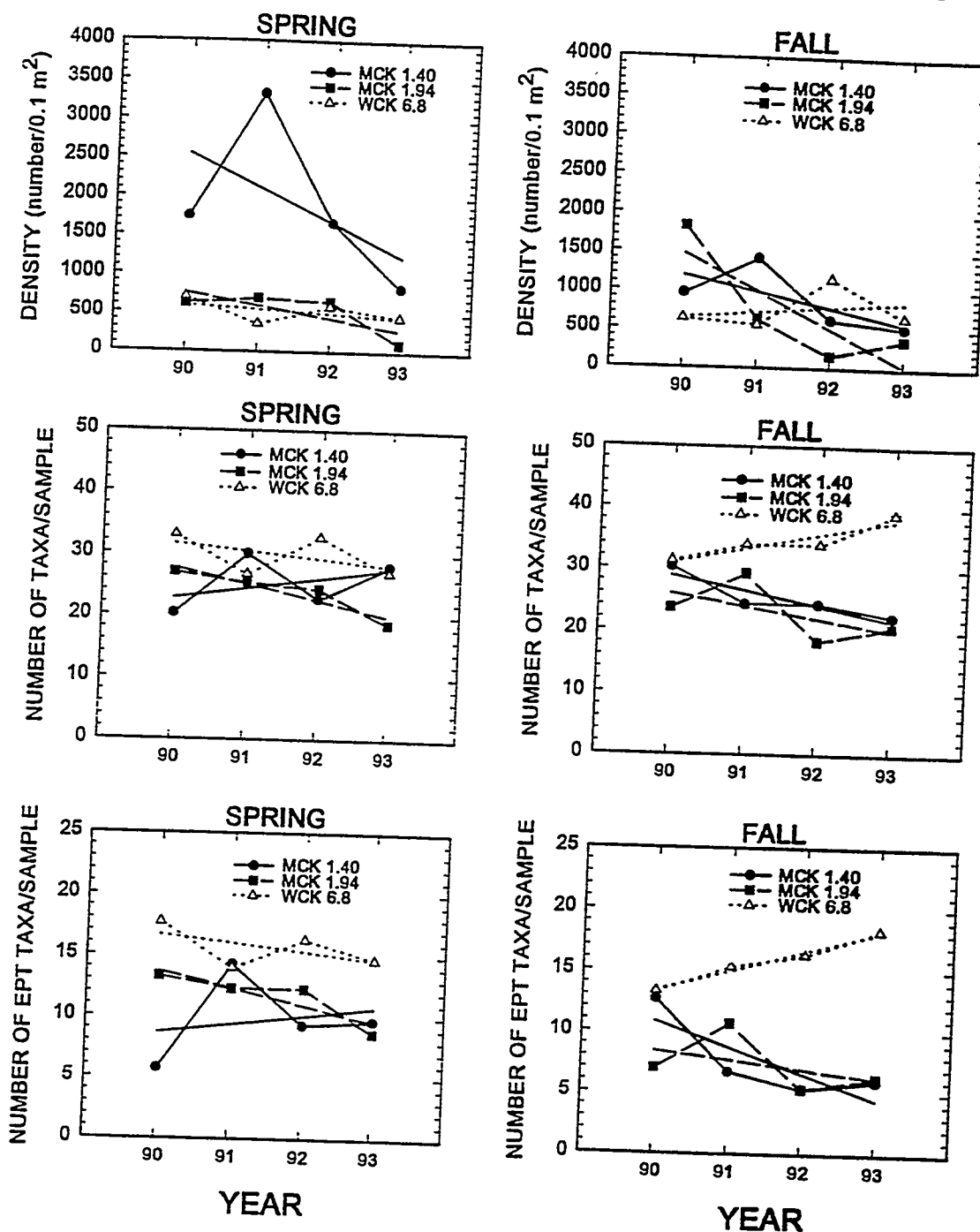


Fig. F.2. Seasonal regression lines for total density, total richness, and richness of the Ephemeroptera, Plecoptera, and Trichoptera (EPT richness) of the benthic macroinvertebrate communities in McCoy Branch and White Oak Creek, April 1990–October 1993. Estimates of error (i.e., standard error) are available in Fig. 7.1. MCK = McCoy Branch kilometer; WCK = White Oak Creek kilometer. Spring = April sampling periods; Fall = October sampling periods.



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