Infrastructure · Water · Environment · Buildings

Imagine the result

Updated Data Analysis and Temporal Trend Evaluations in Biota: 2009 – 2014

Tennessee Valley Authority
Ash Recovery Project
Kingston, Tennessee

October 2015

150930-TNTVA-RPT-255

Updated Data Analysis and Temporal Trend Evaluations in Biota: 2009 – 2014

Tennessee Valley Authority
Kingston Ash Release

Amber Stojak
Staff Environmental Scientist

Suzanne Walls
Senior Ecologist/Project Manager

Prepared for:
Tennessee Valley Authority

Prepared by:
Arcadis U.S., Inc.
114 Lovell Road
Suite 202
Knoxville
Tennessee 37934
Tel 865 675 6700
Fax 865 675 6712

Our Ref.:
TNTVAKIP.LTM5

Date:
October 19, 2015

Tables

Figures

Appendices

Acronyms and Abbreviations

BERA	Baseline Ecological Risk Assessment
COC	constituent of concern
CRM	Clinch River Mile
DQOs	data quality objectives
dw	dry weight
EDDs	electronic data deliverables
ERM	Emory River Mile
KIF	Kingston Fossil Plant
LTM	Long Term Monitoring
MDL	method detection limits
mg/kg	milligram per kilogram
ORNL	Oak Ridge National Laboratory
PLM	polarized light microscopy
QAPP	Quality Assurance Project Plan
QC	quality control
SAP	Sampling and Analysis Plan
site	Tennessee Valley Authority Kingston Fossil Plant Release Site in Roane County, Tennessee
TRM	Tennessee River Mile
TVA	Tennessee Valley Authority
USEPA	U.S. Environmental Protection Agency

1. Introduction

In August 2012, a Baseline Ecological Risk Assessment (BERA) (ARCADIS 2012a) was approved by the U.S. Environmental Protection Agency (USEPA). The BERA evaluated the potential ecological effects on biota from ash residuals in the river system at the Tennessee Valley Authority (TVA) Kingston Fossil Plant (KIF) Release Site, in Roane County, Tennessee (the site, Figure 1). The BERA focused primarily on data collected post-dredging. The BERA was developed in support of the *Kingston Ash Recovery Project, Non-Time Critical Removal Action, River System Engineering Evaluation/Cost Analysis* (TVA 2012), which evaluated alternatives for restoration of the river system impacted by the December 22, 2008 ash release.

In May 2013, a *Long-Term Monitoring Sampling and Analysis Plan* (LTM SAP) (Jacobs 2013) was approved by the USEPA for the TVA KIF Release Site. The LTM SAP described the data quality objectives (DQOs), sampling design, and sampling procedures for data collections necessary to assess the effectiveness of the selected removal action of monitored natural attenuation. This LTM SAP began in 2014, and the changes to sampling frequency or focused study area designs are discussed as part of each of the subsequent sections.

Monitoring for some ecological receptors is ongoing. The purpose of this report is to quantitatively assess the most recent dataset (i.e., 2014), and when possible, to evaluate temporal trends in constituent concentrations from 2009 through 2014. Consistent with the results of the BERA and the LTM SAP, selenium and arsenic were identified as constituents of concern (COCs) and are the focus of discussions for each receptor. In addition, percent ash is also considered a COC in sediment. This report evaluates data from 2009 through 2014 for the following biota:

- Fish,
- Benthic invertebrates, and
- Tree swallows.

The overall conclusions presented here do not change the risk management recommendations provided in the BERA (ARCADIS 2012a) or the recommendations and anticipated long-term monitoring requirements established in the LTM SAP (Jacobs 2013). The data included in this report were reported by analytical laboratories and validated via a quality assurance/quality control review as specified in the *Quality Assurance Project Plan for the Tennessee Valley Authority Kingston Ash Recovery Project, Revision 1* (QAPP) (TVA 2010). A summary of the data validation process and data quality results is presented in the final chapter of this report.

2. Fish

Many species of fish were selected for evaluation in the BERA because they represent various feeding guilds and are ubiquitous in the Emory, Clinch, and Tennessee River locations near the site. Fish may be exposed to ash-related constituents through their gills, ingestion of sediment and water, consumption of aquatic prey, and maternally transferring constituents to eggs. Exposure to ash-related constituents may lead to bioaccumulation over time which may then affect the health of the fish community.

The main study objectives were to 1) compare community metric results among locations and across years; 2) evaluate fish reproductive condition among locations and years; 3) compare concentrations of metals and metalloids in fish tissues; 4) evaluate fish health condition among locations and years; and 5) relate concentrations measured at the study sites to reference area concentrations and literature-derived effects values, when available.

2.1 Spring Sport Fish

Spring sport fish have been studied in the TVA reservoirs and include five locations within the Watts Bar Reservoir. These consist of Emory River mile (ERM) 2.5 and Clinch River mile (CRM) 2.5 and three locations on the Tennessee River (Caney Creek, Blue Springs, and Watts Bar Forebay). A summary of collection and processing methodology can be found in *Evaluation of Spring Sport Fish Survey Results for Watts Bar Reservoir, 2002 – 2011* (Baker 2011a) and a detailed summary of results is presented in Appendix A.

Spring sport fish surveys were conducted at the CRM 2.5 location from 2002 to 2005 and continued from 2009 to 2013. In 2013, these annual surveys were changed to a biennial schedule as part of the LTM SAP. They were sampled most recently in 2015. The ERM 2.5 location was surveyed annually from 2009 until 2013, and then also moved to a biennial schedule and was sampled most recently in 2015. The three Tennessee River locations have been sampled annually from 2002 to 2015. These are considered far-field locations. While results for these three locations are included in Appendix A, they are not discussed below as they are not included in the LTM SAP.

The 2015 catch rates for black bass were highest to date for ERM 2.5 and CRM 2.5. These higher catch rates were largely dominated by an increase in the number of age one largemouth bass, indicating good recruitment of young-of-year fish. Other survey metrics were also similar between 2015 and previous years, such as size class, prevalence of anomalies, and prevalence of parasites. There were some differences in relative weights among sites; however, similar differences have been noted historical both pre- and post-spill. Overall, the results of the 2015 spring sport fish sampling were similar to previous years and do not indicate that residual ash from the 2008 TVA KIF Release Site is posing a long-term risk to sport fish in the Emory and Clinch Rivers.

2.2 Fish Reproduction

Oak Ridge National Laboratory (ORNL) conducted fish reproductive studies that focused on assessing the health and condition of fish ovaries. A detailed description of the sampling locations and collection methods can be found in *Evaluating the Effects of the Kingston Fly Ash Release on Fish Reproduction: Spring 2009 – 2010 Studies* (Greeley et al. 2012) and *2015 ORNL Progress Report for the TVA-Kingston Project* (Appendix B).

Fish reproduction has been evaluated at TVA KIF since 2009. The ovary was chosen for evaluation of fish reproduction because it provides a route for maternal transfer of metals and metalloids to the developing eggs. Evaluations of fish reproduction were not conducted during the 2014 season as the LTM SAP moved to biennial fish evaluations beginning in 2013. While fish were collected from ERM 1.0 during the spring of 2014 for bioaccumulation studies, these samples were archived for potential future use in reproductive analyses and were not evaluated for reproductive measures during the 2014 calendar year.

In 2014, ORNL conducted additional statistical analyses on fish reproductive data collected from 2009 through 2013. The results of these evaluations were cited in a recent ORNL paper submitted for review to the journal *Ecotoxicology* and in a recently published ORNL paper submitted to the journal *Bulletin of Environmental Contamination and Toxicology* (Greeley et al. 2014). Results from these field and laboratory-based studies indicate that the residual ash from the 2008 TVA KIF Release Site is unlikely to pose significant long-term risks to the reproductive success of fish populations in the Watts Bar Reservoir.

2.3 Fish Bioaccumulation

Fish bioaccumulation studies have been ongoing at the TVA KIF Release Site since 2008. A detailed description of the sampling locations and collection methods can be found in *Fish Bioaccumulation Studies Associated with the Kingston Fly Ash Spill, Spring 2009 – Fall 2010* (Adams et al. 2012), *Trace Element Concentrations in Fish: 2010* (ARCADIS 2012b), and in *2015 ORNL Progress Report for the TVA-Kingston Project (Appendix B)*.

Bioaccumulation of metals and metalloids were measured in fillets of largemouth bass, bluegill sunfish, and redear sunfish. Spring sampling was conducted in April and May of 2014. In the years 2009 through 2013, fish were collected from three upstream references (ERM 8.0, Little Emory River mile 2.0, and CRM 8.0), and three impacted locations (ERM 3.0, ERM 0.9, and CRM 1.5). In 2014, fish were only collected from ERM 1.0, as required in the LTM SAP. Fish fillets were collected by TVA and ORNL staffs and/or their contractor, and sent to Pace Analytical Services, Inc., for analysis of metals and metalloids (Appendix B). Spatial and temporal trends for selenium, arsenic, and mercury concentrations in fish fillets were evaluated by ORNL, with details provided in Appendix B, and are summarized below.

Historically, selenium concentrations in all three species' fillets have been higher in individuals collected from impacted locations compared to the reference locations. In 2014, redear sunfish continued to show the highest selenium concentrations compared to other species, which most likely reflects differences in diet. Mean concentrations of selenium in fillets in redear and bluegill sunfish decreased significantly in 2014 compared to previous years. Samples were not collected from reference locations in 2014; however, concentrations at ERM 1.0 in 2014 were comparable to reference concentrations from previous years. Mean selenium concentrations were lowest in largemouth bass compared with the other two fish species. Overall, concentrations of selenium in all species remain below the USEPA proposed ambient water quality criterion of 7.91 milligram per kilogram (mg/kg) (whole-body, dry weight [dw]) in all years of study.

Arsenic and mercury concentrations in fish fillets were also compared to regulatory guidelines and across all years of study. In general, arsenic concentrations remained below levels of concern, with no statistically significant changes from 2013 to 2014. Mercury is a legacy constituent not related to the ash release, but is the focus of fish consumption advisories for the Emory River. Two largemouth samples collected in 2014 had individual mercury concentrations (0.34 and 0.40 mg/kg) above the USEPA human health fish consumption criterion for methyl mercury in fish fillets (0.3 mg/kg wet weight). While the reference site was not sampled in 2014; these concentrations are comparable to results from previous years where some samples also exceeded the USEPA human health fish consumption criterion. Mean concentrations of mercury in 2014 were below the mercury consumption criteria for all three species.

2.4 Fish Health

Fish health studies have been conducted at the TVA KIF Release Site since 2009. A detailed description of the sampling locations and collection methods can be found in *Fish Health Studies Associated with the Kingston Fly Ash Spill, Spring 2009 – Fall 2010* (Adams and Fortner 2012) and in *2015 ORNL Progress Report for the TVA-Kingston Project* (Appendix B).

In 2014, the fish health evaluation was limited to visual inspection of three fish species (bluegill sunfish, redear sunfish, and largemouth bass) collected from ERM 1.0 in conjunction with the spring fish bioaccumulation studies. At least six fish of each of the fish species were processed at ORNL and evaluated for basic health conditions. Blood analyses and a full suite of health parameters were not evaluated in 2014, as specified in the LTM SAP. Based on visual inspection, there was no evidence of unusual internal or external anomalies.

Also in 2014, a more in-depth statistical temporal analysis was conducted on fish health (Appendix B). This evaluation linked measured health metrics with bioaccumulation and reproductive health responses, as well as liver and gill histopathology data. Overall, ORNL's findings suggested a lack of consistent evidence of compromised health correlated with the ash release. While a trend in gill and liver pathology increased immediately post-dredging, this trend then decreased across all sites and species after 2011. Analysis of covariance was used to determine if site, year, arsenic, mercury, and/or selenium had any effect on the composite liver pathology metric. Though individual metals/metalloids correlated with the liver pathology metric, there were no significant differences among sites and years.

3. Benthic Invertebrates

Benthic invertebrates are found living within or on top of sediments in the Emory, Clinch, and Tennessee Rivers. Benthic invertebrates in these rivers consist of mostly oligochaetes (aquatic worms), chironomids (larval midges), burrowing mayfly nymphs, and also crustaceans (crayfish and amphipods), bivalves (mussels and clams), snails, larval flies, leeches, and mites. Because of their close association with the sediments and water, benthic invertebrates have the potential for bioaccumulation of metals and metalloids. They may also transfer these constituents to fish and wildlife consumers. Snails and mayflies serve as a useful receptor in order to understand exposure and potential effects of these constituents on the benthic invertebrate community.

The main study objectives in 2014 were to assess impacts to the benthic community by 1) comparing community metric results among sites and across years; 2) comparing tissue concentrations of metals and metalloids in mayfly nymphs and mayfly adults for evaluating differences among sites and years; and finally 3) relating concentrations measured at the study sites to reference area concentrations and literature derived effects values, when available. A brief discussion of each objective is presented in the subsections below.

3.1 Benthic Invertebrate Community

Benthic invertebrate community evaluations in November and December 2014 were conducted on the Emory, Clinch, and Tennessee Rivers, similar to previous years. A detailed description of the sampling locations and collection methods can be found in *Evaluation of Benthic Macroinvertebrate Communities in the Vicinity of TVA's Kingston Fossil Plant, 2009-2010* (Baker 2011b) and also in Appendix C of this report.

In 2014, two transect locations at ERM 1.0 and ERM 0.7 were monitored for population abundance and diversity. Ten grab samples were collected from each transect and benthic invertebrates within each sample were identified to the lowest possible taxon. The total number of taxa were tallied and used to generate benthic invertebrate community metrics in order to assess the overall health of the benthic invertebrate community. Population density, taxa richness, number of organisms, number of taxa, percent oligochaetes and chironomids, and other metrics were used to assess the benthic invertebrate community. Details on how these metrics are calculated are presented in *Evaluation of Benthic Macroinvertebrate Communities in the Vicinity of TVA's Kingston Fossil Plant, 2009-2010* (Baker 2011b). At each sample location, water depth was also recorded along with a physical description of the sediment in the sample in order to estimate: percent ash, grain size, and substrate type. In addition to the benthic invertebrate community data collections, sediment chemistry data (percent ash, metals, and the percent sand, silt, clay, or gravel) were also collected from the Emory River. The purpose of the co-located data collections was to better interpret the various factors potentially influencing the benthic invertebrate community. Sediment quality associated with ERM 1.0 and ERM 0.7 is discussed in Section 3.3.

Variations in November 2014 Emory River benthic invertebrate community abundance, composition, and diversity among sites were consistent with the variation seen in previous years. These differences were related more to a temporal variation when comparing across the 6 years, or are a reflection of spatial heterogeneity when comparing among sites. While samples were not collected from reference sites in 2014, the differences seen at ERM 1.0 and ERM 0.7 were similar to those observed in the previous years of data collected from the reference sites. These differences are a reflection of natural variability in the benthic invertebrate community found in a large river system. Overall, the results of the 2014 benthic invertebrate community surveys indicate that residual ash from the 2008 TVA KIF release is not causing distinguishable adverse effects on the benthic invertebrate population in the Emory River.

3.2 Benthic Invertebrate Bioaccumulation

A detailed description of the collection methods used for benthic invertebrate bioaccumulation can be found in *Evaluation of Invertebrate Bioaccumulation of Fly Ash Contaminants in the Emory, Clinch, and Tennessee Rivers, 2009-2010* (Smith 2012) and in *2015 ORNL Progress Report for the TVA-Kingston Project* (Appendix B).

In 2014, mayfly adults and nymphs (*Hexagenia bilineata*) were collected from ERM 1.0 and mayfly adults were collected opportunistically as close to ERM 1.0 as possible. Mayfly nymphs were separated into depurated and non-depurated samples, and adult mayflies were separated by sex and life stage. All samples were analyzed for arsenic and selenium.

All mayfly nymph (depurated and non-depurated) and adult selenium concentrations were below the LTM remedial tissue monitoring endpoints (7 mg/kg dw). Concentrations in depurated mayfly nymphs were similar between 2014 and the previous year of data. Non-depurated mayfly nymph concentrations were lower compared to previous years. Selenium in mayfly nymph tissue at ERM 1.0 continued to be approximately 2-times higher than the mean for the four reference sites from 2009 through 2013. Selenium concentrations in mayfly adults were lower in 2014, but still higher than long-term mean concentrations found at the four reference sites.

Similarly, all mayfly nymph (depurated and non-depurated) arsenic concentrations were below the LTM remedial tissue monitoring endpoints (34 to 83 mg/kg). Arsenic in depurated nymphs was approximately 25 to 35% lower in 2014 than in 2013; however, these concentrations were still twice as high as the mean concentration for the reference sites between 2009 and 2013.

3.3 Sediment Quality

Sediment sampling activities were conducted in November 2014 at ERM 1.0 and ERM 0.7 using two types of sediment sampling, as defined in the LTM SAP. The first sampling type included taking a 3-point composite of sediment from the left descending bank, center of channel, and right descending bank. These samples were analyzed for metals (selenium and arsenic), ash content, and grain size. The second sampling type included collecting co-located samples with the benthic invertebrate community surveys at ten discrete sample points from each transect. All sediment samples were collected using a decontaminated WILDCO Ponar Dredge Sampler. Composite samples were placed in high-density polyethylene 3-gallon tubs and were transported to a field laboratory where the overlay water was decanted prior to homogenization. Following homogenization, samples were split into labeled sterile containers for arsenic, selenium, polarized-light microscopy (PLM) analysis of ash content, and grain size. Similarly, co-located sediment samples were also homogenized, but were only analyzed for ash content. Once sub-samples were partitioned to the appropriate containers, the samples were custody sealed and placed on ice. Samples were shipped to their appropriate laboratories for analysis (RJ Lee for PLM analysis and TestAmerica for metals and grain size).

The LTM SAP identified three main COCs at the site with corresponding remedial goals for sediment. These include ash content or percent ash, arsenic, and selenium. The remedial goal for percent ash is 50% or less. The remedial goal for arsenic is a range of 29 to 41 mg/kg, and the remedial goal for selenium is a range of 3.0 to 3.2 mg/kg.

In 2014 composite sediment samples, percent ash range from 6% to 52% at ERM 1.0 and 4% to 53% at ERM 0.7. While one composite sample from each location had an ash content greater than the remedial goal of 50%, the mean percent ash values are less than the remedial goal of 50% at each location (Table 1). Percent ash results in co-located sediment samples range from 1% to 40% at ERM 1.0 and 2% to 42% at ERM 0.7. All results are below the remedial goal of 50% (Table 2).

Selenium results for composite sediment samples at ERM 1.0 range from 1.15 mg/kg to 3.96 mg/kg. While two of these concentrations are above the remedial goal range of selenium in sediment, the mean selenium concentration (2.79 mg/kg) is below the remedial goal range (Table 1). Selenium concentrations at ERM 0.7 range from 2.73 mg/kg to 3.44 mg/kg. One of these concentrations is above the selenium remedial goal; however, the mean concentration (3.07 mg/kg) is within the remedial goal range (Table 1).

Arsenic results for composite samples at ERM 1.0 range from 9.61 mg/kg to 30.4 mg/kg, with a mean concentration of 20.1 mg/kg. These concentrations are all within or below the arsenic remedial goal range. Arsenic results for composite samples at ERM 0.7 range from 12 mg/kg to 20.7 mg/kg, with a mean concentration of 17.5 mg/kg. These values are all below the remedial goal range for arsenic.

Grain size analysis was also conducted on composite sediment samples at ERM 1.0 and ERM 0.7 (Table 1). This analysis indicates that, similar to previous years of study, the sediment at ERM 1.0 is dominated by sand (56%) and silt (29%), and sediment at ERM 0.7 is dominated by silt (61%) and sand (26%).

4. Tree Swallows

Tree swallows (*Tachycineta bicolor*) were selected as a representative aerial-feeding insectivorous bird species for the site. Tree swallows are a breeding migratory resident in Tennessee (Nicholson 1997; Robinson 1990), and forage 100 to 200 meters around their nest during the breeding season. They commonly prey on a variety of insects, and when nest boxes are placed along aquatic areas, they feed primarily on emergent aquatic insects (U.S. Geological Survey 2003; Blancher and McNicol 1991; Quinney and Ankney 1985). As a result, tree swallow tissue residues often reflect the local sediment contamination for those chemicals that transfer into the aquatic emergent insects (McCarty and Winkler 1999; Froese et al. 1998).

In 2014, tree swallow colonies were erected at two locations along the Emory River (ERM 3.0 and ERM 1.4), and at one reference location (Tennessee River mile [TRM] 572.0). Boxes were monitored daily at ERM 1.4 and TRM 572.0 from April through July. Boxes at ERM 3.0 were not monitored due to the construction activity occurring in this area during the breeding season. A detailed description of the collection methods can be found in *Trace Element Concentrations and Productivity in Tree Swallows: 2009-2010* (ARCADIS 2012c) and in Appendix D.

The main study objectives were to 1) determine the extent of maternal transfer of metals and metalloids to the eggs between locations and across years; 2) evaluate tree swallow reproductive success among locations and years; and 3) assess impacts to tree swallows by comparing concentrations measured at the study sites with literature-derived effects values, when available.

4.1 Tree Swallow Reproduction

The total number of eggs (clutch size), the number of eggs that hatched (hatching success), the number of young that survived to day 15 (fledgling success), and the number of females fledglings produced per nesting female (fecundity) were recorded in 2014 at ERM 1.4 and TRM 572.0 colonies. In addition, egg mass and volume were recorded, as well as morphological measures (egg length and width).

Similar to 2013, the 2014 clutch size, hatching success, fledgling success, and fecundity were not statistically significantly different between colonies or years (Appendix D). Additionally, no differences were observed for the egg volume or egg mass between colonies or years.

4.2 Tree Swallow Bioaccumulation

In 2014, tree swallow eggs were collected to evaluate exposure of tree swallows to ash-related COCs. A total of 31 eggs were collected from ERM 1.4 and 32 eggs were collected from TRM 572.0. Following collection, eggs were frozen and shipped to Pace Analytical for trace element analysis. Arsenic was detected in all but two samples; however, concentrations were not significantly different between locations and are not discussed further.

Selenium results showed a statistically significant difference between the colonies, with a slightly higher average concentration at ERM 1.4 (2.21 mg/kg dw) compared to TRM 572.0 (1.80 mg/kg dw). When compared to previous years of data, 2014 concentrations at ERM 1.4 were lower than 2013 (3.65 mg/kg dw in 2013; 2.21 mg/kg dw in 2014) and were within the range of selenium concentrations in eggs collected from other reference sites between 2009 and 2012 (Appendix D). Literature studies of selenium in eggs of other species have been reviewed and suggest threshold effects (EC_{10}) concentrations ranging from 7.7 to 60 mg/kg dw in various species of avian eggs (Janz et al. 2010). Similar to previous years, selenium concentrations in eggs collected from ERM 1.4 in 2014 are well below the most conservative of these literature values, indicating that it is unlikely that selenium is causing adverse effects on the tree swallow population.

5. Data Quality Assurance/Quality Control Results

This section focuses on the evaluation of data quality and usability.

5.1 Analytical Data Review

TVA's contracted laboratories were required to submit three types of deliverables: a limited (Level 1) data package containing sample results and batch quality control (QC) sample results; a fully-documented (Level 4) data package including raw data for all analyses; and electronic data deliverables (EDDs) for storage in TVA's EarthSoft EQuIS® database.

EDDs were subjected to completeness and correctness testing during loading to TVA's EQuIS database; once loaded to the EQuIS database, the data were subjected to verification. As defined in the TVA-KIF-QAPP (TVA 2010), data verification involved comparison of the data loaded in the EQuIS database to the results reported in the Level 1 data package. In addition, data verification included review of the batch QC summary forms for compliance with the applicable methods and for data usability with respect to the project DQOs and the TVA-KIF-QAPP.

Following receipt of the Level 4 data package, data were subjected to validation. As defined in the TVA-KIF-QAPP, data validation included review of raw data and associated QC summary forms for compliance with the applicable methods and for data usability with respect to the appropriate guidance documents. As stated in the QAPP: “Initially, 100% of the chemical analysis data will be reported in full documentation data packages for independent data validation. Depending on the nature and frequency of issues identified during data validation, the percentage of data undergoing full data validation may be reduced to a lesser percentage (such as 20%) or data verification may be substituted. The reduction in full data validation may be matrix specific, laboratory specific, or analyte specific. If after the percentage of full data validation has decreased, a trend in frequency of reporting issues, method non-compliances, or data usability issues is identified, data validation will be conducted for specific data points or the percentage of full data validation percentage may be increased until the issues have been minimized to their initial frequency.” Data validation expands upon the completeness, correctness, and usability assessment performed during verification to include evaluation of instrumental QC analyses, review of sample preparation information, and recalculation of reported results from raw data. A summary of the data review efforts are presented in Table 3.

5.2 Data Quality Summary

Data validation was performed based on the sample results, summary QC data, and raw data provided by the laboratory. Data validation includes a review of the following QC measures (where applicable):

- Sample condition upon laboratory receipt;
- Initial calibration linearity;
- Blank analysis results greater than the method detection limits (MDL);
- Sample preparation and holding times;
- Initial calibration verification/continuing calibration verification standard recoveries;
- MDLs and linear ranges;
- Internal standard recoveries;
- Percent moisture;
- Matrix spike/matrix spike duplicate;
- Laboratory and field duplicate precision;
- Quantitation of positive results;
- Laboratory control sample/laboratory control sample duplicate recoveries and precision;
- Analytical sequence;
- Reporting limit standard recoveries (metals only);
- Serial dilutions (metals only);
- Post-digestion spike/post-digestion spike recoveries and precision (metals only);
- Internal standard recoveries;

- Inductively coupled plasma interference check standard results (metals only);
- Quantitation of positive results;
- MDL verification standards (metals only); and
- Standard reference material recoveries (metals only).

The data met the DQOs defined for this task and were acceptable for use for each of the receptors. Table 4 summarizes the data quality for each receptor based on the review performed and as compared to the data quality measures identified in the TVA-KIF-QAPP.

6. Summary

Overall, the results of the 2014 biota data collections are consistent with the conclusions and risk management recommendations presented in the BERA (Arcadis 2012a). Based on these results, there are no anticipated or recommended changes to the long-term monitoring requirements established in the LTM SAP (Jacobs 2013) for upcoming years at this time.

7. References

Adams, S.M., C.C. Brandt, and A.M. Fortner. 2012. Fish Bioaccumulation Studies Associated with the Kingston Fly Ash Spill, Spring 2009 – Fall 2010. May. (BERA Appendix K)

Adams, S.M. and A.M. Fortner. 2012. Fish Health Studies Associated with the Kingston Fly Ash Spill, Spring 2009 – Fall 2010. May. (BERA Appendix I)

ARCADIS. 2012a. Baseline Ecological Risk Assessment (EPA-AO-050). Revision 1. 21 May 2012.

ARCADIS. 2012b. Trace Element Concentrations in Fish: 2010. May. (BERA Appendix L)

ARCADIS. 2012c. Trace Element Concentrations and Productivity in Tree Swallows: 2009-2010. May. (BERA Appendix Z).

Baker, T.F. 2011a. Evaluation of the Spring Sport Fish Survey Results for Watts Bar Reservoir, 2002-2011. October. (BERA Appendix E).

Baker, T.F. 2011b. Evaluation of Benthic Macroinvertebrate Communities in the Vicinity of TVA's Kingston Fossil Plant, 2009 – 2010. November. (BERA Appendix M).

Blancher, P.J., and D.K. McNicol. 1991. Tree swallow diet in relation to wetland acidity. Can. J. Zool. 66:842-849.

Froese, K., D. Verbrugge, G. Ankley, G. Niemi, C. Larsen, and J. Giesy. 1998. Bioaccumulation of Polychlorinated Biphenyls from Sediments to Aquatic Insects and Tree Swallow Eggs and Nestlings in Saginaw Bay, Michigan, USA. Environmental Toxicology and Chemistry, 17(3): 484-492.

Greeley, M. S, Jr., L. R. Elmore and M. K. McCracken. 2014. Evaluating the effects of the Kingston fly ash release on fish reproduction and early life stages: Long-term exposures to fly ash in the laboratory. ORNL/TM-2013/11. Oak Ridge National Laboratory. Oak Ridge, TN. Available online at http://info.ornl.gov/sites/publications/Files/Pub40944.pdf

Greeley, M.S., S.M. Adams, and M.K. McCracken. 2012. Evaluating the Effects of the Kingston Fly Ash Release on Fish Reproduction: Spring 2009-2010 Studies. May. (BERA Appendix H)

Jacobs. 2013. Non-Time-Critical Removal Action for the River System Long-Term Monitoring Sampling and Analysis Plan (SAP). Document No. EPA-AO-059. Tennessee Valley Authority, Kingston Ash Recovery Project.

Janz, D.M., D.K. DeForest, J.L. Brooks, P.M. Chapman, G. Gilron, D. Hoff, W.A. Hopkins, D.O. McIntyre, C.A. Mebane, V.P. Palace, J.P. Skorupa, and M. Wayland. 2010. Chapter 6: Selenium toxicity in aquatic organisms. Pages 141-232 in: Chapman, P.M., W.J. Adams, M.L. Brooks, C.G. Delos, S.N. Luoma, W.A. Maher, H.M. Ohlendorf, T.S. Presser, and D.P. Shaw (eds.), Ecological Assessment of Selenium in the Aquatic Environment. Pensacola, Florida, USA: CRC Press and Society of Environmental Toxicology and Chemistry.

McCarty, J.P. and D.W. Winkler. 1999. Foraging ecology and diet of tree swallows feeing selectivity of nestlings. The Cooper Ornithological Society. The Condor, 101: 246-254.

Nicholson, C. 1997. Atlas of the Breeding Birds of Tennessee. Knoxville, Tennessee: The University of Tennessee Press.

Quinney, T.E. and C.D. Ankney. 1985. Prey size selection by Tree Swallows. Auk 102: 245-250.

Robinson, J. 1990. An Annotated Checklist of the Birds of Tennessee. Knoxville, Tennessee: The University of Tennessee Press.

Smith, J.G. 2012. Evaluation of Invertebrate Bioaccumulation of Fly Ash Contaminants in the Emory, Clinch, and Tennessee Rivers, 2009 – 2010. May. (BERA Appendix Q).

TVA. 2012. Kingston Ash Recovery Project Non-Time-Critical Removal Action River System Engineering Evaluation/Cost Analysis (EE/CA). EPA-AO-051. August.

TVA. 2010. Quality Assurance Project Plan for the Tennessee Valley Authority Kingston Ash Recovery Project. Revision 1. August. TVA-KIF-QAPP. EPA-AO-014. Prepared by Environmental Standards, Inc. December 18, 2009 (USEPA approval).

U.S. Geological Survey. 2003. Tree Swallows: A Nationwide Sentinel Species for Assessing and Monitoring Aquatic Contamination. Fact Sheet FS 2004-3042.

Tables

Table 1
2014 Emory River Composite Sediment Results

Tennessee Valley Authority
Kingston, Tennessee

Parameter	Remedial Goal	ERM 0.7				ERM 1.0			
		Mean	LB	CC	RB	Mean	LB	CC	RB
Inorganics									
Arsenic, Total (mg/kg)	29 to 41	17.5	19.7	12 J	20.7	20.1	30.4	20.3 J	9.61
Selenium, Total (mg/kg)	3.0 to 3.2	3.07	3.05	2.73 J	3.44	2.79	3.27	3.96 J	1.15
Physical Properties									
Percent Ash (%)	50	35	53	4	49	25	52	18	6
Clay (%)	NA	13	12.2	17.3	9.4	8	7.6	12.4	5
Gravel (%)	NA	0	0	0	0	6	19.1	0	0
Sand (%)	NA	26	22.6	21.7	32.5	56	53.3	34.2	80.3
Sand, Coarse (%)	NA	1	0.6	2.5	1.1	5	15.4	0.7	0.2
Sand, Fine (%)	NA	23	21	17.4	29.7	46	26.6	31.9	78.3
Sand, Medium (%)	NA	2	1	1.8	1.7	5	11.3	1.6	1.8
Silt (%)	NA	61	65.2	61	58.1	29	20	53.4	14.8

Acronyms and Abbreviations:
CC = Center of channel
ERM = Emory River mile
LB = Left descending bank
NA = Not applicable
RB = Right descending bank

Qualifiers:
J = estimated value

Table 2
2014 Emory River Co-Located Sediment Results

Tennessee Valley Authority
Kingston, Tennessee

Parameter	Remedial Goal	Location	Minimum	Maximum	Mean	Drop 1	Drop 2	Drop 3	Drop 4	Drop 5	Drop 6	Drop 7	Drop 8	Drop 9	Drop 10
Percent Ash (%)	50	ERM 0.7	2	42	18	42	34	6	2	8	8	16	15	33	17
		ERM 1.0	1	40	20	1	37	22	20	3	3	18	14	40	40

Acronyms and Abbreviations:
ERM = Emory River mile

Table 3
2014 Analytical Data Review

Tennessee Valley Authority
Kingston, Tennessee

Matrix	Number of COCs	Number of Samples by Matrix	Number of Equipment Blanks by Lab	Number of Analytical Results	Percentage Final-Verified	Percentage Validated
Fish (fillets, livers, and ovaries)	4	52	0	780	73%	27%
Mayfly Adults (whole body)	1	10	1	130	0%	100%
Mayfly Nymphs (depurated, non-depurated)	1	10	1	130	0%	100%
Tree Swallow (egg content)	3	60	0	780	67%	33%
Sediment	5	43	1	93	100%	0%

Notes:

All biota samples were analyzed at Pace Analytical Services, Inc. Sediment samples were analyzed by RJ Lee Group and TestAmerica, Inc. (Nashville, TN; Burlington, VT; and North Canton, OH facilities).

Acronyms and Abbreviations:

COCs = chain of custody

Table 4
2014 Data Quality Review

Tennessee Valley Authority
Kingston, Tennessee

Matrix	Analytical Results (Total Count)	Acceptable (No Qualification) [a]		Acceptable (Estimated) [b]		Blank Qualified [c]		Rejected [d]	
Fish (fillets, livers, and ovaries)	780	605	*78%*	153	*20%*	22	*3%*	0	*0%*
Mayfly Adults (whole body)	130	79	*61%*	48	*37%*	3	*2%*	0	*0%*
Mayfly Nymphs (depurated, non-depurated)	130	79	*61%*	47	*36%*	4	*3%*	0	*0%*
Tree Swallow (egg content)	780	524	*67%*	200	*26%*	56	*7%*	0	*0%*
Sediment	93	88	*95%*	4	*4%*	1	*1%*	0	*0%*

Notes:

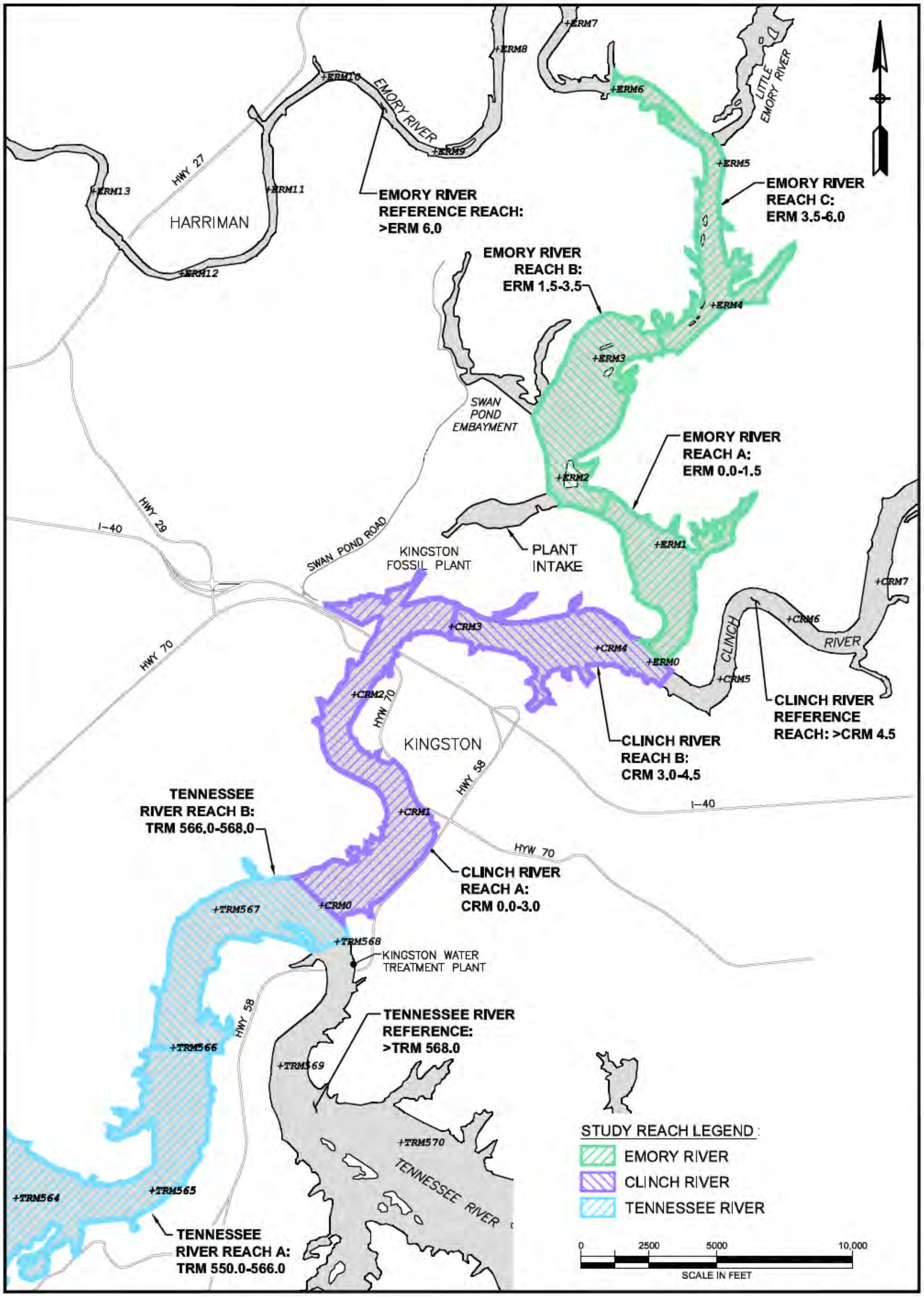
[a] Acceptable, No Qualification – Qualification of data was not warranted based on a review of the applicable quality control measures.

[b] Acceptable, Estimated – Quantitation or detection limit is approximate due to limitations or bias identified during a review of the applicable quality control measures.

[c] Blank Qualified – Result is considered "not-detected" because it was detected in an associated blank at a similar level.

[d] Rejected – Unreliable result or detection limit; analyte may or may not be present in sample.

Figures



ARCADIS

River Reaches

Updated Data Analysis and Temporal Trend Evaluations in Biota: 2009-2014

TENNESSEE VALLEY AUTHORITY

KINGSTON, TENNESSEE

FIGURE

1

ARCADIS

Appendix A

2015 TVA Spring Sport Fish Survey

Spring Sport Fish Survey

The Spring Sport Fish Survey is designed to provide information on fisheries, primarily black bass species (largemouth, smallmouth and spotted bass), in Tennessee Valley Authority (TVA) reservoirs. Surveys were conducted at five locations on Watts Bar Reservoir during April 2015, including Emory River 2.5, Clinch River 2.5, and three Tennessee River locations — Blue Springs, Caney Creek, and Watts Bar Forebay (Figure 1). Spring sport fish surveys have been conducted annually on Watts Bar Reservoir since 2002, with the frequency of sampling varying by location:

- Emory River 2.5 (2009 to 2013, 2015)
- Clinch River 2.5 (2002 to 2005, 2009 to 2013, 2015)
- Caney Creek (2002 to 2015)
- Blue Springs (2002 to 2015)
- Watts Bar Forebay (2002 to 2015)

Sampling methodology consisted of 30 minutes of continuous electrofishing at fixed stations (i.e., transects) in the littoral zones of prominent habitat types represented in the reservoir. At each location, 12 transects were sampled and all black bass collected were identified by species, weighed, measured, enumerated, and noted for general health prior to releasing. These data were used to develop key population metrics for black bass populations, including measures of catch rates, general fish condition (e.g., parasites and anomalies), length frequency, and relative weights.

Black bass catch rates in 2015 were the highest to date for Emory River 2.5 and Clinch River 2.5 and either the highest or second highest for Tennessee River locations (Figure 2, Table 1). The higher catch rates resulted largely from collecting more age-one largemouth bass than in other years (Figure 3), which suggests good recruitment of young-of-year fish from the preceding year. Historically, the black bass population in Watts Bar Reservoir has been comprised of mostly largemouth bass. In 2015, largemouth bass comprised about 95-98% of the population at each location, with the exception of Clinch River 2.5. Consistent with previous years, the black bass population at Clinch River 2.5 was comprised of about 85% largemouth and 14% smallmouth bass. Few spotted bass were collected during the 2015 survey. In 2006, there was a rather abrupt decline in the number of spotted bass collected reservoir wide and their numbers have remained low since.

The 2015 survey results also were similar among several sites with largemouth bass within the 10 to 14 inch range being under represented and the numbers of preferred stock (15 to 19 inches) slightly above average. Although site differences in relative weights existed (Figure 4), these differences remained consistent during both pre-spill and post-spill surveys and similar temporal patterns were evident across all locations for each length category. Additionally, the prevalence of anomalies among black bass in 2015 was relatively low across all locations, with percentages (0.6 to 2.4%) below the respective pre-spill averages (Figure 5, Table 1); the prevalence of anomalies was highest (5.6 to 13.9%) in 2011 for all locations. Similarly, the percentages of black bass with external parasites in 2015 were within the ranges observed during pre-spill surveys (Figure 6). The prevalence of parasites, however, was lowest during the initial surveys, increased consecutively from 2007 through 2009, and then has fluctuated during subsequent surveys.

Similar spatial and temporal patterns in catch rates, length-frequency distributions, relative weights, and the prevalence of parasites and anomalies among locations, both before and after the ash spill, continue to suggest that the patterns observed were associated with factors that extended reservoir-wide (e.g., meteorology) and/or location differences in limnological properties (e.g., primary productivity). When considered along with the results of fish bioaccumulation, health, and reproductive studies conducted by

ORNL, there is no clear evidence that the ash spill has adversely affected black bass populations in Watts Bar Reservoir.

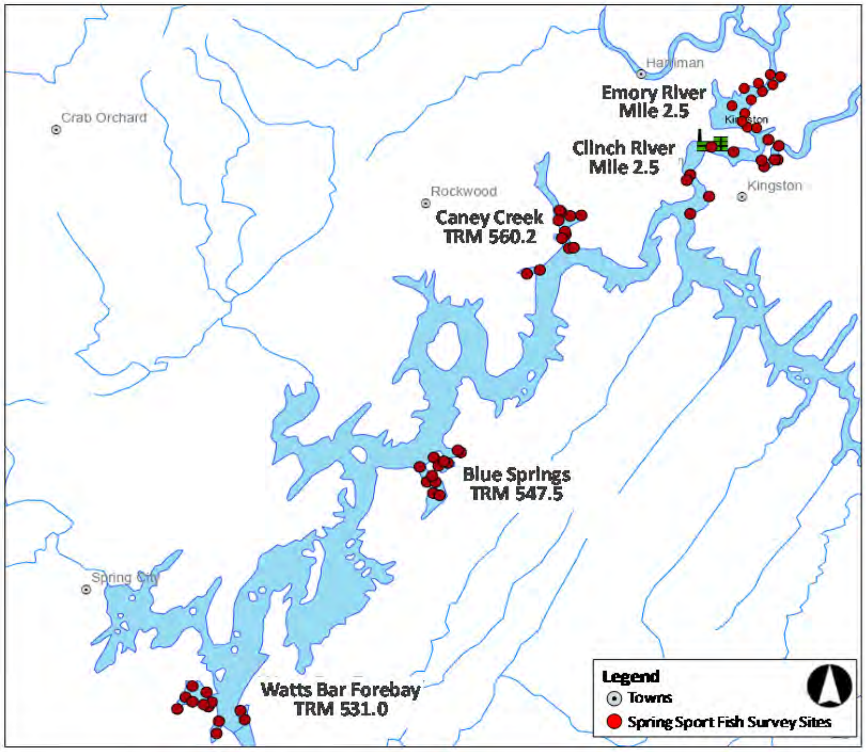


Figure 1. Spring Sport Fish Survey sampling sites on Watts Bar Reservoir.

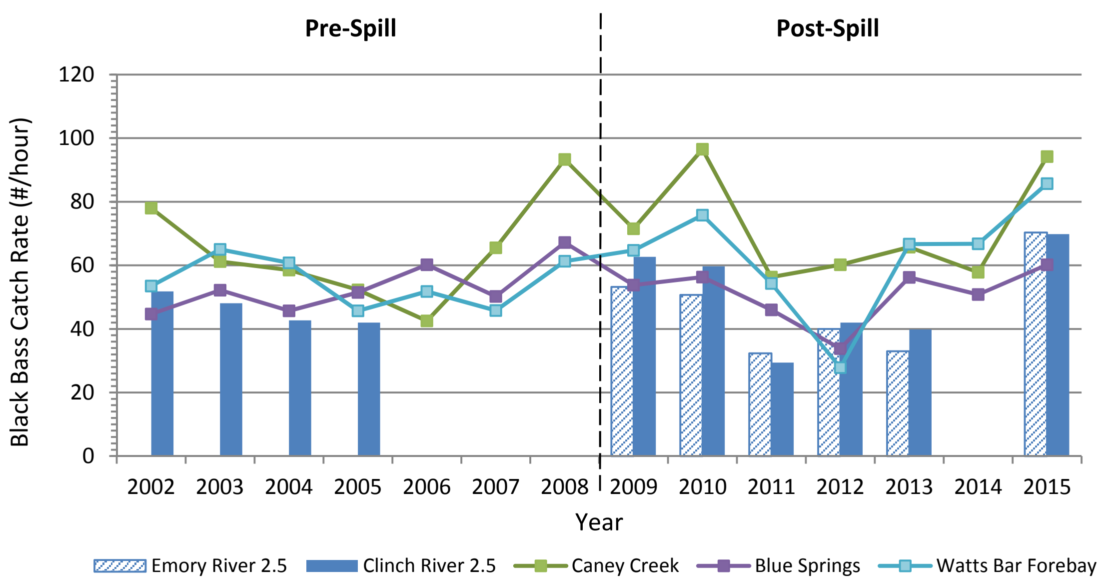
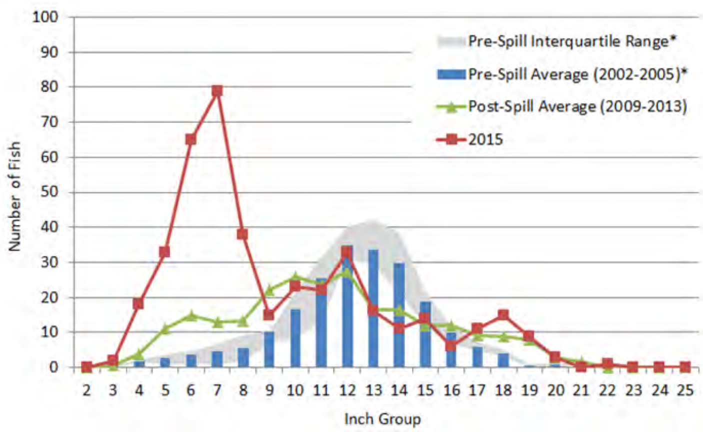
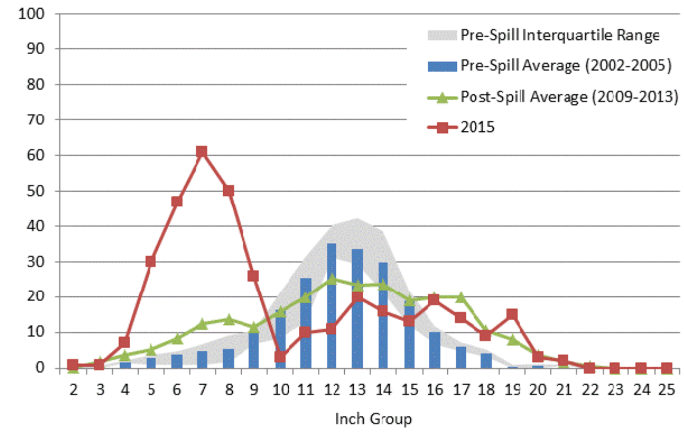


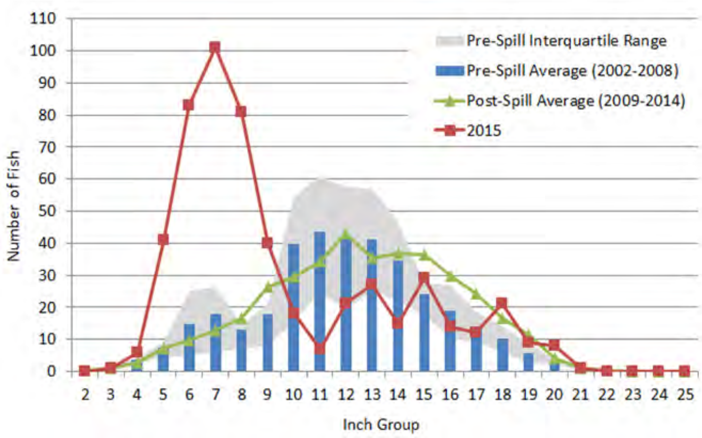
Figure 2. Black bass catch rates during Spring Sport Fish Surveys on Watts Bar Reservoir from 2002 through 2015. The frequency of sampling varied by location.



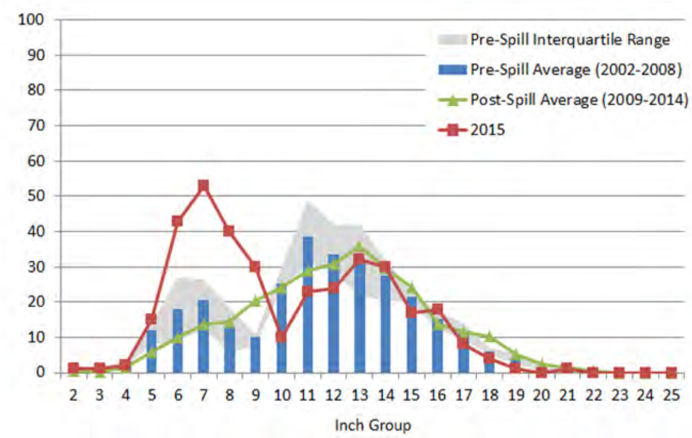
a. Emory River 2.5



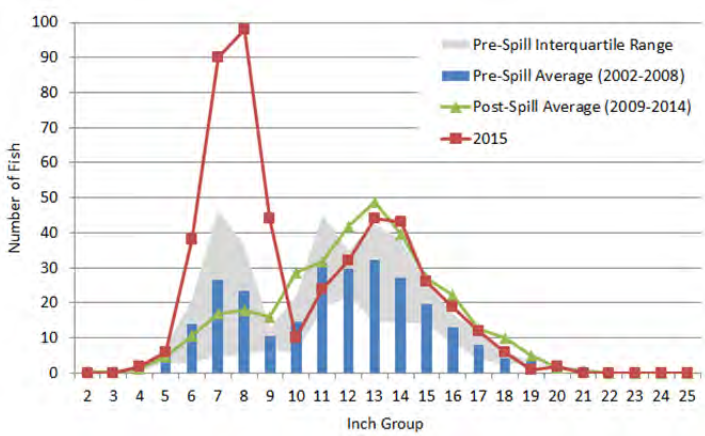
b. Clinch River 2.5



c. Caney Creek

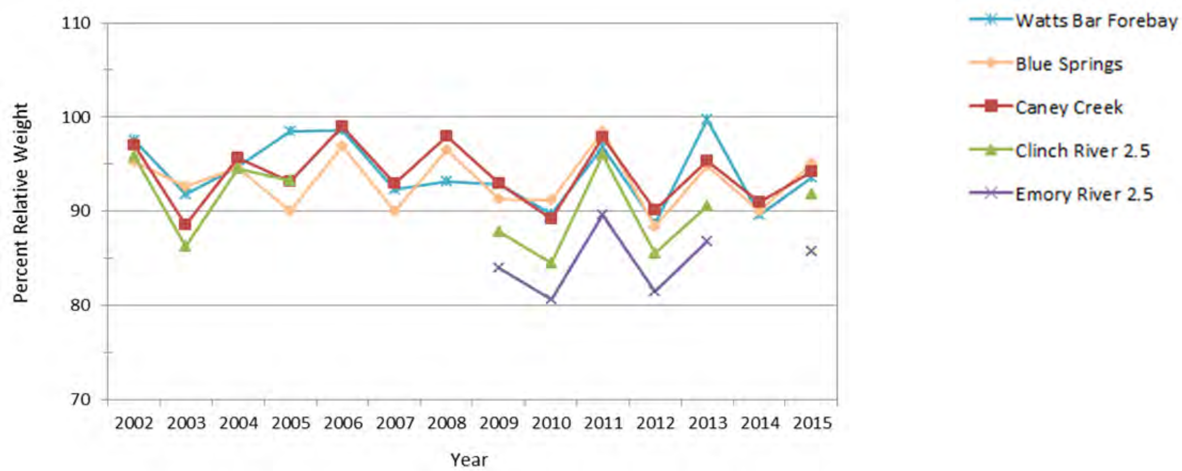


d. Blue Springs

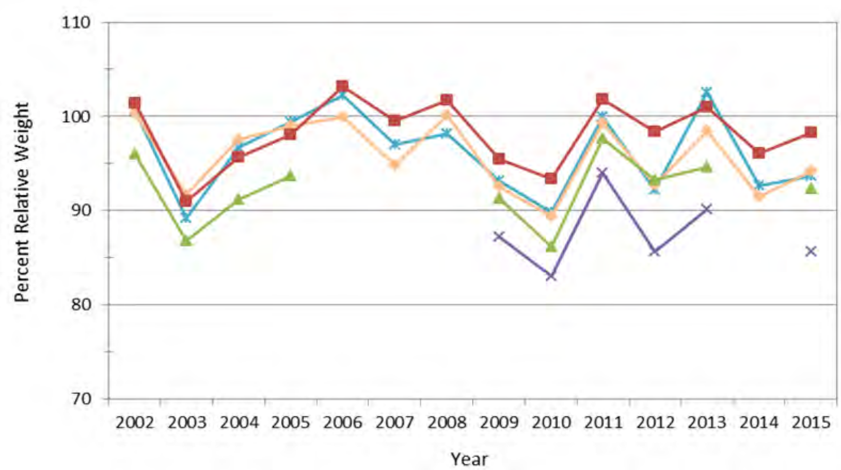


e. Watts Bar Forebay

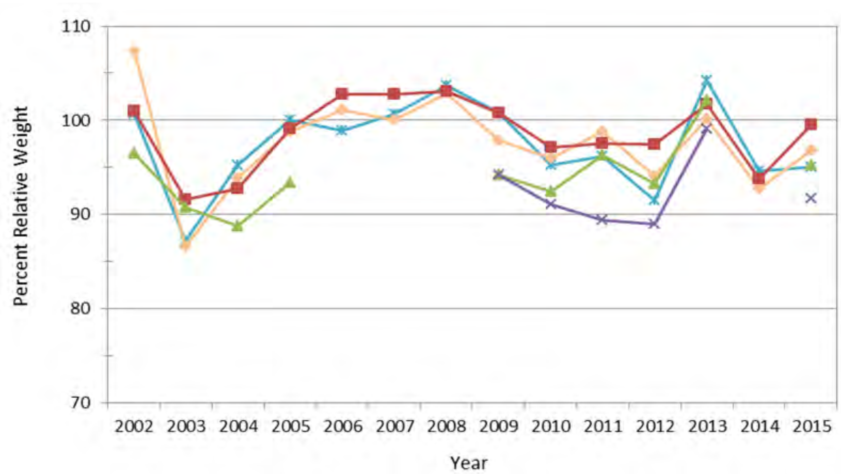
Figure 3. Comparison of length-frequency distributions of largemouth bass collected during pre- and post-spill surveys at five locations on Watts Bar Reservoir. *Surveys were not conducted at Emory River 2.5 prior to the spill; therefore, post-spill results for Emory River 2.5 (Figure 1a) are compared with Clinch River 2.5 pre-spill results (2002-2005).



a. Stock (8 to 11 inches)



b. Quality (12 to 14 inches)



c. Preferred (15 to 19 inches)

Figure 4. Relative weights for three size classes of largemouth bass: a. Stock (8 to 11 inches), b. Quality (12 to 14 inches), and c. Preferred (15 to 19 inches).

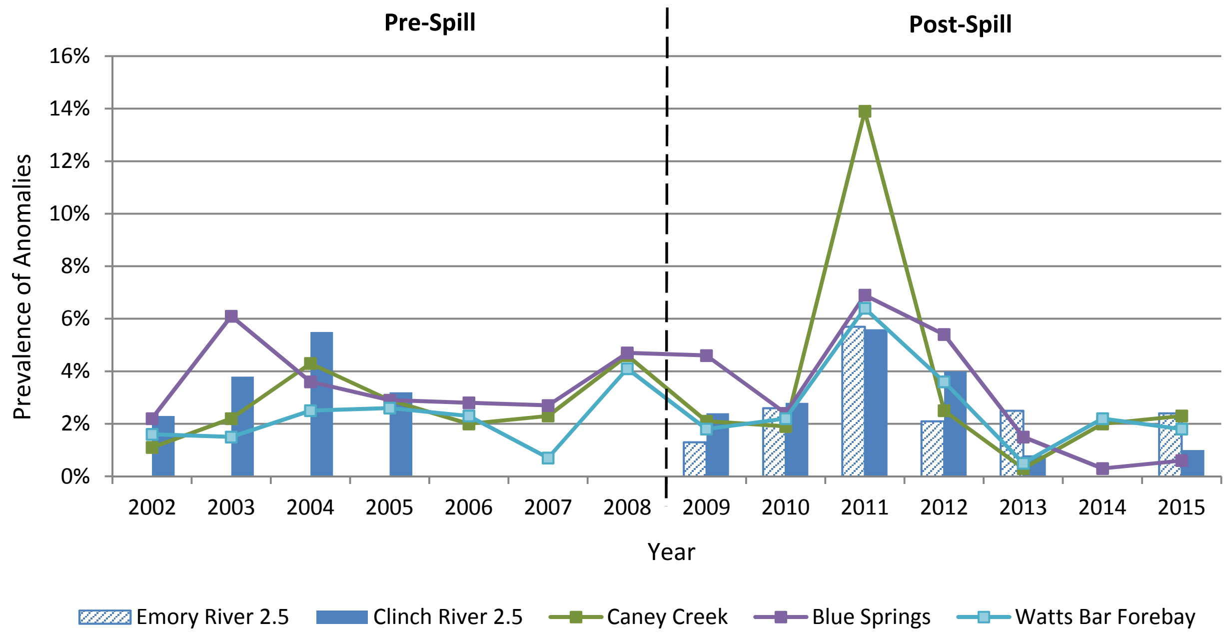


Figure 5. Prevalence of physical anomalies (%) in black bass populations, 2002-2015. The frequency of sampling varied by location.

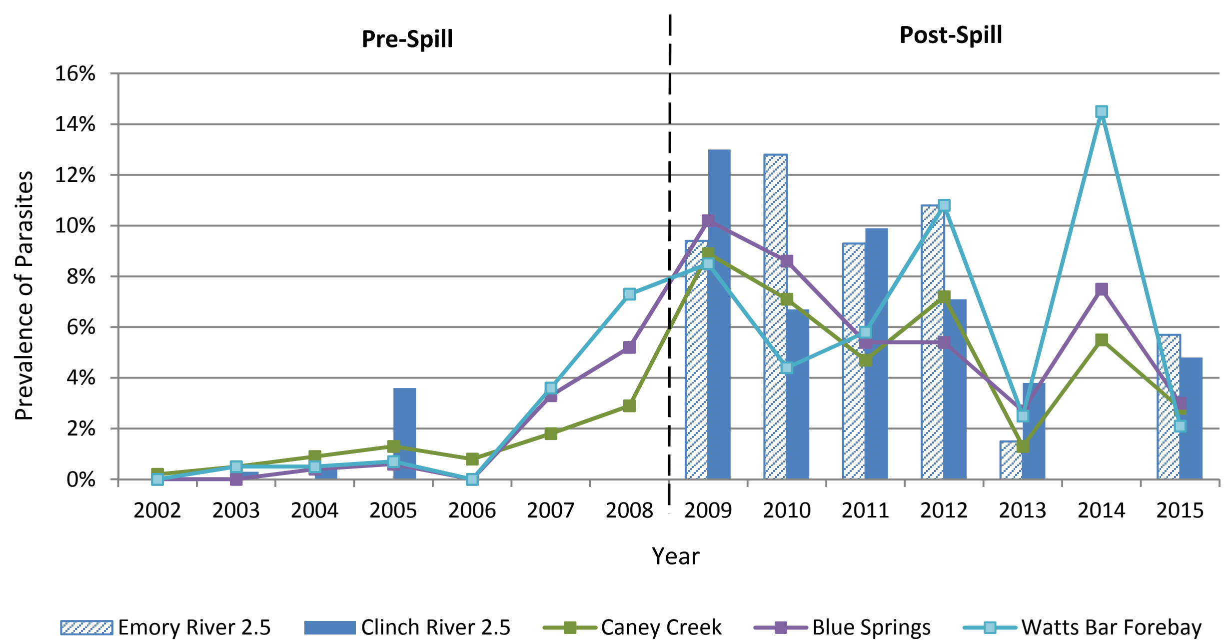


Figure 6. Prevalence of external parasites (%) in black bass populations, 2002-2015. The frequency of sampling varied by location.

Table 1. Summary of Spring Sport Fish Survey results for black bass collected at sampling locations on the Emory, Clinch and Tennessee Rivers, Watts Bar Reservoir, 2002-2015.

Location	Largemouth bass	Smallmouth bass	Spotted bass	Total # of Bass	Catch Rate (no./hr.)	Physical Anomalies		Parasites		Hook Injury	
Emory River 2.5											
2009	304	0	15	319	53.2	4	1.3%	30	9.4%	3	0.9%
2010	297	5	2	304	50.7	8	2.6%	39	12.8%	5	1.6%
2011	192	0	2	194	32.3	11	5.7%	18	9.3%	8	4.1%
2012	234	1	5	240	40.0	5	2.1%	26	10.8%	12	5.0%
2013	193	3	2	198	33.0	5	2.5%	3	1.5%	8	4.0%
2015	414	4	4	422	70.3	10	2.4%	24	5.7%	12	2.8%
Average	**272**	**2**	**5**	**280**	**46.6**	**7**	**2.8%**	**23**	**8.3%**	**8**	**3.1%**
Clinch River 2.5											
2002	247	19	45	311	51.8	7	2.3%	0	0.0%	6	1.9%
2003	206	30	53	289	48.2	11	3.8%	1	0.3%	9	3.1%
2004	192	22	42	256	42.7	14	5.5%	1	0.4%	13	5.1%
2005	199	23	30	252	42.0	8	3.2%	9	3.6%	4	1.6%
2009	344	28	4	376	62.7	9	2.4%	49	13.0%	12	3.2%
2010	314	37	6	357	59.5	10	2.8%	24	6.7%	8	2.2%
2011	157	19	1	177	29.4	10	5.6%	18	9.9%	11	6.1%
2012	220	32	1	253	42.2	10	4.0%	18	7.1%	16	6.3%
2013	213	25	1	239	39.8	2	0.8%	9	3.8%	19	7.9%
2015	358	58	3	419	69.8	4	1.0%	20	4.8%	15	3.6%
Average	**245**	**29**	**19**	**293**	**48.8**	**9**	**3.1%**	**15**	**5.0%**	**11**	**4.1%**
Caney Creek											
2002	446	19	3	468	78.0	5	1.1%	1	0.2%	9	1.9%
2003	320	34	13	367	61.2	8	2.2%	2	0.5%	10	2.7%
2004	321	17	13	351	58.5	15	4.3%	3	0.9%	13	3.7%
2005	285	19	10	314	52.3	9	2.9%	4	1.3%	4	1.3%
2006	211	38	6	255	42.5	5	2.0%	2	0.8%	3	1.2%
2007	346	43	4	393	65.5	9	2.3%	7	1.8%	10	2.5%
2008	539	19	2	560	93.3	26	4.6%	16	2.9%	12	2.1%
2009	403	22	4	429	71.5	9	2.1%	38	8.9%	11	2.6%
2010	545	31	3	579	96.5	11	1.9%	41	7.1%	20	3.5%
2011	307	28	3	338	56.3	47	13.9%	16	4.7%	14	4.1%
2012	333	28	0	361	60.2	9	2.5%	26	7.2%	24	6.6%
2013	365	28	1	394	65.7	1	0.3%	5	1.3%	13	3.3%
2014	322	23	2	347	57.8	7	2.0%	19	5.5%	15	4.3%
2015	535	27	3	565	94.2	13	2.3%	16	2.8%	15	2.7%
Average	**377**	**27**	**5**	**409**	**68.1**	**12**	**3.2%**	**14**	**3.3%**	**12**	**3.0%**

Table 1, continued

Location	Largemouth bass	Smallmouth bass	Spotted bass	Total # of Bass	Catch Rate (no./hr.)	Physical Anomalies		Parasites		Hook Injury	
Blue Springs											
2002	261	6	1	268	44.7	6	2.2%	0	0.0%	3	1.1%
2003	270	21	22	313	52.2	19	6.1%	0	0.0%	1	0.3%
2004	252	14	8	274	45.7	10	3.6%	1	0.4%	5	1.8%
2005	271	25	13	309	51.5	9	2.9%	2	0.6%	5	1.6%
2006	346	14	1	361	60.2	10	2.8%	0	0.0%	2	0.6%
2007	290	11	0	301	50.2	8	2.7%	10	3.3%	5	1.7%
2008	380	17	6	403	67.2	19	4.7%	21	5.2%	17	4.2%
2009	315	8	0	323	53.8	15	4.6%	33	10.2%	8	2.5%
2010	333	5	0	338	56.3	8	2.4%	29	8.6%	19	5.6%
2011	268	7	1	276	46.0	19	6.9%	15	5.4%	13	4.7%
2012	185	18	0	203	33.8	11	5.4%	11	5.4%	9	4.4%
2013	323	14	0	337	56.2	5	1.5%	9	2.7%	12	3.6%
2014	291	11	3	305	50.8	1	0.3%	23	7.5%	9	3.0%
2015	353	8	0	361	60.2	2	0.6%	11	3.0%	6	1.7%
Average	**296**	**13**	**4**	**312**	**52.0**	**10**	**3.3%**	**12**	**3.7%**	**8**	**2.6%**
Watts Bar Forebay											
2002	266	20	35	321	53.5	5	1.6%	0	0.0%	2	0.6%
2003	312	13	65	390	65.0	6	1.5%	2	0.5%	10	2.6%
2004	246	25	94	365	60.8	9	2.5%	2	0.5%	11	3.0%
2005	172	47	55	274	45.7	7	2.6%	2	0.7%	5	1.8%
2006	263	32	16	311	51.8	7	2.3%	0	0.0%	1	0.3%
2007	255	18	2	275	45.8	2	0.7%	10	3.6%	4	1.5%
2008	359	8	1	368	61.3	15	4.1%	27	7.3%	2	0.5%
2009	370	18	0	388	64.7	7	1.8%	33	8.5%	19	4.9%
2010	436	19	0	455	75.8	10	2.2%	20	4.4%	17	3.7%
2011	305	21	0	326	54.3	21	6.4%	19	5.8%	4	1.2%
2012	148	19	0	167	27.8	6	3.6%	18	10.8%	7	4.2%
2013	385	15	0	400	66.7	2	0.5%	10	2.5%	11	2.8%
2014	384	17	0	401	66.8	9	2.2%	58	14.5%	24	6.0%
2015	497	17	0	514	85.7	9	1.8%	11	2.1%	4	0.8%
Average	**314**	**21**	**19**	**354**	**59.0**	**8**	**2.4%**	**15**	**4.4%**	**9**	**2.4%**

ARCADIS

Appendix B

2015 ORNL Progress Report for the TVA-Kingston Project

NOTICE: This document contains information of a preliminary nature and is not intended for release. It is subject to revision or correction and therefore does not represent a final report.

ORNL/TM-2015/245

2015 ORNL Progress Report for the TVA–Kingston Project

Mark Peterson
Mark Greeley
Terry Mathews
Brenda Pracheil
John Smith

September 2015

Draft. Not for public release.

OAK RIDGE NATIONAL LABORATORY
MANAGED BY UT-BATTELLE FOR THE US DEPARTMENT OF ENERGY

ORNL/TM-2015/245

Environmental Sciences Division

2015 ORNL PROGRESS REPORT FOR THE TVA–KINGSTON PROJECT

Mark Peterson
Mark Greeley
Terry Mathews
Brenda Pracheil
John Smith

Date Published: September 2015

Prepared for

Tennessee Valley Authority
400 West Summit Hill Drive
Knoxville, Tennessee 37902-1499

Department of Energy (DOE) Project No. 1610-V083-09

Prepared by
OAK RIDGE NATIONAL LABORATORY
Oak Ridge, TN 37831-6283
managed by
UT-BATTELLE, LLC
for the
US DEPARTMENT OF ENERGY
under contract DE-AC05-00OR22725

CONTENTS

LIST OF FIGURES

LIST OF TABLES

ACRONYMS

ANCOVA	analysis of covariance
CRM	Clinch River Mile
ERM	Emory River Mile
KIF	Kingston Fossil Plant
ORNL	Oak Ridge National Laboratory
TVA	Tennessee Valley Authority

1. INTRODUCTION

In December 2008, an ash dike at the Tennessee Valley Authority (TVA) Kingston Fossil Plant (KIF) ruptured and released a large quantity of coal fly ash into the Emory and Clinch Rivers. Coal ash may contain several contaminants that, if found in large enough quantities in some aquatic systems, can be a human or ecological risk concern. In the case of the Kingston spill, numerous coal ash constituents were studied for years after the event; in particular, selenium, mercury, and arsenic have been a major focus of monitoring and research because of their toxicity or tendency to bioaccumulate in aquatic food chains. To assess the potential impact of the spilled fly ash on humans and the environment, a comprehensive biological monitoring program has been in place since the event. Resident aquatic organisms are collected on a regular basis to determine contaminant exposure and evaluate the risks to humans and wildlife.

Oak Ridge National Laboratory (ORNL) scientists have supported the TVA–Kingston project's biological monitoring and research activities since shortly after the spill. Primary project tasks include bioaccumulation sampling and analysis, including fish and invertebrates, toxicity testing studies, and fish health and reproduction evaluations. ORNL program management activities associated with the core tasks, including data management and quality assurance, have been significant because of the overall project's large scope and the scientific rigor expected of the program. ORNL staff has provided a number of published reports as well as internal assessments and guidance over the years that were used for the project's human and ecological risk assessments, as well as numerous presentations at project and scientific meetings. A key current ORNL effort is to disseminate TVA–Kingston research in open literature publications.

This report provides an update on the TVA–Kingston project as of September 2015; the report includes monitoring results through 2014 and progress on sampling and analysis and publication activities in 2015. The 2014 monitoring and assessment activities for the TVA–Kingston project were focused largely on a more limited set of metrics at the site nearest the spill, with a more extensive sampling of locations, species, and metrics in 2015. The most current results are provided in Sect. 2, Fish Studies (including bioaccumulation results through 2014 and fish health and reproduction results through 2013), and Sect. 3, Invertebrate Bioaccumulation. Sect. 4, 2015 Progress, presents ORNL sampling and analysis progress associated with the 2015 monitoring effort, as well as a list of recent presentations that offer more detailed information about ORNL studies of the Kingston fly ash spill.

2. FISH STUDIES

Fish studies at ORNL are divided into three tasks: (1) fish bioaccumulation, (2) fish health, and (3) fish reproduction. To assess the effects of coal ash exposure on overall and reproductive fish health, we targeted the collection of eight individuals of each of three species (largemouth bass, redear sunfish, and bluegill) from five sites. Our sites included three ash-affected sites—Emory River mile (ERM) 3.0, ERM 1.0, and Clinch River mile (CRM 1.5; Fig. 1) and two reference sites (ERM 8.0 and CRM 8.0). The reference sites were chosen for this study because they are upstream of the coal ash release and are therefore not contaminated by coal ash, but it is important to note that both reference sites have been affected by other legacy contaminant releases. We scaled back sampling protocols in 2014 because contaminant concentrations were not above guidelines or levels of concern and because there was no clear evidence that fish health or reproduction were impacted severely by the ash spill. In 2014, the only site sampled was ERM 1.0, a site downstream of the spill with the highest contaminant concentrations among ash-affected sites.

Fish were collected twice annually in the spring and fall from 2009 to 2013 and in the spring only in 2014. Spring collections occurred in April through June to coincide with the beginning of the breeding seasons of the study species and included only females to investigate possible relationships among metal exposure, fish health, and reproductive fitness. However, because sex determination in the field is not 100% accurate, we sometimes collected more than the target of eight individuals per site. Fall collections occurred in October through November and included both male and female fish. All fish were collected by TVA and ORNL staff and/or their contractors, in most cases using TVA equipment (e.g., electrofishers, dip nets, etc.).

Fish were then transported to the lab, where we collected blood for blood chemistry assessments before euthanizing fish with buffered MS-222. We then conducted external and internal fish health examinations and removed spleen, liver, ovary, and fillet tissue for metal concentration analysis. Portions of ovaries also were removed and preserved for reproductive health assessments.

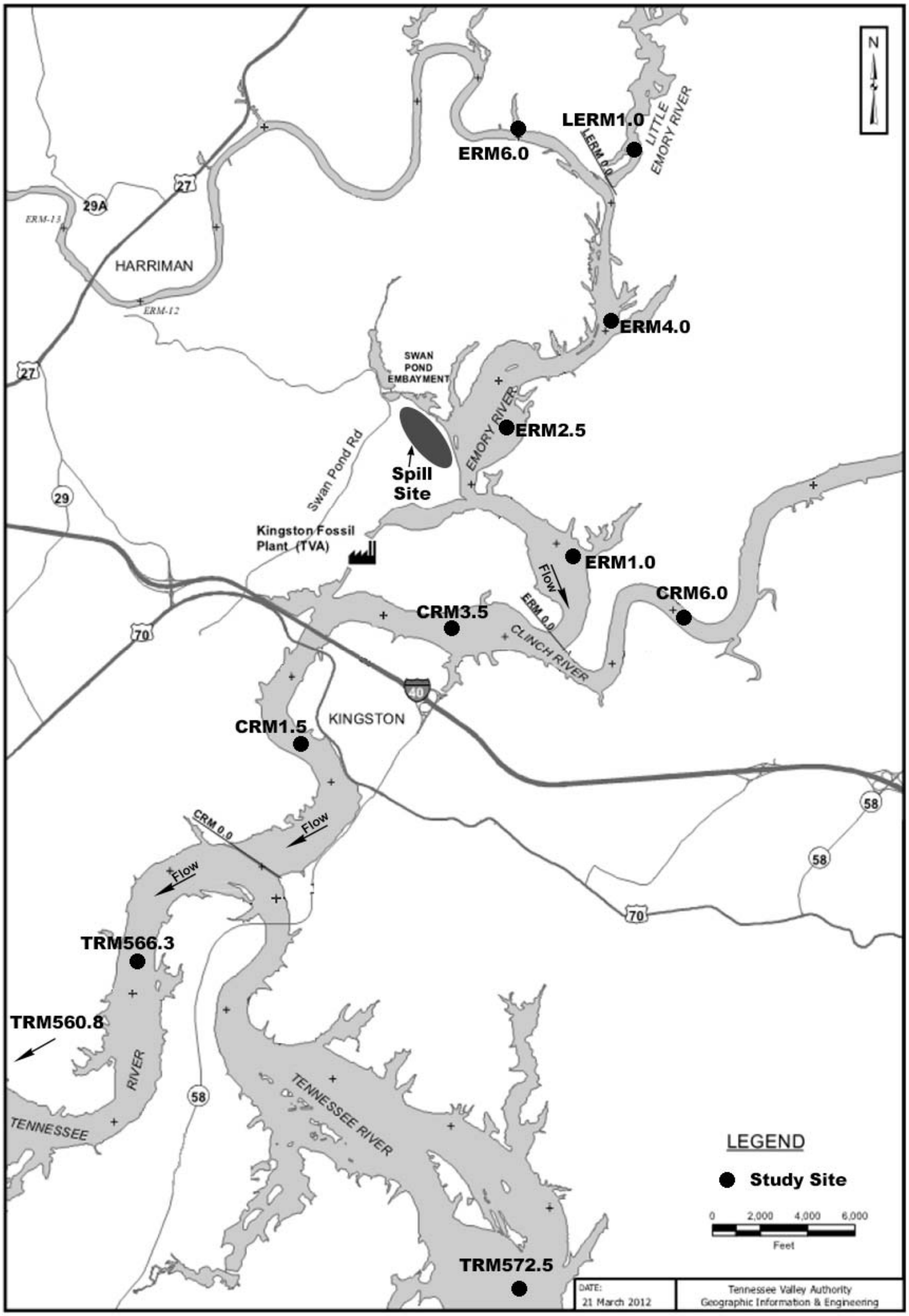


Fig. 1. Study sites for the Tennessee Valley Authority Kingston coal fly ash spill research project (*Source:* Adapted from Tennessee Valley Authority, used with permission).

2.1 FISH BIOACCUMULATION

The objective of the fish bioaccumulation task is to assess exposure to contaminants over time to infer potential risks to humans and wildlife because of the ash spill. As noted in Sect. 1, the primary contaminants of concern in terms of fish bioaccumulation are selenium, arsenic, and mercury.
A preliminary analysis of contaminant concentrations in fish shows that selenium concentrations decreased significantly in redear and bluegill sunfish fillets, dropping from average concentrations of 1.02 and 0.80 mg/kg wet wt in 2013 to 0.71 and 0.63 mg/kg wet wt in 2014 in redear and bluegill, respectively (Tables 1 and 2). These concentrations are significantly lower than those seen previously at this site, and though samples were not collected from reference sites in 2014, the mean concentrations seen in 2014 are comparable to those observed at reference sites in previous years. Mean selenium concentrations in largemouth bass fillets were the lowest of the three species studied and did not change significantly in 2014 (Table 3). Selenium concentrations continue to be well below toxicity and risk guidelines. Mercury and arsenic fillet concentrations remain low (and below regulatory guidelines) and did not change significantly from 2013 to 2014 in any species monitored. Arsenic concentrations were highest in the liver in all three species.

Temporal trends in selected trace element concentrations in fillets of fish collected at ERM 1.0 are shown in Fig. 2. Overall mercury concentrations in largemouth bass concentrations appear to be increasing (both at ERM 1.0 and at other monitored locations, including those not affected by the coal ash spill). These data are not shown here, as ERM 1.0 was the only site monitored in 2014. Temporal trends in mercury in largemouth bass at other sites were presented previously.

Table 1. Arsenic, mercury, and selenium concentrations (µg/g wet wt) in tissues of bluegill sunfish collected at ERM 1.0 in 2014

	Number of samples	Number of detects	Mean concentration	Standard deviation	Minimum	Maximum
			Arsenic			
Fillet	6	6	0.03	0.01	0.02	0.05
Liver	1	1	0.34	NA	0.34	0.34
Ovary	6	6	0.07	0.02	0.05	0.11
			Mercury			
Fillet	6	5	0.03	0.05	0.003	0.12
Liver	1	1	0.12	NA	0.12	0.12
Ovary	6	5	0.01	0.01	0.003	0.02
			Selenium			
Fillet	6	6	0.63	0.13	0.52	0.86
Liver	1	1	2.20	NA	2.20	2.20
Ovary	6	6	1.24	0.25	0.95	1.70

Note: NA = not applicable.

Table 2. Arsenic, mercury, and selenium concentrations (µg/g wet wt) in tissues of redear sunfish collected at ERM 1.0 in 2014

	Number of samples	Number of detects	Mean concentration	Standard deviation	Minimum	Maximum
			Arsenic			
Fillet	6	6	0.15	0.04	0.08	0.20
Liver	1	1	1.50	NA	1.50	1.50
Ovary	6	6	0.58	0.20	0.20	0.75
			Mercury			
Fillet	6	6	0.08	0.03	0.05	0.11
Liver	1	1	0.08	NA	0.08	0.08
Ovary	6	2	0.00	0.00	0.00	0.01
			Selenium			
Fillet	6	6	0.71	0.12	0.60	0.93
Liver	1	1	3.00	NA	3.00	3.00
Ovary	6	6	1.43	0.22	1.10	1.70

Note: NA = not applicable.

Table 3. Arsenic, mercury, and selenium concentrations (µg/g wet wt) in tissues of largemouth bass collected at ERM 1.0 in 2014

	Number of samples	Number of detects	Mean concentration	Standard deviation	Minimum	Maximum
			Arsenic			
Fillet	6	6	0.19	0.04	0.12	0.23
Liver	6	6	0.41	0.10	0.31	0.59
Ovary	6	6	0.27	0.06	0.18	0.34
			Mercury			
Fillet	6	6	0.26	0.09	0.19	0.40
Liver	6	6	0.14	0.05	0.08	0.20
Ovary	6	5	0.02	0.01	0.003	0.03
			Selenium			
Fillet	6	6	0.59	0.08	0.51	0.69
Liver	6	6	1.85	0.29	1.30	2.10
Ovary	6	6	1.15	0.15	1.00	1.40

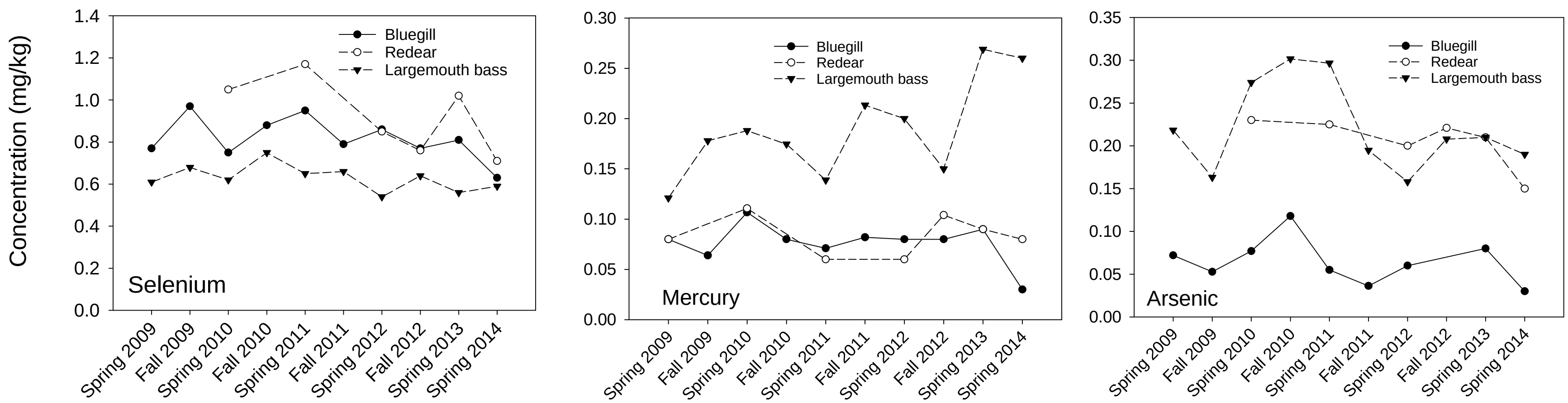


Fig. 2. Mean trace element concentrations in fish fillets (redear, bluegill, and largemouth bass) collected from ERM 1.0, from spring 2009 to spring 2014.

Because many contaminants are accumulated in fish through dietary exposure, we have been examining contaminant concentrations throughout the food chain to help explain bioaccumulation patterns seen in fish. For example, the invertebrate bioaccumulation task has examined metal concentrations in mayfly nymphs and snails both before and after depuration. From these data, we have calculated assimilation efficiencies and will relate these to both exposure concentrations and trophic transfer to fish (see Sect. 3).

Because mercury and selenium are accumulated primarily through dietary exposure, the trophic transfer of these two elements is of particular interest. Selenium has been recognized to mitigate mercury toxicity, and concentrations of these two elements have been shown to be correlated inversely in fish fillets. However, the mechanisms controlling this inverse relationship remain unknown. Selenium can affect mercury bioaccumulation at multiple steps in the food chain, leading to the observed trends in selenium and mercury levels in fish. Very few studies address the effects of selenium on the bioaccumulation and toxicity of mercury in freshwater primary producers or invertebrates, although this likely is a critical link in the understanding of mercury/selenium interactions observed in fish. Over the past 5 years, bioaccumulation monitoring for the TVA–Kingston project has included organisms at multiple trophic levels. Whereas the relationship between mercury and selenium concentrations in fish varies with site and species, there appears to be an overall negative relationship between mercury and selenium concentrations in fish (Fig. 3). This relationship is not observed in periphyton or invertebrates, where there is either no relationship or a positive relationship between mercury and selenium concentrations. Although we are still investigating factors such as size, sex, and depurated vs. non-depurated, these preliminary observations provide evidence that the inverse relationship seen between selenium and mercury in fish may be happening by detoxification mechanisms within fish, rather than within the entire food chain.

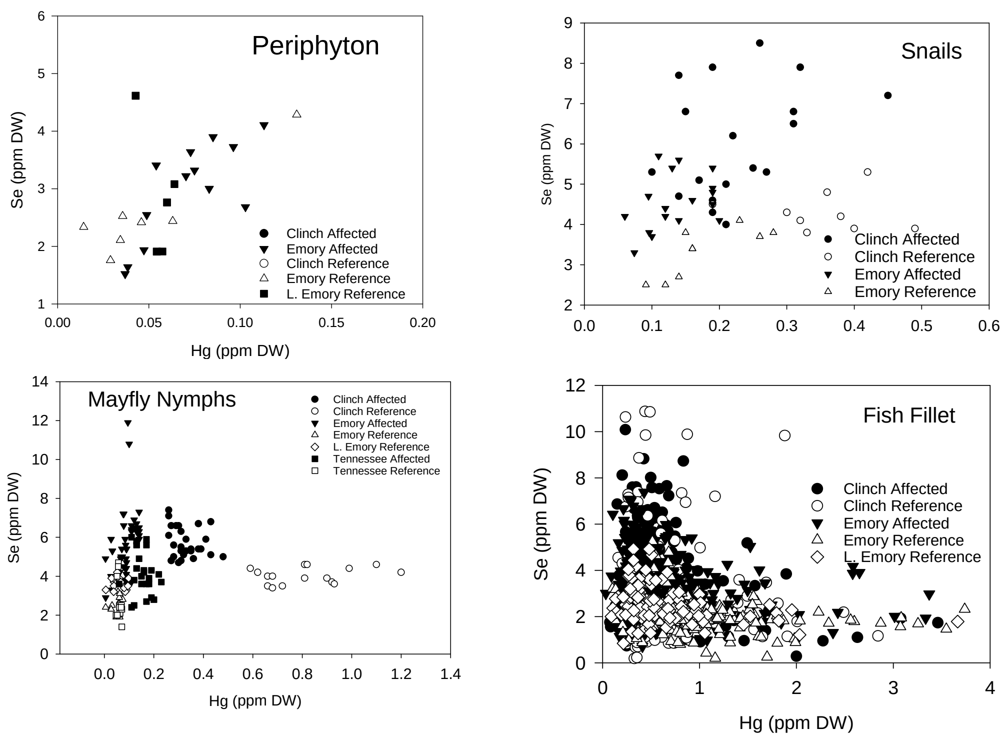


Fig. 3. The relationship between mercury and selenium concentrations in organisms collected around the Kingston ash spill site (2009–2014). ***Note:*** **DW – Dry weight.**

2.2 FISH HEALTH

Efforts during 2014 for this task included the collection of fish samples from one location on the Emory River (ERM 1.0) in collaboration with TVA. Bluegill sunfish, largemouth bass, and redear sunfish were collected at the beginning of their respective breeding seasons during April and May 2014 for the three fish-based ORNL tasks. At least six fish of each of the three study species were processed at ORNL for bioaccumulation analyses, and a visual inspection of fish health conditions was performed. The normal annual fish health assessment, which includes blood analysis, was not performed on 2014 fish per the monitoring plan. The visual inspection revealed that there was no evidence of unusual internal or external anomalies. The results presented in this section focus on the findings from the ORNL fish health studies over the 2009–2013 period.

The effect of coal ash exposure on fish health in natural freshwater communities is largely unknown. Given the large number of possible pathways of effects (e.g., toxicological effect of exposure to multiple metals, physical effects from ash exposure, and food web effects), measurement of only a few health metrics is not likely to give a complete picture. From field studies completed through 2013, a suite of 25 health metrics was measured from 1,300+ fish collected from six sites (three ash affected sites: ERM 3.0, ERM 1.0, and CRM 1.5; and three reference sites: ERM 8.0, Little Emory River mile 2.0, and ERM 8.0).

Health parameters obtained from each fish included the following:

- Measures of bioenergetic condition based on the general condition of the fish and internal organs (liver, ovaries, spleen, and viscera)
- Analysis of blood chemistry and hematology to produce metrics that are indicative of compromised physiological function
- Histopathological analysis of cellular lesions and dysfunction in gill and liver tissue
- Overall health assessment index (disease, parasites, developmental anomalies, etc.) based on visual inspection of external and internal anomalies

We measured metrics that represent a wide range of physiological and energetic responses. We linked these responses to bioaccumulation and reproductive health data using univariate and multivariate statistical techniques. We also statistically linked liver and gill histopathology data to bioaccumulation data using multivariate and univariate techniques. With the data set complete through 2013, individual metrics were evaluated for temporal trends and differences between reference sites and spill sites.

Our findings suggest that whereas fish tissue concentrations of some ash-associated contaminants were elevated at the spill site over the 2009-2013 period, there was no consistent evidence of compromised fish health linked with the spill (Fig. 4). Further, we found relationships between elevated fillet burdens of ash-associated contaminants and some fish metrics, but these relationships were not indicative of exposure to coal ash or spill sites. This study also found little evidence linking the TVA KIF coal ash spill to reproductive effects. Although we did find associations between increased fillet selenium and decreased fecundity, vitellogenic oocytes, and increased atretic oocytes in bluegill, there were no consistent patterns linking decreased fitness to coal ash affected sites (Table 4). Similarly, with the exception of redear where ash affected sites had the highest numbers of atretic oocytes and vitellogenic oocytes as well as the highest fecundity, analyses of covariance (ANCOVA) and post-hoc Tukey's tests did not indicate the coal ash spill had negative effects on any reproductive metric for any species among the sites (also see Sect. 2.3). In fact, the only site effects indicated by ANCOVA showed redear had significantly higher numbers of vitellogenic oocytes in spill sites than in reference sites. The details of this study have been submitted for publication in *Ecotoxicology*.

Similarly, our joint analysis of histopathology and bioaccumulation data did not show consistent trends in gill or liver histopathology data that were related to either reference or spill sites, although we did find an increase in overall gill (Fig. 5) and liver (Fig. 6) pathology immediately post-dredging that generally decreased across all sites and species after 2011.

ANCOVA examination of the effects of site, year, arsenic, mercury, and selenium on the composite liver pathology metric (e.g., the sum of all six liver pathology scores measured) was significant for bluegill, redear, and largemouth bass. In these models, mercury was associated with composite liver pathology for redear and largemouth bass, and selenium was associated with composite liver pathology for bluegill and largemouth bass (Table 5). No ANCOVA examining the effects of site, year, arsenic, mercury, and selenium on the composite gill pathology metric (e.g., the sum of all six gill pathology scores measured) was significant. The details of these analyses have been included in papers submitted to, or in preparation for, peer reviewed journals.

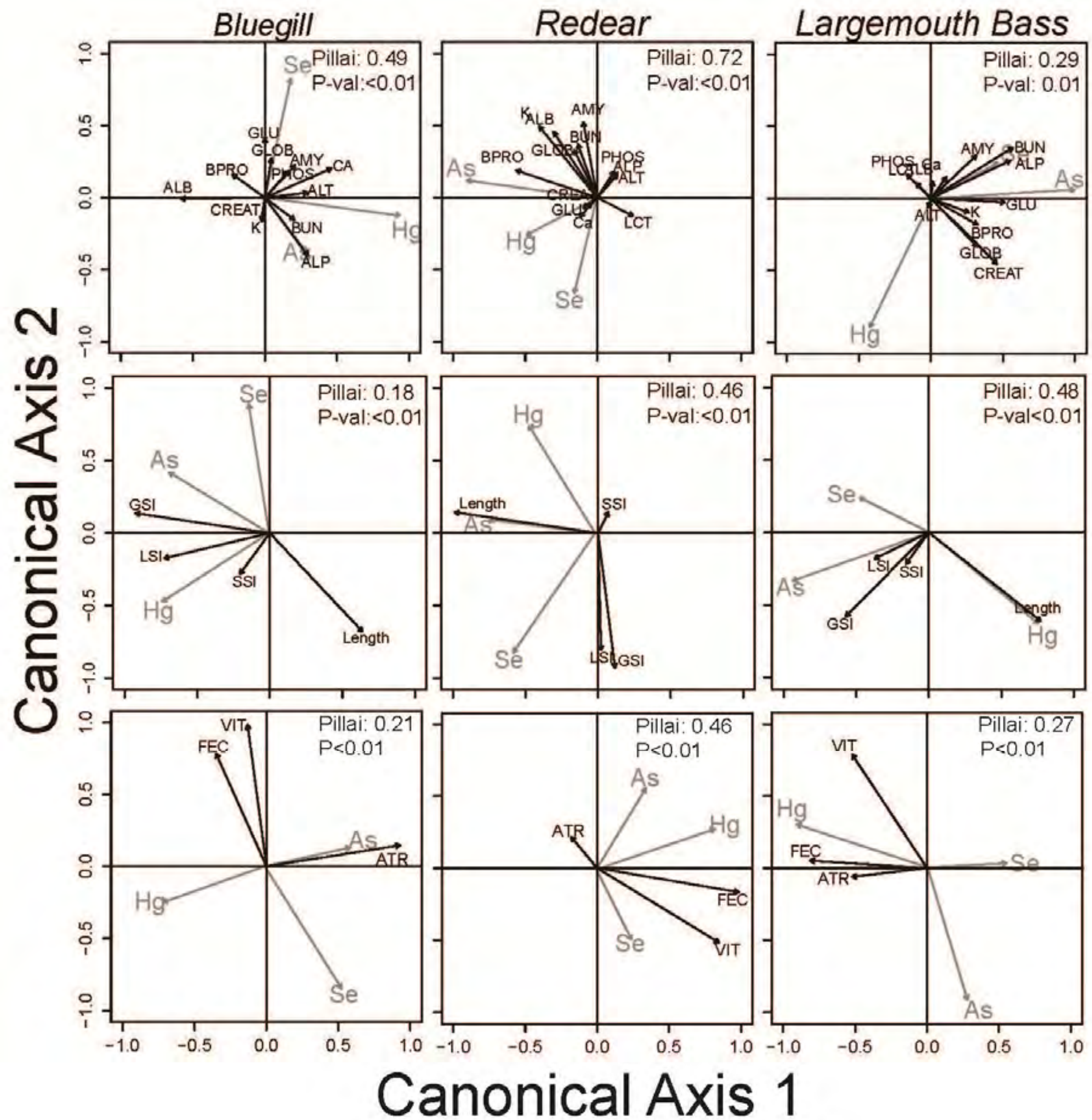


Fig. 4. Scatterplot of loadings (dimensions 1 and 2) from canonical correlation analysis of fillet heavy metal concentrations (gray) and metrics of fish (black) health (top row), condition (middle row), and reproduction (bottom row). Standard elemental abbreviations are used for heavy metals, and fish metric abbreviations are shown in Table 5.

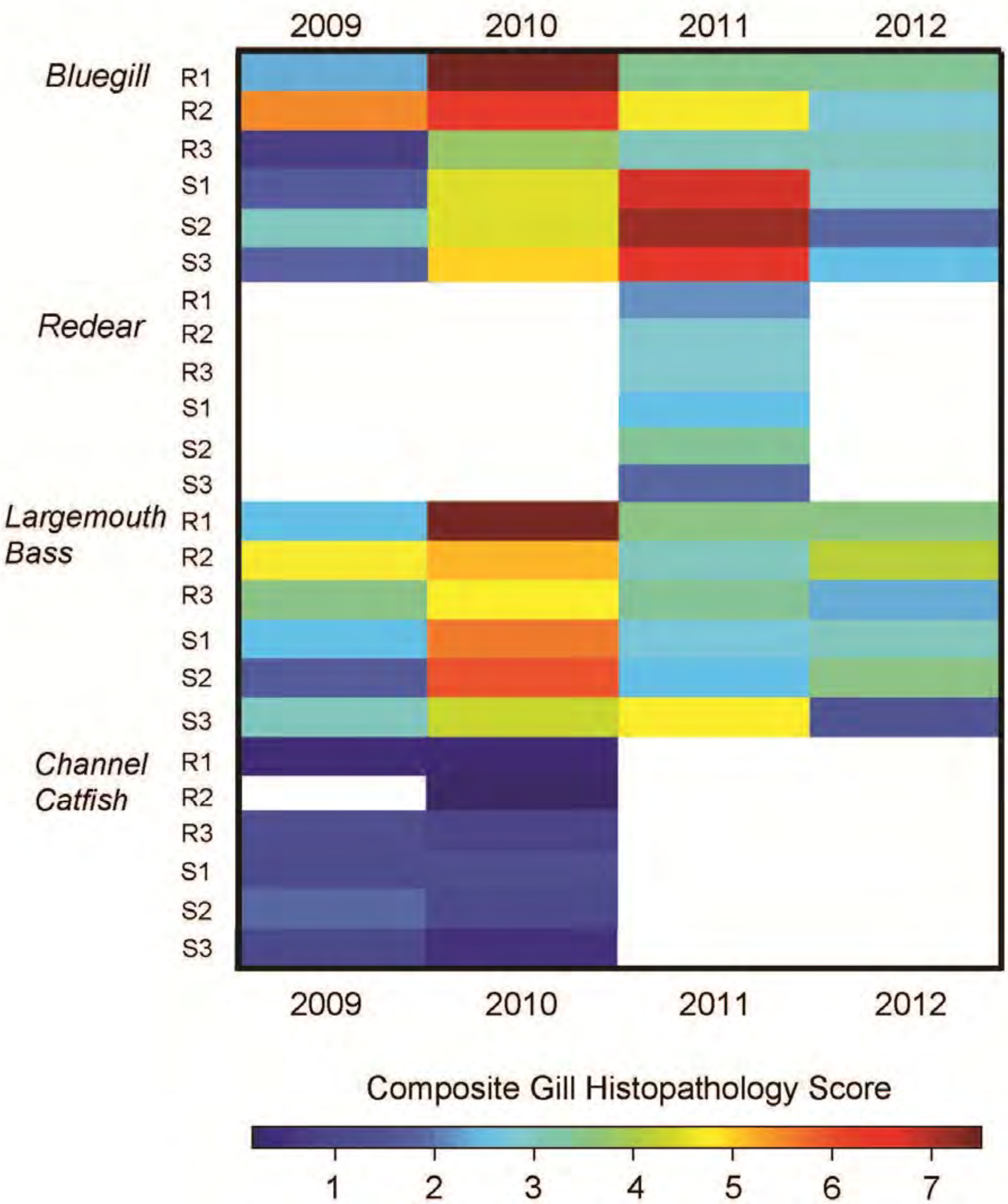


Fig. 5. Composite gill histopathology score calculated as the sum of all six pathology types quantified in this study by year for each fish species at each site. All gill samples were collected from female fish during the spring season.

Table 4. Results for analyses of covariance examining effects of ash-associated metals, fish length, collection site, year, and site x year interaction on fish reproductive health metrics for each fish species where reproductive health was assessed

Response metric	Arsenic	Selenium	Mercury	Length	Site	Year	Site*year
Bluegill							
Atresia $F_{26,133} = 1.90$, $P = 0.0119$	NS	NS	NS	NS	NS	0.0014	NS
Vitellinogen $F_{26,133} = 5.82$, $P < 0.0001$	0.0072	0.0223	NS	< 0.0001	NS	NS	0.0490
Fecundity $F_{26,133} = 3.70$, $P < 0.0001$	0.0270	NS	NS	< 0.0001	NS	NS	NS
Redear							
Atresia $F_{21, 115} = 2.39$, $P = 0.0023$	NS	NS	NS	0.0193	0.0260	0.0003	NS
Vitellinogen $F_{21, 115} = 6.37$, $P < 0.0001$	0.0009	0.0221	0.0196	< 0.0001	0.0384	0.0391	NS
Fecundity $F_{21, 115} = 7.58$, $P < 0.0001$	NS	NS	0.0114	< 0.0001	NS	NS	NS
Largemouth bass							
Atresia $F_{26,140} = 2.67$, $P = 0.0002$	NS	NS	NS	NS	NS	0.0005	0.0218
Vitellinogen $F_{26,140} = 3.07$, $P < 0.0001$	NS	NS	NS	< 0.0001	NS	NS	NS
Fecundity $F_{26,140} = 3.85$, $P < 0.0001$	NS	NS	NS	< 0.0001	NS	< 0.0001	NS

Notes: Overall model results are shown below the response metric as $F_{\text{num d.f., denom d.f.}}$ and P-value. Significant P-values ($\alpha = 0.05$) for each covariate from Type III sum-of-squares are shown in the cells. NS denotes a covariate was not significant.

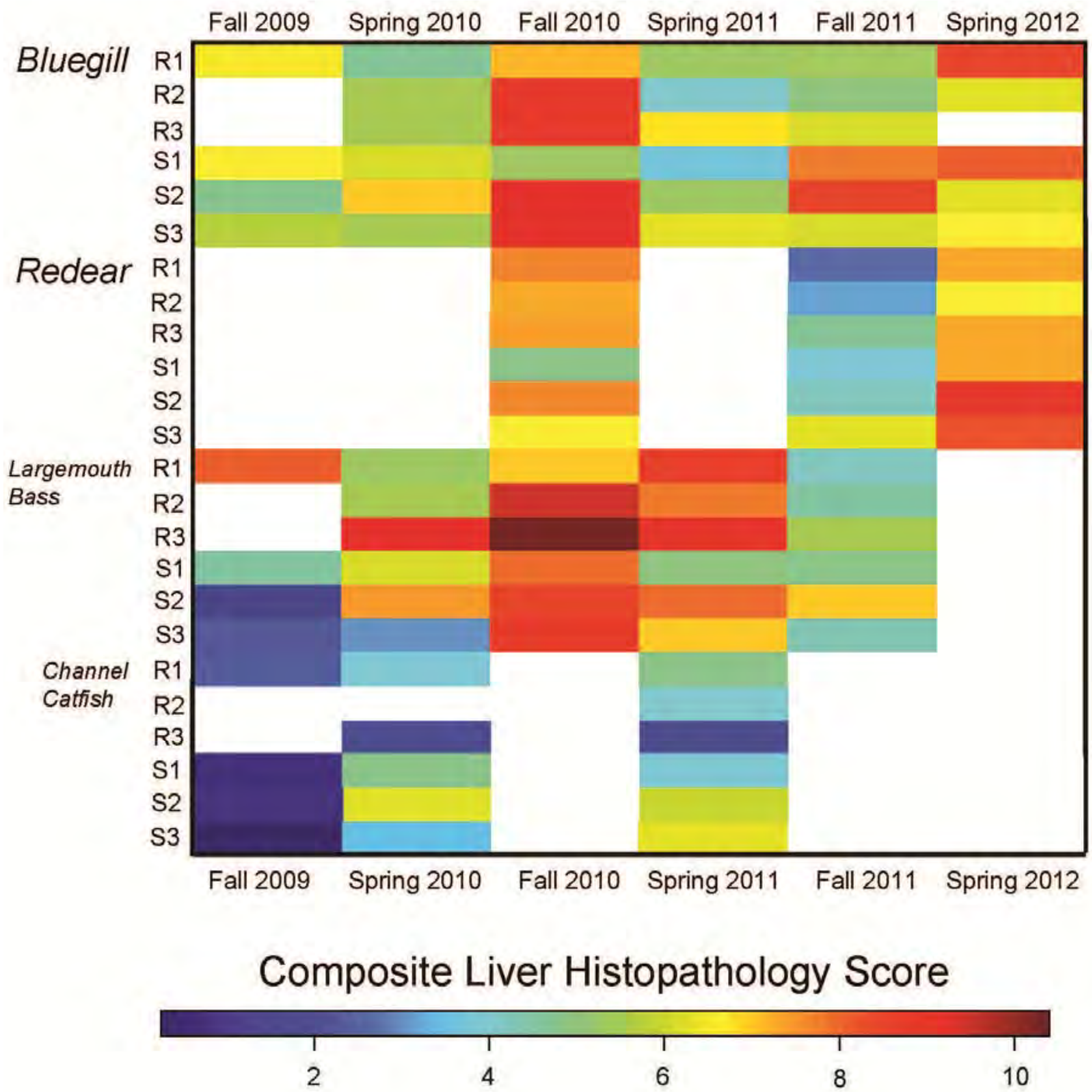


Fig. 6. Composite liver histopathology score calculated as the sum of the scores assigned to all six liver lesion types quantified in this study by year for each fish species and season at each site.

Table 5. Results from general linear models examining the effects of independent variables (listed in the Effect column) on composite liver scores for each fish species

Species	Effect	F-value	P-value
Bluegill	Model	$F_{9,160} = 3.43$	0.0007
	Site	2.71	0.0224
	Year	1.54	0.2166
	Mercury	1.67	0.1983
	Arsenic	3.25	0.0735
	Selenium	0.79	0.3744
Redear	Model	$F_{9, 89} = 3.15$	0.0024
	Site	3.43	0.0071
	Year	0.14	0.7064
	Mercury	4.80	0.0311
	Arsenic	8.89	0.0037
	Selenium	0.16	0.6885
Largemouth bass	Model	$F_{9,122} = 3.70$	0.0004
	Site	2.75	0.0216
	Year	5.20	0.0243
	Mercury	10.11	0.0019
	Arsenic	0.09	0.7625
	Selenium	0.63	0.4289

Notes: Gray shaded rows represent overall model results ($F_{\text{num d.f., denom d.f.}}$), and unshaded rows represent Type III sum of squares F-values and P-values.

2.3 FISH REPRODUCTION

As noted above, bluegill sunfish, largemouth bass, and redear sunfish were collected at ERM 1.0 during April and May 2014 at the beginning of their respective breeding seasons. In addition to bioaccumulation analyses and a visual inspection of fish health, samples of select tissues including ovaries were archived for potential future use.

In addition to the limited 2014 fish sampling and processing activities, progress during 2014 for this task included the completion of all reproductive analyses on fish ovary samples from previous years' sampling events. These analyses consisted of conducting morphometric analyses on remaining ovary samples from female fish collected through spring 2013 for the Kingston coal ash release investigations. These analyses included calculations of fecundity, ovary staging, and determination of overall fish reproductive status and condition; measurements of oocyte/egg size and abundance; assessments of oocyte/egg developmental stages; and measures of oocyte quality such as incidences of oocyte atresia.

A few statistically significant differences or trends between study sites were noted in some of the reproductive parameters evaluated during the 5-year post-spill monitoring efforts. The most notable previously reported example was a delay in the reproductive development of female bluegill sunfish at the spill site in the Emory River during the first breeding season following the ash release in spring 2009. More recently, relatively high incidences of oocyte atresia were observed in some largemouth bass collected from ash-exposed sites during the 2011−2013 breeding seasons, although comparisons with fish collected from reference sites and consideration of the weather and river conditions immediately before sample collections suggest contributing factors other than the ash spill.

The data set for the fish reproduction samples collected from 2009 through 2013 was included in correlation analyses that formed the basis of a recent paper focused on the relationships between metal bioaccumulation and fish health and reproduction that was submitted for review to the journal *Ecotoxicology* (with Brenda Pracheil as primary author). Additional details concerning these analyses are provided in Sect. 2.2. Figure 7, which is adapted from that paper, summarizes year-to-year variation from 2009 through 2013 in select reproductive parameters for two of the sentinel fish species—bluegill sunfish and redear sunfish—employed in the reproductive studies. In general, the results of the field-based fish reproductive collections and analyses, when considered along with the results of fish community monitoring conducted by TVA, continue to suggest that the December 2008 Kingston ash release has had little, if any, ecologically significant adverse effects on the reproduction of fish in areas of the Emory and Clinch Rivers affected by the spill.

In addition to these field-based reproductive assessments, various laboratory-based and/or combined laboratory- and field-based fish studies have also been conducted in the aftermath of the spill to assess the potential for the coal ash release to adversely impact fish reproduction in areas affected by the spill. In 2014, the results of tests conducted to evaluate the potential effects of exposure to ash-laden bottom sediments on fish embryo–larval development were published in the *Bulletin of Environmental Contamination and Toxicology*. The results of related studies—examining the effects of both in situ and laboratory exposures of fish to ash from the spill on the offspring of exposed adults—were published as ORNL technical reports in 2014, with journal articles based on these reports currently in preparation or under revision for journal submission. Table 6, which is adapted from one of these draft journal manuscripts, demonstrates some of the results of a detailed reanalysis begun in 2014 of the results of an in vitro spawning study with redear sunfish that originally was summarized in one of the 2014 ORNL technical reports. This reanalysis includes additional study end-points and reinterpretations to those previously presented in the ORNL Technical Report. The results clearly demonstrate that the eggs and offspring of the redear sunfish—a species of fish that has exhibited the greatest overall bioaccumulation potential of the various species regularly monitored following the ash spill—do not differ significantly between populations at ash-exposed or unexposed reference sites in various measures of early developmental success, including fertilization success, hatching success, embryo–larval survival, and incidences of larval deformities. These and similar results provide evidence, in addition to that obtained from the strictly field-based reproductive assessments, that the residual ash that remains following remediation appears unlikely to pose significant long-term risks to the reproductive success of exposed fish populations in the upper Watts Bar system.

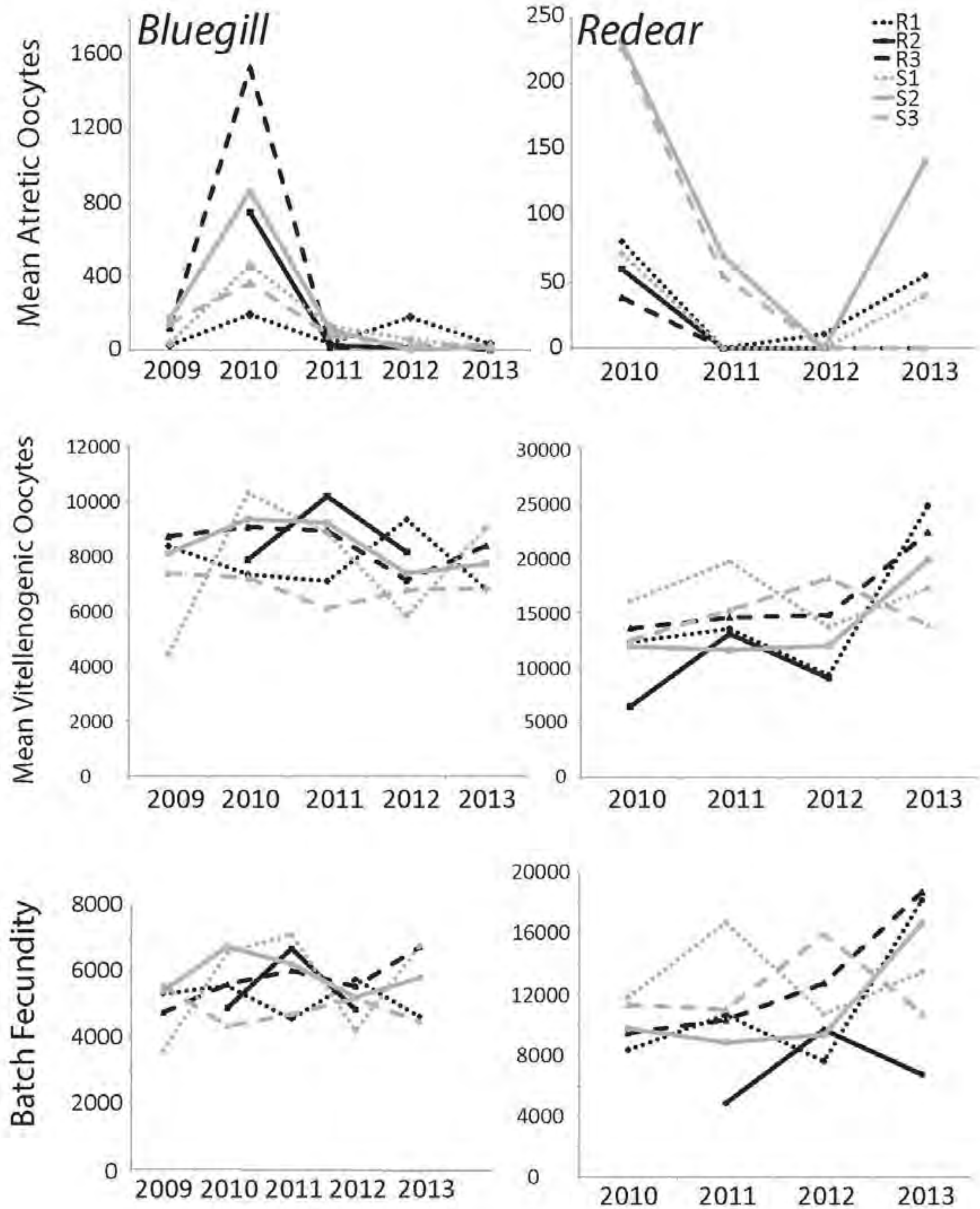


Fig. 7. Year-by-year mean variation in reproductive parameters measured in bluegill and redear sunfish from 2009 through 2013 at various reference and ash-exposed study sites in the Emory and Clinch Rivers. Reference site 1 (R1) = Emory River mile 8.0; R2 = Little Emory River mile 2.0; R3 = Clinch River mile 8.0; study site 1 (S1) = Emory River mile 3.0; S2 = Emory River mile1.0; and S3 = Clinch River mile 1.5.

Table 6. Percent fertilization success, hatching success, survival, and deformities of redear sunfish embryos and larvae in 7-d laboratory tests following in vitro spawning of fish collected from coal ash-exposed and reference sites in the Emory and Clinch Rivers, Tennessee

Results are expressed as mean percentages ± standard errors of the means

Fish sites	Spawning trials	Fertilization success		Hatching success		Survival		Deformities	
Water Source =		***Site***	***Reference***	***Site***	***Reference***	***Site***	***Reference***	***Site***	***Reference***
ERM 8.0	7	98.7 ± 0.4	97.7 ± 0.4	96.1 ± 1.9	95.3 ± 2.9	90.6 ± 3.5	90.4 ± 4.9	1.7 ± 0.9	1.1 ± 0.6
ERM 3.0	14	95.0 ± 1.4	95.9 ± 0.9	97.3 ± 1.6	98.4 ± 0.6	83.9 ± 6.5	81.1 ± 7.5	2.9 ± 0.9	2.4 ± 0.8
ERM 1.0	8	93.6 ± 3.0	95.9 ± 1.3	98.3 ± 0.9	98.3 ± 0.9	79.4 ± 7.6	91.0 ± 2.1	2.0 ± 0.7	2.4 ± 0.7
CRM 8.0	6	93.0 ± 4.2	96.0 ± 0.9	97.2 ± 1.4	99.0 ± 0.5	83.3 ± 4.4	86.8 ± 4.3	3.8 ± 1.5	2.0 ± 0.5
CRM 1.5	8	99.3 ± 0.3	97.1 ± 1.5	98.9 ± 0.4	98.5 ± 0.5	95.8 ± 0.8	92.5 ± 2.2	1.9 ± 0.6	1.0 ± 0.4

Notes: ERM = Emory River mile; CRM = Clinch River mile.

3. INVERTEBRATE BIOACCUMULATION

Activities related to KIF invertebrate bioaccumulation studies in 2014 were much reduced compared with 2009–2013 because 2014 marked the initial year of TVA's long-term monitoring program for the KIF coal ash spill. Assessment of invertebrate bioaccumulation in 2014 was limited to a single site, ERM 1.0, which is the site previously shown to have the highest concentrations of ash-related contaminants. Additionally, bioaccumulation was assessed in only the nymph and adult stages of mayflies (*Hexagenia bilineata*) in 2014. Although several ash-related contaminants were found in snails (*Pleurocera canaliculatum*) during the first 5 years of monitoring, bioaccumulation trends in snails were more variable and ambiguous than those found for mayflies.

Samples of mayfly nymphs were collected from ERM 1.0 in May 2014. As in previous years, samples were collected for analysis of depurated and non-depurated nymphs. Processing of samples was completed in late July and submitted to the analytical laboratory for analysis on August 12, 2014.

The emergence pattern of adult mayflies in 2014 provided opportunities to collect specimens for samples on three separate occasions: June 9, June 18–20, and July 3. The collection of adults on June 9 was one of the earliest times adults have been collected since 2009; however, their abundance was low at that time. Much larger "hatches" occurred during the second and third collections, with the collection in mid-June dominated by male imagos and the collection in July primarily a mixture of male and female subimagos. As has often occurred in past years, there were insufficient numbers/mass of female imagos collected for analysis; thus, only samples of male and female subimagos and male imagos were analyzed. Sample processing was completed in late July, and the samples were submitted to the analytical contract laboratory for analysis on August 12, 2014.

Summary descriptive statistics for mayfly nymph samples (non-depurated and depurated) collected at ERM 1.0 in 2014 are presented in Table 7. Results for non-depurated nymphs from the first 4 years (during the period when samples were analyzed for 26 elements) have shown that arsenic and selenium were both good tracers for the coal ash. Of those elements, results from 2014 indicate that selenium appears to be the only analyte detected at notably lower concentrations in non-depurated nymphs compared with previous years (e.g., 5.43 µg/g dry mass in 2014 vs. 6.63 µg/g dry mass in 2013) (Fig. 8). However, selenium remains somewhat elevated above the long-term average for non-depurated nymphs from the reference sites (5-year average from four reference sites = 3.27 µg/g dry mass). Concentrations of arsenic in non-depurated nymphs were similar to those found in 2013 and continued to be about two times higher than the mean for the four reference sites from 2009 to 2013. In contrast to non-depurated nymphs, concentrations of arsenic in depurated nymphs were 25–35% lower in 2014 than in 2013, although they were still 1.25 to 2.0 times higher than the long-term means for the reference sites. Concentrations of arsenic in both groups of nymphs (10.47 µg/g dry mass for non-depurated and 2.27 µg/g dry mass for depurated nymphs) remain elevated above the long-term mean for reference sites (3.81 µg/g dry mass and 1.31 µg/g dry mass, respectively). However, they were well below the lower range of the Environmental Protection Agency remediation goal set for that metal (i.e., 34 µg/g dry mass).

Summary descriptive statistics for adult mayfly samples (female subimagos, male imagos, and male subimagos) collected at ERM 1.0 in 2014 are presented in Table 8. Results for adult mayflies from the first 5 years of monitoring have indicated that several elements potentially associated with coal ash were above background concentrations derived from reference sites. However, concentrations of most elements were much lower than those found in nymphs, and those found in concentrations similar to or higher than in nymphs were predominantly essential elements such as copper, iron, and zinc. Selenium was the only element of concern found at concentrations that were similar to or in some cases higher than the concentrations in nymphs. Based on mean concentrations calculated from all adult groups combined, selenium concentrations were notably lower in 2014 (5.00 µg/g dry mass) than in 2013 (6.64 µg/g dry mass) but still were higher than the long-term mean for concentrations found in adult mayflies from the four reference sites (3.22 µg/g dry mass) (Fig. 9).

Table 7. Summary statistics for mayfly nymphs from samples collected at ERM 1.0 in 2014

Depurated?	Analyte	Number of samples	Number of detects	Mean	Medium	Minimum	Maximum	Standard deviation	Standard error
No	Arsenic	3	3	10.5	9.8	9.6	12	1.33	0.77
	Barium	3	3	71.5	73.4	61.8	79.4	8.95	5.17
	Cadmium	3	3	1.2	1.2	1.2	1.2	0	0
	Chromium	3	3	9.37	9.3	8.1	10.7	1.30	0.75
	Copper	3	3	18.7	18.7	17.8	19.7	0.95	0.55
	Iron	3	3	9690.0	10000	8470	10600	1098.3	634.1
	Manganese	3	3	592.7	607	466	705	120.1	69.4
	Mercury	3	3	0.123	0.12	0.12	0.13	0.0058	0.0033
	Selenium	3	3	5.43	5.4	5.3	5.6	0.153	0.088
	Strontium	3	3	19.9	18.9	18.1	22.7	2.46	1.42
	Thallium	3	3	0.187	0.19	0.16	0.21	0.025	0.015
	Vanadium	3	3	15.5	15.4	13.6	17.5	1.95	1.13
	Zinc	3	3	176.7	176	176	178	1.15	0.67
Yes	Arsenic	3	3	2.27	2.4	2	2.4	0.231	0.133
	Barium	3	3	13.2	12.3	11.3	16	2.48	1.43
	Cadmium	3	3	1.47	1.5	1.2	1.7	0.252	0.145
	Chromium	3	3	1.08	1.1	0.85	1.3	0.225	0.130
	Copper	3	3	31.8	31.6	30.5	33.4	1.46	0.85
	Iron	3	3	1293.3	1330	1190	1360	90.7	52.4
	Manganese	3	3	134.0	125	123	154	17.3	10.0
	Mercury	3	3	0.128	0.099	0.074	0.21	0.072	0.042
	Selenium	3	3	4.37	4.4	4.1	4.6	0.252	0.145
	Strontium	3	3	6.47	5.9	5.9	7.6	0.981	0.567
	Thallium	3	0						
	Vanadium	3	3	1.93	1.9	1.5	2.4	0.451	0.260
	Zinc	3	3	211.3	213	195	226	15.6	9.0

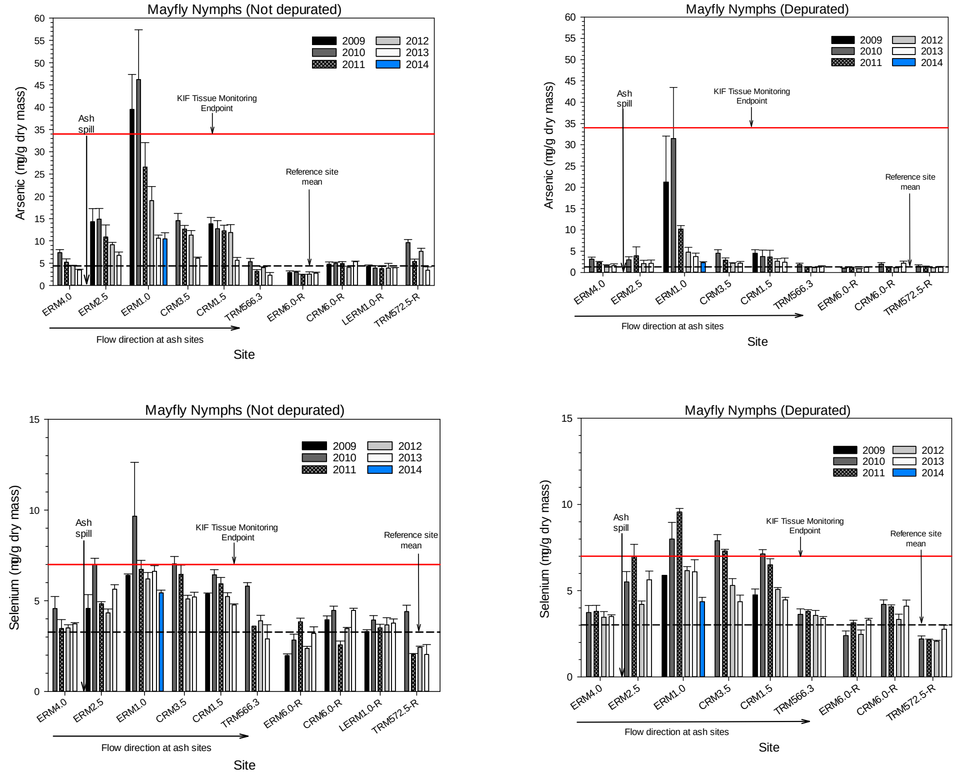
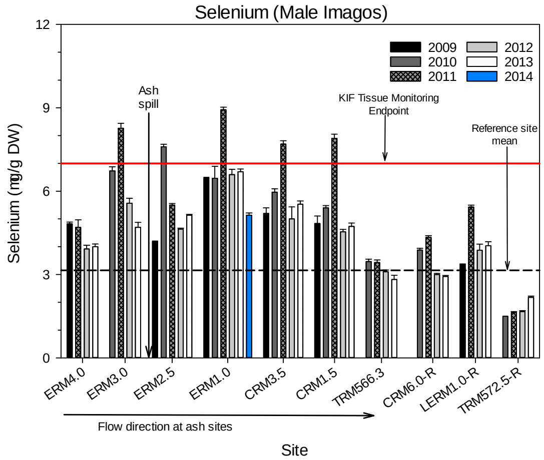


Fig. 8. Mean concentrations (±1 standard deviation) of arsenic and selenium in non-depurated (left) and depurated (right) mayfly nymphs (*Hexagenia bilineata*), 2009–2014; samples were collected at only Emory River mile (ERM) 1.0 in 2014.
Site names followed by "R" are reference sites.

Table 8. Summary statistics for adult mayflies from samples collected at ERM 1.0 in 2014

Analyte	Number of samples	Number of detects	Mean	Medium	Minimum	Maximum	Standard deviation	Standard error
Female subimagos								
Arsenic	3	3	0.197	0.19	0.19	0.21	0.0115	0.0067
Barium	3	3	0.807	0.8	0.7	0.92	0.1102	0.0636
Cadmium	3	3	0.26	0.25	0.18	0.35	0.0854	0.0493
Chromium	3	3	0.12	0.12	0.11	0.13	0.01	0.0058
Copper	3	3	13.7	13.4	13.3	14.4	0.6083	0.3512
Iron	3	3	95.3	93.7	92.3	100	4.1016	2.3681
Manganese	3	3	5.43	4	3.6	8.7	2.8361	1.6374
Mercury	3	3	0.047	0.047	0.042	0.051	0.0045	0.0026
Selenium	3	3	5.37	5.3	5.2	5.6	0.2082	0.1202
Strontium	3	3	0.693	0.72	0.62	0.74	0.0643	0.0371
Thallium	3	0						
Vanadium	3	3	0.074	0.074	0.071	0.078	0.0035	0.0020
Zinc	3	3	228.3	230	215	240	12.6	7.3
Male imagos								
Arsenic	3	3	0.2	0.2	0.18	0.22	0.0200	0.0115
Barium	3	3	1.047	1.1	0.84	1.2	0.186	0.107
Cadmium	3	3	0.447	0.5	0.2	0.64	0.225	0.130
Chromium	3	2	0.220	0.22	0.16	0.28	0.085	0.060
Copper	3	3	28.2	27.8	27.4	29.3	1.002	0.578
Iron	3	3	205.3	208	191	217	13.2	7.6
Manganese	3	3	2.6	2.1	2	3.7	0.954	0.551
Mercury	3	3	0.081	0.081	0.08	0.082	0.0010	0.0006
Selenium	3	3	5.13	5.1	5	5.3	0.153	0.088
Strontium	3	3	0.803	0.74	0.72	0.95	0.127	0.074
Thallium	3	0						
Vanadium	3	3	0.207	0.23	0.13	0.26	0.0681	0.0393
Zinc	3	3	94.6	92.2	91.9	99.6	4.36	2.52
Male subimagos								
Arsenic	3	3	0.203	0.2	0.19	0.22	0.015	0.009
Barium	3	3	1.57	1.5	1.3	1.9	0.31	0.18
Cadmium	3	3	0.24	0.23	0.18	0.31	0.066	0.038
Chromium	3	2	0.125	0.125	0.12	0.13	0.007	0.005
Copper	3	3	25.1	25.6	23.7	25.9	1.19	0.69
Iron	3	3	158.3	158	158	159	0.58	0.33
Manganese	3	3	3.27	2.4	2.3	5.1	1.59	0.92
Mercury	3	3	0.054	0.053	0.052	0.056	0.002	0.001
Selenium	3	3	4.5	4.4	4.4	4.7	0.17	0.10
Strontium	3	3	1.4	1.4	1.4	1.4	0	0
Thallium	3	0						
Vanadium	3	3	0.093	0.092	0.088	0.1	0.006	0.004
Zinc	3	3	97.4	97.3	94.9	100	2.55	1.47



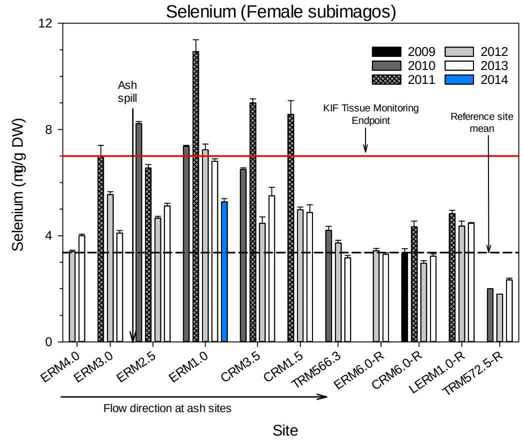


Fig. 9. Mean concentrations (±1 standard deviation) of selenium in adult male imago (left) and female subimago (right) mayflies (*Hexagenia bilineata*), 2009–2014; samples were collected at only Emory River mile (ERM) 1.0 in 2014. *Note:* DW – Dry weight.

Results are shown only for male imagos and female subimagos because those groups provided the most comprehensive temporal coverage for each sex. Sites names followed by "R" are reference sites.

4. 2015 PROGRESS

4.1 FISH

Fish were sampled from five sites on the Emory and Clinch Rivers in April to June 2015 for analysis of metal bioaccumulation, fish health, and fish reproductive fitness. The objective was to collect fish at or near the beginning of their respective breeding seasons—from sample sites that included ERM 8.0, ERM 3.0, ERM 1.0, CRM 8.0, and CRM 1.5—to investigate possible relationships among metal bioaccumulation, fish health, and reproductive fitness. Sites represented ash-affected areas along both rivers (ERM 3.0, ERM 1.0, and CRM 1.5), a primary reference area on the Emory River upstream of the ash release location (ERM 8.0), and a positive control site (CRM 8.0) located on the Clinch River downstream of US Department of Energy facilities in Oak Ridge and upstream of the influence of the Kingston coal ash release. Target sample sizes for fish health and reproductive fitness analyses were eight adult females of each of three fish species—largemouth bass, redear sunfish, and bluegill—that represent different trophic levels and home ranges. Actual numbers of fish collected from each site (Table 9) varied in certain cases because of fish availability or the need to resample to better standardize fish sizes or status within the respective breeding seasons among the various sample sites.

A variety of fish health parameters were evaluated in each fish, including the determination of an overall health assessment index, measures of bioenergetic condition based on the general condition of the fish and internal organs, and analysis of blood chemistry and hematology. For reproductive fitness evaluations, ovary weights and overall condition were assessed. Also, representative ovarian tissue samples were preserved for later morphometric analyses, including determinations of the abundance and developmental stages of the developing oocytes, frequency of oocyte atresia, abundance and condition of mature eggs (if present), and estimates of clutch size and/or total fecundity (as applicable). Six of the sampled fish of each species were filleted for each site, and muscle tissue, ovaries, and livers were processed and submitted for metals analysis.

Table 9. Fish sampling summary for spring 2015

Site	Species	Total samples	Bioaccumulation	Fish health	Fish reproduction
ERM 8.0	Bluegill	7	6	7	7
	Largemouth bass	8	6	8	8
	Redear sunfish	10	6	10	8
ERM 3.0	Bluegill	10	6	10	8
	Largemouth bass	8	6	8	8
	Redear sunfish	10	6	10	8
ERM 1.0	Bluegill	10	6	10	8
	Largemouth bass	8	6	8	8
	Redear sunfish	9	6	9	8
CRM 8.0	Bluegill	9	6	9	8
	Largemouth bass	8	6	8	8
	Redear sunfish	10	6	10	8
CRM 1.5	Bluegill	11	6	11	8
	Largemouth bass	8	6	8	8
	Redear sunfish	12	6	12	8
Totals		138	90	138	119

Notes: ERM = Emory River mile; CRM = Clinch River mile.

4.2 INVERTEBRATES

A summary of the sites and groups of invertebrates sampled in 2015 is given in Table 10. As in 2014, only the nymph and adult stages of mayflies were collected; no samples of snails were collected. Sample collection began in early May and was completed by mid-July. Processing of adult mayfly samples began in July and continued in August; processing of nymph samples began in August and was not complete in time for inclusion in this report.

Table 10. Sample collection sites in 2015

Site	Nymphs		Adults
	Not depurated	Depurated	
ERM 6.0	X	X	X
ERM 4.0	X		X
ERM 3.0	X		X
ERM 2.5	X	X	X
ERM 1.0	X	X	X
CRM 6.0	X		X
CRM 3.5	X	X	X

Notes: ERM = Emory River mile; CRM = Clinch River mile.

4.3 RECENT PUBLICATIONS

- Bevelhimer, M. S., S. M. Adams, A. M. Fortner, M. S. Greeley, and C. C. Brandt. 2014. "Using Ordination and Clustering Techniques to Assess Multi-Metric Fish Health Response Following a Coal Ash Spill." *Environmental Toxicology and Chemistry* 33:1903–1913.
- Greeley Jr., M. S., L. R. Elmore, and M. K. McCracken. 2014. *Evaluating the Effects of the Kingston Fly Ash Release on Fish Reproduction and Early Life Stages: Long-Term Exposures to Fly Ash in the Laboratory*. ORNL/TM-2013/11, Oak Ridge National Laboratory, Oak Ridge, TN. Available from http://info.ornl.gov/sites/publications/Files/Pub40944.pdf.
- Greeley Jr., M. S., L. R. Elmore, and M. K. McCracken. 2014. *Evaluating the Effects of the Kingston Fly Ash Release on Fish Reproduction and Early Life Stages: In Vitro Spawning Study*. ORNL/TM-2013/10, Oak Ridge National Laboratory, Oak Ridge, TN. Available from http://info.ornl.gov/sites/publications/Files/Pub40943.pdf.
- Greeley, M. S., L. R. Elmore, M. K. McCracken, and R. M. Sherrard. 2014. "Effects of Sediment Containing Coal Ash from the Kingston Ash Release on Embryo–Larval Development in the Fathead Minnow, *Pimephales promelas* (Rafinesque, 1820)." *Bulletin of Environmental Contamination and Toxicology* 92:154–159.
- Mathews, T., A. Fortner, R. T. Jett, J. G. Morris, J. Gable, M. Peterson, and N. Carriker. 2014. "Selenium Bioaccumulation in Fish Exposed to Coal Ash at the Tennessee Valley Authority Kingston Spill Site." *Env. Toxicol. Chem.* 33:2273–2279.
- Pracheil, Brenda, and Terry Mathews. 2014. "Assessing Bioaccumulation at Kingston Site," podcast audio, http://www.ornl.gov/ornl/news/features/2014/lasers-fish-ears-and-environmental-change.
- Pracheil, B. M., T. Mathews, M. S. Bevelhimer, M. J. Peterson, M. S. Greeley Jr., A. M. Fortner, and C. A. Murphy. "Relating Fish Health and Reproductive Metrics to Metal Bioaccumulation at the Tennessee Valley Authority Kingston Coal Ash Spill Site." *Ecotoxicology.* Accepted, in revision.
- Rigg, D., M. Wacksman, J. Iannuzzi, T. F. Baker, S. M. Adams, and M. Greeley Jr. 2015. "Assessing Ecological Risks to the Fish Community from Residual Coal Fly Ash in Watts Bar Reservoir, Tennessee." *Integrated Assessment and Management* 11:88–101. doi: 10.1002/ieam.1588.
- Smith, J. G., T. F. Baker, C. A. Murphy, and R. T. Jett. "Spatial and Temporal Trends in Contaminants Concentrations in *Hexagenia* Nymphs Following a Coal Ash Spill at the Tennessee Valley Authority's Kingston Fossil Plant." Accepted for publication in *Environmental Toxicology and Chemistry* March 31, 2015. Revision submitted July 31, 2015.
- Sherrard, R. M., N. E. Carriker, and M. S. Greeley Jr. 2015. "How Toxic Is Coal Ash? A Laboratory Toxicity Case Study." *Integrated Assessment and Management* 11 (5–9). doi: 10.1002/ieam.1587.

ARCADIS

Appendix C

2014 TVA Benthic Invertebrate
Community Survey

Benthic Invertebrate Community

Benthic invertebrate community evaluations in November 2014 were conducted on the lower Emory River at two transect locations (ERM 1.0 and ERM 0.7). Monitoring continues to be performed annually at these locations as part of the long-term monitoring plan because they are within the reach of the Emory River with the highest potential for aquatic life exposure to residual ash. Annual monitoring was reduced to biennial in 2013 at nine additional locations on the Emory (5 sites) and Clinch (4 sites) Rivers (Table 1). All locations will be evaluated again in autumn 2015.

As in previous years, 10 equally-spaced Ponar grab samples were collected along each transect and submitted for laboratory processing and identification of organisms to the lowest practical taxonomic level. The total number of each taxa was tallied and used to generate benthic invertebrate community metrics in order to assess the status/response of the community. Water depth also was recorded for each sample along with estimates of proportions of substrate types. In addition, a sample of sediment co-located with each benthic community sample was collected and analyzed for percent ash.

The 2014 benthic community results for ERM 1.0 and ERM 0.7 are consistent with previous years as the community metrics do not show substantial impacts attributable to the ash release. Invertebrate population density and taxa richness at both sites were similar to previous years as were the dominant taxa groups and proportions among feeding guilds and organism habits (Table 2; Figures 1-6). Additionally, results for ash-impacted sites in both the Emory and Clinch Rivers over the 2009-2014 period do not indicate a trend of decreasing invertebrate abundance or decreasing richness and the temporal variations seen at these sites are also evident at reference sites. Furthermore, the most recent analysis (ANCOVA) examining the effects of site, sampling period (2012-2014), and substrate types on benthic community metrics did not indicate a significant negative relationship with percentage ash composition (Table 3). Combined, these results indicate that the structure and function of the benthic community has not been substantially altered and that any adverse effects of the residual ash in the river system are apparently small enough that the long-term viability of the population is not impacted.

Percent ash in sediments co-located with benthic community samples in 2014 ranged from 1 to 40% (mean 19.8%) at ERM 1.0 and 2 to 42% (mean 17.7%) at ERM 0.7 (Figure 7). Laboratory sediment toxicity tests indicate that only river sediments containing greater than 40% ash are likely to cause toxicity to benthic fauna (ARCADIS 2012[1]). For the 10 co-located sediment samples collected at each location annually from 2012 through 2014, the frequency of samples with ash composition equal to or greater than this threshold (40% ash) declined from 7 and 6 samples respectively at ERM 0.7 and ERM 1.0 in 2012 to only 1 or 2 samples at each location in 2013 and 2014. As residual ash is distributed unevenly through the system, these results suggest that ash and natural sediments are becoming more intermixed within the upper 6 inches of sediment.

---

[1] ARCADIS. 2012. River system baseline ecological risk assessment, Tennessee Valley Authority, Kingston Ash Recovery Project. EPA-AO-050. May.

Table 1. Benthic invertebrate community and co-located sediment sampling sites, 2009-2014.
(X-Benthic invertebrate; XX-Benthic invertebrate and co-located sediments)

Sampling Period	1 January 2009	2 December 2009	3 December 2010 - January 2011	4 December 2011 - January 2012	5 November- December 2012	6 December 2013	7 November 2014
River Mile							
ERM 6.0*	X	X	X	XX	XX	XX	.
ERM 5.0	X	X	X	XX	XX	.	.
ERM 4.1	.	X	X	XX	XX	XX	.
ERM 3.5	.	.	.	XX	XX	.	.
ERM 3.0	.	.	X	XX	XX	XX	.
ERM 2.6	.	.	X	XX	XX	XX	.
ERM 2.2	.	X	X	XX	XX	XX	.
ERM 1.0	X	X	X	XX	XX	XX	XX
ERM 0.7	.	.	.	XX	XX	XX	XX
CRM 8.7*	X	X	X	X	X	.	.
CRM 6.0*	X	X	X	X	X	XX	.
CRM 4.0	X	X	X	X	X	XX	.
CRM 3.0	X	X	X	X	X	XX	.
CRM 1.5	X	X	X	X	X	XX	.
CRM 0.5	X	X	X	X	X	.	.
TRM 573.9*	X	X	X	X	.	.	.
TRM 566.3	X	X	X	X	.	.	.
TRM 560.8[1]	X	X	X	X	X	.	.

* - Reference Site.

1 - TRM 560.8 is sampled as part of TVA's Valley-wide monitoring program. Samples were collected at this site in November 2008, 2009 and 2010; December 2011; and October 2012.

Table 2. Benthic invertebrate community metric results for long-term monitoring locations on the Emory River, 2009-2014

	Transect	**ERM 6.0**						**ERM 4.1**					**ERM 3.0**				**ERM 2.6**			
	Sample Period	**1**	**2**	**3**	**4**	**5**	**6**	**2**	**3**	**4**	**5**	**6**	**3**	**4**	**5**	**6**	**3**	**4**	**5**	**6**
	Sample Period	**Jan-09**	**Dec-09**	**Dec-10**	**Dec-11**	**Dec-12**	**Dec-13**	**Dec-09**	**Dec-10**	**Dec-11**	**Dec-12**	**Dec-13**	**Dec-10**	**Dec-11**	**Dec-12**	**Dec-13**	**Dec-10**	**Dec-11**	**Dec-12**	**Dec-13**
	Number of Samples	**10**	**10**	**10**	**10**	**10**	**10**	**10**	**10**	**10**	**10**	**10**	**20**	**10**	**10**	**10**	**20**	**10**	**10**	**10**
Average Abundance (# / m^2)	Population	752	737	452	707	1612	1135	988	2142	1300	1547	1748	2433	2258	1558	3390	2093	2157	2175	3690
	Oligochaetes	273	110	48	402	335	317	255	507	592	517	722	1108	1407	332	1668	628	743	735	1995
	Chironomids	375	415	227	247	987	557	552	1107	422	645	747	1056	697	518	1053	1219	948	662	993
	Hexagenia	9	12	5	3	5	12	10	35	37	28	52	20	13	33	40	55	55	58	65
	Sphaeriidae	0	0	0	0	0	0	5	0	22	80	15	94	40	415	337	53	255	402	423
Average Composition	% Oligochaetes	35	15	19	55	19	24	23	26	41	30	34	41	61	19	39	23	33	31	41
	% Chironomids	50	57	48	35	59	48	56	48	36	40	44	47	31	34	35	56	43	33	32
Taxa Richness	Total Richness	26	23	23	28	27	43	45	41	57	26	38	47	43	32	44	39.5	41	30	37
	Average Richness	8.6	7.7	5.9	8.0	7.6	12.8	11.5	11.3	12.3	11.4	13.4	13.5	12.8	11.7	16.7	13.9	13.9	12.2	13.1
	Total EPT Richness	2	2	3	1	2	5	6	5	5	2	3	3	2	2	4	3	1	2	2
	Average EPT Richness	0.5	0.5	0.4	0.2	0.3	1.1	1.0	1.1	0.8	0.8	1.1	0.7	0.3	1.0	1.5	1.0	0.7	0.8	0.9
Feeding	% Filterer	2	10	7	5	5	5	20	7	11	8	6	8	10	26	16	7	21	20	16
	% Gatherer	76	57	53	65	70	57	54	63	56	60	68	57	71	42	56	53	52	52	59
	% Predator	21	33	29	17	23	32	24	26	29	29	25	28	10	25	27	33	23	26	24
	% Scraper	--	0	0	5	--	0	0	0	2	0	--	0	1	0	0	2	0	0	0
	% Shredder	0	0	5	7	1	3	1	1	2	2	1	5	7	1	1	5	3	0	--
	% Piercer	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--
	% Parasite	1	--	5	0	2	2	0	3	1	1	1	0	1	6	1	0	0	2	1
	% Omnivore	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--

	Transect	**ERM 2.2**					**ERM 1.0**							**ERM 0.7**			
	Sample Period	**2**	**3**	**4**	**5**	**6**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**4**	**5**	**6**	**7**
	Sample Period	**Dec-09**	**Dec-10**	**Dec-11**	**Dec-12**	**Dec-13**	**Jan-09**	**Dec-09**	**Dec-10**	**Dec-11**	**Dec-12**	**Dec-13**	**Nov-14**	**Dec-11**	**Dec-12**	**Dec-13**	**Nov-14**
	Number of Samples	**10**	**20**	**10**	**10**	**10**	**10**	**10**	**20**	**10**	**10**	**10**	**10**	**10**	**10**	**10**	**10**
Average Abundance (# / m^2)	Population	1315	2073	1823	3203	4483	967	1108	1577	2832	1462	1155	1857	2648	2182	2333	2667
	Oligochaetes	260	543	1190	1335	2520	373	465	689	1025	277	210	892	847	590	1083	1328
	Chironomids	378	1026	508	893	1417	320	318	718	1230	632	507	475	1347	1153	853	940
	Hexagenia	168	126	23	63	73	35	115	39	168	77	143	63	193	128	177	97
	Sphaeriidae	360	104	42	657	32	163	128	64	242	372	205	287	145	70	130	160
Average Composition	% Oligochaetes	12	14	46	30	49	33	47	36	36	20	21	50	35	23	41	51
	% Chironomids	27	50	43	34	35	37	28	49	41	45	45	25	47	48	36	34
Taxa Richness	Total Richness	47	40.5	48	32	45	23	24	39.5	55	40	30	41	43	41	34	40
	Average Richness	11.0	12.5	13.4	12.7	16.1	9.0	9.2	11.2	17.6	12.6	12.7	12.8	15.1	16.0	13.8	13.5
	Total EPT Richness	4	2.5	3	2	3	1	3	3.5	3	2	2	3	2	3	2	2
	Average EPT Richness	1.5	1.0	0.6	1.0	1.1	0.6	0.9	0.9	1.1	1.1	0.9	0.9	0.6	1.5	1.0	0.9
Feeding	% Filterer	32	13	12	24	5	19	8	6	15	29	16	19	10	16	7	11
	% Gatherer	43	41	59	51	67	50	68	65	54	46	56	65	56	62	72	76
	% Predator	23	42	24	22	26	29	23	25	19	19	26	14	21	14	20	12
	% Scraper	0	0	3	0	1	0	--	0	0	1	1	0	1	4	0	1
	% Shredder	1	2	2	1	1	1	0	4	11	3	1	1	12	2	1	1
	% Piercer	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--
	% Parasite	0	2	0	1	1	1	--	1	0	2	1	1	0	1	1	0
	% Omnivore	--	--	--	--	--	--	--	--	--	--	--	0	--	--	--	0

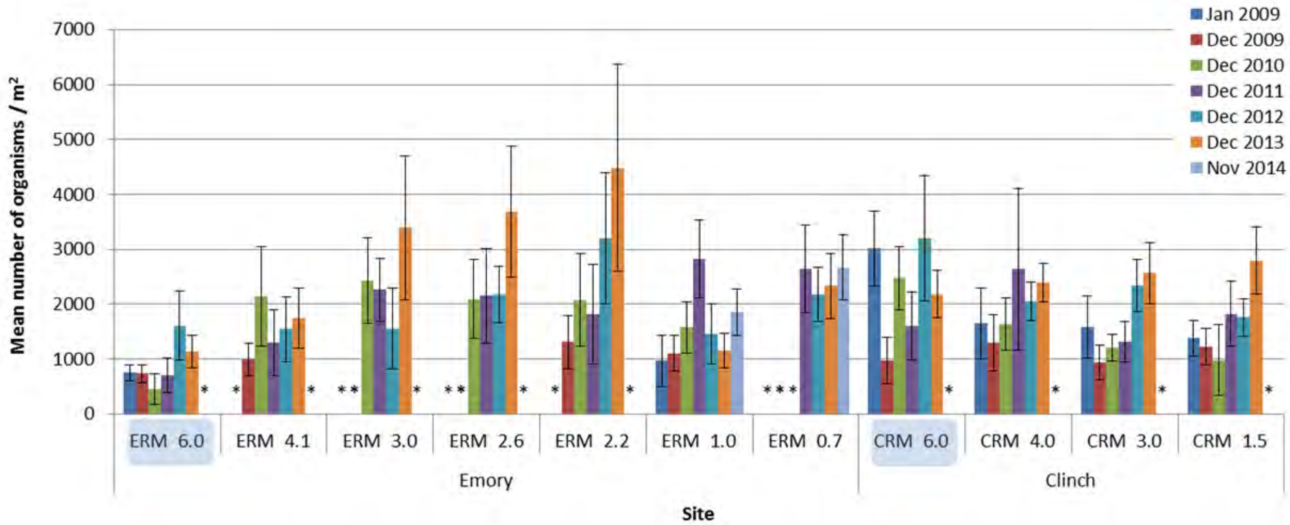
Sample periods are defined as follows: 1=January 2009; 2=December 2009; 3= December 2010-January 2011; 4=December 2011-January 2012; 5=December 2012; 6=December 2013; 7=November 2014
Abbreviations: EPT = Ephemeroptera, Plecoptera, and Trichoptera; ERM = Emory River Mile

Table 3. Summary of ANCOVA results examining effects of predominate substrate types, site, and sampling period on benthic invertebrate community metrics, 2012-2014. Overall model results are shown for each response metric as $F_{\text{num d.f., denom d.f.}}$ and P-value. Significant P-values (α=0.05) for each covariate from type III sum-of-squares are shown. Site x Period interaction was not significant for any ANCOVA.

Response Metric	Site_Group	Model Results	Ash	Detritus	Fines	Sand	Gravel	Site	Sampling Period
Population Density	Group 1	$F_{8,51}$=7.99, P<0.0001	--	0.0002 (+)	0.0115 (+)	--	0.0210 (+)	0.0139	--
	Group 2	$F_{8,49}$=8.27, P<0.0001	--	<0.0001 (+)	0.0059 (+)	--	0.0334 (+)	--	0.0003
Oligochaete Density	Group 1	$F_{8,51}$=11.60, P<0.0001	--	<0.0001 (+)	--	0.0013 (+)	--	0.0416	--
	Group 2	$F_{8,49}$=18.20, P<0.0001	--	<0.0001 (+)	--	0.0282 (+)	--	--	<0.0001
Chironomid Density	Group 1	$F_{8,51}$=6.78, P<0.0001	--	<0.0001 (+)	0.0308 (+)	--	0.0114 (+)	--	0.0043
	Group 2	$F_{8,49}$=6.07, P<0.0001	--	0.0001 (+)	--	--	--	--	0.0001
Hexagenia Density	Group 1	$F_{8,51}$=5.10, P=0.0001	--	0.0002 (-)	--	<0.0001 (-)	--	--	--
	Group 2	--	--	--	--	--	--	--	--
Sphaeriid Density	Group 1	$F_{8,51}$=6.35, P<0.0001	--	0.0467 (-)	0.0017 (+)	0.0008 (-)	--	0.0242	--
	Group 2	$F_{8,49}$=9.24, P<0.0001	--	--	<0.0001 (+)	--	--	--	--
Total Richness	Group 1	$F_{8,51}$=3.46, P=0.0030	--	--	0.0349 (+)	--	0.0004 (+)	--	--
	Group 2	$F_{8,49}$=2.72, P=0.0144	--	--	0.0232 (+)	0.0252 (+)	0.0106 (+)	--	0.0389
Gatherer	Group 1	$F_{8,51}$=9.98, P<0.0001	--	<0.0001 (+)	--	--	0.0329 (+)	0.0006	--
	Group 2	$F_{8,49}$=14.4, P<0.0001	--	<0.0001 (+)	--	--	--	--	<0.0001
Predator	Group 1	$F_{8,51}$=2.59, P=0.0187	--	0.0217 (+)	0.0130 (+)	--	--	--	--
	Group 2	$F_{8,49}$=3.86, P=0.0014	0.0014 (+)	0.0373 (+)	0.0462 (+)	--	--	--	0.0018
Filterer	Group 1	$F_{8,51}$=5.31, P<0.0001	--	--	0.0143 (+)	0.0013 (-)	--	--	0.0047
	Group 2	$F_{8,49}$=7.74, P<0.0001	--	--	<0.0001 (+)	--	0.0407 (+)	--	--
Burrower	Group 1	$F_{8,51}$=7.28, P<0.0001	--	0.0010 (+)	0.0230 (+)	--	--	0.0332	--
	Group 2	$F_{8,49}$=7.47, P<0.0001	--	<0.0001 (+)	0.0103 (+)	--	--	--	0.0018

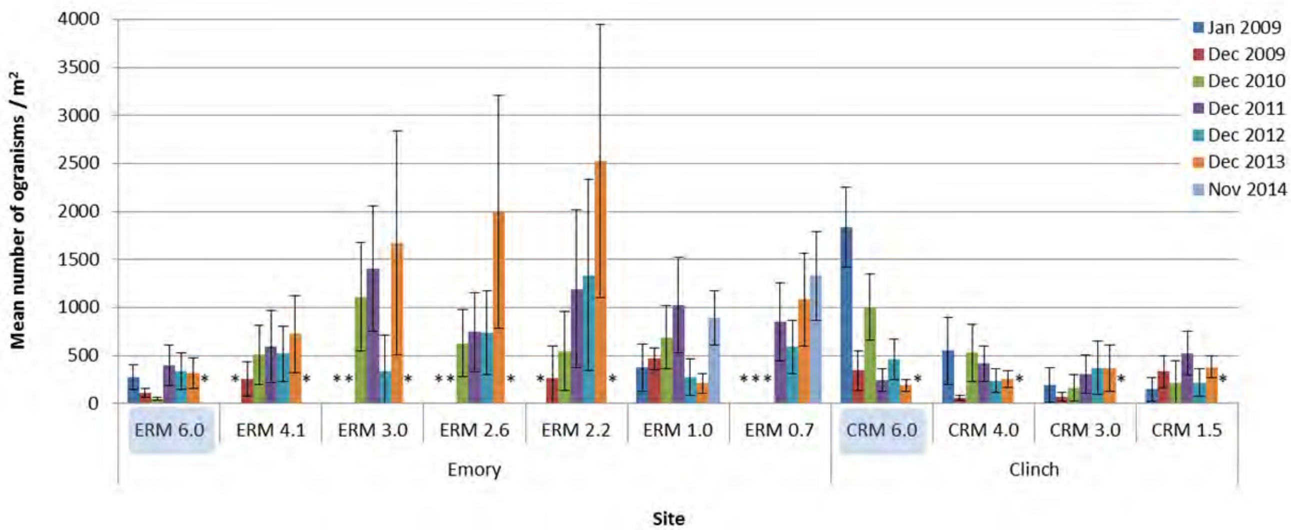
Group 1: ERM 1.0 and ERM 0.7 sampled in 2012, 2013, and 2014; Group 2: ERM 3.0, 2.6, and 2.2 sampled in 2012 and 2013. (+) positive correlation; (-) negative correlation

Analysis was performed on the 2012 to 2014 dataset for long-term monitoring sites in the Emory River. These data include results for co-located sediments for each benthic community sample (i.e., 10 samples per transect). ERM 6.0 and ERM 4.1 were excluded from the analysis because few samples had detectable ash and detection yielded low ash content. Given the complex nature of the river system, the remaining five sites were grouped based on proximity of sampling locations, similarities of river cross-sections, and estimated risk for exposure to residual ash. Two site-groups were formed, one consisting of ERM 3.0, 2.6, and 2.2 and one consisting of ERM 1.0 and ERM 0.7. Because differences between ERM 0.7 and ERM 1.0 were evident in the ANCOVA for several metrics; these sites were also assessed individually (results not shown); no significant relationship was found between percentage ash composition and benthic community metrics.



Reference Sites
Error Bars = 95% Confidence Interval
* Indicates sampling event(s) site was not sampled

Figure 1. Mean benthic invertebrate population densities at long-term monitoring locations in the Emory and Clinch Rivers, 2009-2014; only Emory River Miles (ERM) 1.0 and 0.7 were sampled in 2014.



Reference Sites
Error Bars = 95% Confidence Interval
* Indicates sampling event(s) site was not sampled

Figure 2. Mean densities of oligochaetes at long-term monitoring locations in the Emory and Clinch Rivers, 2009-2014; only Emory River Miles (ERM) 1.0 and 0.7 were sampled in 2014.

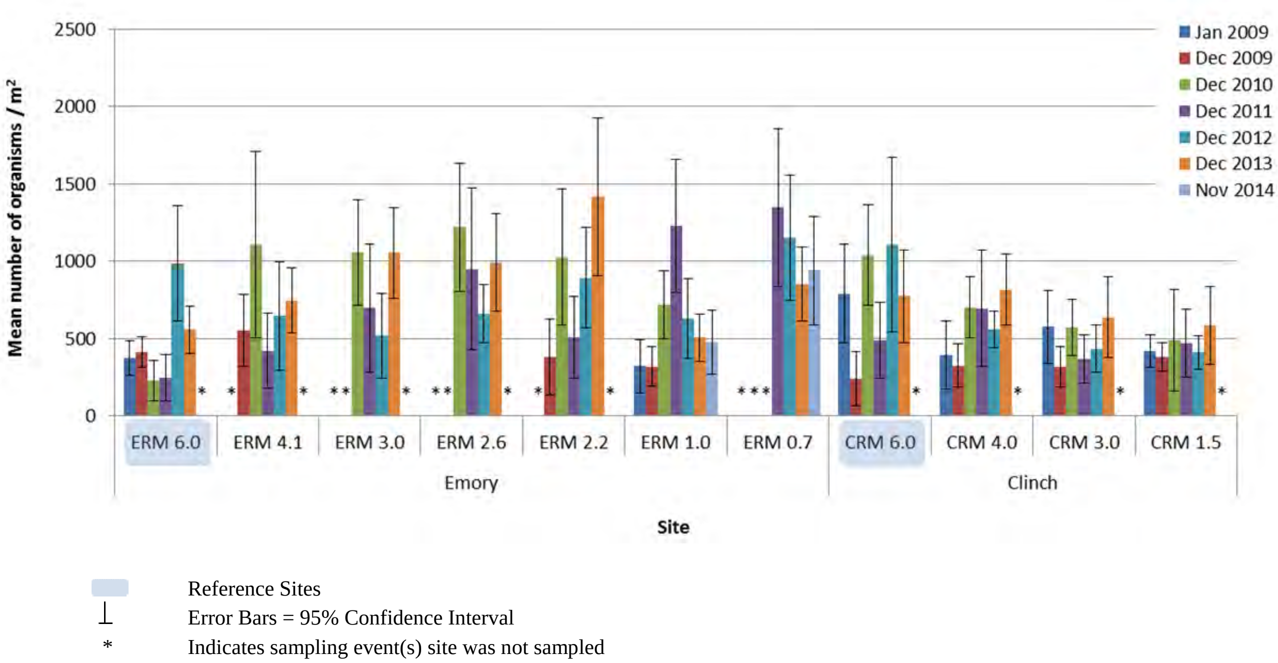


Figure 3. Mean densities of chironomids at long-term monitoring locations in the Emory and Clinch Rivers, 2009-2014; only Emory River Miles (ERM) 1.0 and 0.7 were sampled in 2014.

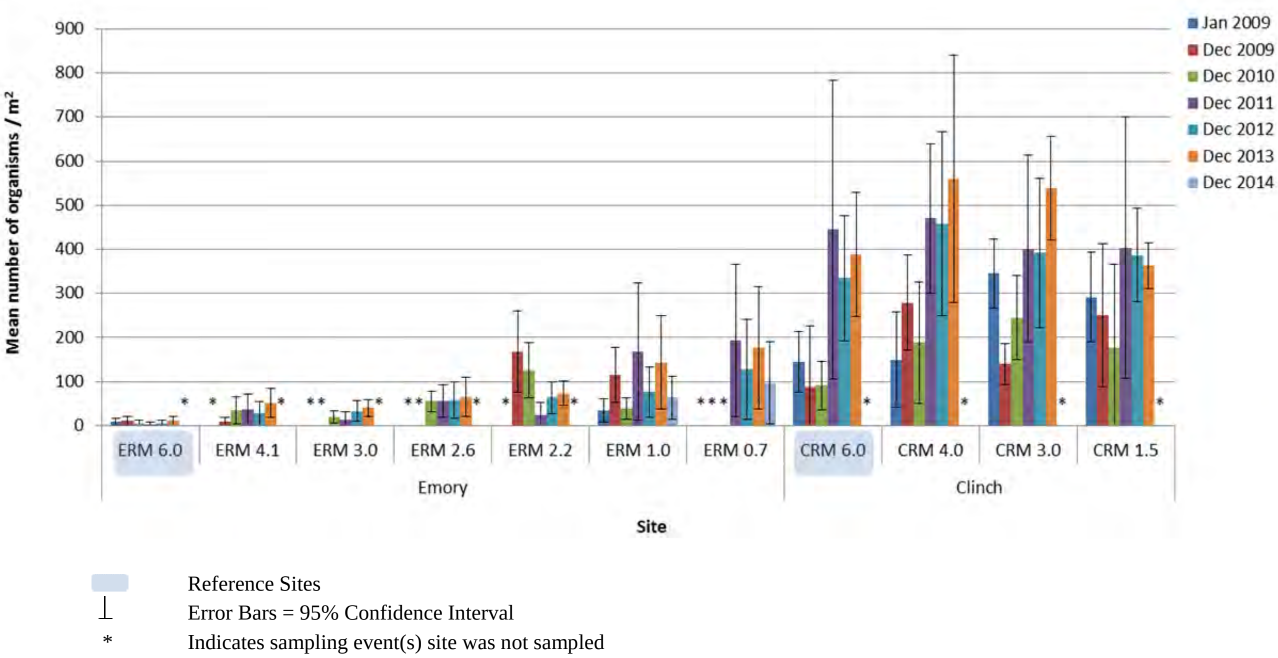
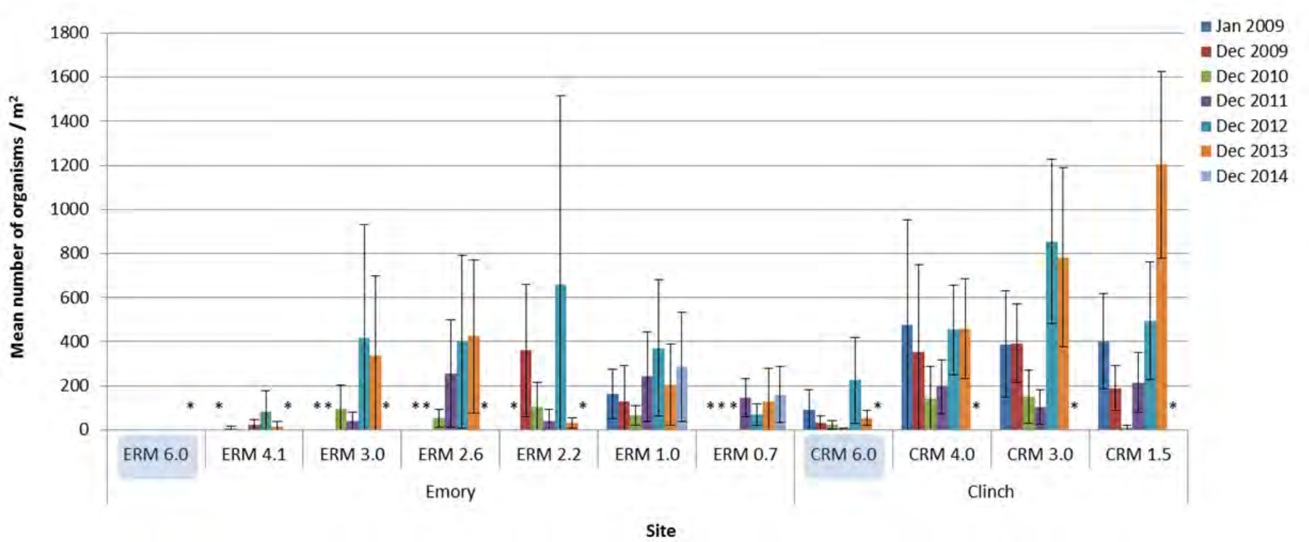
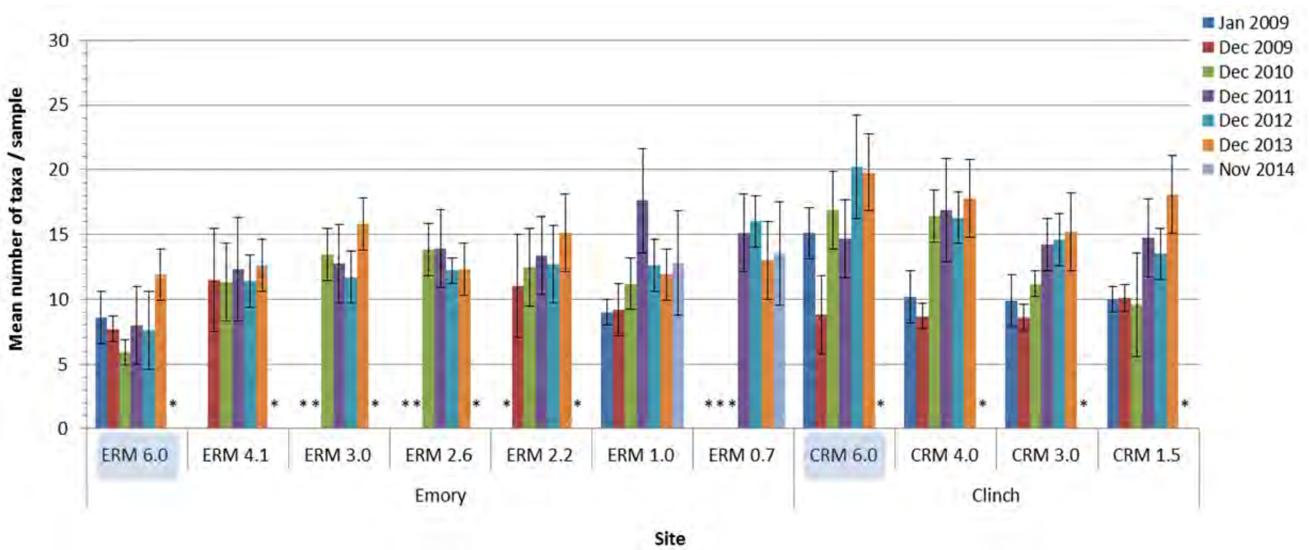


Figure 4. Mean densities of *Hexagenia* at long-term monitoring locations in the Emory and Clinch Rivers, 2009-2014; only Emory River Miles (ERM) 1.0 and 0.7 were sampled in 2014.



Reference Sites
Error Bars = 95% Confidence Interval
* Indicates sampling event(s) site was not sampled

Figure 5. Mean densities of sphaeriid clams at long-term monitoring locations in the Emory and Clinch Rivers, 2009-2014; only Emory River Miles (ERM) 1.0 and 0.7 were sampled in 2014.



Reference Sites
Error Bars = 95% Confidence Interval
* Indicates sampling event(s) site was not sampled

Figure 6. Mean number of benthic invertebrate taxa at long-term monitoring locations in the Emory and Clinch Rivers, 2009-2014; only Emory River Miles (ERM) 1.0 and 0.7 were sampled in 2014.

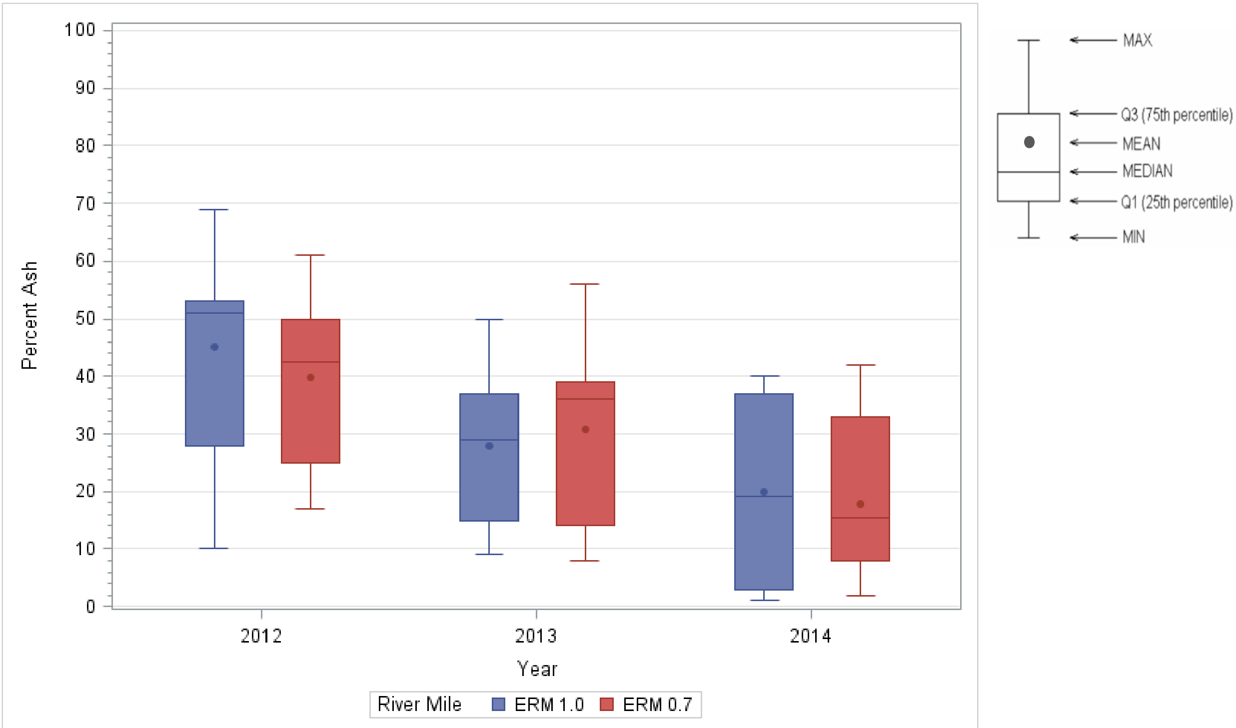


Figure 7. Percentage ash composition in sediment samples collected co-located with benthic invertebrate community samples at ERM 1.0 and ERM 0.7, 2012-2014.

Appendix D

2014 ARCADIS Tree Swallow Data Analysis Memorandum

ARCADIS
Infrastructure, environment, buildings

Imagine the result

TVA

2014 Tree Swallow Data Analysis Memorandum

Kingston, Roane County, Tennessee

August 2015

ARCADIS

Amber Stojak
Staff Environmental Scientist

Suzanne J. Walls
Senior Ecologist

2014 Tree Swallow Data Analysis Memorandum

Kingston, Tennessee

Prepared for:
Tennessee Valley Authority

Prepared by:
ARCADIS
114 Lovell Road
Suite 202
Knoxville
Tennessee 37917
Tel 865 675 6700
Fax 865 675 6712

Project Number:
TNTVAKIP.LTM5

Date:
August 2015

Tables

Figures

ARCADIS

List of Acronyms and Abbreviations

COPECs	Constituents of Potential Ecological Concern
ERM	Emory River Mile
mg/kg dw	milligrams per kilogram dry weight
TRM	Tennessee River Mile
site	Tennessee Valley Authority Kingston Fossil Plant

1. Tree Swallows

Tree swallows (*Tachycineta bicolor*) were selected as a representative aerial-feeding insectivorous bird species for Tennessee Valley Authority Kingston Fossil Plant (the site). Tree swallows are a breeding migratory resident in Tennessee, inhabiting standing cavities of dead trees, bluebird boxes, or other artificial structures (Nicholson 1997; Robinson 1990), and foraging 100 to 200 meters around their nest during the breeding season. They commonly prey on a variety of insects, and when nest boxes are placed along aquatic areas, they feed primarily on emergent aquatic insects (U.S. Geological Survey 2003; Blancher and McNicol 1991; Quinney and Ankney 1985). As a result, tree swallow tissue residues often reflect the local sediment contamination for those chemicals that transfer into the aquatic emergent insects (McCarty and Winkler 1999; Froese et al. 1998). Tree swallows serve as a useful receptor in order to understand exposure and potential effects of these constituents on the aerial-feeding insectivorous bird and mammal communities.

In 2014, tree swallow colonies were erected at two locations along the Emory River at Emory River Mile (ERM) 1.4 and ERM 3.0, and at one reference location Tennessee River Mile (TRM) 572.0 (Figure 1). Boxes were monitored daily at ERM 1.4 and TRM 572.0 from April through July. ERM 3.0 was not monitored due to the construction activity occurring in this area during the breeding season. A detailed description of the collection methods can be found in *Trace Element Concentrations and Productivity in Tree Swallows: 2009-2010* (Appendix Z, ARCADIS 2012).

The main study objectives were to 1) determine the extent of maternal transfer of metals and metalloids to the eggs between locations and across years; 2) evaluate tree swallow reproductive success among locations and years; and 3) assess impacts to tree swallows by comparing concentrations measured at the study sites with literature-derived effects values, when available.

1.1 Tree Swallow Reproduction

The total number of eggs (clutch size), the number of eggs that hatched (hatching success), the number of young that survived to day 15 (fledgling success), and the number of females fledglings produced per nesting female (fecundity) were recorded in 2014 at ERM 1.4 and TRM 572.0 colonies. In addition, egg mass and volume were recorded, as well as morphological measures (egg length and width).

In 2014, clutch size and hatching success were slightly lower at ERM 1.4 compared with TRM 572.0 (Figure 2); however, these differences were not statistically significant when evaluated using a Mann-Whitney Rank Sum Test (Sigma Plot version 12.5). ERM 1.4 had slightly greater fledgling success and fecundity compared with TRM 572.0 (Figure 2); however, differences in fledgling success and fecundity were not statistically significantly ($p>0.05$) different. No differences were observed for the egg volume or egg mass between colonies ($p>0.05$).

1.2 Tree Swallow Bioaccumulation

In 2014, tree swallow eggs were collected to evaluate exposure of tree swallows to ash-related constituents of potential ecological concern (COPECs). Eggs were of particular interest as some ash-related COPECs can be maternally transferred from the adult female to her young. When possible, the first egg laid from each available nest was collected within 3 days of clutch completion. A total of 31 eggs were collected from ERM 1.4 and 32 eggs were collected from TRM 572.0. Eggs were frozen and prepared for trace element analysis. Concentrations of arsenic and selenium in egg tissue collected in 2014 are presented in Table 1 and Figure 3. Arsenic was detected in all but two samples; however, concentrations were not significantly different between locations and are not discussed further.

A Mann-Whitney Rank Sum Test was used to compare selenium concentrations in eggs from ERM 1.4 to selenium concentrations in eggs from TRM 572.0. The results showed a statistically significant difference between the colonies ($p<0.001$), with slightly higher average concentration at ERM 1.4 (2.21 milligrams per kilogram dry weight [mg/kg dw]) compared to TRM 572.0 (1.80 mg/kg dw). When compared to previous years of data, 2014 selenium concentrations at ERM 1.4 were lower than 2013 and were similar to selenium concentrations from reference sites collected between 2009 and 2014 (Figure 2). Literature studies of selenium in eggs of other species have recently been reviewed and suggest threshold effects (EC_{10}) concentrations ranging from 7.7 to 60 mg/kg dw in various species of avian eggs (Janz et al. 2010). Similar to previous years, selenium concentrations in eggs collected from ERM 1.4 in 2014 were well below the most conservative of these literature values, indicating that it is unlikely that selenium is causing adverse effects on the tree swallow population.

ARCADIS

2. References

ARCADIS. 2012. Trace Element Concentrations and Productivity in Tree Swallows: 2009-2010. May 2012. (River System Baseline Ecological Risk Assessment - Appendix Z).

Blancher, P.J., and D.K. McNicol. 1991. Tree swallow diet in relation to wetland acidity. Can. J. Zool. 66:842-849.

Froese, K., D. Verbrugge, G. Ankley, G. Niemi, C. Larsen, and J. Giesy. 1998. Bioaccumulation of Polychlorinated Biphenyls from Sediments to Aquatic Insects and Tree Swallow Eggs and Nestlings in Saginaw Bay, Michigan, USA. Environmental Toxicology and Chemistry, 17(3): 484-492.

Janz, D.M., D.K. DeForest, J.L. Brooks, P.M. Chapman, G. Gilron, D. Hoff, W.A. Hopkins, D.O. McIntyre, C.A. Mebane, V.P. Palace, J.P. Skorupa, and M. Wayland. 2010. Chapter 6: Selenium toxicity in aquatic organisms. Pages 141-232 in: Chapman, P.M., W.J. Adams, M.L. Brooks, C.G. Delos, S.N. Luoma, W.A. Maher, H.M. Ohlendorf, T.S. Presser, and D.P. Shaw (eds.), Ecological Assessment of Selenium in the Aquatic Environment. Pensacola, Florida, USA: CRC Press and Society of Environmental Toxicology and Chemistry.

McCarty, J.P. and D.W. Winkler. 1999. Foraging ecology and diet of tree swallows feeding selectivity of nestlings. The Cooper Ornithological Society. The Condor, 101: 246-254.

Nicholson, C. 1997. Atlas of the Breeding Birds of Tennessee. Knoxville, Tennessee: The University of Tennessee Press.

Quinney, T.E. and C.D. Ankney. 1985. Prey size selection by Tree Swallows. Auk 102: 245-250.

Robinson, J. 1990. An Annotated Checklist of the Birds of Tennessee. Knoxville, Tennessee: The University of Tennessee Press.

U.S. Geological Survey. 2003. Tree Swallows: A Nationwide Sentinel Species for Assessing and Monitoring Aquatic Contamination. Fact Sheet FS 2004-3042.

ARCADIS

Tables

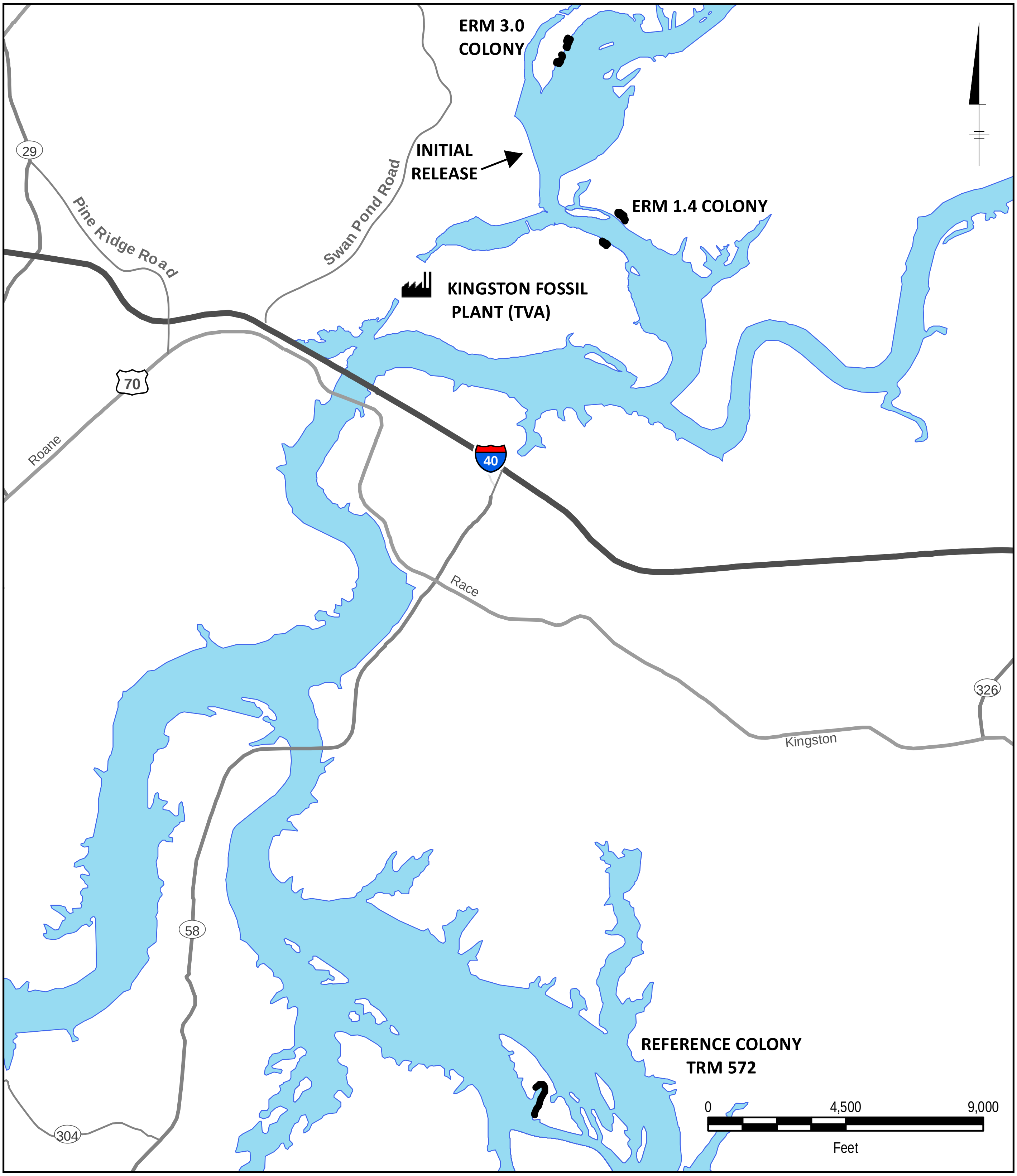
Table 1
Tree Swallow Egg Summary Statistics for 2014
Tennessee Valley Authority
Kingston, Tennessee

Analyte	Location	Number of Detects	Number of Samples	Mean [a] ± SD	Range	SE
Arsenic	Emory River ERM 1.4	30	30	0.91 ± 0.39	0.44 - 2.23	0.07
	Tennessee River TRM 572.0	28	30	0.88 ± 0.20	0.33 - 1.25	0.04
Selenium	Emory River ERM 1.4	24	30	2.21 ± 0.53	1.25 - 3.26	0.10
	Tennessee River TRM 572.0	13	30	1.80 ± 0.05	1.36 - 3.91	0.09

Footnotes:
a. Mean calculations include reporting limits substituted for non-detects; concentrations presented in mg/kg dw.

Acronyms and Abbreviations:
ERM = Emory River Mile
mg/kg dw = milligrams per kilogram dry weight
SD = standard deviation
SE = standard error
TRM = Tennessee River Mile

Figures



Service Layer Credits:

Legend

- 2014 Tree Swallow Box Location

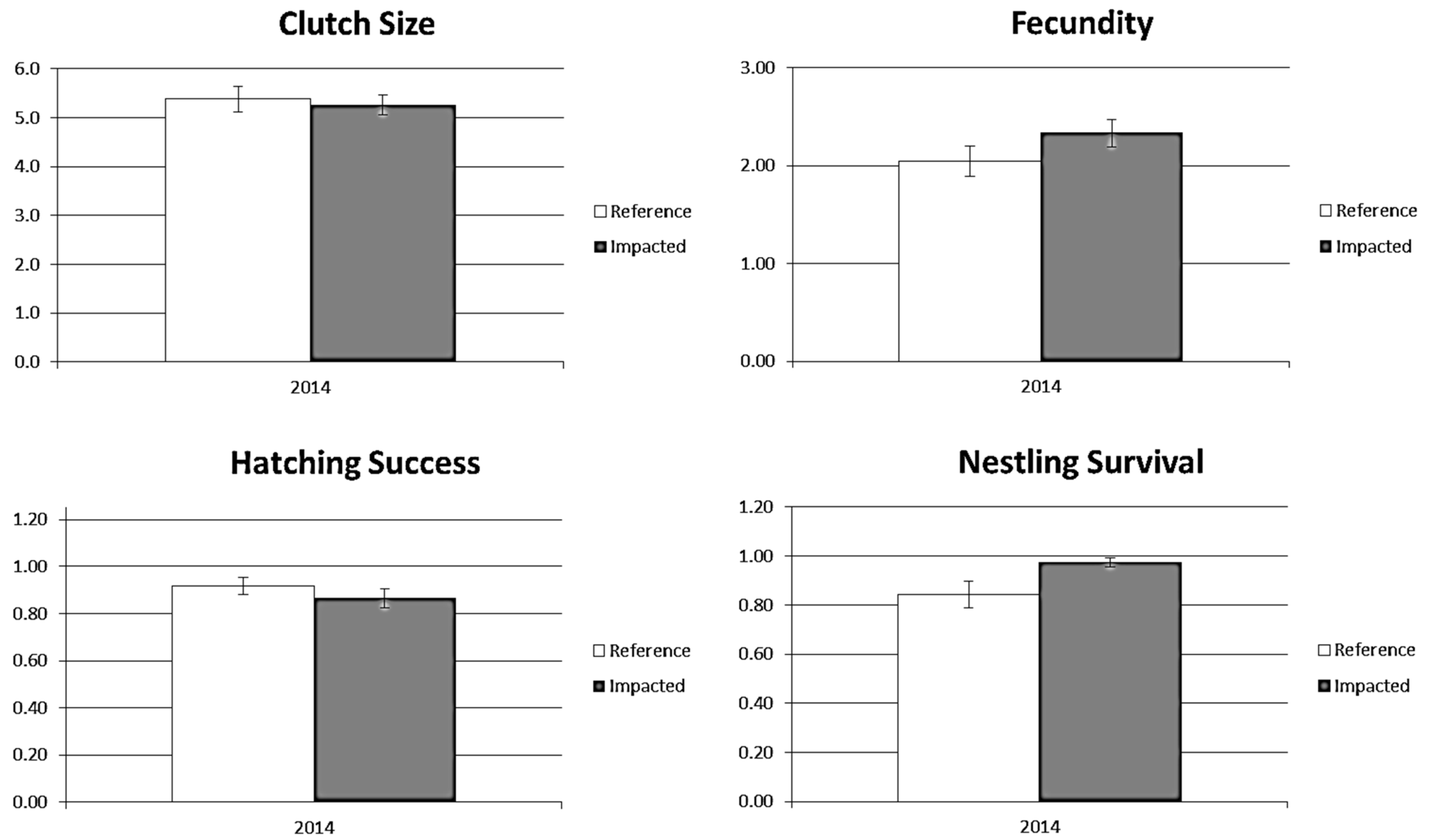
TENNESSEE VALLEY AUTHORITY
KINGSTON, TENNESSEE
2014 TREE SWALLOW DATA ANALYSIS MEMORANDUM

2014 Tree Swallow Box Locations

ARCADIS

FIGURE
1

CITY: (KNOXVILLE) DIV/GROUP:(ENV/GIS) DB: ccsmith LD: C.SMITH PIC: PM: S.WALLS TM: A.STOJAK
PROJECT: PATH: Z:\GISPROJECTS_ENV\TVA_Kingston\2014BoxLocs.mxd SAVED: 9/30/2015 BY: ccsmith



ARCADIS

Reproductive Metrics in Tree Swallows: 2014

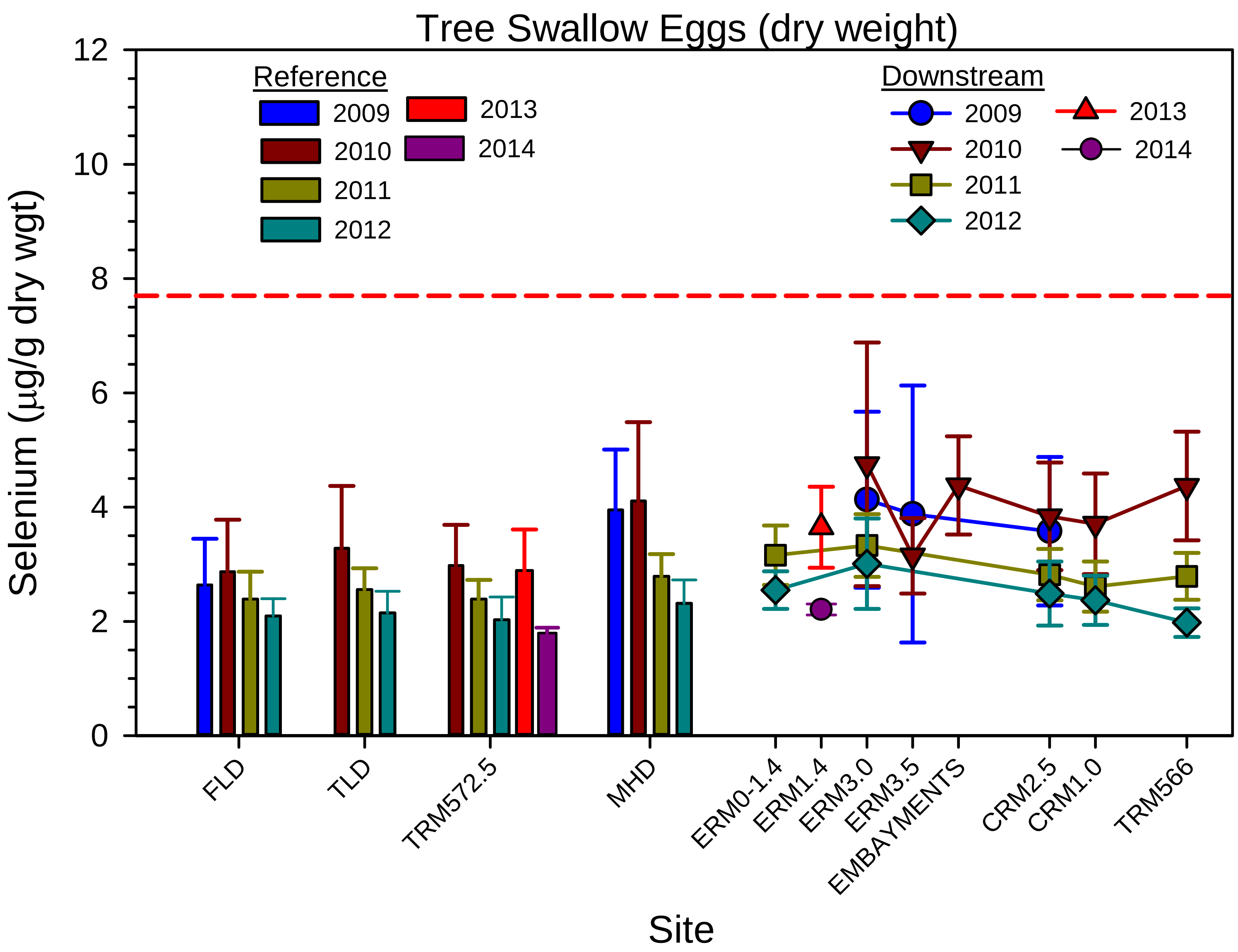
2014 Tree Swallow Data Analysis Memorandum

TENNESSEE VALLEY AUTHORITY

KINGSTON, TENNESSEE

FIGURE

2



ARCADIS

Selenium Concentrations in Tree Swallow Eggs: 2009-2014

2014 Tree Swallow Data Analysis Memorandum

TENNESSEE VALLEY AUTHORITY

KINGSTON, TENNESSEE

FIGURE

3