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To the Graduate Council:

I am submitting herewith a thesis written by Suzanne Jane Young entitled "A Comprehensive Evaluation of Benthic Invertebrate Communities in the Emory River, Watts Bar Reservoir, TN." I have examined the final electronic copy of this thesis for form and content and recommend that it be accepted in partial fulfillment of the requirements for the degree of Master of Science, with a major in Entomology and Plant Pathology.

Jerome F. Grant, Major Professor

We have read this thesis and recommend its acceptance:

J. Kevin Moulton, Teresa J. Mathews

Accepted for the Council: Carolyn R. Hodges

Vice Provost and Dean of the Graduate School

(Original signatures are on file with official student records.)

A Comprehensive Evaluation of Benthic Invertebrate Communities in the Emory River, Watts Bar Reservoir, TN

> A Thesis Presented for the Master of Science Degree The University of Tennessee, Knoxville

> > Suzanne Jane Young August 2015

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ABSTRACT

The release of fly ash at the Tennessee Valley Authority (TVA) Kingston Fossil Plant (KIF) on 22 December 2008 discharged approximately 4.1 million cubic meters of coal ash into the adjacent aquatic and terrestrial systems. Previous benthic invertebrate investigations conducted by TVA and collaborative researchers concluded that benthic invertebrates in the Emory River were at moderate risk from ash-related constituents. primarily arsenic, in ash-contaminated sediment that remained in the Emory River following extensive dredging efforts. These conclusions were based on the observation of statistically significant reductions in growth and biomass in laboratory toxicity tests with Emory River sediment. Benthic invertebrate community survey results from 2010, however, did not support this conclusion. These previous surveys evaluated benthic invertebrate community data and sediment data across a large spatial scale, providing an "area-wide" interpretation of the relationships between the benthic invertebrate community results to the ash release. In the present research, co-located sediment and benthic invertebrate community samples were collected from nine locations in the Emory River. Community metric results were compared among samples, locations, and previous years and to co-located sediment chemistry and physical sediment properties. Temporal trends were also evaluated over a 5-year period of time at two locations to gauge if an initial impact and/or recovery could be determined. Despite this refined investigation, no trends or significant differences were identified between ash-impacted locations compared to the reference location, and no evidence of an initial impact or subsequent recovery trends were established. Furthermore, no significant relationships could be established between benthic invertebrate community metrics and sediment chemistry results. This information is important for the informed monitoring, remediation,

iii

and damage assessment of the benthic invertebrate community at the Kingston Ash Recovery site. This research also increases our knowledge of benthic invertebrate tolerance to metal mixtures in sediment of natural systems.

TABLE OF CONTETS

CHAPTER 1 INTRODUCTION	1
1.1 Benthic Invertebrates as Biological Monitors and Indicators	1
1.2 Effects of Heavy Metals and Coal Ash on Benthic Invertebrates	6
1.3 Tennessee Valley Authority Background and Kingston Ash Release	9
1.4 Kingston Ash Release Site Conditions and Clean-Up	10
1.5 Emory River Hydrology and Sediment Characterization	12
1.6 Benthic Invertebrate Communities in the Emory River	14
1.7 Previous Benthic Invertebrate Studies in the Emory River	16
1.8 Objectives and Justification for Research	18
CHAPTER 2 SPATIAL ANALYSIS OF BENTHIC INVERTEBRATE	
COMMUNITIES IN THE EMORY RIVER AFTER DREDGING WATTS BAR	
RESERVOIR ROANE COUNTY IN	21
Abstract	22
2.1 Introduction	23
2.2 Materials and Methods	25
2.2.1 Study Site	.25
2.2.2 Sample Collection	
2.2.3 Benthic Invertebrate Metric Calculations	
2.2.4 Statistical Analysis	
2.3 Results	29
2.3.1 Benthic Invertebrate Community Metrics	29
2.3.2 Chemical Analysis	30
2.3.3 Substrate Composition	32
2.3.4 Relationships between Community Metrics and Sediment	33
2.4 Discussion	33
2.5 Summary	38
CHAPTER 3 TEMPORAL RESPONSES OF BENTHIC INVERTEBRATE	
COMMUNITIES IN THE EMORY RIVER IMPACTED BY COAL FLY ASH OVE	-R
FIVE YEARS WATTS BAR RESERVOIR ROANE COUNTY IN	40
Abstract	.41
3.1 Introduction	.42
3.2 Materials and Methods	.45
3.2.1 Study Site	45
3.2.2 Sample Collection	46
3.2.3 Benthic Invertebrate Metric Calculations	47
3.2.4 Statistical Analysis	49
3.3 Results	49
3.4 Discussion	53
3.5 Summary	58

CHAPTE 4.1 4.2	ER 4 CONCLUSIONS AND FUTURE RESEARCH NEEDS Conclusions Future Research Needs	60 60 64
REFERE	ENCES	65
APPENI Tables Figure	DICESs s	79
VITA		117

LIST OF TABLES

Table 1. Summary of the benthic invertebrate community composition in the Emory River following the TVA Kingston ash release, Watts Bar Reservoir,	
Roane County, TN ¹	1
Table 2. Average benthic invertebrate community metrics calculated in the Emor	y
River, Watts Bar Reservoir, Roane County, TN'8	3
Table 3. Average concentrations of ash-related metals (mg/kg) in submerged	
sediment collected in the Emory River, Watts Bar Reservoir, Roane County,	
TN ¹ 8	4
Table 4. Chemical and physical characteristics at ERM 6.0 (reference location) and ERM 1.0 (impacted location), Watts Bar Reservoir, Roane County, TN ¹	
	5
Table 5. Qualitative substrate composition at ERM 6.0 (reference location) and	
ERM 1.0 (impacted location), Watts Bar Reservoir, Roane County, TN ¹ 8	6
Table 6. List of benthic invertebrate species collected at ERM 6.0 (reference	
location) and ERM 1.0 (impacted location). Watts Bar Reservoir, Roane	
County TN ¹	7
Table 7 Summary of benthic invertebrate metrics at FRM 6.0 (reference	•
location) and FRM 1.0 (impacted location) Watts Bar Reservoir Roane	
County TN ¹	2
	-

LIST OF FIGURES

Figure 1. Location of Tennessee Valley Authority Kingston Ash Fossil Plant, Roane County, TN
Figure 2. Bathymetric survey presenting the most current prediction of ash deposition in the Emory River, Watts Bar Reservoir, Roane County, TN95
Figure 3. Location of spatial study sites in the Emory River, Watts Bar Reservoir, Roane County, TN
Figure 4. Overview of dredged (green shading) and un-dredged (orange shading) ash deposits in the Emory River, Watts Bar Reservoir, Roane County, TN 97
Figure 5. Benthic invertebrate community composition during Periods 4 and 5 in the Emory River, Watts Bar Reservoir, Roane County, TN
Figure 6. Average total abundance of individuals collected per transect at reference (†) and impacted locations in the Emory River, Watts Bar
Figure 7. Average total number of distinct taxa (richness) collected per transect at reference (t) and impacted locations in the Emory River. Watts Bar
Reservoir, Roane County, TN
impacted locations in the Emory River, Watts Bar Reservoir, Roane County, TN
Figure 9. Average North Carolina Biotic Index (NCBI) for tolerance of taxa collected per transect at reference (†) and impacted locations in the Emory Piver Watts Bar Reservoir, Roane County, TN
Figure 10. Concentrations of metals in sediment (mg/kg) collected per transect at reference (†) and impacted locations in the Emory River, Watts Bar
Reservoir, Roane County, TN
Watts Bar Reservoir, Roane County, TN
Reservoir, Roane County, TN
(ERM) 6.0 and ERM 1.0 during 5-year study (2009-2012), Emory River, Watts Bar Reservoir, Roane County, TN
Figure 14. Distribution of feeding guilds at (A) Emory River mile (ERM) 6.0 (reference location) and (B) ERM 1.0 (impacted location) during all periods
of study, Emory River Watts Bar Reservoir, Roane County, TN
or otady, Emory rate, watte bar reservoir, roane county, rre

Figure 17. Average (± SE) total number of distinct taxa (richness) collected per sample at Emory River mile (ERM) 6.0 (reference location) and ERM 1.0 (impacted location), Emory River, Watts Bar Reservoir, Roane County, TN..

Figure 21. Total abundance of chironomid and Tubificidae taxa collected per period at Emory River mile (ERM) 6.0 (reference location) and ERM 1.0 (impacted location), Emory River, Watts Bar Reservoir, Roane County, TN.. 114

Figure 22. Total abundance of non-chironomid and oligochaete taxa collected per period at Emory River mile (ERM) 6.0 (reference location) and ERM 1.0 (impacted location), Emory River, Watts Bar Reservoir, Roane County, TN.. 115

Figure 23. Total number of non-chironomid and oligochaete distinct taxa	
(richness) collected per period at Emory River mile (ERM) 6.0 (reference	
location) and ERM 1.0 (impacted location), Emory River, Watts Bar	
Reservoir, Roane County, TN	.116

CHAPTER 1 INTRODUCTION

Despite numerous environmental laws and regulations meant to prevent or control natural disasters, accidents causing adverse effects to environmental resources continue to occur. The release of fly ash at the Tennessee Valley Authority (TVA) Kingston Fossil Plant (KIF) on 22 December 2008 discharged an unprecedented amount of coal ash slurry into the adjacent aquatic and terrestrial systems of the Emory River. Immediately following the Kingston ash spill, on the basis of a few hastily-collected samples several researchers predicted dire effects on the aquatic ecology in the region as a result of bioaccumulation of ash-related constituents (Chattanooga Times Free Press, Emory River at 'tipping point', May 19, 2009). Metals and metalloids, including arsenic and selenium, are the primary constituents of potential concern for coal fly ash. The initial response focused on public protection and stabilization of the released ash, but rapidly evolved to include comprehensive monitoring of ambient media and ecological receptors. The size and complexity of the potentially affected ecosystems necessitated a comprehensive environmental monitoring program, which TVA continues to perform in cooperation with numerous federal, state, and academic organizations to evaluate the potential for adverse environmental effects from the Kingston fly ash spill. As discussed in the chapters to follow, benthic invertebrate communities in the Emory River were of particular concern given their importance to the health and function of both aquatic and terrestrial systems.

1.1 Benthic Invertebrates as Biological Monitors and Indicators

Benthic invertebrates are organisms that dwell in or attach to the sediments on the river bottom, near the sediment-water interface and on top of sediments. These

organisms are vital to cycling nutrients and processing organic matter in aquatic systems. Various species of fish and wildlife also feed on benthic invertebrates, making them an essential part of the aquatic and riparian food chain. The distribution and abundance of benthic invertebrates are largely defined by the habitat and feeding requirements of the various taxa and the environmental complexity of river beds. Two of the most important factors influencing benthic invertebrate community composition include the availability of food and the substrate type (Hawkins et al. 1982, Downes et al. 2000, Jones et al. 1999). Benthic invertebrates are commonly grouped based on their mode of feeding. The five main categories for functional feeding include: collectorgatherer, scraper-grazer, predator, shredder, and filterer. The physical environment (e.g., substrate and current velocity) typically dictates the types of organisms that will be present (Wallace and Webster 1996).

The quality of an aquatic ecosystem is often determined by the presence or absence of environmental stressors. A stressor is defined as a factor that is outside of the normal range, due to the influence of anthropogenic influence (Townsend et al. 2008). Benthic invertebrates have long been used as indicators of stream or water quality in both lenthic and lotic systems, as these organisms are in direct contact with surface water and sediment (Li et al. 2010; Wallace and Webster 1996; Sundermann et al. 2013; Clements 1994; Allan 2004; Maret et al. 2003; Gebler 2004; Clements 1999). Benthic invertebrate species may respond differently to chemical contaminants than to other types of environmental stressors, such as nutrient enrichment and changes to stream hydrology or habitat structure. As our understanding of indicator species assemblages and rapid bioassessment protocols has increased, the use of benthic invertebrates for biomonitoring has also increased (Clements 1994). Identifying and

understanding the relationships between aquatic organisms and environmental stressors is critical for "the effective management, restoration, and preservation of aquatic ecosystems" (Burton and Johnston 2010).

Benthic invertebrates are used in biomonitoring for a number of reasons. They are made up by a diverse group of organisms found in all freshwater ecosystems. These organisms are relatively immobile; consequently, they are closely associated with sediments and other local conditions. Benthic invertebrates accumulate metals and other contaminants, which are often times bound to the sediments in which they live, and can transfer these contaminants into aquatic and riparian food chains by serving as prey for upper trophic level receptors. Many of these organisms are in immature stages of development, so reproductive cycles and sexual differences need not be accounted for, and life-spans range from several months to multiple years, allowing for accumulation of contaminants (Li et al. 2010; Goodyear and McNeill 1999, Kiffney and Clements 1994). Benthic invertebrate communities are often described and summarized by calculating metrics or indices. These metrics are used to analyze the community data and are often categorized as: composition/abundance, richness/diversity, sensitivity/tolerance, or function (Sundermann et al. 2013; Barbour et al. 1995).

The Tennessee Department of Environment and Conservation (TDEC) commonly uses benthic invertebrate metrics to evaluate aquatic systems (TDEC 2011). Seven metrics, including taxa richness, total Ephemeroptera, Plecoptera, and Tricoptera (EPT) taxa, percent EPT taxa (excluding *Cheumatopsyche*), percent chironomids and oligochaetes, North Carolina Biotic Index (NCBI) for tolerance, percent clingers, and percent nutrient tolerant organisms, are typically used to evaluate streams and rivers within the state. TVA has also created a multi-metric evaluation of larger rivers, known

as the Reservoir Benthic Index (RBI). The RBI evaluates benthic invertebrate communities in reservoir systems, as these communities differ greatly from free-flowing river systems. Reservoirs are man-made systems, and as such, are difficult to compare to other rivers or upstream reference locations because the physical habitat has been altered to meet the needs of human use. TVA also selected seven metrics to evaluate reservoirs; however, they vary based on the type of reservoir being evaluated (e.g., run-of-river versus tributary reservoirs). Watts Bar Reservoir, a run-of-river reservoir, would be evaluated based on the following metrics: taxa richness, total number of EPT taxa, long-lived taxa (i.e., the presence or absence of at least one long-lived organism, such as *Corbicula* or *Hexagenia*), percent oligochaetes, percent of the two most dominant taxa, density (excluding chironomids and oligochaetes), and zero-samples (proportions of samples with no organisms present). A scoring criteria was established for each metric using 6 years of collections from several reservoirs, rating each metric from excellent to very poor (Baker 2006).

Changes in a number of benthic invertebrate community metrics are often associated with either increased or decreased human impact. Metrics that commonly decrease with increasing impact include: total taxa richness; number of intolerant species; EPT taxa richness; sediment-surface taxa richness; total abundance; and the proportion of individuals that feed as shredders, grazer-scrapers, and predators (Kerans et al. 1992). On the contrary, metrics that generally increase with human impact include: the proportion of *Corbicula*, oligochaetes, and chironomids; the proportion of individuals in the two most abundant taxa; the proportion of omnivorous individuals; and the proportion of individuals feeding as detritivores, filterers, or gatherers (Kerans et al. 1992). Clements (1999) found that predator-prey interactions within benthic invertebrate

communities may also be impacted by environmental stressors, such as exposure to contaminants, as some species are more sensitive to contaminants than others. Tolerance values, ranging from 0 (very intolerant) to 10 (very tolerant) are widely used to evaluate the ability of different benthic invertebrate taxa to occur in aquatic ecosystems with varying water quality (Wallace and Webster 1996). In systems with historical contamination, benthic invertebrate communities with more tolerant individuals may be the result of individual physiological adjustments or adaptations, or from more tolerant species simply replacing those species that are sensitive to contamination (Clements 1999, Burton and Johnston 2010).

Benthic invertebrates are also used as indicators of sediment quality and contaminant transfer from aquatic to riparian ecosystems by evaluating upper trophic level consumers that feed on these insects. Aquatic- and riparian-feeding receptors, such as birds, bats, and predatory invertebrates (e.g. spiders), feed on emergent benthic invertebrates exposed to sediment for a period of their lifespan. Walls et al. (2015) found that tree swallows, insectivorous passerines whose diet consists primarily of emergent aquatic insects, had higher concentrations of selenium in eggs collected from colonies closer to the Kingston ash release compared to reference colonies. Similarly, a study by Otter et al. (2013) reported concentrations of selenium in Tetragnathid spiders, receptors that feed over bodies of water, were higher in ash-associated sites compared to reference sites following the ash release. Another study by Custer et al. (2003) used tree swallows nesting along a polychlorinated biphenyl (PCB) contaminated portion of the Housatonic River to evaluate hatching success. Here, reduced hatching success was associated with increased PCB concentrations in benthic invertebrate tissues and sediments.

1.2 Effects of Heavy Metals and Coal Ash on Benthic Invertebrates

Heavy metals may persist in aquatic ecosystems even after the source of contamination has been removed, with sediments often acting as a "sink" for various types of contaminants (Ho and Burgess 2013). Given that benthic invertebrates often live, feed, or breed within sediments, contamination within the sediment can be a primary stressor to these aquatic organisms throughout a portion of their lifespan (Courtney and Clements 2002; Burton and Johnston 2010). Benthic invertebrates can bioaccumulate contaminants within their tissues, providing possible toxic body burdens for themselves, as well as creating a route for trophic transfer of these contaminants to their predators (Goodyear and McNeill 1999; Cherry and Guthrie 1977; Maret et al. 2003; Rowe et al. 2002). In a study by Cain et al. (2011), several species of mayflies were found to accumulate high concentrations of cadmium and copper. While the different species accumulated these metals at varying rates, the study indicated that consumption of periphyton was the leading exposure over uptake from the aqueous phase. Similarly, Culioli et al. (2009) discovered higher concentrations of arsenic and antimony in benthic invertebrate taxa downstream of mining activity, with levels of accumulation relating to the feeding behavior, specific habit, and position of the different taxa in the food chain. Conley et al. (2009) found that selenium concentrations reduced fecundity in adult female mayflies, and also caused a reduction in adult body mass. Mouthpart deformities of one chironomid species, *Chironomis riparius*, were found when exposed to concentrations of copper and zinc (Di Veroli et al. 2014).

Field observations of the community structure and function can often times be linked with this contamination. Streams with heavy-metal pollution frequently have

benthic invertebrate communities characterized by reduced abundance and species richness, as well as a host of more tolerant taxa (e.g., chironomids) and fewer sensitive taxa (e.g., mayflies), due to toxicity of select metals (Cortelezzi et al. 2011; Jones et al. 1999, Courtney and Clements 2002; Harper and Peckarsky 2005; Pollard and Yuan 2006; Hickey and Clements 1998). A study by Maret et al. (2003) evaluating benthic invertebrate communities located downstream of mining activities found that elevated metal concentrations in surface water and sediment (e.g., cadmium, lead, and zinc) were directly related to reduced total taxa richness and density and EPT taxa richness and density. Clements (1994) found that heavy metal pollution impacted the distribution and composition of the benthic invertebrate community in the Arkansas River in Colorado; however, other factors, such as differences in environmental conditions and recolonization ability, also likely influenced the community characteristics.

Coal fly ash and effluent from settling ponds contain heavy metals that can provide a potential source of contamination to aquatic ecosystems. Releases of coal ash and effluent in streams have negatively impacted benthic invertebrate densities and species richness by causing both physical disturbance and chemical toxicity (Rowe 2014; Rowe et al. 2002; Smith 2003; Cherry et al. 1979; Cairns et al. 1970; Specht et al. 1984; Guthrie and Cherry 1979). Furthermore, coal ash has been shown to influence the composition of community structure. Physical effects include smothering and increased turbidity to stream habitats, as particles that are suspended in the water column may clog or damage the respiratory organs of aquatic invertebrates (Burton and Johnston 2010). Releases of fly ash can create environmental conditions similar to conditions caused by sedimentation or siltation (Smith 2003). A study conducted on fly ash effluent discharged from a settling pond into a stream found that the reduction of benthic

invertebrate density was the most severe during the period when suspended solids were highest. Chemical toxicity was also attributed to the heavy metals found in fly ash (Rowe 2014; Mayfield et al. 2013; Rowe et al. 2002; Winner et al. 1980; Cherry and Guthrie 1977; Clements et al. 1988). Harper and Peckarsky (2005) found that abundance and taxa richness were both reduced following the release of coal into a small stream in New York. Two years following the release, benthic invertebrate communities continued to demonstrate impacts. While no significant effects (p=0.871) were identified for EPT taxa, other less sensitive invertebrates within the community had declined. These impacts were thought to be the result of the clean-up, which included changes to the stream banks and modified channel. A study by Cairns et al. (1970) evaluating an accidental release of coal ash effluent into the Clinch River found similar kinds of organisms at impacted and reference sites, but reduced densities of the various organisms in impacted areas compared to reference areas. However, Cairns et al. (1970) also noted that the benthic invertebrate communities were quickly recovering just 2 years after the release. Another study by Smith (2003) evaluating a small stream in Tennessee receiving discharges of coal fly ash found that benthic invertebrate density and taxa richness began to recover as soon as coal ash discharges were reduced and then stopped. Furthermore, Smith (2003) noted that the rate and extent of community recovery depended on a number of factors, including the type of disturbance, habitat conditions, the season the disturbance ended, and the potential for colonizing invertebrates to reoccupy the area.

While the dynamics of metal pollution to benthic invertebrate communities have been studied for decades, interactions between heavy metals and benthic community structure are still highly uncertain. Smith (2003) stated that the recovery of the benthic

invertebrate community in McCoy Branch, a small headwater stream that received heavy metals from decades of ash slurry discharge before operational changes removed the source, could not be attributed to the physical reduction of ash or the chemical contamination associated with the ash. A study by Winner et al. (1980) compared two benthic invertebrate communities with different types of heavy-metal pollution stress. One community was exposed to copper at relatively low, constant concentrations, while the other had highly variable concentrations of copper, chromium, and zinc. Both benthic invertebrate communities demonstrated similar patterns of decreased diversity and dominance of tolerant taxa, indicating that continuous low-level stress may have an overall comparable biological impact to mixtures of metals found with potentially greater intensity. Winner et al. (1980) concluded that as chemical stress decreases in a system, changes in substrate composition and seasonal variability begin to account for more of the differences found in community composition. Tolerant organisms, such as chironomids, may still be dominant in some areas; however, this occurrence is not as predictable. In many cases, metal pollution is not the only environmental stressor; as a result, it is difficult to demonstrate with confidence the contribution of heavy metals to ecological effects observed within the benthic invertebrate community in the field (De Jonge et al. 2013). Distinguishing natural variation in benthic invertebrate communities from variation caused by anthropogenic disturbances is also an on-going problem (Clements 1994). Additional research is needed to address these uncertainties.

1.3 Tennessee Valley Authority Background and Kingston Ash Release

TVA, an independent corporation owned by the federal government, provides power to the majority of Tennessee and some portions of the surrounding southern

states. The TVA KIF, one of TVA's larger fossil plants, was built at the confluence of the Emory and Clinch Rivers within Watts Bar Reservoir located in Roane County, Tennessee (Figure 1) in 1955. The KIF produces electricity by burning coal, which heats water drawn from the surrounding rivers. The heated water produces steam, which is then directed into a turbine connected to a generator. As the generator spins, electricity is produced. Ash, a by-product of the coal-burning process, has historically been stored in unlined containment areas onsite. An unprecedented release of coal fly ash occurred at this facility in December 2008, when an ash containment area wall failed. Approximately 4.1 million cubic meters (m³) of coal ash was released into the Emory River and overbank areas, covering approximately 121 hectares. While the released ash mainly consisted of fine aluminosilicate particles, it also contained trace amounts of heavy metals such as arsenic, copper, mercury, nickel, selenium, and zinc, which occur naturally in coal (Jacobs 2010; Gieré et al. 2003; Tishmack and Burns 2004; Rowe et al. 2002).

1.4 Kingston Ash Release Site Conditions and Clean-Up

Given the force and sheer volume of the released ash, field surveys and subsequent laboratory analyses on sediment indicated that ash was initially pushed and deposited upriver as far as Emory River mile (ERM) 5.75, and following several heavy rains with high river flow events, was distributed downriver into the Clinch and Tennessee Rivers. In the immediate vicinity of the spill, at ERM 2.2, ash filled the main river channel, with thicknesses of ash approximately 10 meters (m) deep (Figure 1). Upstream of the spill, ash deposits appeared to rapidly decrease beyond ERM 3.5, approximately 1.5 river miles from the initial release. Similarly, ash deposition in the downstream direction also quickly diminished below ERM 1.0. While pockets of ash

occurred in the lower Emory and Clinch Rivers, only small amounts of ash (generally less than 5 centimeters (cm)) were found downstream of Clinch River mile (CRM) 2.0. Ash was also detected in the Tennessee River at Tennessee River mile (TRM) 566 (approximately 2 river miles south of the confluence with the Clinch River and 8 river miles downstream of the initial spill location); however, deposition was limited to 1 to 3 cm (Jacobs 2010).

Approximately 3 months after the release, dredging efforts began in the Emory River. The initial dredging pilot program was conducted from March to July, 2009. Beginning in August, 2009, Phase I production dredging was implemented to reduce the potential for upstream flooding and downriver migration of ash by removing ash from the river channel as quickly as possible. Phase II precision dredging began in February 2010. While this phase of dredging continued to minimize downriver movement of ash, it was also intended to return the river channel to pre-release elevations. All dredging efforts were completed by August, 2010, in total removing approximately 2.7 million m³ of released ash and sediment from portions of the Emory River. Throughout the entire dredging process, both hydraulic and mechanical dredges were used, and engineering and operational controls were implemented to reduce the levels of suspended solids generated during the dredging operations. These controls included the use of silt curtains, and the reduction of cutter head speeds, rates of advance, and reverse cutter head rotation (ARCADIS 2012; Jacobs 2011). Efforts were also made to reduce the disturbance of legacy sediment located between ERM 0.0 and ERM 1.75. This portion of the Emory River, as well as sediments in the KIF intake channel, was not dredged due to the presence of cesium-137 in underlying sediment samples. Unrelated to the ash

release, cesium-137 is the result of historical releases from U.S. Department of Energy (USDOE) facilities on the Oak Ridge Reservation (ARCADIS 2012).

To date, approximately 407,000 m³ of ash remain in the river system, as described in the U.S. Environmental Protection Agency (USEPA)-approved *Kingston Ash Recovery Project, On-Scene Coordinator Report for the Time-Critical Removal Action at the TVA Kingston Fossil Fuel Plant Release Site, Roane County, Tennessee* (Jacobs 2011). This estimate of residual ash was based on interpretations of data from multiple sources, including pre- and post-release river bathymetric data, dredging logs, visual surveys, and VibeCore™ data; consequently, these estimates include some uncertainty. Additional sediment samples have been collected to refine the distribution of residual ash in the river system. Using interpretations of bathymetric survey information and results of VibeCore™ sampling data, the most current prediction of ash deposition suggests that residual ash may be present in distinct pockets, as well as intermixed or imbedded with submerged natural river sediments (Figure 2) (Jacobs 2010).

1.5 Emory River Hydrology and Sediment Characterization

The Emory River is one of the major tributaries that drains into Watts Bar Reservoir. This reservoir is contained within portions of Loudon, Meigs, Rhea, and Roane counties in eastern Tennessee. Watts Bar Reservoir was created in 1942 with the construction of the Watts Bar Dam and holds approximately 15,783 hectares of surface water (TVA 2009). The drainage basin associated with the reservoir encompasses approximately 45,000 square kilometers (km²) in Tennessee, North Carolina, and Virginia, and includes almost 3,000 km of streams that drain directly to the reservoir. Watts Bar Reservoir contains three main branches, including:

• Emory River: 19.3 km of navigable water

- Clinch River: 37.0 km of navigable water from the confluence of the Clinch and Tennessee Rivers to Melton Hill Dam (CRM 23.1)
- Tennessee River: 115.9 km of navigable water from the Watts Bar Dam (TRM 529.9) to Fort Loudoun Dam (TRM 602.3).

Watts Bar Reservoir is considered a "run-of-river" reservoir, meaning that the waters within it have a short retention time (approximately 18 days) and that the winter drawdown only reduces the depths by approximately 1 m. Consequently, sediment within Watts Bar Reservoir is classified as seasonally-exposed sediment or submerged sediment. Seasonally-exposed sediment refers to the sediment that is exposed to the air during the winter months when water levels are low. Submerged sediment refers to the sediment that is below water year-round (Baker 2006; ARCADIS 2012).

Submerged sediments in the Emory River vary in substrate type and thickness. As the river changes from riverine to lacustrine (moving from upstream to downstream), the classification of sediment substrate follows suit. Upstream portions of the river bottom, above ERM 6.0, are characterized with bedrock, hard-packed sediments, silts, clays, sands, and detritus (leaves, twigs, and other natural organic materials). This portion of the river was used as a reference area following the spill because no observable released ash was found in the sediments. Moving downstream, closer to the initial release (ERM 6.0 to ERM 3.5), sediments are comprised of increasing ash content, along with hard-packed clays and bedrock near overbank areas. This portion of the river is prone to scouring during heavy rain events. Areas that are not scoured are comprised of gravels, fine silts, detritus, or sands with some coal particles. The sediments in the area of the river immediately impacted by the release, ERM 3.5 to ERM 1.5, are highly variable. The general composition includes fine clays and silts

mixed with ash. Some areas near ERM 3.0 also include fine silts and sands with detritus or hard-packed clay. This section of the river channel widens considerably, allowing for more deposition to occur. The lower, undredged sediment of the Emory River (ERM 1.5 to ERM 0.0) consists of fine silts, detritus, and ash (ARCADIS 2012). This section of the river is immediately upstream of the confluence with the Clinch River, and contains sediment contaminated with cesium-137 from historical releases from USDOE.

Submerged sediment is an important component of aquatic ecosystems. It provides habitat for a variety of aquatic organisms, such as benthic invertebrates, which come in direct contact with the sediments. Wildlife that inhabit or forage in the river system (e.g., great blue heron, killdeer, and muskrats) may be indirectly exposed to submerged sediments through incidental ingestion in their diet. These types of ecological exposures typically occur only in the upper 15 cm of submerged sediments; therefore, this portion of sediment has been the focus of sampling and ecological studies previously conducted at the site and in most literature studies (ARCADIS 2012).

1.6 Benthic Invertebrate Communities in the Emory River

Dominant benthic invertebrate taxa found in the Emory River include Diptera, Oligochaete, and Ephemeroptera. Predominant dipteran taxa include non-biting midges (Chironomidae), phantom midges (*Chaoborus*), and biting midges (Ceratopogonidae). Oligochaete taxa consist mainly of aquatic worms and tubificids (e.g., Tubificidae and Lumbriculidae). Burrowing mayflies (*Hexagenia*) are the primary Ephemeroptera taxa; however, occasional gatherer/collector mayflies (*Caenis* and *Callibaetis*) have also been observed. A variety of freshwater bivalves, including fingernail clams (*Musculium transversum*) and Asiatic clams (*Corbicula fluminea*) are also present throughout the river, as well as freshwater snails (e.g., Hydrobiidae and Viviparidae) and leeches

(Glossiphoniidae) (Baker 2006; Buys et al. 2015; ARCADIS 2012). Historical surveys conducted in Watts Bar Reservoir identified a number of protected or sensitive invertebrate species. Alabama lamp mussel (*Lampsilis virescens*), dromedary pearly mussel (*Dromus dromas*), purple bean (*Villosa perpurpurea*), fine-rayed pigtoe (*Fusconaia cuneolus*), and Anthony's river snail (*Athearnia anthonyi*) are species of protected mollusks that have historically been observed within Watts Bar Reservoir area and its tributaries; however, these species have not been found in the past 30 years. Their absence is likely a result of the construction of Watts Bar Dam (TVA 2009).

Benthic invertebrate community composition and structure can offer insight to the general habitat conditions present within the river bottom and may also indicate if environmental stressors are affecting the quality of habitat used by these communities (Jones et al. 1999). Sediment grain size and texture, spatial distribution, substrate diversity, and organic content are all environmental factors that influence benthic invertebrate communities (Jones et al. 1999, Jahnig and Lorenz 2008; Lepori et al. 2005; Boyero 2003). In a field study by Cummins and Lauff (1968), substrate particle size was identified as the most likely "common denominator" in various benthic invertebrate community compositions. A study by Jones et al. (1999) in the Clinch River, Tennessee, found that variation in habitat explained more than 50% of the variance observed in the diversity of benthic invertebrate communities. Due to the sedentary nature of these organisms and their direct contact and exposure to surface water and sediment, benthic invertebrates are often the most sensitive receptor group to metals and other stressors in sediments and related porewaters. Changes from expected benthic invertebrate communities may be the result of environmental stressors, including the presence of chemical constituents in surface water or sediment, increased sedimentation, or

hydrological changes. Consequently, they are often used as indicators of the quality associated within a given stream or river bottom (Courtney and Clements 2002; ARCADIS 2012).

1.7 Previous Benthic Invertebrate Studies in the Emory River

Post-spill investigations for benthic invertebrates conducted by TVA and collaborative researchers concluded that benthic invertebrates continue to be at moderate risk in the Emory River from ash-related constituents, primarily arsenic, in the residual ash-contaminated sediment (ARCADIS 2012; Carriker et al. 2015). A number of studies were conducted on benthic invertebrates in the Emory River in order to reach this conclusion of moderate risk; however, the results of the sediment toxicity tests were the driving line of evidence. Statistically significant reductions in growth and biomass in Hyalella azteca (p<0.05) and significantly decreased emergence and survival of Chironomus dilutus (p<0.05) were observed in sediment toxicity tests conducted with Emory River sediment in Spring 2011. The majority of these effects were sub-lethal, indicating that effects are not likely to be immediate or severe, but could result in impacts to the population over time (Stojak et al. 2015). This evidence was augmented by findings that showed ash-related metal concentrations were present in benthic invertebrate tissue, sediment, and porewater at concentrations potentially associated with adverse effects to benthic invertebrates. Benthic invertebrate tissue concentrations indicated that arsenic, and to some degree selenium, may be bioaccumulating in invertebrates such as *Hexagenia* sp., a burrowing mayfly commonly found in the Emory River (Smith et al. 2015; Conley et al. 2009). Sediment concentrations indicated that ash-related constituents may pose a low risk based on exceedances of conservative benchmarks. Similarly, porewater concentrations indicated that ash-related constituents

may also pose a low risk based on exceedances of ambient water quality criteria (ARCADIS 2012).

Benthic invertebrate community survey results, however, did not support this conclusion. Prior to the ash release at KIF, benthic invertebrate communities were evaluated as part of TVA's Valley-Wide Vital Signs Monitoring Program in 31 different reservoirs managed by TVA; however, of the three rivers impacted by the ash release, only the Tennessee River was monitored prior to the spill (Carriker 1999). No historical invertebrate data are available as a baseline comparison for the Emory and Clinch Rivers. Following the release in 2008, benthic invertebrates were collected in 2009, 2010, and 2011 to evaluate potential impacts from ash and ash-related contaminants in the Emory, Clinch, and Tennessee Rivers. The surveys conducted in 2010 and 2011 provided no substantive evidence that the community composition has been negatively impacted. Macroinvertebrate density and taxa richness in the immediate area of the ash release were similar to or even greater than other locations in the river system. These data did not indicate a trend of decreasing macroinvertebrate abundance or decreasing richness. Combined, these results showed no obvious patterns of persistent adverse impacts from the ash release and differences were associated with habitat variation. The community composition was strongly correlated with substrate type rather than ash-related constituents. Despite this contrary evidence, it is possible that over time reductions in growth and biomass could result in a measurable impact on reproduction or community structure (Buys et al. 2015). For this reason, risk management actions were recommended for the protection of the benthic invertebrate community (Carriker et al. 2015).

1.8 Objectives and Justification for Research

Previous investigations conducted on benthic invertebrates in the Emory River have evaluated non-co-located benthic invertebrate community data and sediment data across a larger spatial scale; chronic and long-term sediment toxicity tests; bioaccumulation of ash-related constituents in invertebrate tissue; and ambient media concentrations. The sediment toxicity tests indicated toxicity to sensitive laboratory organisms when sediment concentrations had approximately 40% ash; however, singlespecies toxicity tests often times overestimate or do not adequately reflect the effect of contaminants on natural communities in the field (Kiffney and Clements 1994). As a result, sediment toxicity tests are used in conjunction with other lines of evidence to evaluate a benthic invertebrate community. While areas remain in the Emory River with ash percentages at or above the 40% range, the benthic invertebrate community results showed no differences associated with the ash release in the impacted Emory River transects compared to the upstream Emory River reference transect (Buys et al. 2015). The previous evaluations of the benthic invertebrate community data and non-co-located sediment data provided an "area-wide" interpretation of the relationships between the benthic invertebrate community metric results to the ash release; however, given the discrepancy between the sediment toxicity test results and the benthic invertebrate community results additional evaluation is warranted.

Spatial variability of individual benthic invertebrate organisms and communities is commonly studied in aquatic ecosystems. Understanding the natural variability that occurs in both substrate composition and benthic invertebrate communities at a site is critical before conclusions can be made on human disturbance (Gebler 2004). A previous study conducted by Boyero (2003) found that substrate composition had a

significant effect on benthic invertebrate abundance, richness, and evenness on a sample scale; however, these effects were not found to be significant when data were evaluated by larger segments or reaches. Similarly, Downes et al. (1993) found high variability in smaller stream segments was not represented when combining these areas into larger segments. As a result, the spatial scale at which evaluations are conducted are of particular interest.

The purpose of this proposed research is to assess the differences in the benthic invertebrate community in the Emory River, 4 years following the ash spill. The objectives are to: 1) compare community metric results among samples, locations, and previous years, 2) compare community metric results to co-located sediment chemistry and physical sediment properties, and 3) determine if the geographic spatial scale of the evaluation influences the overall interpretation of the differences in community results. This information is important for the informed monitoring, remediation, and damage assessment of the benthic invertebrate community at the Kingston Ash Recovery site. This recovery is currently being monitored as part of TVA's long-term monitoring program (Carriker et al. 2015). Results from this research could alter the understanding of the recovery of the benthic invertebrate community in the Emory River; as a result, changes in the sampling design and/or frequency of benthic invertebrate community collections in the long-term monitoring program may be warranted if relationships are established between community metrics and percent ash in sediment. This research will increase our knowledge of benthic invertebrate tolerance to metal mixtures in sediment of natural systems. While scientific literature is available for disturbances of stream benthic invertebrate communities, there are a limited number of studies for benthic invertebrate communities in large reservoir systems. Consequently, this research may

increase our knowledge of benthic invertebrate tolerance to metal mixtures in sediment if no relationships between community metrics and percent ash or ash-related contaminants are found.

A number of hypotheses have been proposed from this research. Previous studies have indicated strong relationships between community relationships and sediment substrate type, which are expected to continue. In addition, given the natural sedimentation processes and river system recovery that is likely to continue, it is hypothesized that benthic community metrics from impacted areas will have higher numbers of organisms and more taxa diversity compared to previous years. Finally, given the results of the laboratory sediment toxicity testing which indicated toxicity to benthic invertebrates when sediments contained >40% ash, it is hypothesized that benthic community and to previous and it is hypothesized that benthic community and EPT taxa as well as a stronger percent of taxa tolerant to anthropogenic disturbance and pollution.

CHAPTER 2 SPATIAL ANALYSIS OF BENTHIC INVERTEBRATE COMMUNITIES IN THE EMORY RIVER AFTER DREDGING, WATTS BAR RESERVOIR, ROANE COUNTY, TN

Abstract

The release of fly ash at the TVA KIF on 22 December 2008 discharged approximately 4.1 million cubic meters of coal ash into the adjacent aquatic and terrestrial systems. Previous benthic invertebrate investigations conducted by TVA and collaborative researchers concluded that benthic invertebrates in the Emory River were at moderate risk from ash-related constituents, primarily arsenic, in ash-contaminated sediment that remained in the Emory River following extensive dredging efforts. These conclusions were based on the observation of statistically significant reductions in growth and biomass in toxicity tests with Emory River sediment. Benthic invertebrate community survey results from 2010, however, did not support this conclusion. These previous surveys evaluated benthic invertebrate community data and sediment data across a large spatial scale, providing an "area-wide" interpretation of the relationships between the benthic invertebrate community results to the ash release. In this study, colocated sediment and benthic invertebrate community samples were collected over a 2year period from nine locations in the Emory River. Benthic invertebrate community metric results including taxa abundance, taxa richness, Shannon Diversity, and tolerance were compared among samples, locations, and years. These metrics were also evaluated with the co-located sediment chemistry and physical sediment properties. Despite this refined investigation, no trends or significant differences were identified between ash-impacted locations compared to the reference location. Furthermore, no significant relationships could be established between benthic invertebrate community metrics and sediment chemistry results. This information is important for the informed monitoring, remediation, and damage assessment of the benthic invertebrate community at the Kingston Ash Recovery site. This research also increases our knowledge of benthic invertebrate tolerance to metal mixtures in sediment of natural systems.

2.1 Introduction

Previous research has demonstrated impacts to aquatic- and riparian-feeding organisms associated with releases of coal combustion residues (CCRs) into aquatic ecosystems (Cherry and Guthrie 1977; Rowe et al. 2002; Smith 2003; Ruhl et al. 2012). The release of CCRs into aquatic systems not only physically changes the habitat through sedimentation and turbidity, but may also chemically alter the ecological conditions by changing water pH and introducing high concentrations of contaminants (Rowe et al. 2002). Fly ash, one of the main components of CCRs, contains several trace elements (primarily arsenic, cadmium, copper, lead, nickel, selenium, strontium, and zinc) (Rivera et al. 2015). Uptake and bioaccumulation of these trace elements from surface waters, sediments, and prey items have resulted in various effects in aquaticand riparian-feeding organisms. A comprehensive body of literature, including both field and laboratory studies, has documented these impacts to fish, amphibians, reptiles, and birds (Rowe et al. 2002; Rowe 2014). One laboratory study evaluating lake chubsuckers (*Erimyzon sucetta*) exposed to sediments with coal ash found significantly (p<0.001) higher body burden concentrations of selenium, strontium, and vanadium after four months of exposure (Hopkins et al. 2000). This exposure resulted in 25% mortality of the exposed fish during the study (Hopkins et al. 2000). Similarly, eggs of eastern narrowmouth toads (Gastrophryne carolinensis) collected near a coal-burning power plant contained elevated concentrations of selenium and strontium and were linked to reduced hatching success (by approximately 11%). Hopkins et al. (1999) also studied trace element concentrations in banded water snakes (Nerodia fasciata) near polluted coal combustion waste sites. Higher concentrations of arsenic and selenium in snakes from the polluted study site were found compared to those captured in reference locations,

which was attributed to ingestion of contaminated dietary items of the snake (Hopkins et al. 1999). Beck et al. (2013) and Walls et al. (2015) found that colonies of tree swallows (*Tachycineta bicolor*) nesting near the TVA Kingston ash release were exposed to higher concentrations of ash-related elements such as selenium in their diet, resulting in higher concentrations of these elements in tree swallow egg tissues.

Perhaps one of the most studied groups of organisms exposed to CCR releases are benthic invertebrates. These organisms are in direct contact with the sediment and often have the highest potential for exposure to contaminants. Benthic invertebrates can accumulate ash-related metals within their tissues, leading to body burdens which can be a source of contaminants for higher level trophic feeding wildlife (Goodyear and McNeill 1999, Cherry and Guthrie 1977, Maret et al. 2003; Rowe et al. 2002). A study of heavy metals accumulation from coal ash found concentrations of barium, copper, manganese, mercury, and zinc in invertebrate tissue from within a coal ash basin (Guthrie and Cherry 1979). Similarly, Conley et al. (2009) found that elevated exposure to selenium reduced fecundity and also caused a reduction in adult body mass. While the dynamics of metal pollution to benthic invertebrate communities have been studied for decades, long-term, multi-generational effects of exposure to heavy metals on benthic community structure are still highly uncertain.

Initial evaluations of the benthic invertebrate community in the Emory River suggested potential short-term impacts, but noted that the community quickly recovered after the majority of ash was removed from the river system (Buys et al. 2015). However, Buys et al. (2015) noted few to no measurable impacts on the benthic community related to the ash. The majority of these findings were based on a similarity analysis conducted across large stretches of the Emory River. The purpose of the present investigation is to

provide a more comprehensive assessment of benthic invertebrate community responses to ash and metals in the Emory River using co-located sediment samples and benthic invertebrate community samples. This investigation began approximately 8 months after a substantial dredging effort of released ash was completed.

2.2 Materials and Methods

2.2.1 Study Site

An unprecedented release of coal ash (4.1 million m³) from the TVA KIF occurred on 22 December 2008 in Watts Bar Reservoir in Roane County, Tennessee (Figure 3). Following the release, TVA used hydraulic and mechanical dredges to remove mass amounts of ash from sediments in large segments of the Emory River (Figure 4). The released ash contains trace amounts of heavy metals that naturally occur in the coal and remain after the combustion process. Following more than 1 year of dredging, approximately 300,000 m³ of ash were estimated to remain in the Emory River between ERM 0.0 and ERM 6.0 (Jacobs 2012). The current study evaluated samples collected approximately 8 months and 18 months after dredging was completed.

2.2.2 Sample Collection

Co-located benthic invertebrate community samples and sediment were collected in January 2012 (Period 4) and December 2012 (Period 5) from nine locations in the Emory River (Figure 3). These collection months were selected to coincide with previous years of monitoring, which typically occur in the late fall or early winter. One reference location (ERM 6.0) with no recordable ash deposition and eight impacted locations (ERM 5.0, 4.1, 3.5, 3.0, 2.6, 2.2, 1.0, and 0.7) with varying amounts of ash deposition were sampled. Ten samples, evenly spaced across the width of the channel, were collected at each of the nine locations during both periods of collection. Each sample consisted of a
benthic invertebrate community survey and a co-located submerged sediment sample, both of which were individually collected using a Ponar dredge (0.05 m²). Benthic invertebrate community survey samples were rinsed through a 0.6 mm mesh screen. The remaining contents were placed into a container (the size and number of containers) depended on the amount of material; containers include 0.5 and 1 liter glass jars) and preserved in a 10% formalin solution. Samples were sorted at Pennington and Associates, Inc. Laboratory, where they were preserved in 85% ethanol and identified to the lowest taxon possible. Each co-located submerged sediment sample was collected and homogenized, removing twigs and other large debris. All submerged sediment samples were analyzed for percent ash using Polarized Light Microscopy. At each transect, a minimum of three sediment samples were randomly selected and analyzed for metals using inductively coupled plasma mass spectrometry (ICP-MS) (23 analytes including: aluminum, antimony, arsenic, barium, beryllium, boron, cadmium, chromium, cobalt, copper, iron, potassium, mercury, manganese, molybdenum, nickel, selenium, lead, silver, strontium, thallium, vanadium, and zinc). All sediment samples were analyzed for sediment grain size and total organic carbon. The grain size analysis classified substrates as ash, silt, sand, clay, or gravel.

2.2.3 Benthic Invertebrate Metric Calculations

Data from benthic invertebrate community surveys were analyzed using a series of metrics to evaluate abundance, richness, diversity, and tolerance of organisms in the Emory River community. All metrics were calculated for each sample from all nine locations for both periods of study. Specific metrics included: 1) number of total taxa (abundance); 2) number of distinct taxa (richness); 3) Shannon Diversity; and 4) NCBI to evaluate organism tolerance. Abundance metrics may include counts of all organisms,

relative abundance of various taxonomic groups (e.g., orders, families, etc.), and others. These metrics provide information about the identity of organisms within a community and also help to recognize ecological patterns or specific environmental conditions present (Barbour et al. 1995). Abundance metrics may increase or decrease depending on the environmental stressor. Richness metrics describe the number of different or distinct taxa in a community. Generally, the number of taxa decreases as water quality declines (Merritt et al. 2008). Shannon Diversity accounts for abundance and evenness of the species present, and was calculated using the following equation, where *i* is the proportion of species relative to the total number of species (*pi*). The resulting product is summed across species and multiplied by -1 (Peet 1975).

Shannon Diversity = $-\sum p_i \ln p_i$

NCBI includes tolerance scores for taxa, such as species, genus, and family, from the North Carolina Department of Environment and Natural Resources (TDEC 2011). If a North Carolina tolerance score has not been assigned for a taxon, values from USEPA's Rapid Bioassessment protocols were substituted in the following order: Southeast, Midwest, Upper Midwest, Mid-Atlantic, and Northwest. If no genus level tolerance values are available from any of these sources, the family level tolerance value from North Carolina was substituted. Organisms with no tolerance value were excluded from this metric. The following equation was used to calculate NCBI (TDEC 2011), where x_i is the number of individuals within a taxon, t_i is the tolerance value of a taxon, and N is the total number of individuals in the subsample that have been assigned a tolerance value:

2.2.4 Statistical Analysis

For all benthic invertebrate community metrics, metals, and substrate types with normal or lognormal distribution, two-way analysis of variance tests were conducted for similar locations (e.g., sites identified as sampled in both January and December), evaluating period or year, location, and the interaction of year and location using parametric tests (using SAS, v. 9.4). Means were separated with Tukey-Kramer at 5% significance level. The data for each of these variables were tested for normality and homogeneity of variance to ensure a parametric test was appropriate.

To evaluate whether constituents were related to reductions in benthic invertebrate community metrics, concentrations of metals in sediment, percent ash, sediment grain size, percent dredgefull, and water depth were correlated with community metrics measures (total abundance, total richness, NCBI, and Shannon Diversity) using Spearman (rank correlation) coefficients.

Variable selection modeling was also conducted prior to running multiple regression analyses to determine if potential relationships exist between each benthic invertebrate community metrics and physical and chemical sediment data. The goal of variable selection modeling was to identify important sediment variables that can predict patterns in benthic invertebrate community structure. Concentrations of 22 ash-related metals (aluminum, antimony, arsenic, barium, beryllium, boron, cadmium, chromium, cobalt, copper, iron, lead, manganese, mercury, molybdenum, nickel, selenium, silver, strontium, thallium, vanadium, and zinc) and percent ash were included in the variable selection model, along with the sediment grain size and water depth. Metals detected in less than 25% of the samples (antimony, boron, molybdenum, selenium, silver, and thallium) were excluded from the analysis. Also, multicollinearity and non-significant

variables were removed. Following the variable selection modeling, a multiple regression analysis was conducted to see if the selected variables could predict benthic invertebrate community metrics, such as total abundance or richness.

2.3 Results

A spatial analysis of benthic invertebrate community metrics, chemical concentrations, and substrate composition are discussed in the subsections below. Relationships between these components and their ability to make future predictions of the benthic invertebrate community are also presented.

2.3.1 Benthic Invertebrate Community Metrics

Benthic invertebrate community metrics are summarized below. A total of 180 samples with 17,404 organisms, representing five phyla, eight classes, 19 orders, and 42 families, were collected during the 2 years of study (Table 1). Included in Insecta were the following number of distinct families: five Diptera, five Ephemeroptera, three Plecoptera, three Tricoptera, two Coleoptera, two Odonata, and one Megaloptera. The five most common and abundant taxa across all locations during both years included Tubificidae, *Chironomus* sp., *Musculium transversum, Chaoborus punctipennis*, and *Procladius* sp.

The benthic invertebrate community composition for each transect is presented in Figure 5. Total abundance of organisms collected was not significantly different (p>0.05) among locations or between periods (Figure 6,). The average total abundance of organisms collected from ash-impacted locations with ash ranged from 41 to 158 organisms, while the average total abundance from the reference location (ERM 6.0) was 42 and 125 organisms (Table 2). The highest number of organisms collected in a single sample was 324 from ERM 2.2 during Period 5, and the lowest number of

organisms collected was four from ERM 3.5 and ERM 5.0 during Period 4. Similarly, the total number of distinct taxa (richness) was not significantly different (p>0.05) among locations or between periods (Figure 7). The total richness of organisms collected from locations with ash ranged from seven to 17 distinct taxa, while the average reference richness numbers were eight and nine. The highest and lowest numbers of distinct taxa were both recorded in Period 4, with the highest at ERM 1.0 (30) and the lowest at the reference ERM 6.0 (1). Shannon Diversity was not significantly different (p>0.05) among locations or between periods. Average diversity scores in ash impacted locations were similar, ranging from 1.3 to 2.2, compared to the reference location, which ranged from 1.4 to 1.5 (Figure 8). Similarly, NCBI values were not significantly different among locations (p>0.05) but were different between periods. Higher tolerance scores were recorded in Period 5 compared to Period 4 (p<0.05), indicating that more sensitive species were observed closer to the end of dredging (during Period 4) than after more time had passed (Figure 9). When specific locations and periods were evaluated, however, these differences were limited to NCBI scores from Period 4 at ERM 1.0 and ERM 3.5 (NCBI score of 7.0 and 6.6, respectively) compared to the reference in Period 5 (NCBI score of 8.8).

2.3.2 Chemical Analysis

Of the 23 metals analyzed in sediment samples, only eight constituents (arsenic, barium, beryllium, boron, chromium, selenium, strontium, and vanadium) were previously associated with benthic invertebrate measurement endpoints in sediment toxicity tests for the Emory River (Stojak et al. 2015). As a result, only these eight constituents are discussed within this evaluation (Table 3, Figure 10A through 10F). No significant differences (p>0.05) among location or between period of study were

identified for boron or selenium. Arsenic (p < 0.05), barium (p < 0.05), beryllium (p < 0.05), and strontium (p<0.05) concentrations were significantly different among locations but did not differ between periods of study. Mean arsenic concentrations were significantly (p<0.05) higher at ERM 0.7, ERM 1.0, ERM 2.2, and ERM 3.0 compared to the reference locations (Figure 10A). Mean arsenic concentrations were at least four times higher at ash-impacted locations (16.6 to 29.8 milligrams per kilogram (mg/kg)) compared to the reference location (3.7 mg/kg). Mean barium concentrations were higher at ERM 0.7 compared to the reference location and two of the upstream impacted locations (ERM 4.1 and ERM 5.0). The mean barium concentration at ERM 0.7 was more than twice that of the reference location with a concentration of 148.6 mg/kg compared to 67.8 mg/kg, respectively (Figure 10B). No location-specific significant differences (p>0.05) were identified for beryllium concentrations (Figure 10C); however, mean concentrations at the impacted locations (0.8 to 1.5 mg/kg) were higher than those means recorded at the reference location (0.6 mg/kg). Mean strontium concentrations were significantly (p<0.05) higher at ERM 0.7, ERM 1.0, ERM 2.2, and ERM 3.0 compared to the reference locations (Figure 10D). Mean strontium concentrations from these impacted locations ranged from 48.5 to 93.8 mg/kg compared to 10.9 mg/kg at the reference location.

Mean concentrations of chromium were significantly different by location (p<0.05) and period of study (p<0.05). Chromium concentrations during Period 5 were found to be significantly (p<0.05) higher compared to Period 4 (Figure 10E). This difference appears to be driven by mean chromium concentrations at ERM 3.0, ERM 3.5, and ERM 4.1. Mean concentrations of vanadium also differed by location (p<0.05) and period of study (p<0.05). Mean vanadium concentrations were higher at ERM 0.7,

ERM 1.0, ERM 2.2, ERM 2.6, and ERM 3.0 compared to the furthest upstream impacted location ERM 5.0 (p<0.05), but were similar to the reference location (p>0.05). Mean vanadium concentrations from these impacted locations ranged from 27.6 to 34.5 mg/kg compared to 12.0 mg/kg at ERM 5.0 (Figure 10F). Vanadium concentrations were also higher during Period 5 compared to Period 4 (p<0.05), which was likely driven by mean vanadium concentrations at ERM 3.0, ERM 3.5, and ERM 4.1.

2.3.3 Substrate Composition

Average substrate compositions in Period 4 and Period 5 are depicted in Figure 11 (A and B, respectively). Silt and sand dominated the substrates at all of the locations, accounting for 80 to 95% of the sediments. Mean percentages of silt and sand were significantly different among locations (p<0.05) but did not differ between periods of study (p>0.05). Silt was more prevalent at ERM 0.7, ERM 1.0, and ERM 2.6 compared to the reference location. On the contrary, sand was more prevalent at the reference compared to ERM 2.6. Measurements of percent ash in the substrates were also significantly different among locations (p<0.05), with higher concentrations of ash at ERM 0.7, ERM 1.0, ERM 2.2, ERM 2.6, ERM 3.0, and ERM 3.5 compared to the reference location (Figure 11). Percent of ash from these impacted locations ranged from 19 to 30%, compared to no observable ash in the reference location. Percent ash also differed by period of study (p=0.001), with higher percentages of ash observed in Period 5 compared to Period 4. No significant differences (p<0.05) among locations or between period of study were identified for clay or gravel. Clay accounted for roughly 10 to 20% of the substrates in most of the locations, while gravel typically accounted for less than 5%.

2.3.4 Relationships between Community Metrics and Sediment

Each of the benthic invertebrate community metrics were significantly correlated (Spearman rank) with ash-related metals, sediment grain size, percent dredgefull, and/or water depth. Specifically, the total abundance was moderately correlated to the percent dredgefull (r = 0.62, p<0.05). Total richness was correlated moderately but negatively to water depth (r = -0.60, p<0.05). NCBI scores were moderately correlated to manganese, gravel, and water depth (r = 0.40, r = 0.52, and r = 0.61, p<0.05). NCBI was also moderately but weakly correlated with silt (r = -0.41, p<0.05). Shannon Diversity was moderately but negatively correlated with barium, manganese, gravel, and water depth (r = -0.46, and r = -0.39, p<0.05). Notably, no combination of ash-related constituents was negatively correlated with total abundance or total richness, which supports the lack of differences between location metrics.

Despite the correlations identified above, results of the variable selection modeling indicated that no one sediment variable or set of sediment variables (including 17 ash-related metals, percent ash, water depth, percent dredge full, and sediment grain size) could predict total abundance, total richness, NCBI, or Shannon Diversity in Emory River samples.

2.4 Discussion

Initial predictions of long-term impacts to the benthic invertebrate community and subsequent effects to the aquatic ecosystem were catastrophic (Lisenby et al. 2009; Chattanooga Times Free Press, Emory River at 'tipping point', May 19, 2009). However, the results of this investigation do not support those claims. A review of previous studies evaluating exposure of benthic invertebrate communities to CCRs also contradicts the findings of the current study. Specht et al. (1984), Cherry et al. (1979), Harper and

Peckarsky (2005), and Smith (2003) each reported initial impacts to benthic invertebrate communities with reduced richness and abundance measures. In each of these studies, abundance and richness metrics improved within several months to 2 years after the majority of the coal combustion source material was removed from each aquatic system. In this study, spatial differences were expected due to the difference in the amount of residual ash and ash-related metals at each location and the effects that dredging the river bottom likely had to the benthic invertebrate community at several of the transects (Figure 4). While spatial differences were observed in percent ash, arsenic, barium, strontium, and vanadium, with higher concentrations at locations closest to the initial release, these differences did not lead to measureable variances in the abundance of benthic organisms or in the number of distinct taxa, diversity, or tolerance of collected individuals.

The rapid recovery or overall lack of impact of invertebrate assemblages in the Emory River locations suggests that 1) the benthic invertebrate community in the Emory River pre-release was comprised of highly tolerant organisms, 2) abundant upstream sources of recolonizing organisms populated the impacted portions of the river, or 3) the ash provided a similar silty substrate that was previously present in much of the impacted river reach.

A review of literature has indicated that aquatic systems most likely to recover following a disturbance are those systems with more irregular and unpredictable water flows with either periods of low or no water to frequent flooding (Mackay 1992; Poff and Ward 1990). Mackay (1992) stated that lotic invertebrate communities that experience more frequent disturbance are likely to be more resilient to environmental disturbances. The Emory River typically flows at 700 to 1,300 cubic feet per second (cfs) but is also

subject to flash flooding during storm events with 110,000 cfs marking a 10-year flood. During the main period of dredging following the TVA Kingston ash release, from May 2009 to May 2010, the river experienced four storm events with flows between 50,000 and 70,000 cfs (Jacobs 2012). These reoccurring high flow rates still cause channel scouring, transport, and deposition of sediments from the narrow sections of the river to the wider sections of the river (Scott 2014).

Given the fluctuation of water flow, the invertebrate community in the Emory River may have been accustomed to unstable substrate conditions prior to the ash release. Furthermore, the benthic invertebrates found in the Emory River were also likely adapted to reservoir conditions, including softer substrates and low dissolved oxygen (Baker 2006). During Periods 4 and 5, the Emory River benthic invertebrate community was dominated by chironomids and oligochaete taxa. Pre-release benthic invertebrate community data are not available for the Emory River; however, assemblage data collected in similar "run-of-the-river" reservoirs monitored by TVA (including Kentucky, Pickwick, Wheeler, Guntersville, Chickamauga, and Fort Loudoun reservoirs) were reviewed. Benthic invertebrate community surveys conducted in these reservoirs found similar dominant taxa compared to those collected in the Emory River, including predominately chironomid and oligochaete taxa. Previous literature has documented the tolerance of chironomids and oligochaetes to various forms of environmental disturbances and pollution (Mousavi et al. 2003; Waterhouse and Farrell 1985; Lenat 1983; Beck 1977). The abundance of these taxa may have subdued some initial impacts of the ash release. Furthermore, the total abundance (reported as density [number of organisms/m²) was almost twice as high in impacted Emory River transects compared to other reservoirs, with 1,700.73 number/m² compared to 963.8 number/m². Total taxa

richness was similar, with an average of 13.6 distinct taxa collected per transect in similar reservoirs compared to 12.6 taxa collected in the ash impacted Emory River locations (unpublished data; T. Baker, personal communication, September 25, 2013).

Following the 2008 release and subsequent year of dredging, the disturbed stretches of the Emory River would likely have provided unoccupied habitat that could be quickly recolonized from unimpacted sources. Previous studies have indicated that following a localized disruption of the community, even one that is severe, stream invertebrates will begin to populate the substrate as soon as shelter and food become available (Mackay 1992). Drift, swimming, crawling, and flight are the primary methods of redistribution of benthic invertebrates in streams (Williams and Hynes 1976; Minshall and Petersen 1985). While some benthic species are extremely mobile and may have crawled from side channels or neighboring drainages that were not impacted by the ash, the dominant taxa found in the Emory River following the spill (i.e., chironomids and oligochaetes) are typically sedentary; as a result, these organisms likely drifted into the impacted areas. Chironomids and oligochaetes are commonly associated with drifting as a means of redistribution and also possess other characteristics that make them early colonizers, including their small body size, high rate of reproduction and short life span (Oliver 1971; Kennedy 1966). Smith (2003) noted that benthic communities were dominated by common, tolerant taxa during the first 2 years of sampling following the ash release. These taxa included chironomids, hydropsychid caddisflies, and Baetis mayflies, all of which have been known to tolerate moderately disturbed conditions. However, several taxa associated with good water quality were collected roughly 5 years after ash discharges to the McCoy Branch watershed had ceased. Furthermore, the

frequent high flows commonly occurring on the Emory River also encourage drift of invertebrates.

Finally, the released ash, which is comprised primarily of silica particles, provided a silty substrate that was likely similar to the substrate in the Emory River prior to the ash release. Benthic invertebrate community abundance and diversity are closely related to substrate diversity (Henley et al. 2000). Baker (2011) described the pre-released substrate in the Emory River between ERM 0.0 and ERM 6.0 as predominately fines (silt and clay) or a mix of fines, sand, and detritus. Fly ash has a spherical shape, unlike the irregular particle shapes found in quartz-based sediments, and also has a lower particle density than in native sediments. Given these properties, fly ash consolidates and compacts when allowed to settle, much like sedimentation processes of fine sediment (Rivera et al. 2015).

Given this similarity and the dominance of silty substrate in the Emory River, the released ash likely did not result in the same degree of habitat alteration as would have occurred if ash was released into a stream or river with predominately cobble or bolder substrate. In a system with large cobble, the ash would have filled all of the interstitial space, reducing habitat availability and increasing the instability of substrate (Henley et al. 2000). Smith (2003) related the effects of released ash to that of conditions caused by sedimentation or siltation, which are typically associated with negatively affecting the ability of invertebrates to breathe and gather food. Similarly, Cherry et al. (1979) determined turbidity and the associated smothering effects of coal ash passing through the aquatic system to be the leading factors in eliminating or reducing populations of even the most tolerant aquatic taxa. However, the substrate in this portion of the Emory River was predominantly silty, with little to no stable substrate.

While the released ash would have initially smothered the benthic invertebrates inhabiting the sediment, it likely did not significantly alter the type of habitat available for these organisms to colonize. Harper and Peckarsky (2005) stated that the longest recovery times are generally associated with stressors leading to long-term alterations in physical stream habitat. Where sedimentation is actively occurring and substrates are predominately soft and silty, sediment-intolerant taxa become increasing displaced by sediment-tolerant taxa (Relyea et al. 2000). Moderate additions of sediment provide conditions that are tolerated by highly mobile taxa and taxa that are specifically adapted for living in deposited sediments, such as oligochaeta and some Chironomidae (Mackay 1992, Wiederholm 1984). Consequently, the benthic community that existed in the vicinity of the TVA Kingston facility had already adapted to reservoir conditions and was likely predominately composed of organisms that tolerated or preferred soft substrates and hence were less sensitive to this sedimentation-like release.

2.5 Summary

In the case study presented here, co-located benthic invertebrate community samples and submerged sediment samples were collected in order to identify relationships between community metrics and ash-related variables. These relationships would then be used to predict community results in future evaluations. Contrary to previous literature, benthic invertebrate community results of this spatial evaluation could not be tied to ash-related variables, such as percent ash or ash-related metals concentrations. Some hypotheses were made to explain this occurrence, including a pre-release benthic invertebrate community that was highly tolerant of environmental disturbance (i.e., flooding), abundant upstream sources of recolonizing organisms that could quickly fill the newly dredged or smothered habitat, and ash providing a similar

substrate that was previously present in much of the impacted river reach. The results from Periods 4 and 5 found the benthic invertebrate community dominated by oligochaetes and chironomid taxa. While pre-release data are not available for this section of the Emory River, data collected from other similar locations were also dominated by chironomid and oligochaete taxa. These organisms prefer soft, silty substrates and are often distributed through drift mechanisms. They can also be highly tolerant to environmental stressors. Given the known substrate types prior to the 2008 release, it is likely that the lack of differences in the community richness, tolerance, and diversity are due largely in part to the tolerance and adaptability of the Emory River organisms that were historically occurring in this reservoir system.

CHAPTER 3 TEMPORAL RESPONSES OF BENTHIC INVERTEBRATE COMMUNITIES IN THE EMORY RIVER IMPACTED BY COAL FLY ASH OVER FIVE YEARS, WATTS BAR RESERVOIR, ROANE COUNTY, TN

Abstract

The release of fly ash at the TVA KIF on 22 December 2008 discharged approximately 4.1 million m³ of coal ash into the adjacent aquatic and terrestrial systems. Previous benthic invertebrate investigations conducted by TVA and collaborative researchers concluded that benthic invertebrates in the Emory River were at moderate risk from ash-related constituents, primarily arsenic, in ash-contaminated sediment that remained in the Emory River following extensive dredging efforts. These conclusions were based on the observation of statistically significant reductions in growth and biomass in toxicity tests with Emory River sediment. Benthic invertebrate community survey results from 2010, however, did not support this conclusion. These previous surveys evaluated benthic invertebrate community data 1 year after dredging in the Emory River was complete. In this study, benthic invertebrate community metric results including taxa abundance, taxa richness, EPT richness, Shannon Diversity, tolerance, feeding guilds, and organism habits were compared over a 5-year period of study at one ash-impacted location and one reference location. Despite this long-term investigation, no trends indicating benthic invertebrate community recovery were noted over time. In addition, no significant differences were identified between the ashimpacted location compared to the reference location. This information is important for the informed monitoring, remediation, and damage assessment of the benthic invertebrate community at the Kingston Ash Recovery site. This research also increases our knowledge of benthic invertebrate tolerance to environmental disturbances in sediment of natural systems.

3.1 Introduction

Biomonitoring of benthic invertebrates in stream systems is commonly used to assess degradation of water and sediment quality caused by various environmental stressors, ranging from land development to chemical pollution (Goodyear and McNeill 1999; Maret et al. 2003). Benthic invertebrates are relatively sedentary and are closely linked with the sediments; as a result, this diverse group of organisms is fairly representative of local surface water and sediment conditions. Determining the reasons for community characteristic differences or changes among benthic invertebrates, however, is difficult given the number of factors influencing these organisms. Some benthic invertebrate species are associated with specific substrate type, water depth, water flow, and the amount of oxygen available. Likewise, benthic invertebrate community structures can be strongly influenced by the physical habitat (Jones et al. 1999; Jahnig and Lorenz 2008; Lepori et al. 2005; Boyero 2003). As a result, differences in community characteristics may occur naturally throughout water systems. Changes in community characteristics may also be due to species-specific differences in sensitivity to various environmental stressors, as individual benthic invertebrate taxa exhibit a range of sensitivities to pollutants.

Previous research has shown that some species of caddisflies, chironomids, and oligochaetes are relatively tolerant to metal concentrations, and others, such as mayflies, show a higher sensitivity to metals (Kiffney and Clements 1994; Cain et al. 2004; Courtney and Clements 2002). Individual taxa sensitivities may result in reduced abundance and species diversity, or ultimately lead to local elimination of some sensitive species (Courtney and Clements 2002; Cain et al. 2004; Clements 1994). The mechanisms explaining metals toxicity and tolerance to individual species are generally

understood. However, their expression in community-level effects is not well documented, and identifying exact cause and effect relationships between pollutants and changes in benthic invertebrate community compositions remains unclear (Clements et al. 1988; Clements 1999).

Over the past decade, trace metals and other constituents found in CCRs are environmental stressors that have caused increasing concern, particularly following the TVA Kingston ash release in 2008. CCRs include various solid materials that are produced as by-products during the coal-burning process at coal-fired electric power plants. These by-products include fly ash, bottom ash, boiler slag, flue gas desulfurization residues, and fluidized bed combustion wastes (Mayfield et al. 2013). Approximately 118 million metric tons of coal ash are produced in the United States each year. The majority of this waste is stored onsite at the facility in wet impoundments, such as surface impoundments (i.e., ash ponds or lagoons), or in dry landfills. These types of storage methods present potential risk to ecological resources, as coal ash contains trace amounts of heavy metals, such as arsenic, copper, mercury, nickel, selenium, and zinc (Gieré et al. 2003; Tishmack and Burns 2004; Rowe et al. 2002). While these metals occur naturally in coal, ash stored in both wet and dry facilities is subject to leaching and seeping of these contaminants from the impoundments or landfills into groundwater, aquifers, or nearby water systems (Lemly 2010; Lemly and Skorupa 2012). The USEPA has recently proposed new regulations for the disposal of CCRs that will potentially change the current rules for surface water impoundments (USEPA 2010).

Previous studies of CCR impacts on benthic invertebrate communities have focused on releases of ash or ash by-products into small streams and tributaries or into

closed systems (Smith 2003; Harper and Peckarsky 2005; Cairns et al. 1970; Specht et al. 1984; Cherry et al. 1979). Each of these studies identified initial impacts to the benthic invertebrate communities to some degree but recovery was typically noted within a relatively short period of time (2 to 5 years). A benthic invertebrate community study conducted after the TVA Kingston Ash Recovery Project, however, did not follow these same trends. Buys et al. (2015) found no discernable impacts to the benthic invertebrate community in the Emory River, despite the unprecedented release of approximately 4.1 million m³ of coal ash released from a TVA containment cell into the Emory River in December 2008. Because some sections of the river were not accessible immediately following the release, the findings were based on data collected 2 years post-release, after large sections of the Emory River had been mechanically and hydraulically dredged to remove ash from the riverbed (Buys et al. 2015). Benthic invertebrate communities are closely linked to the substrates in which they occur; consequently, the physical changes to the riverbed from dredging, in addition to the time delay in the evaluation, may have masked ash-related impacts in the benthic invertebrate community in the Emory River.

The purpose of the present investigation is to provide a comprehensive temporal assessment of changes in the benthic invertebrate community to ash and ash-related metals in an un-dredged portion of the Emory River. Total abundance of taxa, taxa richness, and other community metrics were used to compare 5 years of study, beginning immediately after the ash release, to determine if any trends or patterns of benthic community degradation or subsequent recovery could be detected. It was hypothesized that taxa abundance, richness, and diversity would be lowest immediately following the release at the downstream study site and would gradually increase as

mixing and capping of the ash by native sediments occurred through natural attenuation, returning the sediment to pre-release conditions.

3.2 Materials and Methods

3.2.1 Study Site

Field studies were conducted at two locations in the Emory River, in Watts Bar Reservoir, Roane County, Tennessee (Figure 12), one upstream and one downstream of the initial release of coal fly ash. The ash was discharged into the Emory River near ERM 2.2 when a containment wall failed. The force and volume of released ash, along with subsequent storms, pushed the ash upstream almost 5 km and downstream more than 32 km into lower parts of the Emory River as well as into the Clinch and Tennessee Rivers. The first collections of benthic invertebrate communities began in January 2009 (Period 1), and continued in December 2009 (Period 2), January 2010 (Period 3), December 2011 to January 2012 (Period 4), and November to December 2012 (Period 5).

An upstream location at ERM 6.0 was selected as a reference site, as it was the closest area to the spill (approximately 5.6 km upstream) that did not have fly ash in the substrate (Figure 12). Water depths at ERM 6.0 range from 2.1 to 10.7 m; however, the main channel encompasses the majority of the channel width. Specific water quality parameters for ERM 6.0 are shown in Table 4. The substrate is dominated by silt, sand, and detritus (Table 5). A downstream location at ERM 1.0 was also sampled. Since the time of the release, ERM 1.0 is the only impacted benthic invertebrate sampling location in the Emory River that has been monitored annually but was not dredged during the spill clean-up. Despite the large volume of ash deposited in this area, ERM 1.0 was excluded from dredging because of historical sediment contamination of cesium-147

from the USDOE facilities on the Oak Ridge Reservation (ARCADIS 2012).

Consequently, ERM 1.0 is the only location sampled on the Emory River that was determined to have large deposits of ash but that incurred no physical change to the riverbed from dredging (Figure 2). ERM 1.0 is approximately 2.4 km downstream of the initial ash release. Five years after the release, 129,000 m³ of ash (of the 4.1 million m³ ash) were estimated to remain in the lower section of the Emory River, between ERM 1.8 to ERM 0.0. Water depths at ERM 1.0 range from 2.4 to 9.1 m. Water quality parameters for ERM 1.0 are also shown in Table 4. After the release, ERM 1.0 substrates were dominated by silt, ash, and detritus (Table 5).

3.2.2 Sample Collection

Benthic invertebrate community samples were collected following the TVA Reservoir Vital Signs Monitoring Program methodology, which was established in 1990 to evaluate reservoirs and their tributaries (Kerans and Karr 1994). At each location, a line-of-site transect was established across the river channel. Ten samples, evenly spaced across the width of the channel, were collected at each location during each period of study. Benthic invertebrate community surveys were collected using a Ponar dredge (0.05 m²). Water depth and dominant substrate composition were recorded for each collection. Samples were rinsed through a 0.6 mm mesh screen. The remaining contents were placed into a container (the size and number of containers depended on the amount of material; containers include 0.5 and 1 liter glass jars) and the remaining contents were preserved in a 10% formalin solution. Samples were then sorted at Pennington and Associates, Inc. Laboratory, where they were preserved in 85% ethanol and identified to the lowest taxon possible.

3.2.3 Benthic Invertebrate Metric Calculations

Metrics are commonly used by USEPA (1999; 2006) and TDEC (2011) to quantify characteristics of benthic invertebrate community structure and function. The benthic invertebrate community metrics selected for this study were based on metrics used in Tennessee's *Quality System Standard Operating Procedure for Macroinvertebrate Stream Surveys* (TDEC 2011), TVA's Vital Signs Monitoring Program for reservoirs (Baker 2006), and other scientific literature (Kerans et al. 1992). The metrics presented in this evaluation included: 1) number of total taxa (abundance); 2) number of distinct taxa (richness); 3) number of distinct EPT taxa; 4) NCBI; and 5) Shannon Diversity. Together, these selected metrics describe the abundance and diversity of the taxa present, as well as consider the occurrence of sensitive and tolerant species.

Metrics often change in predictable ways with increased levels of anthropogenic influences or disturbances (Barbour et al. 1995). The total number of taxa or total abundance, the total number of distinct taxa or total richness, and the total number of distinct EPT taxa are typically expected to decrease in response to disturbances to benthic invertebrate communities (Kerans and Karr 1994). For consistency with past evaluations of the Emory River benthic invertebrate communities, taxa of Copepoda, Collembola, Daphnids, Ostracoda, and Hydrozoa were not included in the benthic community metric calculations. In addition, some specimens could only be identified at high taxonomic levels (i.e., family) due to specimen condition, size, or age (i.e., early instar). These damaged or early instar specimens may not be identifiable or have developed diagnostic characteristics that distinguish them from other specimens. As a result, some specimens are only counted as distinct taxa (i.e., taxa richness) if no other

specimen(s) of the genera are identified at lower taxonomic levels in the same sample. The NCBI is a tolerance index similar to the Hilsenhoff Biotic Index (Hilsenhoff 1987) that is based on a scoring scale of 0 (sensitive) to 10 (tolerant). The purpose of the NCBI index is to evaluate the tolerance of species that occur in a sample relative to their abundance in that sample. NCBI scores in benthic invertebrate communities generally increase when habitats are disturbed as sensitive species are replaced by those more resilient to changing conditions (Hilsenhoff 1987; TDEC 2011). Shannon Diversity is another metric that accounts for species diversity and abundance of each species. Larger numbers calculated in this index indicate greater diversity within the sample. Shannon Diversity is another metric likely to decrease when benthic invertebrate communities are disturbed, as sensitive species are unable to withstand changes to the environment (Krebs 1999; USEPA 2006).

Feeding guilds and organism habits were also evaluated for all genera. Feeding guild metrics categorize different feeding strategies (i.e., predator, scraper, shredder, gatherer, filterer, parasite, and piercer). They are often determined by the type and availability of food, and may change when environmental stressors are present (USEPA 1999). Organism habit describes the way an individual moves or maintains its position in the water or sediment (i.e., burrower, climber, clinger, sprawler, and swimmer). Similar to feeding guilds, organism habit can also be influenced by environmental stressors (USEPA 1999). Typically, the percent of shredders (feeding guild) and the percent of clingers (organism habit) decrease as disturbance to benthic invertebrate communities increase (Merritt et al. 2008; TDEC 2011).

3.2.4 Statistical Analysis

A completely randomized design (CRD) was used to evaluate benthic invertebrate community metrics at ERM 1.0 and at ERM 6.0 to determine if metrics differed among periods within each location. A mixed-model analysis of variance was conducted (using SAS, v. 9.4), and means were separated with Tukey-Kramer at 5% significance level. Locations were then compared in a CRD with split-plot treatment arrangement to evaluate differences between the impacted and reference locations. Factors included location, year, and number of samples (replicates within each location). Location and year were fixed factors while the number of samples per location was considered a random factor. Again, a mixed-model analysis of variance was conducted (using SAS, v. 9.4), and means were separated with Tukey-Kramer at 5% significance level.

3.3 Results

Benthic invertebrates collected at ERM 6.0 and ERM 1.0 included similar community compositions. A list of all species collected from ERM 6.0 and ERM 1.0, along with the period of collection is presented in Table 6. Throughout the 5 years of study, a total of 2,931 individual organisms were collected from ERM 6.0, including 60 distinct taxa. The community was comprised primarily of chironomids, oligochaetes, and other non-chironomid Diptera, with the following three dominant genera: *Chironomus* sp., Tubificidae, and *Chaoborus punctipennis*. Chironomids and oligochaetes accounted for 53% and 27%, respectively, of the total collected organisms (Figure 13). The average total abundance of organisms collected was statistically significantly different among periods (p<0.05). The number of individuals ranged from 27 to 97, with the highest abundance in Periods 1 and 5 (Table 7). During Period 3, the average total abundance

of individuals was significantly lower (p<0.05) than those collected in all other periods. The average number of distinct genera collected per period ranged from 5.9 in Period 3 to 8.6 in Period 1 (Table 7). While the highest richness was observed in Period 1, taxa richness at ERM 6.0 was statistically similar among all periods (p>0.05). The benthic invertebrate communities at ERM 6.0 also had similar diversity and number of distinct EPT taxa among periods (p>0.05) (Table 4). NCBI tolerance scores were statistically different among periods (p<0.05). Period 4 had significantly lower NCBI scores than other periods of collection (p<0.05); however, the scores for all periods were relatively high, ranging from 7.2 to 8.6 (Table 7), indicating that the community was mainly comprised of tolerant species. Feeding guilds included mainly gatherers and predators, although filterers, parasites, scrapers and shredders were also occasionally collected (Figure 14). Organism habits were dominated by burrowers and sprawlers; climbers and clingers were found only sporadically (Figure 15).

A total of 4,613 individual organisms were collected from ERM 1.0 during the study, including 73 distinct invertebrate taxa (Table 6). Throughout the 5 years of collection, the community was comprised primarily of chironomids, oligochaetes, and Bivalvia. The three dominant genera include Tubificidae, *Chironomus* sp., and *Musculium transversum*. Chironomids and oligochaetes accounted for 43% and 33%, respectively, of the total collected organisms (Figure 13). The average total abundance of benthic invertebrates collected at ERM 1.0 differed among periods (p<0.05), with average numbers ranging from 58 to 170 individuals. Significantly higher abundance occurred in Period 4 with almost two times the number of individuals collected than in any other period (p<0.05) (Table 7). Similarly, the total number of distinct taxa at ERM 1.0 was significantly different among periods (p<0.05), ranging from 9 to 17.5

genera per period. Significantly more genera were also collected in Period 4 (p<0.05). The benthic invertebrate communities at ERM 1.0 were generally similar between the five periods of study with respect to diversity, total number of distinct EPT taxa, and NCBI tolerance scores (p>0.05) (Table 7). Feeding guilds included mainly gatherers, predators, and filterers (Figure 14). Parasites, scrapers, and shredders were also occasionally collected. Organism habits were dominated by burrowers. Sprawlers and climbers were also relatively common and clingers were occasionally found (Figure 15).

When metrics were compared between locations, significant differences were noted for all five calculations. Total abundance, total richness, and Shannon Diversity were significantly (p<0.05) higher at ERM 1.0 compared to ERM 6.0. These differences were mainly driven by significantly higher abundance (Figure 16), richness (Figure 17), and diversity (Figure 18) in ERM 1.0 Period 4 collections compared to ERM 6.0 (Tukey, p<0.05). Total distinct EPT taxa was higher at ERM 1.0 (p<0.05) but no period specific differences were noted (Figure 19). Collections of EPT taxa were uncommon in both locations, with the exception of the burrowing mayfly Hexagenia sp. NCBI tolerance scores were significantly (p<0.05) lower at ERM 1.0 compared to ERM 6.0 (Figure 20), indicating that more sensitive taxa were collected from the impacted location than from the reference location. When individual differences were evaluated between periods, the first (Period 1) and last (Period 5) years of study had significantly lower NCBI ratings at ERM 1.0 compared to ERM 6.0 (Tukey, p<0.05). The lower NCBI ratings during these two periods were likely driven by the presence of the sensitive chironomidae genera, Epoicocladius sp., which occurred at ERM 1.0 but was not found at ERM 6.0. However, the overall scores for both locations throughout the five periods were relatively high (7 or

greater) indicating that communities in both locations were dominated by tolerant species.

Chironomidae and oligochaete taxa largely dominated the overall abundance and richness of both ERM 6.0 and ERM 1.0 (Figure 13). When total abundance of these taxa were evaluated, they mirrored the same patterns as the total community abundance, with the highest abundance observed in Period 4 at ERM 1.0 (p<0.05) (Figure 21). When these dominant taxa were removed from the metric calculations, similar results were observed as more taxa, both in abundance and richness, were collected at ERM 1.0 than at ERM 6.0 during all five periods. At ERM 1.0, total abundance of non-dominant taxa was lowest in Period 1, increased slightly in Period 2, decreased in Period 3, and then increased dramatically during Periods 4 and 5 (Figure 22). Non-chironomid and oligochaete taxa abundance at ERM 6.0 was low but constant during Periods 1, 2, and 3; dropped slightly in Period 4, and then increased during Period 5. No significant differences were noted. Trends in taxa richness were similar. While the number of distinct taxa was not statistically significantly different among periods, the lowest richness for both ERM 1.0 and ERM 6.0 was observed during the first two periods and then gradually increased during Periods 3, 4, and 5 (Figure 23).

Qualitative evaluations of feeding guilds (Figure 14) and organism habits (Figure 15) were also considered. Of the six organism feeding guilds present (gatherer, predator, filterer, shredder, scraper, and parasite), gatherers were most prevalent at both locations during all periods of study. Predators were consistently second highest in frequency, with only one exception. While filterers were found in less than 2% of any ERM 6.0 period of study, they ranged from 3 to 31% of the total distribution at ERM 1.0. Scrapers and parasites were seldom found at either location. ERM 6.0 and ERM 1.0

were both dominated by burrowers across all periods. Sprawlers were the second most prevalent habit for both locations. Climbers were present at each location during all periods; however, they were found more consistently at ERM 6.0 across all periods than at ERM 1.0. At ERM 1.0, these organisms made up less than 1% of the overall distribution during the first two periods but were more prevalent during Periods 3, 4, and 5 (11, 13, and 4%, respectively). Clingers were also present at each location during all periods but they made up less than 2% of the distribution during each timeframe.

3.4 Discussion

While benthic invertebrate communities in previous studies with coal ash exposure have shown a variety of responses based on the extent of the release and characteristics of the receiving body of water, all of these responses have included some notable reduction of diversity and abundance (Webster et al. 1986), changes in community composition from sensitive to tolerant species (Cherry et al. 1984), and/or sublethal effects such as reduced metabolic rates and dispersal (Hopkins et al. 2004; Rowe et al. 2002; Webster et al. 1986). Since pre-release data are not available for the Emory River, it is difficult to judge whether a true recovery has occurred following the Kingston ash release; however, the 5 years of results observed at ERM 1.0 following the Kingston release contrast starkly to previously documented patterns of benthic invertebrate metrics recorded in other aquatic systems impacted by CCRs. In a study by Cherry et al. (1979), sedimentation from an overflowing coal ash settling basin smothered the downgradient benthic invertebrate community in surrounding swamp streams, reducing community abundance. Cherry et al. (1979) recorded 50 to 98% decline of invertebrate densities immediately following the spill, including tolerant species that had previously inhabited the drainage system where coal ash contaminants

occurred. However, when the overflow of ash was reduced and water quality improved, the downgradient communities showed recovery within 1 year (Cherry et al. 1979; 1984).

Similarly, a study by Smith (2003) evaluated benthic invertebrate community recovery in a study of McCoy Branch, Tennessee, a stream that received over 20 years of effluent discharge of 80% fly ash and 20% bottom ash. Benthic invertebrate communities were sampled biannually over a period of 6.5 years, beginning while ash discharges were still occurring and ending after discharges had been eliminated. Smith (2003) noted significantly lower density and taxonomic richness in impacted sites, and an absence of pollution-sensitive taxa (i.e., mayflies and caddisflies). As operational changes were made to reduce ash discharges, community metrics improved and many peaked after 2 years of effluent reduction. Furthermore, Specht et al. (1984) studied effluent discharges to Adair Run, a tributary to the New River. Initial improvements were noted to the community 2 months after fly ash discharges were removed from the stream and complete recovery was determined after 10 months. These recovery results were similar to other stream studies of benthic invertebrate communities (Cherry et al. 1979; Harper and Peckarsky 2005; and Cairns et al. 1970), which also evaluated CCR disturbances to aquatic systems.

In contrast to these previously reported case studies, overall abundance (Figure 16) and taxa richness (Figure 17) following the TVA Kingston ash release at ERM 1.0 were similar among four of the five periods, with significant increases only occurring in Period 4 (p<0.05). Period 1 samples were collected less than 1 month after the initial release occurred, and would likely have experienced the most severe "smothering" effect; however, no statistically significant differences (p>0.05) were observed after dredging operations removed approximately 90% of the upstream ash

deposits. During this period, ash was measured in more than one-half of the samples and overall depths of ash deposits ranged from 0.15 m to 3 m (Table 5).

The community composition at ERM 1.0 was dominated by chironomids and oligochaete taxa throughout the study. Chironomids represent a diverse group of aquatic dipterans, comprised of five subfamilies: Tanypodinae, Podonominae, Diamesinae, Chironominae, and Orthocladiinae. Chironomid larvae occur in a range of habitats, with different habits and feeding behaviors. Oligochaetes are segmented aquatic worms that are common in sediment and detritus, generally preferring silt laden substrates. Most species feed by ingesting sediments and other microorganisms and plant matter. Because these taxa are common in the upstream Emory River reference and prefer silty substrates like those found at ERM 1.0, they may have recolonized earlier and more rapidly than other taxa. The life cycle of chironomids vary, with some species producing several generations in one year to others that produce only one generation per year (Pennak 1989). Oligochaete life cycles typically take 1 to 2 years (Pennak 1989; Brinkhurst and Jamieson 1972). Chironomids and oligochaetes have been known to tolerate disturbances from various forms of pollution (Mousavi et al. 2003; Waterhouse and Farrell 1985; Lenat 1983; Beck 1977). Consequently, the abundance of chironomid and oligochaete taxa may have masked some initial impacts of the ash release. Mackay (1992) stated that chironomids were one of the earliest arrivals on empty substrates, using drift from upstream locations as their primary mode of relocation. Furthermore, many of the dominant chironomid genera observed at ERM 1.0 are free-living taxa that are prone to drift (Beck 1977).

When chironomids and oligochaetes were removed from the abundance and richness metrics, trends observed in non-dominant taxa were more similar to those

recorded in the literature, although still not producing statistically significant differences among periods (Figures 22 and 23). The average number of total "non-chironomid and oligochaete" organisms collected per drop gradually increased over the 5 years of study, beginning in Period 3, after dredging of upstream river segments was complete (Figure 22). Total richness gradually increased throughout the five periods of study, with the lowest richness recorded in Period 1 and the highest richness in Period 5 (Figure 23).

While some potential trends may indicate a recovery or shifting of species, evaluations of individual species should be considered with caution as they can be unpredictable (Smith 2003). Rather, feeding guilds and organism habits were evaluated as these provide a more holistic picture of the benthic invertebrate community structure and are less susceptible to erratic change. The evaluation of feeding guilds at ERM 1.0 indicated that filter feeders may have been impacted from the initial release. During the first three periods of study, these organisms made up 17% of the total community distribution but slowly declined during the dredging timeframe (Figure 14). However, after dredging was completed and all engineering controls (i.e., silt curtains) were removed from the Emory River, the distribution of filter feeders in Periods 4 and 5 steadily increased, ending at just over 30% of the total distribution. While filter feeders often dominate benthic invertebrate communities in ponds and impoundments, these organisms are particularly sensitive to sedimentation and increased turbidity, due to the potential for clogging and damage of their and breathing apparatus (Richardson and Mackay 1991). As such, they may have initially experienced stress from the increased turbidity of water when ash was released into the Emory River and when the dredging process was occurring. Similarly, the evaluation of organism habit also indicated some

recovery of climber species (Figure 15). During the first two periods of study, these organisms made up less than 1% of the overall distribution. After dredging was complete, their distribution jumped to 11% and was notably higher during the last two periods of study (13 and 4%). These differences may be attributed to the increase in submerged plants, roots, and other woody debris that were likely more prevalent after dredging was complete and engineering controls were removed from the river.

One reason benthic invertebrate community metrics may not have reflected the predicted decline was that dredging of the released ash removed the majority of ash from the Emory River shortly after the spill occurred (within 18 months). Although ERM 1.0 was not included in the areas of dredging, reducing the amount of ash upstream would also reduce the amount of ash that migrated downstream during high flow events. As a result, natural sedimentation and mixing of native sediments with released ash occurred more quickly than if migration of upstream ash continued to deposit layers of ash downstream. Dredging limited the period of exposure of ash and ash-related constituents to benthic invertebrates, which likely reduced the biouptake of metals into invertebrates and other biota.

Furthermore, this section of the Emory River within the Watts Bar Reservoir is a primarily lotic system. Given the relatively short retention time of water within the reservoir, contaminants associated with the residual ash are rapidly washed downstream and diluted. The retention time of water in the Emory River is rather short (averages 90 days or less) (Baker 2006) compared to retention times of other reservoirs that may hold water for 6 months to several years (Rueda et al. 2006). This process may also have reduced exposure and accumulation of ash-related constituents, thereby preventing constituents from reaching levels that cause adverse effects in the water, sediment, or

food web (Rueda et al. 2006; Carriker et al. 2015). In contrast, selenium and other metals associated with CCRs have been shown to cause significant ecological effects in more lentic systems (Lemly 1997). Selenium-contaminated wastewater from a coal facility in North Carolina contaminated Belews Lake for over a decade (Lemly 1997). In this study, it was determined that two of the key factors influencing the reservoir's inability to recover were the long hydrologic retention time and the slow sedimentation rate. Belews Lake typically holds water for about 4 years (1,500 days) and accumulates <0.5 cm of sediment per year. Consequently, selenium continued to be bioavailable in sediments and biota for years after the input of selenium ceased. Ten years following the release, impacts to the aquatic ecosystem were still evident (Lemly 1997).

Finally, the majority of the previously discussed case studies involved releases of coal ash effluent or caustic water associated with coal plant processes (Cairns et al. 1970; Cherry et al. 1979; Cherry et al. 1984; Smith 2003; Specht et al. 1984; and Lemly 1997). The ash released from the Kingston facility, however, was predominately fly ash that had been stored in a dry landfill for more than 20 years. As a result, the ash-related metals from the Kingston ash may not have been as available to biotic uptake as those occurring in the more acidic effluent. Furthermore, studies evaluating metal-leaching from ash indicate that under natural conditions, the more weathered the ash, the lower the concentrations of leached metals (Meima and Comans 1999).

3.5 Summary

Determining community differences caused by anthropogenic disturbances compared to the natural variation in community structures continues to present a challenge in benthic invertebrate biomonitoring studies (Mousavi et al. 2003; Clements 1994). This post-release evaluation of benthic invertebrate communities exposed to

CCRs in the Emory River highlights these challenges and further demonstrates the need for additional research of benthic invertebrate community composition and structure in rivers and reservoir systems. Although numerous studies have shown severe, initial impacts of invertebrates exposed to CCRs in smaller stream systems or man-made impoundments (Smith 2003; Cherry et al. 1979; Harper and Peckarsky 2005; Cairns et al. 1970; Cherry and Guthrie 1977), these same impacts were not clearly detected during this study. Some evidence of recovery at the ash impacted location ERM 1.0 was observed when evaluating general trends of abundance and richness metrics; however, the overall diversity and tolerance of the benthic community were similar to upstream locations and did not show temporal trends over the 5 years of study. It is unclear whether these results were due to the presence of tolerant organisms prior to the release that were able to cope with the increased environmental stressors associated with the spill, or if the specific characteristics associated with the released ash in this reservoir system did not subject the benthic invertebrates to lethal conditions. Since no pre-release data are available to determine if communities return to a pre-disturbed state, benthic invertebrate communities may need to be monitored over a period longer than 5 years to see if abundance or richness metrics continue to increase or indicate other patterns of recovery.

CHAPTER 4 CONCLUSIONS AND FUTURE RESEARCH NEEDS

4.1 Conclusions

Benthic invertebrate communities have long been used to evaluate environmental stresses to aquatic ecosystems. These organisms are in direct contact with surface water and sediment and are relatively immobile, which closely associates them to sediments and other local conditions (Li et al. 2010; Wallace and Webster 1996; Sundermann et al. 2013; Clements 1994; Allan 2004; Maret et al. 2003; Gebler 2004; Clements 1999). Not only can benthic invertebrates accumulate metals and other contaminants which are bound to the sediments in which they live, but they can also transfer these contaminants into aquatic and riparian food chains by serving as prey for upper trophic level receptors. Benthic invertebrate communities are often described and summarized by calculating metrics or indices. These metrics are used to analyze the community data and are often categorized as: composition/abundance, richness/diversity, sensitivity/tolerance, or function (Sundermann et al. 2013; Barbour et al. 1995).

Benthic invertebrate species may respond differently to chemical contaminants than to other types of environmental stressors, such as nutrient enrichment and changes to stream hydrology or habitat structure. As our understanding of indicator species assemblages and rapid bioassessment protocols has increased, the use of benthic invertebrates for biomonitoring has also increased (Clements 1994). Identifying and understanding the relationships between aquatic organisms and environmental stressors is critical for "the effective management, restoration, and preservation of aquatic ecosystems" (Burton and Johnston 2010).

While a considerable body of literature exists documenting the impact and recovery of benthic invertebrate communities to CCR releases into aquatic systems, the case study presented here did not follow the trends previously recorded at other spill sites. Previous evaluations of benthic invertebrate community metrics have demonstrated severe, initial impacts of invertebrates exposed to CCRs. These studies focused on small stream systems or man-made impoundments (Smith 2003; Cherry et al. 1979; Harper and Peckarsky 2005; Cairns et al. 1970; Cherry and Guthrie 1977). Contrary to these recordings, the benthic invertebrate community metrics measured following the TVA Kingston ash release showed little to no change in abundance, richness, diversity, or tolerance over a 5-year period of time. Furthermore, few relationships could be established between these metrics and any of the measured sediment variables (including: 17 ash-related metals concentrations, percent ash, water depth, percent dredgefull, and sediment grain size) in co-located sediment samples. No single variable or group of variables could predict, with even weak confidence, increases or decreases in any of the four benthic invertebrate community metrics.

Determining community differences caused by anthropogenic disturbances compared to the natural variation in community structures continues to present a challenge in benthic invertebrate biomonitoring studies (Mousavi et al. 2003; Clements 1994). It is unclear why the results from the present study differ from so many of the previous evaluations; however, several ideas have been considered.

One potential explanation for the lack of observed effects on the benthic invertebrate community in the Emory River was the initial properties of the released ash and immediate dredging of the riverbed. The ash released from the Kingston facility was predominately weathered fly ash. The majority of the previously discussed case studies,
however, involved releases of coal ash effluent or caustic water associated with coal plant processes (Cairns et al. 1970; Cherry et al. 1979; Cherry et al. 1984; Smith 2003; Specht et al. 1984; and Lemly 1997). Studies evaluating metal-leaching from ash indicate that under natural conditions, the more weathered the ash, the lower the concentrations of leached metals (Meima and Comans 1999). As a result, the ash-related metals from the Kingston ash may not have been as available to biotic uptake as those occurring in the more acidic effluent. In addition, the majority of the ash was removed from the Emory River within 18 months of the initial release. While not all areas of the river were dredged, reducing the overall amount of ash in the Emory River would also reduce the amount of ash that migrated downstream during high flow events. Furthermore, the impacted section of the Emory River is also within the Watts Bar Reservoir. The retention time of water in the Emory River is relatively short (averages 90 days or less) (Baker 2006) compared to retention times of other reservoirs that may hold water for 6 months to several years (Rueda et al. 2006). As a result, contaminants associated with the residual ash were likely washed downstream and/or diluted. Together, these processes may have reduced exposure and accumulation of ash-related constituents, thereby preventing constituents from reaching levels that cause adverse effects in the water, sediment, or benthic invertebrate tissue (Rueda et al. 2006; Carriker et al. 2015).

Following the 2008 release and subsequent year of dredging, the disturbed stretches of the Emory River would likely have provided unoccupied habitat that could be quickly recolonized from unimpacted sources. Previous studies have indicated that following a localized disruption of the community, even one that is severe, stream invertebrates will begin to populate the substrate as soon as shelter and food become available (Mackay 1992). In addition, aquatic systems most likely to recover following a

62

disturbance are often those systems with more irregular and unpredictable water flows with either periods of low or no water to frequent flooding (Mackay 1992; Poff and Ward 1990). While the Emory River typically flows at less than 1,500 cfs, it is also subject to flash flooding during storm events. The frequent high flows commonly occurring on the Emory River not only enable drift of invertebrates, but may also promote invertebrate communities that are more likely to be resilient to environmental disturbances (MacKay 1992).

Finally, benthic invertebrate community abundance and diversity are closely related to substrate diversity (Henley et al. 2000). The benthic invertebrates found in the Emory River were likely adapted to reservoir conditions, including softer, fine substrates (predominately silt and clay) as described in Baker (2011). The released ash, which is comprised primarily of silica particles that are spherical in shape, consolidates and compacts when allowed to settle, much like sedimentation processes of fine sediment (Rivera et al. 2015). Given this similarity and the dominance of silty substrate in the Emory River, the released ash likely did not result in the same degree of habitat alteration as would have occurred if ash was released into a stream or river with predominately cobble or bolder substrate. Where sedimentation is actively occurring and substrates are predominately soft and silty, sediment-intolerant taxa become increasing displaced by sediment-tolerant taxa (Relyea et al. 2000). Moderate additions of sediment provide conditions that are tolerated by highly mobile taxa and taxa that are specifically adapted for living in deposited sediments, such as oligochaeta and some chironomidae (Mackay 1992, Wiederholm 1984). Consequently, the benthic community that existed in the vicinity of the TVA Kingston facility had already adapted to reservoir conditions and was likely

63

predominately composed of organisms that tolerated or preferred soft substrates and hence were less sensitive to this sedimentation-like release.

4.2 Future Research Needs

This post-release evaluation of benthic invertebrate communities exposed to CCRs in the Emory River highlights the challenges associated with distinguishing natural variability and environmental stress in aquatic ecosystems. Since no data recording the benthic invertebrate community are available for this area of the Emory River, it is difficult to determine if communities have returned to a pre-disturbed state. Furthermore, community data for reservoir systems remain undocumented, providing few comparisons for typical metrics measured in these unique environments. These issues demonstrate the need for additional research of benthic invertebrate community composition and structure in large rivers and reservoir systems. In addition, benthic invertebrate communities in the Emory River may need to be monitored over a period longer than 5 years to see if abundance or richness metrics continue to increase or indicate other patterns of recovery.

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69

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77

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APPENDICES

Tables

Table 1. Summary of the benthic invertebrate community composition in the Emory River following the TVA Kingston ash release, Watts Bar Reservoir, Roane County, TN¹

Phylum	Class	Order	Family
Platyhelminthes	Turbellaria	Tricladida	Planariidae
Arthropoda	Insecta	Coleoptera	Elmidae
			Hydrophilidae
		Diptera	Ceratopogonidae
			Chaoboridae
			Chironomidae
			Simuliidae
			Tipulidae
		Ephemeroptera	Ameletidae
			Caenidae
			Ephemerellidae
			Ephemeridae
			Heptageniidae
		Megaloptera	Sialidae
		Odonata	Coenagrionidae
			Gomphidae
		Plecoptera	Capniidae
			Chloroperlidae
			Taeniopterygidae
		Trichoptera	Hydroptilidae
			Leptoceridae
			Polycentropodidae
	Arachnida	Acariformes	Arrenuridae
			Hygrobatidae
			Lebertiidae
			Pionidae
			Unionicolidae
	Malacostraca	Amphipoda	Talitridae
Mollusca	Gastropoda	Architaenioglossa	Viviparidae
		Basommatophora	Ancylidae
			Physidae
		Neotaenioglossa	Hydrobiidae
			Pleuroceridae
	Bivalvia	Unionoida	Unionidae
		Veneroida	Corbiculidae
			Sphaeriidae

Table 1. Continued.

Phylum	Class	Order	Family
Annelida	Hirudinea	Pharyngobdellida	Erpobdellidae
		Rhynchobdellida	Glossiphoniidae
	Oligochaeta	Haplotaxida	Enchytraeidae
			Naididae
			Tubificidae
		Lumbriculida	Lumbriculidae
Nematoda	-	-	-

1 – Sample periods included Period 4 (December 2011 - January 2012) and Period 5 (November - December 2012). Samples were collected from Emory River mile (ERM) 6.0 (reference) and ERM 0.7, 1.0, 2.2, 2.6, 3.0, 3.5, 4.1, and 5.0 (impacted).

Location ²	Period	Total Abundance	Total Richness	Shannon Diversity	NCBI ³
ERM 0.7	4	124	14	2.0	7.3
	5	145	14	1.7	7.2
ERM 1.0	4	134	17	2.0	7.0
	5	113	14	1.9	7.5
ERM 2.2	4	74	12	2.0	7.3
	5	158	10	1.3	7.5
ERM 2.6	4	113	15	2.1	7.5
	5	101	11	2.0	7.6
ERM 3.0	4	128	13	1.8	7.5
	5	101	11	1.7	7.8
ERM 3.5	4	90	14	1.8	6.6
	5	57	7	1.5	8.0
ERM 4.1	4	65	11	1.8	7.4
	5	64	10	1.6	8.0
ERM 5.0	4	89	11	1.6	7.6
	5	41	13	2.2	7.7
ERM 6.0 ⁴	4	42	8	1.5	7.2
	5	125	9	1.4	8.8

Table 2. Average benthic invertebrate community metrics calculated in the Emory River, Watts Bar Reservoir, Roane County, TN^1

1 – Sample periods included Period 4 (December 2011 - January 2012) and Period 5
2 – ERM = Emory River mile.
3 – NCBI = North Carolina Biotic Index.

4 – ERM 6.0 = Reference location.

Location ²		Arsenic	Barium	Beryllium	Boron	Chromium	Selenium	Strontium	Vanadium
ERM 0.7	Period 4	17.9	156.04	1.44	10.78	15.39	2.55	82.8	30.3
	Period 5	16.3	141.13	1.28	8.36	14.57	1.83	77.8	27.7
	Average	17.1	148.6	1.4	9.6	15.0	2.2	80.3	29.0
ERM 1.0	Period 4	17.3	163.58	1.52	11.35	16.17	2.95	82.0	31.6
	Period 5	16.0	117.55	1.14	7.03	12.53	1.76	58.8	23.7
	Average	16.6	140.6	1.3	9.2	14.3	2.4	70.4	27.6
ERM 2.2	Period 4	32.8	170.20	1.55	16.44	13.18	3.29	103.8	35.2
	Period 5	26.8	157.83	1.51	9.78	17.50	2.10	83.8	33.9
	Average	29.8	164.0	1.5	13.1	15.3	2.7	93.8	34.5
ERM 2.6	Period 4	35.8	161.58	1.57	15.17	17.15	3.15	77.7	35.4
	Period 5	14.8	106.55	1.22	8.95	15.84	1.82	54.2	27.1
	Average	25.3	134.1	1.4	12.1	16.5	2.5	65.9	31.2
ERM 3.0	Period 4	14.4	99.20	1.06	8.17	10.12	2.13	30.2	18.1
	Period 5	27.8	161.48	1.50	14.36	21.75	1.91	66.9	37.3
	Average	21.1	130.3	1.3	11.3	15.9	2.0	48.5	27.7
ERM 3.5	Period 4	14.5	65.12	0.84	9.31	6.40	1.64	39.8	14.5
	Period 5	36.1	157.43	1.75	11.83	20.45	2.32	85.5	39.1
	Average	25.3	111.3	1.3	10.6	13.4	2.0	62.7	26.8
ERM 4.1	Period 4	3.4	37.50	0.60	5.92	4.48	1.48	6.4	7.3
	Period 5	8.3	105.90	1.12	8.52	20.38	2.13	24.3	27.2
	Average	5.8	71.7	0.9	7.2	12.4	1.8	15.4	17.2
ERM 5.0	Period 4	15.1	68.85	0.95	9.96	8.13	2.27	31.7	15.9
	Period 5	4.9	52.33	0.75	6.80	6.28	3.06	8.9	8.2
	Average	10.0	60.6	0.8	8.4	7.2	2.7	20.3	12.0
ERM 6.0 ³	Period 4	3.1	48.28	0.72	6.66	5.77	1.67	6.7	8.8
	Period 5	4.3	87.37	0.57	23.09	27.54	2.90	15.0	19.0
	Average	3.7	67.8	0.6	14.9	16.7	2.3	10.9	13.9

Table 3. Average concentrations of ash-related metals (mg/kg) in submerged sediment collected in the Emory River, Watts Bar Reservoir, Roane County, TN¹

1 – Sample periods included Period 4 (December 2011 - January 2012) and Period 5 (November - December 2012). 2 – ERM = Emory River mile. 3 – ERM 6.0 = Reference location.

Table 4. Chemical and physical characteristics at ERM 6.0 (reference location) and ERM 1.0 (impacted location), Watts Bar Reservoir, Roane County, TN¹

Parameter	ERM 6.0	ERM 1.0
Water Depth (meter)	7.09	6.99
Range (meter) ²	(0.46 - 3.2)	(1.6 - 11.4)
Channel Width (meter)	150	700
Temperature (°C)	5.39	8.6
Dissolved Oxygen (mg/liter)	12.46	10.13
Turbidity	3.0	4.4
рН	7.02	7.63
Conductivity	55	273

Parameters recorded during Period 4 sampling event, unless otherwise noted; ERM = Emory River Mile.
 Water depth range recorded during each period of study for all samples.

Location	Period ²	Ash	Clay	Silt	Sand	Gravel	Detritus
ERM 6.0	1	0.0	0.0	74	17	0.0	8.5
		(0-0)	(0-0)	(25 - 95)	(0 - 70)	(0-0)	(5 - 20)
	2	0.0	0.0	49	22.0	4.5	24
		(0-0)	(0-0)	(5 - 90)	(0 - 60)	(0 - 40)	(5 - 90)
	3	0.0	0.0	43	37	3.0	18
		(0-0)	(0-0)	(0 - 80)	(0 - 85)	(0 - 15)	(10 - 45)
	4	0.0	12	43	39	2.5	3.7
		(0-0)	(0 - 90)	(0 - 95)	(0 - 99)	(0 - 20)	(0 - 15)
	5	0.0	0.0	80	17	1.0	2.9
		(0-0)	(0-0)	(0 - 100)	(0 - 100)	(0 - 5)	(0 - 15)
ERM 1.0	1	29	0.0	51	0.0	0.0	20
		(0 - 85)	(0-0)	(0 - 93)	(0-0)	(0-0)	(5 - 40)
	2	38	0.0	36	1.5	1.5	22
		(0 - 85)	(0-0)	(5 - 75)	(0 - 15)	(0 - 15)	(0 - 60)
	3	24	0.0	63	9.0	0.5	4.0
		(0 - 70)	(0-0)	(25 - 100)	(0 - 65	(0 - 5)	(0 - 20)
	4	9.9	0.0	81	10	4.0	4.7
		(0 - 24)	(0-0)	(5 - 98)	(0 - 90)	(0 - 40)	(0 - 10)
	5	27	3.0	46	15	0.0	8.0
		(0 - 60)	(0 - 30)	(15 - 95)	(0 - 80)	(0-0)	(0 - 25)

Table 5. Qualitative substrate composition at ERM 6.0 (reference location) and ERM 1.0 (impacted location), Watts Bar Reservoir, Roane County, TN¹

1 - Composition of samples (%) was characterized based on visual observations. Percentages may not add to 100 due to rounding. ERM = Emory River mile. 2 – Sample Periods are defined as follows:

Period 1: January 2009

Period 2: December 2009

Period 3: December 2010 - January 2011

Period 4: December 2011 - January 2012

Period 5: November - December 2012

Table 6. List of benthic invertebrate species collected at ERM 6.0 (reference location) and ERM 1.0 (impacted location), Watts Bar Reservoir, Roane County, TN¹

	Period of Collection				
Таха	Period	Period	Period	Period	Period
	1	2	3	4	5
Arachnida					
Acariformes					
Hygrobatidae					
Atractides sp.					x/o
Lebertildae					
Lebertia sp.				0	
Unionicolidae					
Neumania sp.				х	X
Unionicola sp.	х		х	х	x/o
NA					
Acariformes sp.			х		
Bivalvia					
Unionoida					
Unionidae					
Unionidae					x / o
Veneroida					
Corbiculidae					
Corbicula fluminea	x / o	x / o	x / o	x / o	x / o
Sphaeriidae					
Musculium transversum	х	х	х	х	х
Pisidium compressum				х	
Pisidium sp.		х	х	х	х
Gastropoda					
Basommatophora					
Ancylidae					
Ferrissia rivularis		0			
Neotaenioglossa					
Hydrobiidae					
Amnicola limosa	х				х
Pleuroceridae					
Pleurocera canaliculata					х
Hirudinea					
Rhynchobdellida					
Glossiphoniidae					
Actinobdella inequiannulata			х		х

Table 6. Continued.

	d of Colle	of Collection			
Таха	Period	Period	Period	Period	Period
A stime to de lla ser	1	2	3	4	5
Actinobdella sp.				X	
Helobdella stagnalis	X				
Glossiphoniidae	X	х			
Insecta					
Coleoptera					
Elmidae					
Dubiraphia sp.	0	0	Х	x/o	х/о
Microcylloepus pusillus			х	0	
Stenelmis sp.				х	
Diptera					
Ceratopogonidae					
Ceratopogonidae	x / o	х	x / o	x/o	x/o
Chaoboridae					
Chaoborus punctipennis	x / o	x/o	x / o	x / o	x/o
Chironomidae					
Ablabesmyia annulata	x / o	x / o	х	х	x / o
Ablabesmyia mallochi		х		х	
Ablabesmyia peleensis				х	х
Antillocladius sp.				х	
Axarus sp.			х		
Cardiocladius obscurus				х	
Chironomus crassicaudatus				х	
Chironomus decorus gp.				x/o	
Chironomus sp.	x/o	x/o	x/o		x/o
Chironominae subfamily				х	
Cladopelma sp.			х		
Cladotanytarsus sp.				x/o	x/o
Clinotanypus sp.	о				
Coelotanypus sp.	x/o	о	х	х	х
Coelotanypus tricolor				х	
Cricotopus bicinctus				о	
Cricotopus sylvestris gp.				х	
Cryptochironomus sp.	x/o	x/o	x/o	x/o	x/o
Dicrotendipes modestus				x	x
, Dicrotendipes neomodestus			x/o	x	x
Dicrotendipes simpsoni					x
Dicrotendipes sp.	о				
Endochironomus nigricans	o				

	Period of Collection					
Таха	Period	Period	Period	Period	Period	
	1	2	3	4	5	
Epoicocladius flavens		х		x/o	х	
Epoicocladius sp.	х	х	х			
Eukiefferiella claripennis gp.				0		
Glyptotendipes sp.	х/о		x/o	x/o	0	
Hydrobaenus sp.				x/o	х	
Lipiniella sp.	0					
Microchironomus sp.				х	x / o	
Microtendipes pedellus gp.		x / o	0	x / o	х	
Nilothauma sp.			х			
Orthocladius sp.				х		
Pagastiella sp.	0	0		x/o	0	
Paracladopelma gp.	х					
Paracladopelma undine				х		
Paralauterborniella		о	х	x/o	о	
nigrohalteralis						
Paratanytarsus sp.				х		
Paratendipes albimanus				х		
Phaenopsectra obediens gp.				0		
Phaenopsectra punctipes gp.				0		
Polypedilum halterale	х	0	x/o	x/o	x / o	
Polypedilum scalaenum gp.		х				
Procladius sp.	x / o	x / o	x / o	x / o	x / o	
Pseudochironomus sp.			x / o	х		
Rheotanytarsus exiguus gp.	0		0			
Smittia sp.			х			
Stenochironomus sp.			х			
Stictochironomus caffrarius gp.				х	х	
Stictochironomus devinctus			0		0	
Tanytarsus sp.	о	x/o	х	x/o	x/o	
Tribelos fuscicorne				х		
Tribelos jucundum	о	x/o	x/o	x/o	х	
Tvetenia vitracies	о					
Zalutschia sp.	x/o				x	
Ephemeroptera						
Ephemeridae						
Hexagenia sp.	x/o	x/o	x/o	x/o	x/o	
Heptageniidae	_	_	-	-	-	
Heptageniidae				x		

Table 6. Continued.

Table 6. Continued.

	d of Colle	I of Collection			
Таха	Period	Period	Period	Period	Period
Magalantara	1	2	3	4	5
Niegaloptera					
Sialis sp.				X	
Taeniopteryx sp.			0		
Irichoptera					
Hydropsychidae					
Cheumatopsyche sp.		Х	х		
Hydroptilidae					
Hydroptila sp.			0		
Leptoceridae					
Oecetis sp.	0	х		х	x/o
Polycentropodidae					
Nyctiophylax sp.			х		
Polycentropus sp.		0			
Malacostraca					
Isopoda					
Asellidae					
Lirceus sp.		х			
Nematoda					
Nematoda	x / o		0	x / o	x / o
Oligochaeta					
Haplotaxida					
Enchytraeidae					
Enchytraeidae			х	х	
Naididae					
Arcteonais Iomondi		0			
Dero sp.		x/o		х	
Nais pardalis					х
Nais sp.				х	
Slavina appendiculata		о		о	
Naididae	х	о		x/o	о
Tubificidae					
Aulodrilus piqueti			о	x	x
Branchiura sowerbyi	x/o	x	x	x/o	x/o
Limnodrilus cervix			x/o		x/o
Limnodrilus claparedianus	x/o	x/o	о	x/o	о
	A/U	~/ 0	0	~/ 0	0

90

Table 6. Continued.

	Period of Collection				
Таха	Period	Period	Period	Period	Period
	1	2	3	4	5
Limnodrilus hoffmeisteri	x/o	x/o	x / o	Х	0
Limnodrilus sp.		0	х	0	x / o
Quistadrilus multisetosus			0		
Tubificidae	x/o	x/o	x/o	x/o	x / o
Lumbriculida					
Lumbriculidae					
Lumbriculidae		0		х	
Platyhelminthes					x

1 – ERM = Emory River mile; gp = group; o = present at ERM 6.0 (Reference site); sp = species; x = present at ERM 1.0 (Impacted site); x / o = present at both ERM 6.0 and ERM 1.0

2 – Sample Periods are defined as follows: Period 1: January 2009 Period 2: December 2009 Period 2: December 2009 - January 2011 Period 4: December 2011 - January 2012 Period 5: November - December 2012

Location	Period	Total Individuals	Number of Distinct Taxa	Shannon Diversity Index	Total EPT Richness ²	NCBI Tolerance Score ³
ERM 6.0	1	82.7 ab	8.6 a	1.5 a	0.5 a	8.4 a
	2	44.2 bc	7.7 a	1.7 a	0.5 a	7.9 ab
	3	27.1 c	5.9 a	1.4 a	0.3 a	8.3 a
	4	42.4 bc	8.0 a	1.5 a	0.2 a	7.2 b
	5	96.7 a	7.7 a	1.4 a	0.3 a	8.6 a
ERM 1.0	1	58 b	9.0 b	1.8 a	0.6 a	7.2 a
	2	66.5 b	9.2 b	1.8 a	0.9 a	7.4 a
	3	79.2 b	11.0 b	1.8 a	0.9 a	7.8 a
	4	169.9 a	17.5 a	2.0 a	1.1 a	7.0 a
	5	87.7 b	12.6 ab	1.9 a	1.1 a	7.2 a

Table 7. Summary of benthic invertebrate metrics at ERM 6.0 (reference location) and ERM 1.0 (impacted location), Watts Bar Reservoir, Roane County, TN¹

1 – Two-way ANOVA comparing periods was applied to five years of data for ERM 6.0 and ERM 1.0. The mean for each period is shown in this table. If the ANOVA was significant, significant differences among means were determined using post-hoc Tukey test and are denoted with different letters (p<0.05). ERM = Emory River mile.</p>

2 - EPT = Ephemeroptera, Plecoptera, Tricoptera

3 – NCBI = North Carolina Biotic Index

Figures



Figure 1. Location of Tennessee Valley Authority Kingston Ash Fossil Plant, Roane County, TN. ERM = Emory River mile.



Figure 2. Bathymetric survey presenting the most current prediction of ash deposition in the Emory River, Watts Bar Reservoir, Roane County, TN.



Figure 3. Location of spatial study sites in the Emory River, Watts Bar Reservoir, Roane County, TN. Transect line (red) at Emory River mile (ERM) 6.0 (reference location) and ERM 0.7, 1.0, 2.2, 2.6, 3.0, 3.5, 4.1, and 5.0 (impacted locations).



Figure 4. Overview of dredged (green shading) and un-dredged (orange shading) ash deposits in the Emory River, Watts Bar Reservoir, Roane County, TN. ERM = Emory River mile.


Figure 5. Benthic invertebrate community composition during Periods 4 and 5 in the Emory River, Watts Bar Reservoir, Roane County, TN. Sites include Emory River mile (ERM) 6.0 (reference location) and ERM 0.7, 1.0, 2.2, 2.6, 3.0, 3.5, 4.1, and 5.0 (impacted locations). Asterisk (*) includes species of Chaoboridae, Ceratopogonidae, Simuliidae, and Tuplidae; (**) includes species of alderflies (Megaloptera), amphipods, caddisflies (Tricoptera), dragonflies (Odonata), leeches (Hirundinea), mussels, nematodes, non-parasitic flatworms (planarians), stoneflies (Plecoptera) and water beetles.



Figure 6. Average total abundance of individuals collected per transect at reference (†) and impacted locations in the Emory River, Watts Bar Reservoir, Roane County, TN. No significant difference noted between impacted and reference locations (p>0.05). ERM = Emory River mile.

99



Figure 7. Average total number of distinct taxa (richness) collected per transect at reference (†) and impacted locations in the Emory River, Watts Bar Reservoir, Roane County, TN. No significant difference noted between impacted and reference locations (p>0.05). ERM = Emory River mile.



Figure 8. Average Shannon Diversity collected per transect at reference (†) and impacted locations in the Emory River, Watts Bar Reservoir, Roane County, TN. No significant difference noted between impacted and reference locations (p>0.05). ERM = Emory River mile.



Figure 9. Average North Carolina Biotic Index (NCBI) for tolerance of taxa collected per transect at reference (†) and impacted locations in the Emory River, Watts Bar Reservoir, Roane County, TN. Bars with different letters differ significantly (p<0.05). ERM = Emory River mile.

Period 4

Period 5



Figure 10. Concentrations of metals in sediment (mg/kg) collected per transect at reference (†) and impacted locations in the Emory River, Watts Bar Reservoir, Roane County, TN. Bars with different letters differ significantly (p<0.05). ERM = Emory River mile.



Figure 11. Substrate composition collected per transect during Period 4 (A) and Period 5 (B) at reference (†) and impacted locations in the Emory River, Watts Bar Reservoir, Roane County, TN. ERM = Emory River mile.



Figure 12. Location of temporal study sites in the Emory River, Watts Bar Reservoir, Roane County, TN. Transect line (red) at Emory River mile (ERM) 6.0 (reference location) and ERM 1.0 (impacted location).



Figure 13. Benthic invertebrate community composition at Emory River mile (ERM) 6.0 and ERM 1.0 during 5-year study (2009-2012), Emory River, Watts Bar Reservoir, Roane County, TN. Asterisk (*) includes mites, mussels, snails, stoneflies (Plecoptera) and water beetles in ERM 6.0 and alderflies (Megaloptera), leeches (Hirundinea), mites, mussels, nonparasitic flatworms (planarians), snails, and water beetles in ERM 1.0.



Figure 14. Distribution of feeding guilds at (A) Emory River mile (ERM) 6.0 (reference location) and (B) ERM 1.0 (impacted location) during all periods of study, Emory River Watts Bar Reservoir, Roane County, TN. Asterisk (*) includes scrapers and parasites for both locations.



Figure 15. Distribution of organism habit at (A) Emory River mile (ERM) 6.0 (reference location) and (B) ERM 1.0 (impacted location) during all periods of study, Emory River, Watts Bar Reservoir, Roane County, TN.







Figure 17. Average (\pm SE) total number of distinct taxa (richness) collected per sample at Emory River mile (ERM) 6.0 (reference location) and ERM 1.0 (impacted location), Emory River, Watts Bar Reservoir, Roane County, TN. Bars with different letters differ significantly (p<0.05).



Figure 18. Average (\pm SE) Shannon Diversity collected per sample at Emory River mile (ERM) 6.0 (reference location) and ERM 1.0 (impacted location), Emory River, Watts Bar Reservoir, Roane County, TN. Bars with different letters differ significantly (p<0.05).



Figure 19. Average (\pm SE) total number of Ephemeroptera, Plecoptera, and Tricoptera (EPT) taxa collected per sample at Emory River mile (ERM) 6.0 (reference location) and ERM 1.0 (impacted location), Emory River, Watts Bar Reservoir, Roane County, TN. No significant differences (p<0.05) were noted between impacted and reference location.



Figure 20. Average (± SE) North Carolina Biotic Index for tolerance of taxa collected per sample at Emory River mile (ERM) 6.0 (reference location) and ERM 1.0 (impacted location), Emory River, Watts Bar Reservoir, Roane County, TN. Bars with different letters differ significantly (p<0.05).



Figure 21. Total abundance of chironomid and Tubificidae taxa collected per period at Emory River mile (ERM) 6.0 (reference location) and ERM 1.0 (impacted location), Emory River, Watts Bar Reservoir, Roane County, TN. Asterisk (*) indicates a significant difference between impacted and reference location (p<0.05).



Figure 22. Total abundance of non-chironomid and oligochaete taxa collected per period at Emory River mile (ERM) 6.0 (reference location) and ERM 1.0 (impacted location), Emory River, Watts Bar Reservoir, Roane County, TN. Asterisk (*) indicates a significant difference between impacted and reference location (p<0.05).



Figure 23. Total number of non-chironomid and oligochaete distinct taxa (richness) collected per period at Emory River mile (ERM) 6.0 (reference location) and ERM 1.0 (impacted location), Emory River, Watts Bar Reservoir, Roane County, TN. No significant difference noted between impacted and reference location (p>0.05).

VITA

Suzanne Jane Young was born in Glenville, Pennsylvania. After finishing high school in 2001, Suzanne entered Allegheny College in Meadville, Pennsylvania. She received a Bachelor of Science in Biology and Environmental Politics in May 2005. She began employment as an ecologist at ARCADIS U.S., Inc. in June 2005, where she continues to work today. In January 2011, Suzanne entered the Graduate School of the University of Tennessee, Knoxville. She received a Master of Science in Entomology and Plant Pathology in August 2015.