KINGSTON FOSSIL PLANT ASH SPILL & *CORBICULA FLUMINEA:* ASSESSING THE EFFECTIVENESS OF AN INVASIVE BIVALVE AS A BIOMONITOR OF COAL ASH POLLUTION

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DEDICATION

To my parents, Emma and Thomas Johnson,

who probably never suspected (nor wished) that their little preschool dropout would end up staying in college for the better part of a decade.

Thanks for everything, Mom & Dad.

ACKNOWLEDGEMENTS

I would like to thank my advisor, Mike McKinney, for putting up with me for all these years, for the countless office chats, and for teaching me to "leave a little room for serendipity." My committee members, Melanie Mayes and Ryan Otter, provided fresh perspectives and invaluable insight on the project. I am grateful to Colin Sumrall who, aside from being an incredible teacher and storyteller, provided office space in his lab along with a semester of funding. I am also grateful to the Chemistry Department of UTK for sharing their ICP-OES and even more grateful to Geoff Gilleaudeau for teaching me how to use the blasted thing. I would especially like to thank Rob Lindbom, Bobby Brown and Vince Pontello of Tennessee Wildlife Resources Agency for their enormous help with fieldwork. Those two days boating around the Emory, Clinch and Tennessee Rivers were the highlight of this project. I was lucky to have Grant Mincy as a stellar field buddy, several top-notch officemates, and my very own Set-playing Donnie-the-Distractor. I would also like my roommate, Mathew Halter, to acknowledge that I will henceforth answer to nothing but Master. Finally, I am more than grateful to James Fordyce for teaching me everything I know about statistics, R and acorns (and, in his attempts to do so, for only making me cry once), for giving me a chunk of prime real estate in his lab, and, most important, for teaching me that sometimes you just have to take the day off to watch the birds.

ABSTRACT

The goal of this study was to assess the effectiveness of using the invasive Asian clam, *Corbicula fluminea*, as a biomonitor for trace element bioavailability in the river system affected by the Kingston Fossil Plant coal ash spill of December 2008. I collected *Corbicula* from five different sites (n=20 per site) and analyzed both the soft body tissues and shells via inductively coupled plasma-optical emission spectroscopy (ICP-OES). I then compared the concentrations of a suite of elements in *Corbicula fluminea* among sampling sites and examined the correlation between soft tissue and shell concentrations. The multiple analysis of variance (MANOVA) for both the soft tissue and shell data showed significant ($p<0.0001$) differences among sampling sites. Subsequent univariate ANOVAs showed significant differences among sites for a number of the elements, and post-hoc Tukey's HSD tests revealed how the sites differ. This study demonstrates that trace elements associated with coal mining and coal combustion are bioavailable to *Corbicula fluminea* in the river system in the vicinity of the Kingston Fossil Plant. Given the species' ubiquitous presence in this aquatic system, *Corbicula fluminea* could be used as a component of the long term monitoring of the 2008 ash spill.

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LIST OF ABBREVIATIONS

AMD	Acid Mine Drainage
ANOVA	Analysis of Variance
CDA	Canonical Discriminant Analysis
EPA	Environmental Protection Agency
ICP-OES	Inductively Coupled Plasma-Optical Emission Spectrometer
MANOVA	Multivariate Analysis of Variance
PCA	Principal Component Analysis
ppm	parts per million
TDEC	Tennessee Department of Environment and Conservation
Tukey's HSD	Tukey's honestly significant difference test
TVA	Tennessee Valley Authority
TWRA	Tennessee Wildlife Resources Agency

CHAPTER 1 - INTRODUCTION

1.1 Kingston Fossil Plant Ash Spill

On December 22, 2008, the coal ash impoundment of the Tennessee Valley Authority (TVA) Kingston Fossil Plant near Harriman, Tennessee ruptured, resulting in the release of over 4.1 million cubic meters of coal ash slurry into the adjacent area (Figure 1) (Ruhl et al. 2009, Gottlieb et al. 2010). The slurry spilled through the ruptured earthen dike and covered approximately 1 million square meters of both terrestrial and aquatic habitat with up to 10 meters of coal ash (Tuberty et al. 2009). An estimated 25% of the ash flowed directly into the Emory River with such force that it was able to travel 3.2 kilometers upstream (Tuberty et al. 2009). The confluence of the Emory River and the Clinch River is 3.2 kilometers downstream of the spill, and the Clinch River flows into Watts Bar Lake of the Tennessee River another 6.4 river kilometers downstream (Tuberty et al. 2009).

The Kingston Fossil Plant's coal ash retention pond contained over 50 years of both fly ash and bottom ash, which are two byproducts of coal combustion (Ruhl et al. 2009, Tuberty et al. 2009). While the major components of coal ash are similar to natural soils and rocks (Bednar et al. 2010), both fly ash and bottom ash are highly enriched in trace elements (Ruhl et al. 2009). The exact chemical composition of the ash depends on the type of coal used and the manner in which that coal is burned (Gottlieb et al. 2010). Ruhl et al. report the average concentration (mg/kg) of elements in the TVA coal ash and the ratio of those concentrations to that of soil samples from Kingston, Tennessee, along with the Hg content of coal ash and river sediments near the spill (Table 1 & Table 2) (2009). The results show that, relative to the background

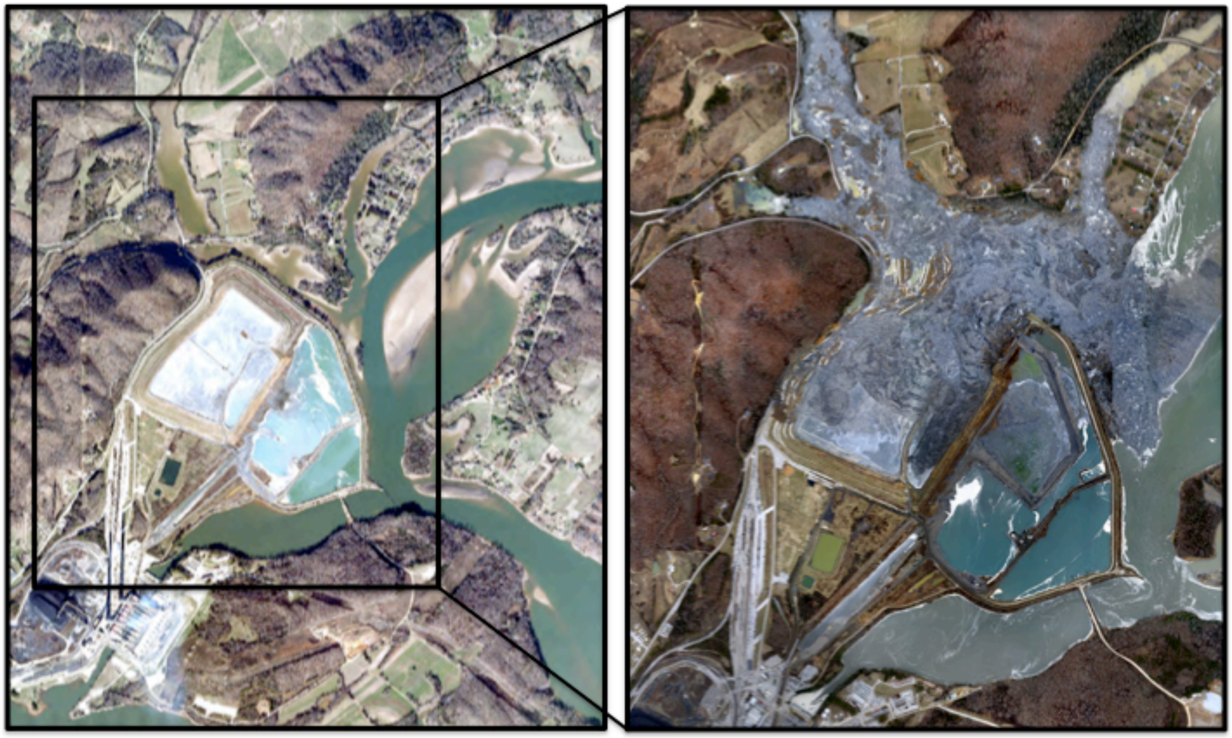


FIGURE 1. Aerial view of the Kingston Fossil Plant before and on the day following the ash spill (TVA 2008).

TABLE 1. Average Metal Concentrations (mg/kg) in TVA Coal Ash (n=12) and Background Soil (n=12) in Kingston, TN (Ruhl et al. 2009).

MATERIAL	Al	As	Ba	Be	Cd	Ca	Cr	Co	Cu	Fe	Pb	Li	Mg	Mn	Mo	Ni	Se	Sr	V	Zn
COAL ASH (mg/kg)	14109	74.6	354.2	3.1	0.03	3325	24.8	13.5	46.2	13333	19	24.6	1616	102	2.1	23	0.2	201	76.7	40.4
STD	7264	20.4	248.8	1.8	0.08	1142	7.6	6	16.6	2807	6.6	6.8	1531	54	2	8	0.6	39.3	30.6	12.5
SOIL (mg/kg)	9367	3.5	73.8	0.3	0.01	1418	11.9	7.3	15.9	15200	16.5	5.3	1211	1056	0.2	4.4	0.9	6.8	20.9	29.7
STD	3485	2.3	51.9	0.3	0.21	933	5.3	7.8	35.2	6729	8.4	1.9	5580	1007	0.4	4.7	1.8	4.3	4.4	11.7
ASH/SOIL RATIO	1.5	21.4	4.8	9.4	3	2.3	2.1	1.9	2.9	0.9	1.1	4.6	1.3	0.1	13.4	5.2	0.2	29.8	3.7	1.4

TABLE 2. Hg Results (μg/kg) in Coal Ash and River Sediments Associated with the Spill Area in Kingston, TN (Ruhl et al. 2009).

Site Description	**Total Hg (μg/kg average ± SD) n=3**	
	7-Feb-09	28-Mar-09
Upstream Emory River	43±0.5	10 (n=1)
Upstream Upper Clinch River	NA	16±5
Upstream Lower Clinch River	NA	54±11
Upstream Emory River near spill site	29.7±3	22±0.2
Spilled ash pile	139±5	NA
Spilled ash pile	145±12	NA
Emory River across from spill	53±3	NA
Confluence of Emory and Clinch Rivers	130±5	104±12
Downstream Clinch River (I-40)	115±9	92±32
Downstream Clinch River	NA	81±38
Clinch River near TN River confluence	NA	51±10
Background soil, Tennessee		
Lower Clinch River (n=9)	45±12	
Upper TN hydrological unit (n=73)	47±27	
Roane County	56±23	

soil, the coal ash was marginally enriched in calcium, magnesium, and aluminum, and considerably enriched in strontium, arsenic, barium, nickel, lithium, vanadium, copper, and chromium (Table 1) (Ruhl et al. 2009). The mercury content in river sediments downstream of the spill is significantly higher than reference sites upstream (Table 2) (Ruhl et al. 2009).

The Environmental Protection Agency (EPA), the Tennessee Department of Environment and Conservation (TDEC), and the Tennessee Valley Authority (TVA) were the three major groups involved with sampling and analysis efforts following the ash spill (Delos et al. 2009). As of 2009, the reported downstream surface water monitoring data suggested that trace metals from the ash spill were not readily dissolving into the surface water (Delos et al. 2009). Between March 2009 and June 2010, hydraulic cutterhead dredges were used to pump ash from the river bottom into an ash recovery ditch and subsequent stilling ponds (Bednar et al. 2013). This remediation effort undertaken by TVA removed over 2.6 million cubic meters of coal ash and sediment from the Emory River (Bednar et al. 2013). However, because much of the coal ash is now incorporated into the sediment of the Emory, Clinch and Tennessee Rivers, the aqueous concentrations of metals may increase in the future as ash particles are mobilized or transformed by rain events, dredging, or biological activities (Delos et al. 2009).

For example, as of 2009, Hg was only found in trace amounts in the surface water downstream of the spill, but the Hg content of downstream river sediments was at an elevated level similar to levels in the coal ash itself (Ruhl et al. 2009). Anaerobic bacteria can transform Hg in the sediment to methylmercury, a bioavailable form of mercury that accumulates in food webs (Ruhl et al. 2009). The presence of sulfate, an essential nutrient for sulfate-reducing bacteria and also a component of coal ash, may increase the rate of mercury methylation in a system (Ruhl et al. 2009). Therefore, the amount of bioavailable mercury downstream from the Kingston coal ash spill may increase in the future (Ruhl et al. 2009).

The coal ash release has the potential to impact biological health in a number of ways (Ruhl et al. 2009). The fine particles may be resuspended into the atmosphere, impacting the health of surrounding communities with airborne toxins (Ruhl et al. 2009). In aquatic systems, toxins in the coal ash may leach into surface waters, and coal ash toxins incorporated into the river sediment may be resuspended or transformed into bioavailable forms at a later date (Ruhl et al. 2009). The presence of coal ash alters aquatic habitat both physically and chemically via sedimentation and changes in turbidity, pH, conductivity, and the concentration of trace elements (Rowe et al. 2002). Both plants and animals living in coal ash affected habitats accumulate trace elements from the water and sediments, which may result in lethal or sublethal effects including stunted growth rates, reproductive failure or death (Rowe et al. 2002).

Because many of the trace elements in coal ash bioaccumulate in the food web, analysis of animal tissue offers a more accurate assessment of the amount of bioavailable toxins over time than analysis of surface waters or sediment alone (Delos et al. 2009). The current monitoring methods associated with the Kingston Fossil Plant spill include sediment and surface water quality testing and the use of fish tissue and aquatic insects as biomonitors (Delos et al. 2009, Ruhl et al. 2010, Otter et al. 2012). The following work focuses on assessing the effectiveness of using the soft body tissue and shells of an invasive freshwater bivalve as an indicator of bioavailable toxins from the coal ash spill over time.

1.2 Biomonitors

Water quality can be assessed using a number of physical, chemical or biological parameters (Phillips 1977). Direct analysis of water or sediment, while certainly useful, can be problematic (Phillips 1977). The concentration of contaminants in the water and sediments of a river can vary drastically over time due to precipitation, changes in season, and sporadic release of industrial toxins, so multiple water and sediment samples are required over a long period of time to accurately represent water quality, and even data such as this may not represent the toxins that are actually biologically available (Phillips 1977).

Biological indicator organisms offer an attractive alternative to water and sediment testing. Bioindicators may be used to qualitatively assess water quality based on relative abundance (Holt and Miller 2011), growth rates, survival and feeding patterns (Bucci et al. 2009). Often the presence or absence of pollution tolerant and pollution sensitive species are used as a proxy for the overall health of a water body, but this sort of method cannot pinpoint the ultimate cause of water quality decline (Holt and Miller 2011). A biomonitor is an organism that provides quantitative information about its environment based on the accumulation of toxins in its tissues (Holt and Miller 2011) and the fact that contaminant levels in biota tend to correlate with levels of environmental toxins (Ion, de Lafontaine et al. 1997). Fish, aquatic insects, and mollusks are often collected and analyzed for contaminant content, but the use of fish can be problematic in a fluvial system, however, as they are not restricted in mobility to a narrow range, making it impossible to precisely pinpoint where accumulated toxins originated (Phillips 1977).

1.3 Bivalves as biomonitors

Both freshwater and saltwater mussels are used worldwide to monitor the bioavailability of trace elements in aquatic systems (Kwan et al. 2003). Mussels are attractive biomonitors due to the ease of their collection, wide distribution, and largely sedentary lifestyle (Kwan et al. 2003). More important, their ability to filter large quantities of water leads to the rapid uptake and concentration of bioavailable trace elements to a greater degree than other common biomonitors (Ion et al. 1997, Kwan et al. 2003).

Unfortunately, an estimated 50% of native bivalves in North America are either extinct or on the brink of extinction, and 72% of native freshwater mussels are listed as extinct, endangered, threatened, or of special concern (Hoftyzer et al. 2008), making unionoids one of the most threatened groups of organisms in the United States (Figure 2) (Grabarkiewicz and Davis 2008). Given this fact, the use of native freshwater mussels in toxicity studies that require sacrificing multiple individuals from a population is less than ideal. A suitable organism for biological monitoring is generally abundant, widespread, sedentary, and able to survive in polluted conditions while accumulating toxins from the environment (Phillips 1977), and *Corbicula fluminea* possesses all of these traits (Graney Jr et al. 1983).

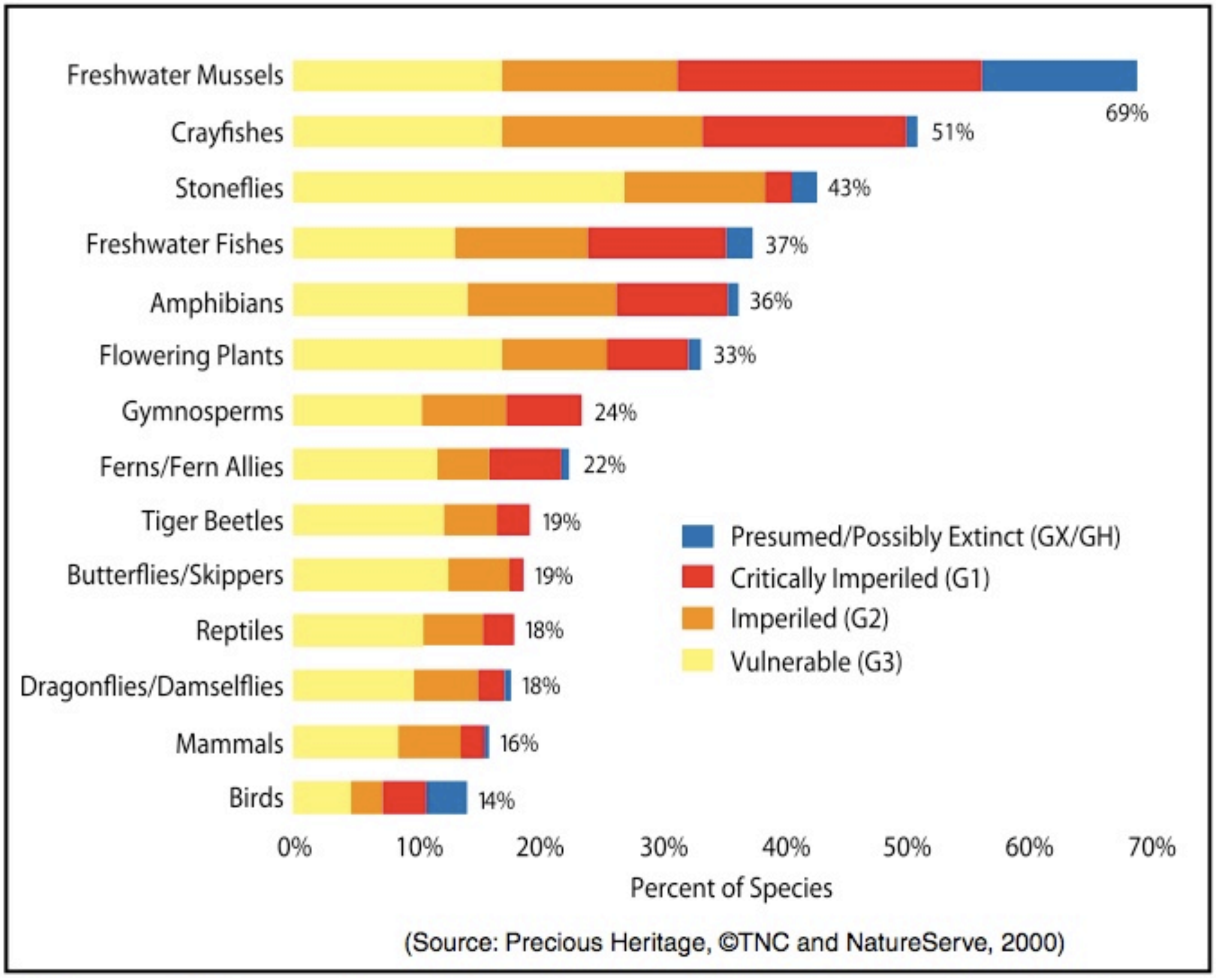


FIGURE 2. Proportion of species at risk by plant and animal group (Grabarkiewicz and Davis 2008).

1.4 Corbicula fluminea

Corbicula fluminea is a freshwater clam native to Southeast Asia that is now one of the most highly invasive species in freshwater ecosystems worldwide (Robinson and Wellborn 1988). The success of *Corbicula fluminea*'s invasion outside of its native range is largely due to certain aspects of the species' life history (Sousa et al. 2008). The hermaphroditic bivalve is capable of self-fertilization, and unlike the native freshwater mussels of North America, *Corbicula fluminea* does not require a host fish for successful reproduction (Doherty 1990). *Corbicula fluminea* grows rapidly and may reach sexual maturity in as little as 3 months (Sousa et al. 2008). The life span of the species ranges considerably from 1 to up to 5 years, and depending on environmental conditions, there may be up to two reproductive periods per year (Sousa et al. 2008). *Corbicula fluminea* have a high fecundity rate with estimates ranging from 8000 to 69000 juveniles per individual per year (Robinson and Wellborn 1988). Larvae are incubated within the branchial tubes of the adult before they are released into the water column (Sousa et al. 2008). At the time of release, the juveniles are a mere 250 μm in size, but they already have a fully formed shell, adductor muscles, foot, gills, statocysts, and digestive system (Sousa et al. 2008). Juveniles also possess a byssal thread that allows them to anchor to sediments or other hard surfaces, but if they remain suspended in the water column they can be transported for considerable distances leading to further dispersion of the species (Sousa et al. 2008).

The first documented occurrences of the *Corbicula* genus in North America were on Vancouver Island in 1924 and later in the Columbia River of Washington in 1938 (Strayer 1999). The subsequent dispersion of Corbicula across the United States was aided by both human activities and the organism's natural capacity for rapid dispersion (Sousa et al. 2008). The first documented occurrence of the invasive clam in Tennessee waters was in 1959 in the Tennessee River in Hardin County (Sinclair and Isom 1961). The current range of *Corbicula* in the United States excludes only the Northern Plains states and the majority of New England where low water temperatures may be preventing further establishment of the species (Figure 3) (Strayer 1999). Therefore, *Corbicula* are attractive as biomonitors due to their extensive global range, abundance, and ability to concentrate heavy metals from the water (Doherty 1990).

1.5 Past work on Corbicula fluminea as biomonitors

Corbicula fluminea have been used as biomonitors in a number of previous laboratory and field studies (Graney Jr et al. 1983, Doherty 1990, Soucek et al. 2001, Peltier et al. 2008, Peltier et al. 2009, Shoults-Wilson et al. 2009, Lewbart et al. 2010, Shoults-Wilson et al. 2010), but the majority of these studies involve soft tissue analysis alone. In his extensive review of the literature dealing with *Corbicula* as biomonitors, Doherty stated that only one study had, at that time, looked at the correlation between simultaneous bioaccumulation in both soft tissue and shells (1990). *Corbicula fluminea* shells, which are usually entirely made up of the calcium carbonate polymorph aragonite (Spann et al. 2010), may be useful in biomonitoring, as other elements can act as metabolic substitutes for calcium (Brown et al. 2005). Because trace elements are incorporated into the shell structure by active bioaccumulation during shell growth rather than sorption from the water column into preexisting layers (Bellotto and Miekeley 2007), analysis of shell element concentration offers a record of trace element

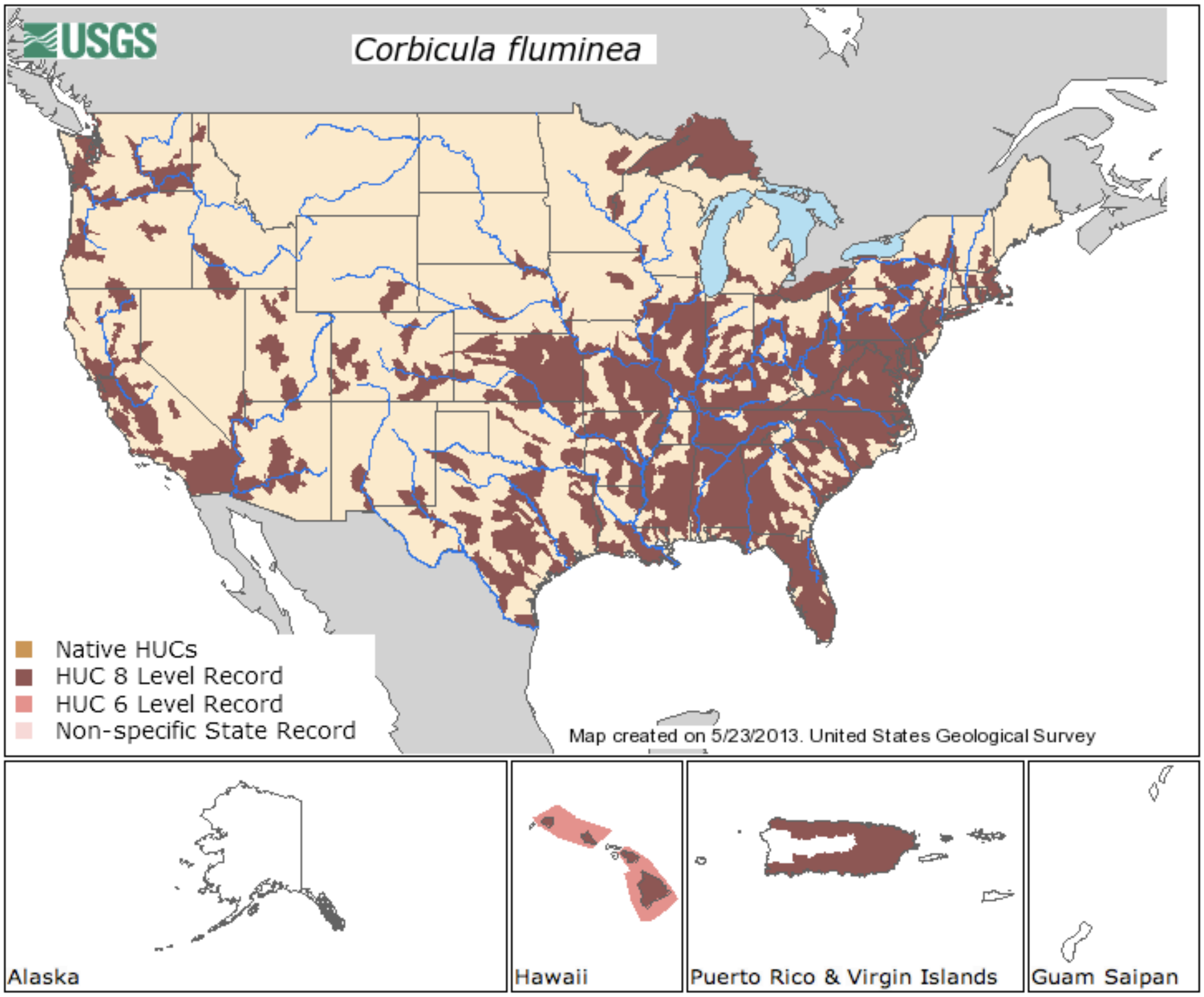


FIGURE 3. Current range of the invasive Asian clam, *Corbicula fluminea,* in the continental United States (Foster et al. 2012).

bioavailability over an individual's lifetime.

Due to its impressive invasion history, *Corbicula fluminea* should never be transplanted for toxicity studies (or for any other reason) into *Corbicula*-free water systems (Doherty 1990). However, because the species is already established in the Emory, Clinch and Tennessee Rivers, in situ *Corbicula fluminea* may be an effective biomonitor as part of a long-term monitoring scheme to assess the persistence of coal ash toxins near the Kingston Fossil Plant. The goals of this study were to address the following questions in an effort to assess the effectiveness of using both the shells and soft tissue of *Corbicula fluminea* as a biomonitor of coal ash toxins in a lotic system: Do shell metal concentrations correlate with tissue metal concentrations? Do metal concentrations in the shells and soft tissue of *Corbicula fluminea* vary significantly among sites? I expected to find that the concentration of coal ash related elements would be significantly higher in those clams collected from sites downstream of the Kingston Fossil Plant.

CHAPTER 2 - METHODS

2.1 Methods overview

I collected *Corbicula fluminea* from 5 sites in the river system affected by the Kingston Fossil Plant ash spill. I then used an inductively coupled plasma-optical emission spectrometer (ICP-OES) to analyze 100 soft body tissue samples and 100 shell samples, or 20 *Corbicula fluminea* per site, for the following elements: Ag, Al, As, Ba, Be, Cd, Co, Cr, Cs, Cu, Fe, Ga, Hg, K, Li, Mg, Mn, Na, Ni, Pb, Rb, Se, Sr, U, V, and Zn. Samples are introduced into an ICP-OES for elemental analysis in liquid form, and, as such, I liquefied my samples via acid digestion before running them through the machine.

2.2 Study area

I chose five *Corbicula fluminea* sampling sites in the Emory, Clinch and Tennessee Rivers in the vicinity of the Kingston Fossil Plant. The sampling sites include two reference sites upstream of the spill including one in the Clinch River (Site A) and one in the Emory River (Site B), one site immediately downstream from the spill in the Emory River (Site C), one site just downstream of the confluence of the Emory and Clinch Rivers (Site D), and one site farther downstream in the Tennessee River (Site E) (Figure 4 & Table A-1).

2.3 Field work

In shallow water, *Corbicula* can easily be collected by hand while wading, but the water depth of the chosen sites required collection via scuba diving. On September 12-13,

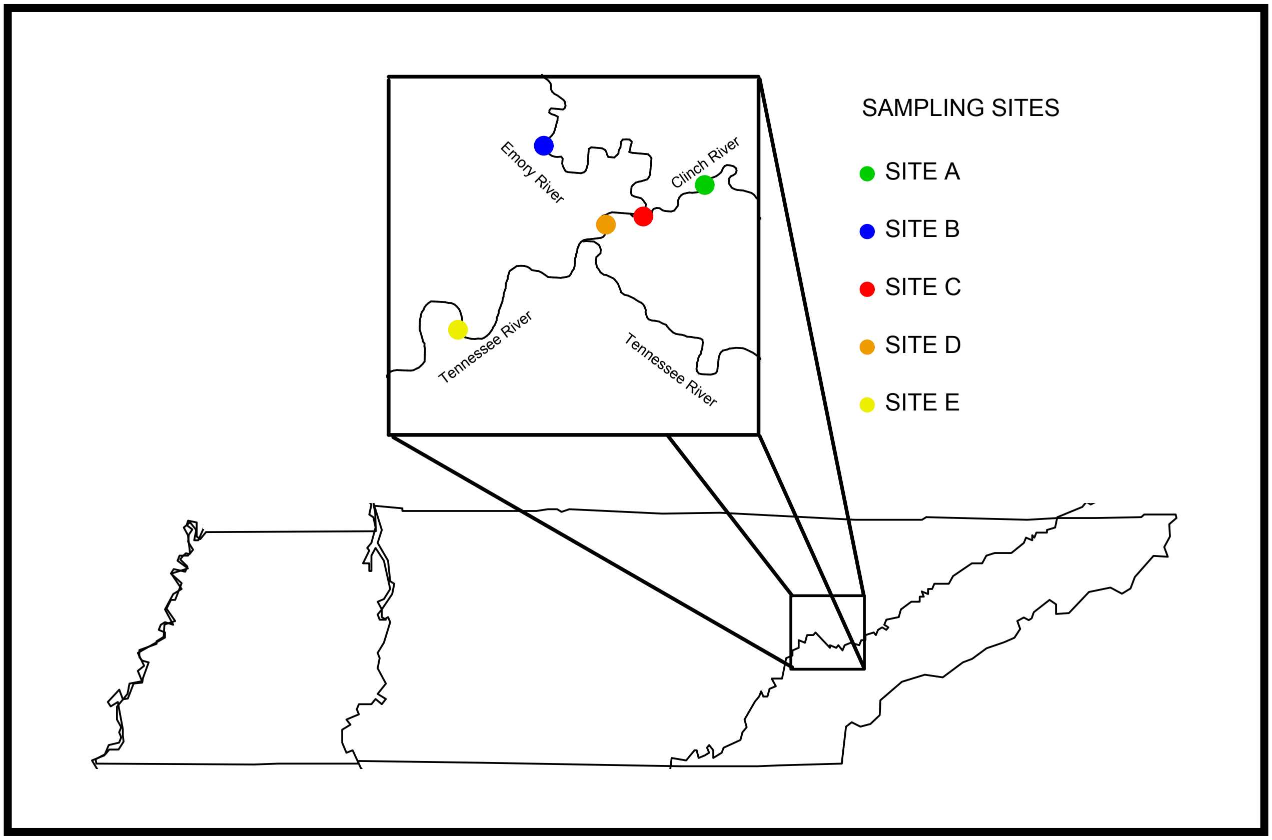


FIGURE 4. Sampling sites for the collection of *Corbicula fluminea* on September 12-13, 2012. Site C is immediately downstream from the ash spill origin.

2012, Tennessee Wildlife Resources Agency (TWRA) provided a dive boat, boat captain Bobby Brown, and two scuba divers to facilitate the collection of *Corbicula fluminea*. The boat was equipped with a hookah diving system, and the two divers, Rob Lindbom and Vince Pontello of TWRA, were able to collect at least 20 mature *Corbicula fluminea* from each of the five sites.

I placed the clams from each site in labeled Ziploc bags after each dive. I poked multiple small holes in the bags to allow aerated water to flow through for respiration. Each bag was placed in a cooler of river water, and small handfuls of ice were periodically added to the water to keep the temperature near that of the river and reduce the stress of the clams. After each field day, I placed an aerator in the cooler to keep the clams alive overnight, thereby giving the individuals time to expel the contents of their guts to ensure that gut content would not contribute to elemental concentrations. I removed the clams from the Ziploc bags roughly 30 hours after collection. I then rinsed the clams in deionized water and hand dried them using paper towels. I then transferred the clams from each site to a clean, labeled Ziploc bag and placed the bags in a freezer to await processing and analysis. Thus, the clams were killed by freezing as in Bellotto et al. (2007).

2.4 Sample processing and ICP-OES analysis

Before I began preparing the samples for ICP-OES analysis, I recorded morphometric data for each *Corbicula fluminea* specimen. Using Mitutoyo Absolute Digmatic digital calipers, I measured the length (anterior to posterior), height (right valve to left valve at umbo), and width (dorsal to ventral at umbo) of each clam. Using a stainless steel tapered micro spatula, I then gently opened the valves and separated the frozen soft body tissue from the shells. I placed the soft body and the valves of each clam into separate, pre-labeled 15 ml sterile polypropylene centrifuge tubes, and placed the sample tubes back in the freezer. I did these steps in batches of five clams per site until I had processed a total of 20 *Corbicula fluminea* from each site.

I then freeze dried the shell and soft body samples in their uncapped centrifuge tubes for 48 hours in a LABCONCO Freezone 4.5 freeze dry system. Using an OHAUS ANALYTICAL Plus balance, I measured the dry weights to the nearest 0.01 mg of the shells and soft body tissue and returned the samples to their respective centrifuge tubes.

I began the acid digestion of the shell samples by adding 5 ml of 5% trace metal grade nitric acid to each tube. Because there was little dissolution after 24 hours, I then added 100% nitric acid to each tube in 0.25 ml increments until the shell was completely dissolved and no further reaction was seen. The proteinaceous periostracum remained intact after the calcium carbonate had completely dissolved. To dissolve the soft body tissue, I added concentrated nitric acid to the tubes in 1 ml increments until no reaction was seen. I centrifuged the dissolved samples at 5000 RPM for seven minutes in an Eppendorf 5804 centrifuge, decanted the supernatant into fresh, pre-labeled polypropylene tubes, recorded the final volume, and set these tubes aside as concentrated stock solution.

I then made a standard dilution series for the ICP-OES using SPEX CertiPrep Claritas PPT standards, including a multi-element solution with 10 mg/L each of Ag, Al, As, Ba, Be, Ca, Cd, Co, Cr, Cs, Cu, Fe, Ga, K, Li, Mg, Mn, Na, Ni, Pb, Rb, Se, Sr, Tl, U, V, and Zn in a 5% HNO_3 matrix and a solution with 10 mg/L of Hg in a 5% HNO_3 matrix. I diluted the standards with ultrapure MilliQ water to make 5 standards with concentrations of 5 ppm, 1.25 ppm, 0.5 ppm, 0.25 ppm, and 0.125 ppm of all the elements. I used these standards to set a calibration curve for element concentration for the ICP-OES.

I prepared a dilution series of one of the body samples and one of the shell samples in order to determine the appropriate dilution of the stock solutions for analysis. Based on the ICP-OES analysis of these dilutions, I determined that a 1:10 dilution of the body stock solutions and a 2:10 dilution of the shell stock solutions were appropriate. For the shell samples, I used a 2μl – 1000μl Gilson Pipetman set to 1000μl to add 8ml of ultrapure MilliQ water to 100 sterile, pre-labeled 15 ml centrifuge tubes. I then added 2ml of the appropriate shell stock solution to each of these tubes. I repeated this for the body samples using 9ml of MilliQ water and 1ml of stock solution.

Using the ICP-OES software, Winlab32, I created a new analytical method for the machine using the 0.25 ppm standard to adjust each element's detection wavelength. Using a blank of ultrapure MilliQ water and the 1.25 ppm, 0.5 ppm, 0.25 ppm, and 0.125 ppm standards, I calibrated the ICP-OES and began running the prepared samples through the machine.

2.5 Statistical Analysis

The ICP-OES reported concentrations of the elements in the dilute samples in mg/L, or parts per million (ppm). To then calculate the concentration in the soft tissue or shell in μg/g dry weight, I used the following equations:

μg element/g of shell = ((ICP-OES ppm)*5*(stock solution L)*1000)/g of shell

μg element/g of body = ((ICP-OES ppm)*10*(stock solution L)*1000)/g of body

Due to a malfunction with the ICP-OES, I was forced to drop all of the data obtained on one day in the lab. Therefore, shell samples 1-5 for all of the sites are missing from the dataset. All zeros reported in the results indicate a concentration under the detection limit of the ICP-OES of that element in the sample and should not necessarily be interpreted as absolute zeros. I dropped from the analyses those elements whose concentrations were consistently below the detectable level in order to better meet the multivariate normality assumptions of a MANOVA. Conversely, the concentrations of Sr and Na in a number of the shells were above the detectable limit and are therefore reported as "saturated." Thus, the elements included in the statistical analyses include the following:

Soft body data (n=20 per site): Ag, Al, As, Ba, Cd, Cu, Fe, Li, Mg, Mn, Ni, Pb, Rb, Se, Sr

Shell data (n=15 per site): Al, As, Ba, Cd, Cu, Fe, Mg, Mn, Ni, Pb, Se

I used a combination of JMP Pro 10 and R64 software to perform the statistical analyses. I log-transformed the data and used those values for the analyses when appropriate. To determine the significance of observed differences in clam size among sites, I first subjected the morphometric data (including the shell measurements, body dry weights, and shell dry weights) to univariate analyses of variance (ANOVAs) followed by Tukey's honestly significant difference (Tukey's HSD) post-hoc tests. I then examined the correlations among metals in both the soft body and shell data sets as well as the correlation of concentrations between the shells and soft tissue. I then performed a principal component analysis (PCA) on the correlations to explore the overall variation in the data. To test for differences among the sampling sites, the log-transformed data were then analyzed using a multivariate analysis of variance (MANOVA) framework followed by subsequent univariate ANOVAs for each metal to test the effect of site. Those elements that were significantly different ($p<0.05$) in the ANOVA were then subjected to Tukey's HSD post-hoc tests.

CHAPTER 3 - RESULTS

3.1 Morphometric analyses and clam age classes

The shell length, width and height along with the shell and body dry weights are reported for each clam (Table A-2). The clams ranged in length from 10.93 mm to 27.18 mm with an average length of 15.61 mm. The length, width and height of the shells were highly correlated (length to width: 0.9416; length to height: 0.8952. width to height: 0.977) (Table A-3). Principal component (PC) 1 of the PCA on the shell size data accounted for 95.875% of the variation (Table A-4). The results of the morphometric ANOVAs and Tukey's HSD post-hoc tests are reported in Table A-5 and show that the *Corbicula fluminea* collected from the Tennessee River were significantly larger ($p<0.05$) than those from the other four sites based on shell length and shell dry weight (Figure 5 & Figure 6). It has been demonstrated in past work that the concentration of elements in *Corbicula* is correlated with size (Peltier et al. 2008), and, as such, I decided to run all further analyses twice: once including all five sites and again excluding the Tennessee River site. The results and discussion will focus on the latter, but results from the former are included in Appendix B. As the clams from the other four sites were not significantly different in size, this exclusion minimizes the effects of size on the analyses.

Using size and age data from Mouthon 2001, I used the average shell lengths reported by cohort of *Corbicula fluminea* collected from the Saone River to determine age classes (less than one year (1-), one to two years (1+), two to three years (2+), three to four years (3+), and greater than four years old (4+)) based on shell length. The age classes and corresponding shell lengths (SL) are as follows:

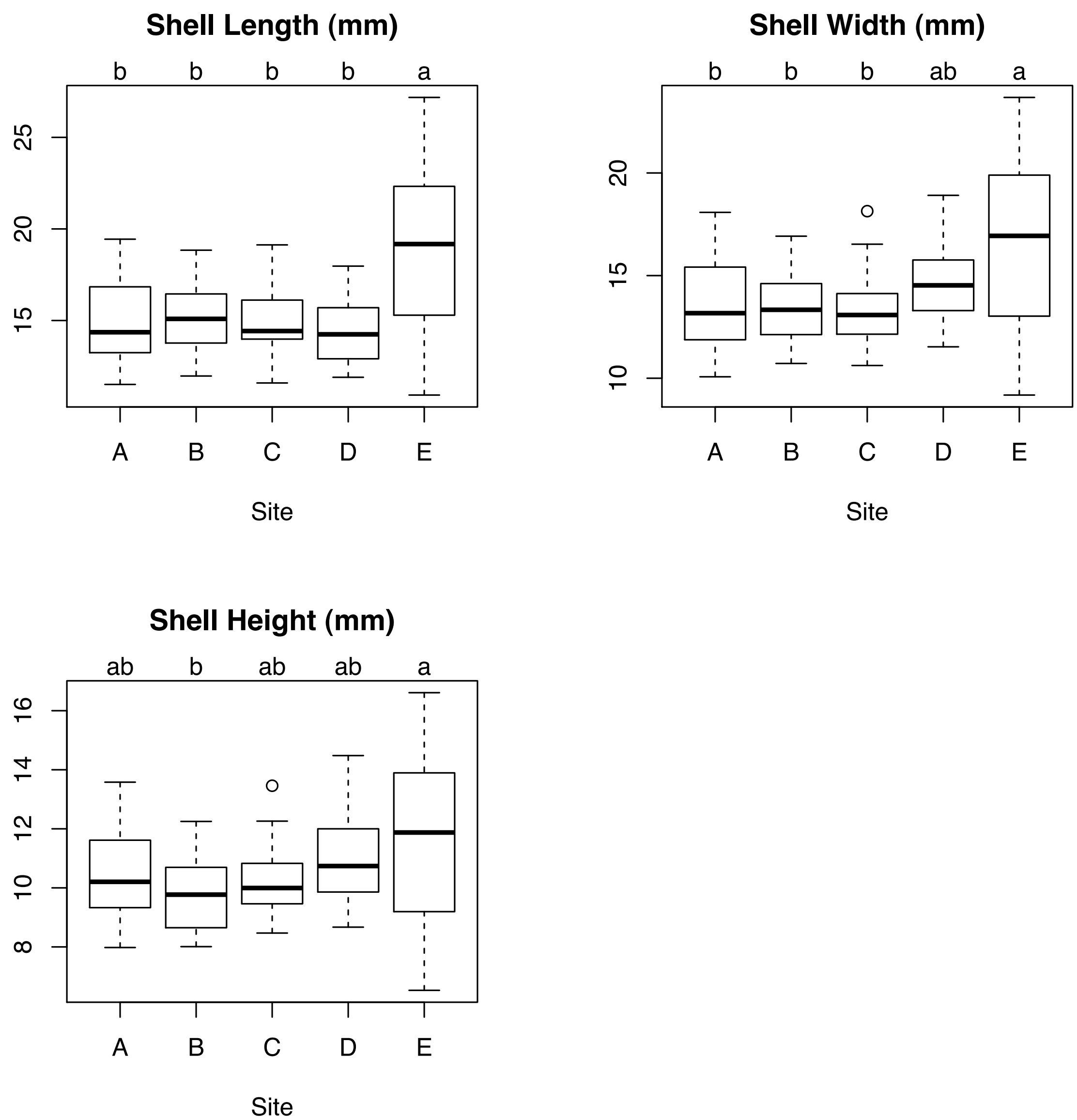


FIGURE 5. Box plots including Tukey's HSD results for shell length, shell width and shell height. Sites not connected by the same letter are significantly different.

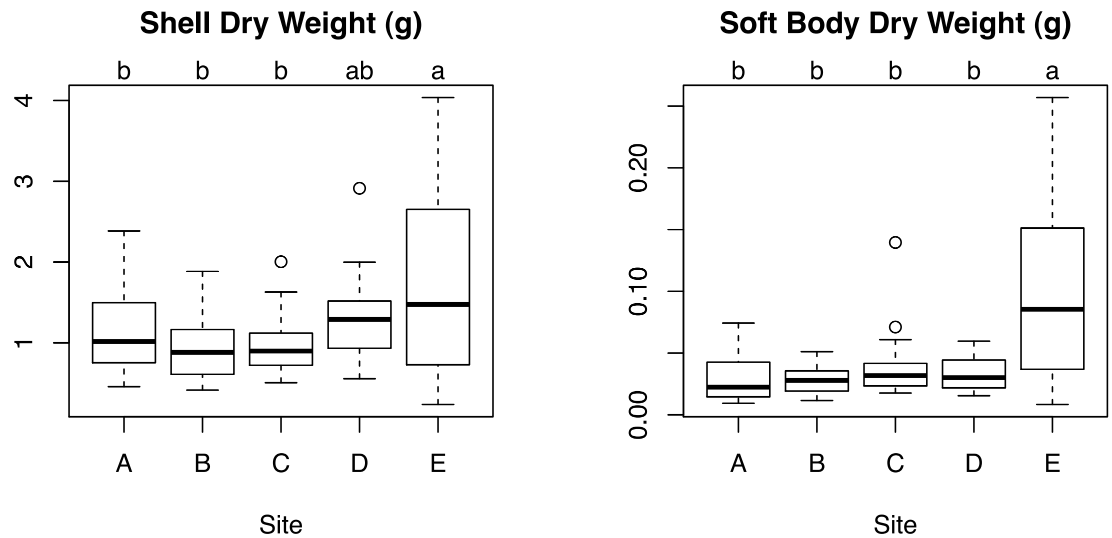


FIGURE 6. Box plots including Tukey's HSD results for body dry weight and shell dry weight. Sites not connected by the same letter are significantly different.

1-	0 mm ≤ SL < 8.583 mm
1+	8.583 mm ≤ SL < 15.76 mm
2+	15.76 mm ≤ SL < 22.12 mm
3+	22.12 mm ≤ SL < 26.07 mm
4+	26.07 mm ≤ SL

Based on these age classes, 61 of the 100 sampled *Corbicula* fall into the 1+ class, 34 into the 2+, 4 into the 3+, and 1 into the 4+. The five individuals older than three years of age were all collected from Site E in the Tennessee River.

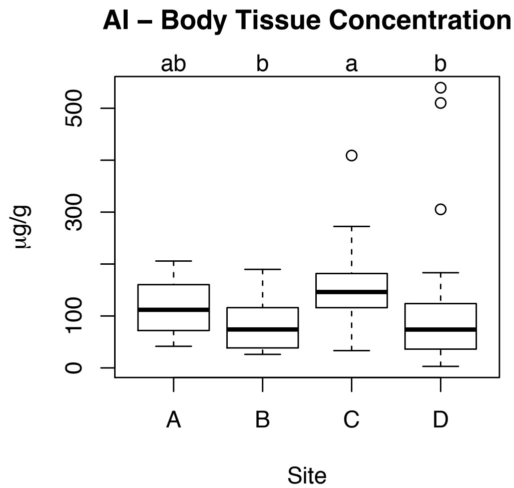
3.2 ICP-OES data

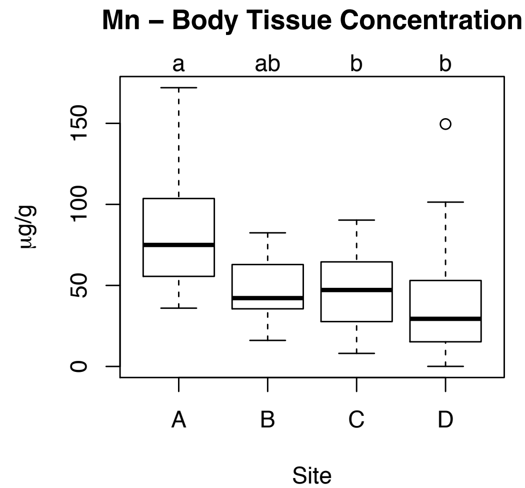
Soft body tissue and shell concentrations of 25 different elements are reported in μg/g dry weight, or parts per million (ppm) (Table A-6 & Table A-7). Box plots for each element included in the analyses showing concentrations by site are given in Figure 7 and Figure 8 for the soft body tissue and shell results, respectively. The box plots are ordered according to the results of the univariate ANOVAs, which are further discussed in sections 3.8 and 3.9.

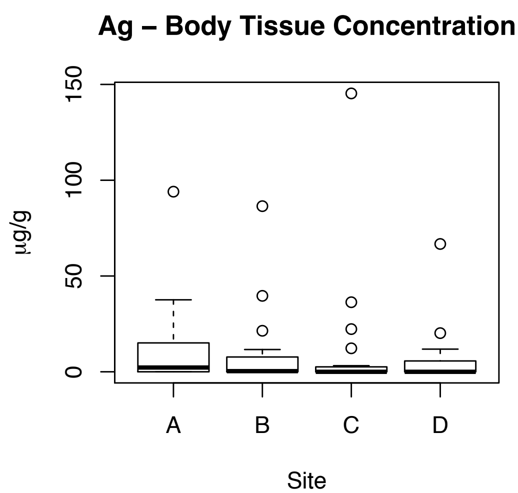
3.3 Correlation among elements

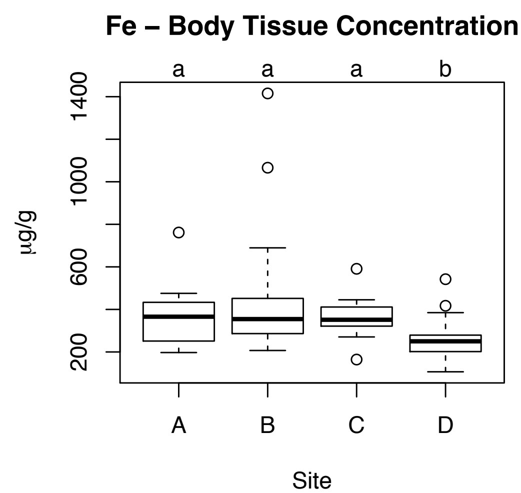
The correlation matrices for the soft body tissue and shell data are provided in Table A-7 & A-8 and represented visually in Figures 9 & 10. Larger and darker circles indicate a stronger correlation. Blue circles indicate positive correlations, while red circles indicate negative correlations.

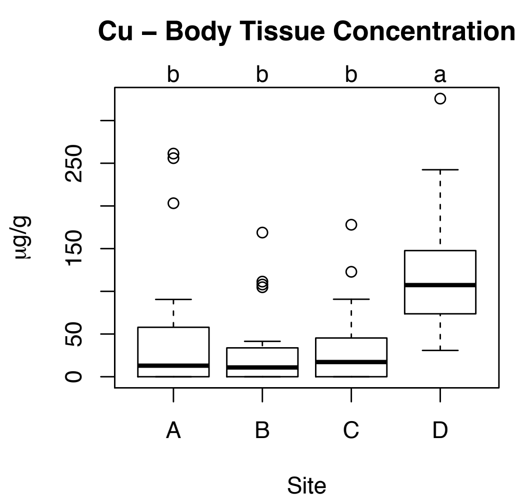
FIGURE 7. Body tissue concentration box plots including results from Tukey's HSD post-hoc tests. Sites not connected by the same letter are significantly different.

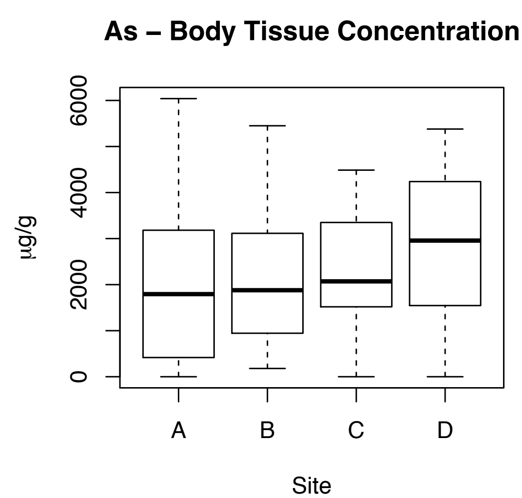

Al – Body Tissue Concentration
ab
b
a
b
µg/g
0
100
300
500
A
B
C
D
Site

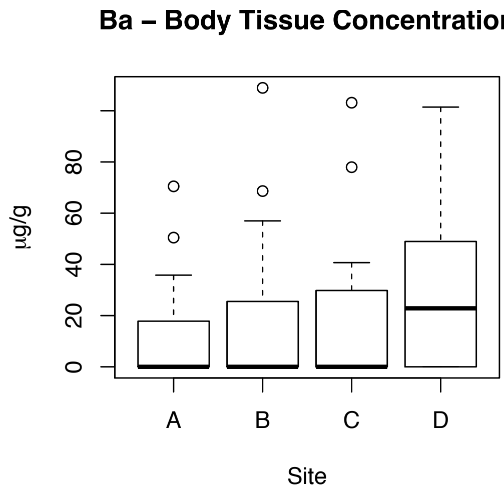

Mn – Body Tissue Concentration
a
ab
b
b
µg/g
0
50
100
150
A
B
C
D
Site

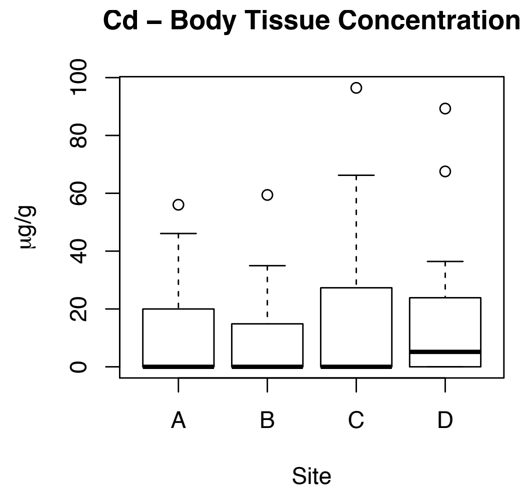

Ag – Body Tissue Concentration
µg/g
0
50
100
150
A
B
C
D
Site

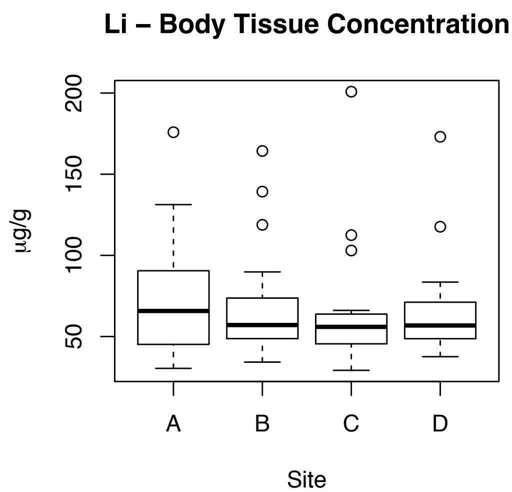

Fe – Body Tissue Concentration
a
a
a
b
µg/g
200
600
1000
1400
A
B
C
D
Site

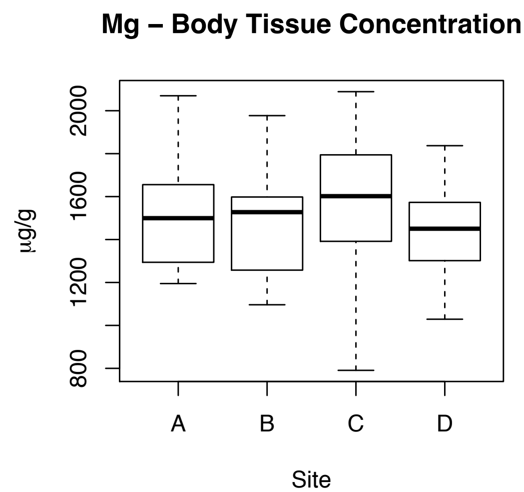

Cu – Body Tissue Concentration
b
b
b
a
µg/g
0
50
150
250
A
B
C
D
Site

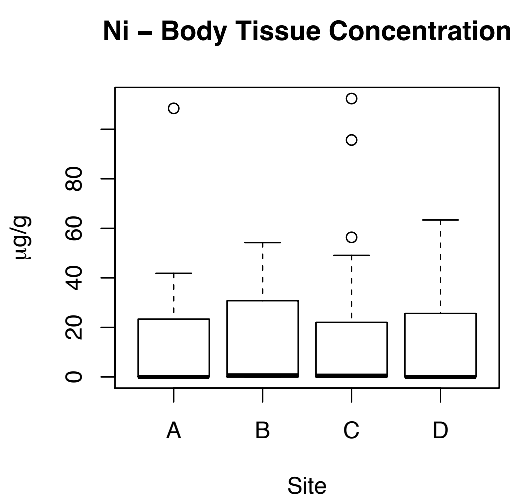

As – Body Tissue Concentration
µg/g
0
2000
4000
6000
A
B
C
D
Site

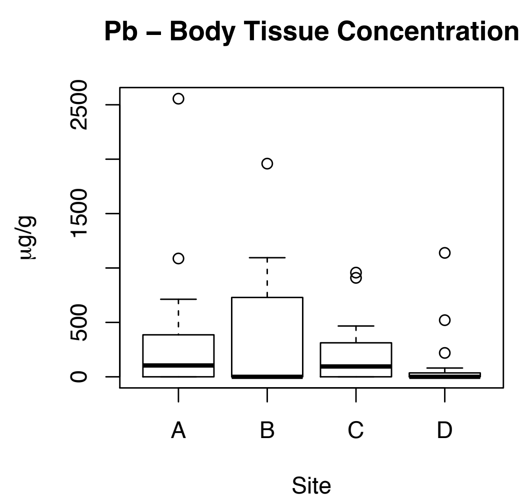

Ba – Body Tissue Concentration
µg/g
0
20
40
60
80
A
B
C
D
Site

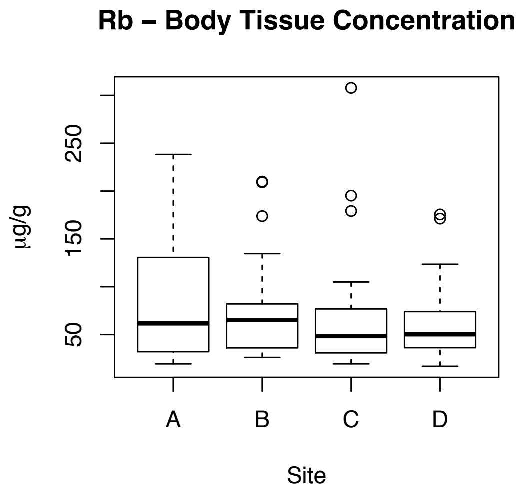

Cd – Body Tissue Concentration
µg/g
0
20
40
60
80
100
A
B
C
D
Site

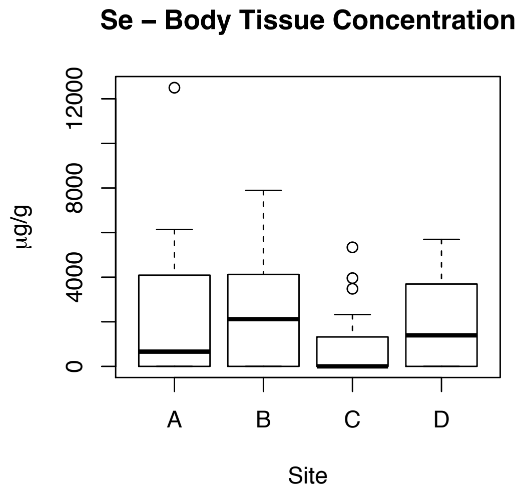

Li – Body Tissue Concentration
µg/g
50
100
150
200
A
B
C
D
Site


Mg – Body Tissue Concentration
µg/g
800
1200
1600
2000
A
B
C
D
Site


Ni – Body Tissue Concentration
µg/g
0
20
40
60
80
A
B
C
D
Site


Pb – Body Tissue Concentration
µg/g
0
500
1500
2500
A
B
C
D
Site


Rb – Body Tissue Concentration
µg/g
50
150
250
A
B
C
D
Site


Se – Body Tissue Concentration
µg/g
0
4000
8000
12000
A
B
C
D
Site

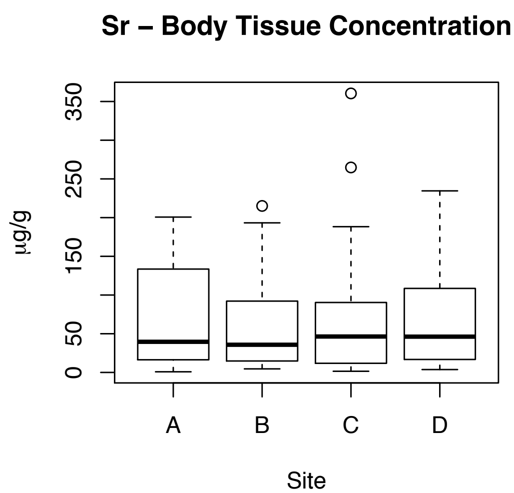
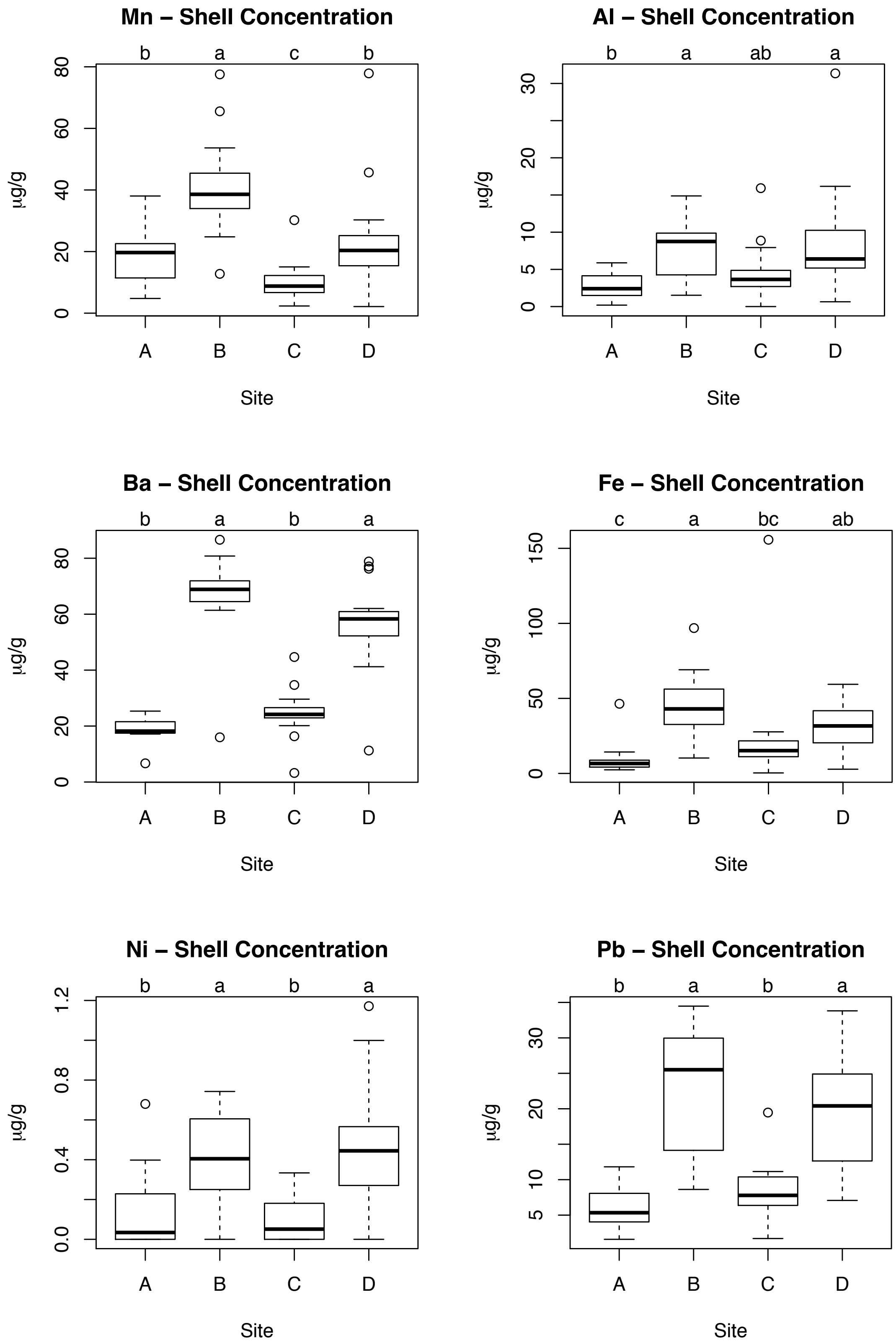
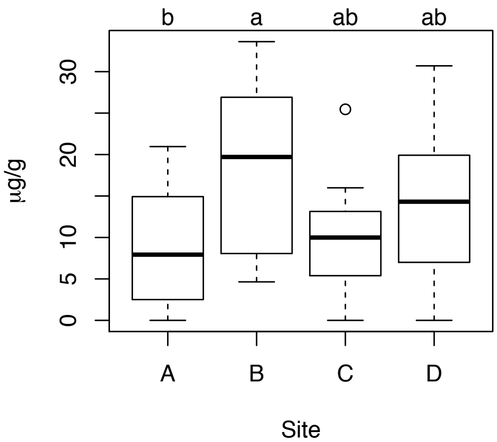

Sr – Body Tissue Concentration
µg/g
0
50
150
250
350
A
B
C
D
Site

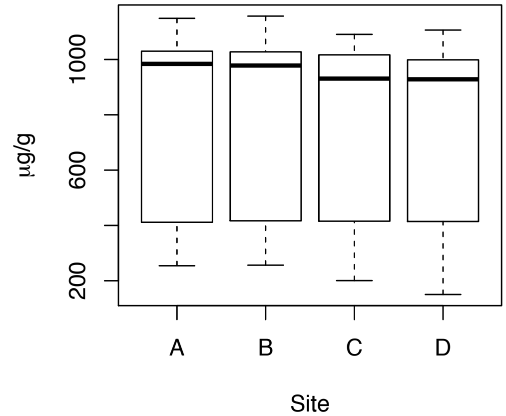
FIGURE 8. Shell concentration box plots including results from Tukey's HSD post-hoc tests. Sites not connected by the same letter are significantly different.


Mn – Shell Concentration
b a c b
µg/g
0 20 40 60 80
A B C D
Site
Al – Shell Concentration
b a ab a
µg/g
0 5 10 20 30
A B C D
Site
Ba – Shell Concentration
b a b a
µg/g
0 20 40 60 80
A B C D
Site
Fe – Shell Concentration
c a bc ab
µg/g
0 50 100 150
A B C D
Site
Ni – Shell Concentration
b a b a
µg/g
0.0 0.4 0.8 1.2
A B C D
Site
Pb – Shell Concentration
b a b a
µg/g
5 10 20 30
A B C D
Site

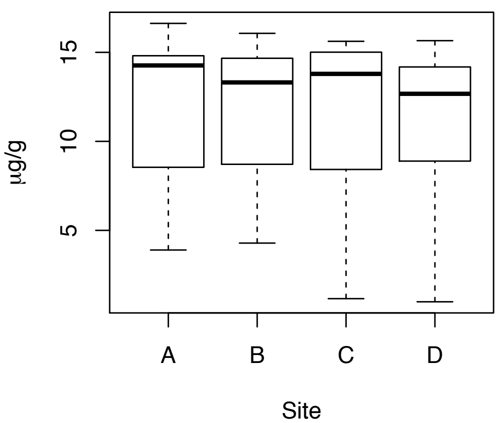
Se – Shell Concentration



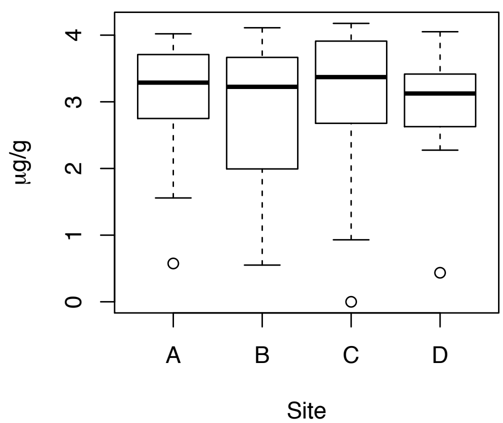
As – Shell Concentration



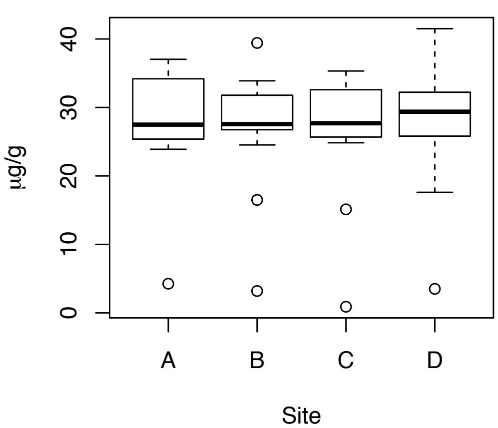
Cd – Shell Concentration



Cu – Shell Concentration



Mg – Shell Concentration



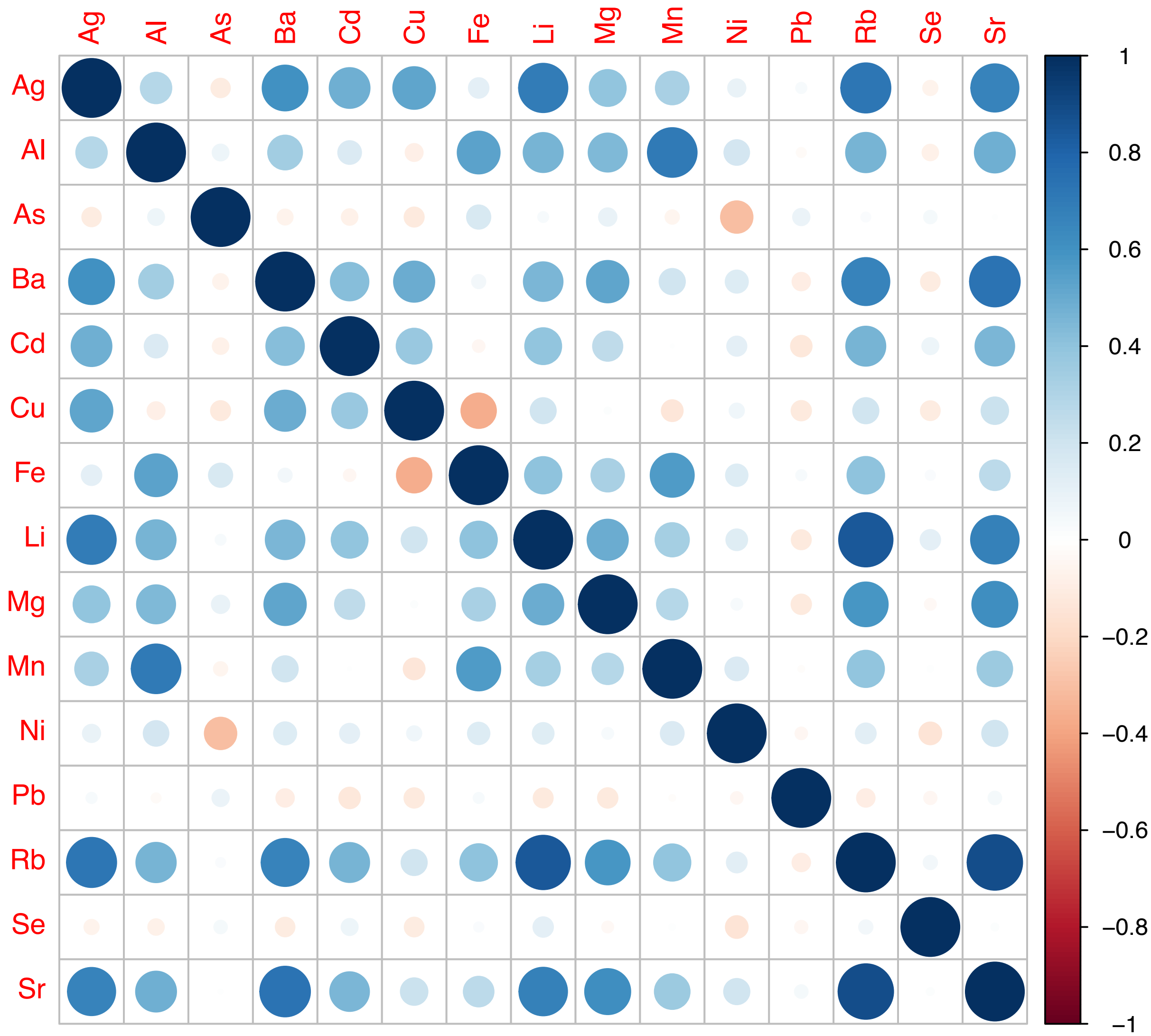


FIGURE 9. Correlation between elements in the *Corbicula fluminea* soft body tissue. Larger and darker circles indicate a stronger correlation. Blue circles indicate positive correlations, while red circles indicate negative correlations.

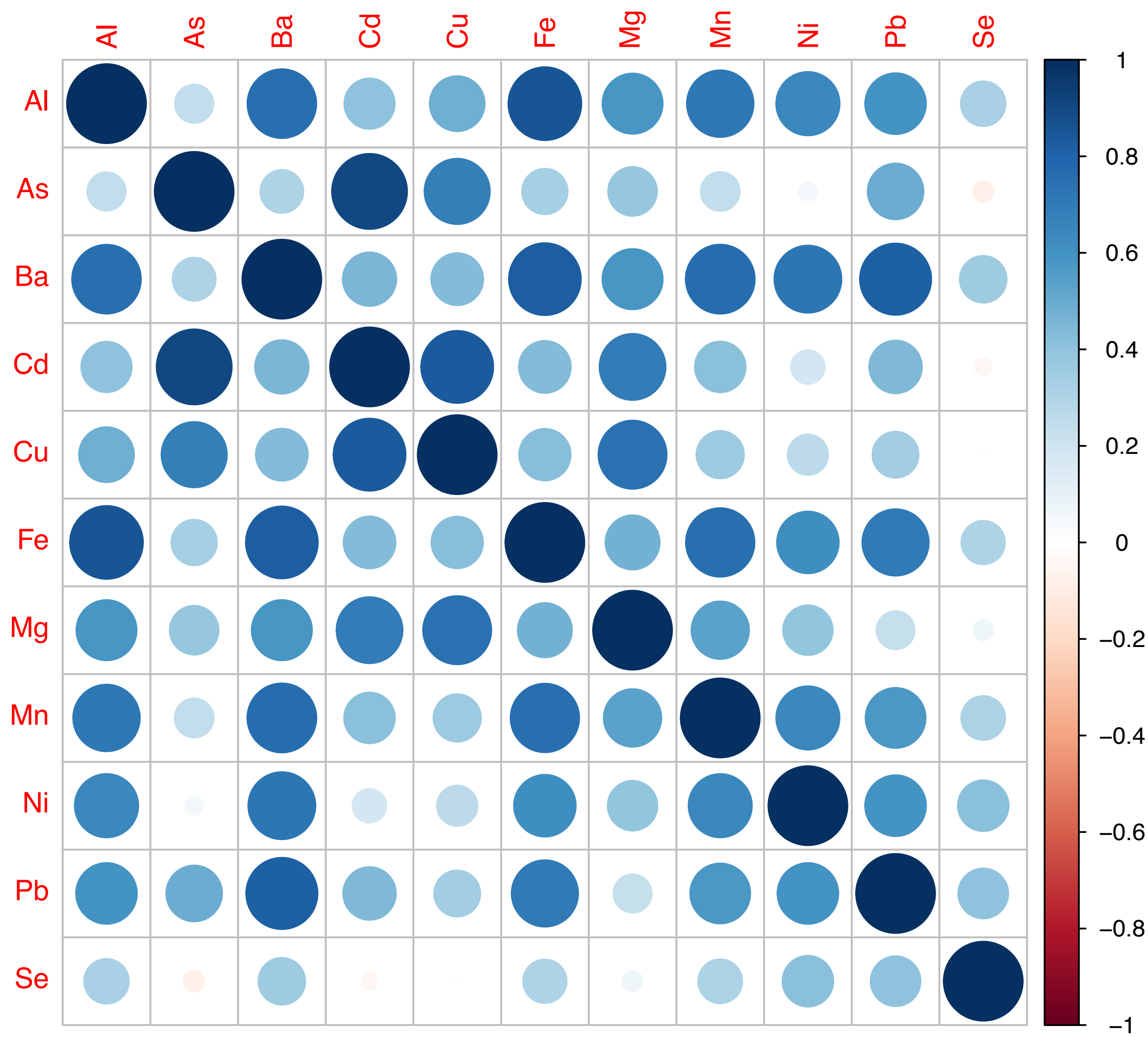


FIGURE 10. Correlation between elements in the *Corbicula fluminea* shells. Larger and darker circles indicate a stronger correlation. Blue circles indicate positive correlations, while red circles indicate negative correlations.

3.4 Correlation between shell and soft body tissue concentrations

The correlation between the concentration of elements in the soft body tissue data and that in the shell data is tabulated in Table A-10 along with the more conservative nonparametric Spearman's Rank correlations. Ba, Cu, K, Mg, Na and Rb are significantly positively correlated between the shells and soft tissue, while Pb is significantly negatively correlated. The Spearman's rank correlation differs only in that the Cu correlation ceases to be significant.

3.5 Principal Component Analysis - Soft Body Tissue

A principal component analysis (PCA) is used to reduce the dimensionality of the data and describe the overall variation in a multivariate dataset. The results of the soft body tissue PCA are shown in Table A-11, and principal component (PC) 1 and PC 2 are plotted in Figure 11. This plot of PC scores offers a two-dimensional representation of the data (Figure 11). PC 1 describes 35.31% of the total variation in the dataset. Aside from As and Pb, which are weakly negative, nearly all of the loadings on PC1 are relatively high positive values with Rb, Sr, Li, Ag and Ba being the most important sources of the overall variation in the dataset based on the loading matrix (Table A-11). PC2 describes 14.98% of the variation with Cu, Fe, Mn, Al and Cd as important sources of variation. While PC1 does not offer much visual separation of the sampling sites, PC2, plotted on the y-axis, shows a weak separation of Site D from the other three sites (Figure 11). Ni, As, and Se are the most important sources of variation on PC3, which describes 9.307% of the variation. PC4 describes 7.467% of the variation in the dataset

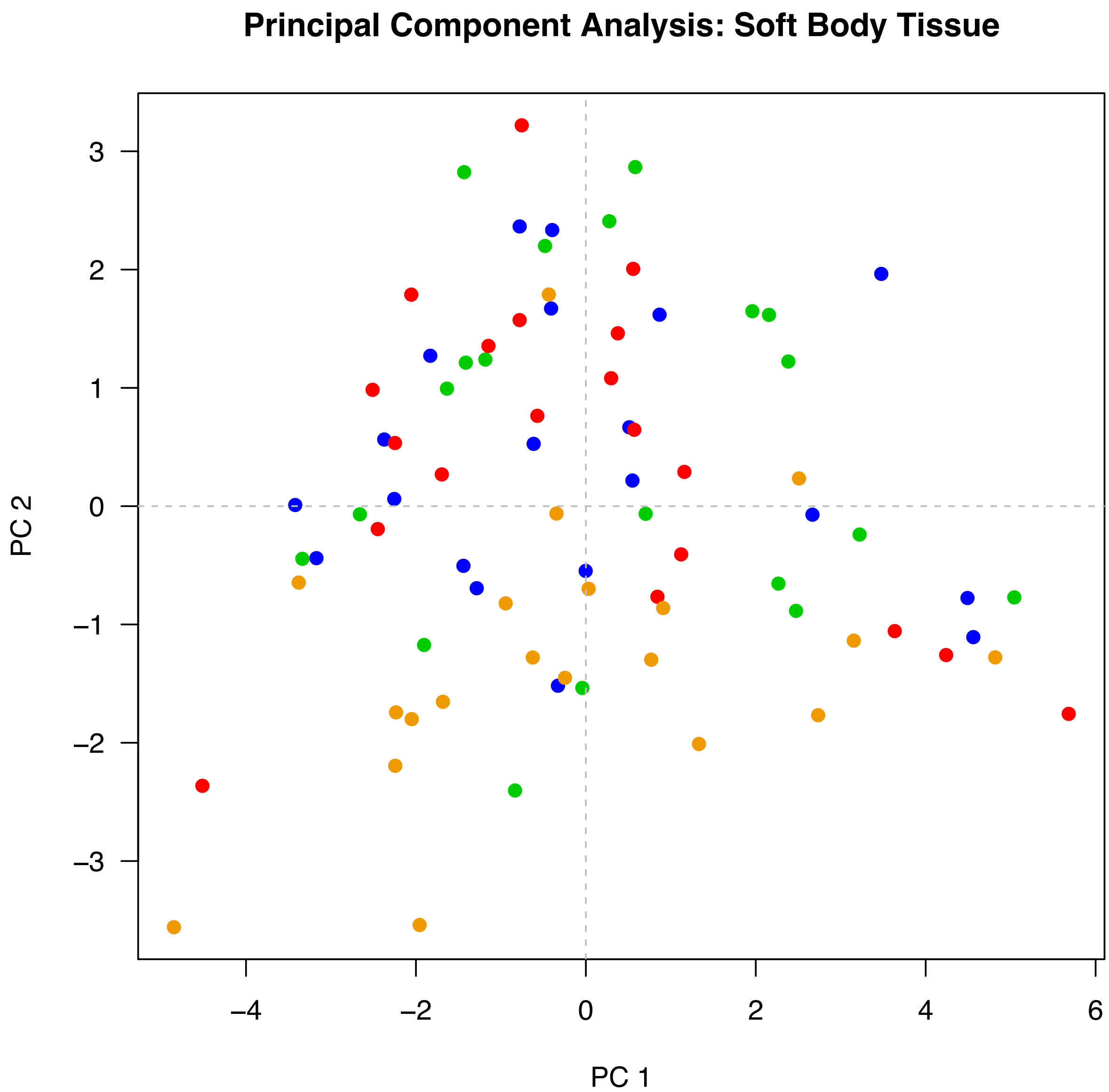


FIGURE 11. Principal component analysis (PCA) of log-normalized *Corbicula fluminea* soft body tissue data from Sites A (green), B (blue), C (red), and D (orange).

with strongly negative Se and strongly positive Pb as important sources of variation on that axis.

3.6 Principal Component Analysis - Shells

The results of the shell data PCA are tabulated in Table A-12 and shown graphically in Figure 12. PC 1 describes 54.182% of the variation, and the loadings are all strongly positive. Ba, Fe, Al, Mn, and Pb are the most important sources of the overall variation in the shell dataset based on the PC loadings. 19.476% of the variation is described by PC2. As, Cd, Cu and Se are the most highly loaded on this axis. The plot of PC1 and PC2 shows marked clustering of the sampling sites on both axes with Sites B and D scores largely clustering away from Sites A and C (Figure 12). PC3 accounts for 8.248% of the variation, and Pb, Mg and As are the most important sources of variation on this axis.

3.7 Multivariate Analysis of Variance (MANOVA)

According to the MANOVAs, concentrations in both the soft body tissues and shells differed significantly among the sampling sites (Wilks' Lambda, $p<0.0001$) (Table A-15, A-16, A-17 & A-18). Plots of the canonical scores (Table A-19) show differentiation among sampling sites (Figures 13 & 14), but interpretation of canonical coefficients in a MANOVA context is sensitive to the assumption of multivariate normality in the data. Thus, I performed subsequent univariate ANOVAs on each of the elements as post-hoc tests to determine which were important in discriminating among sample sites, as ANOVA is more robust to deviations from normality for hypothesis testing purposes.

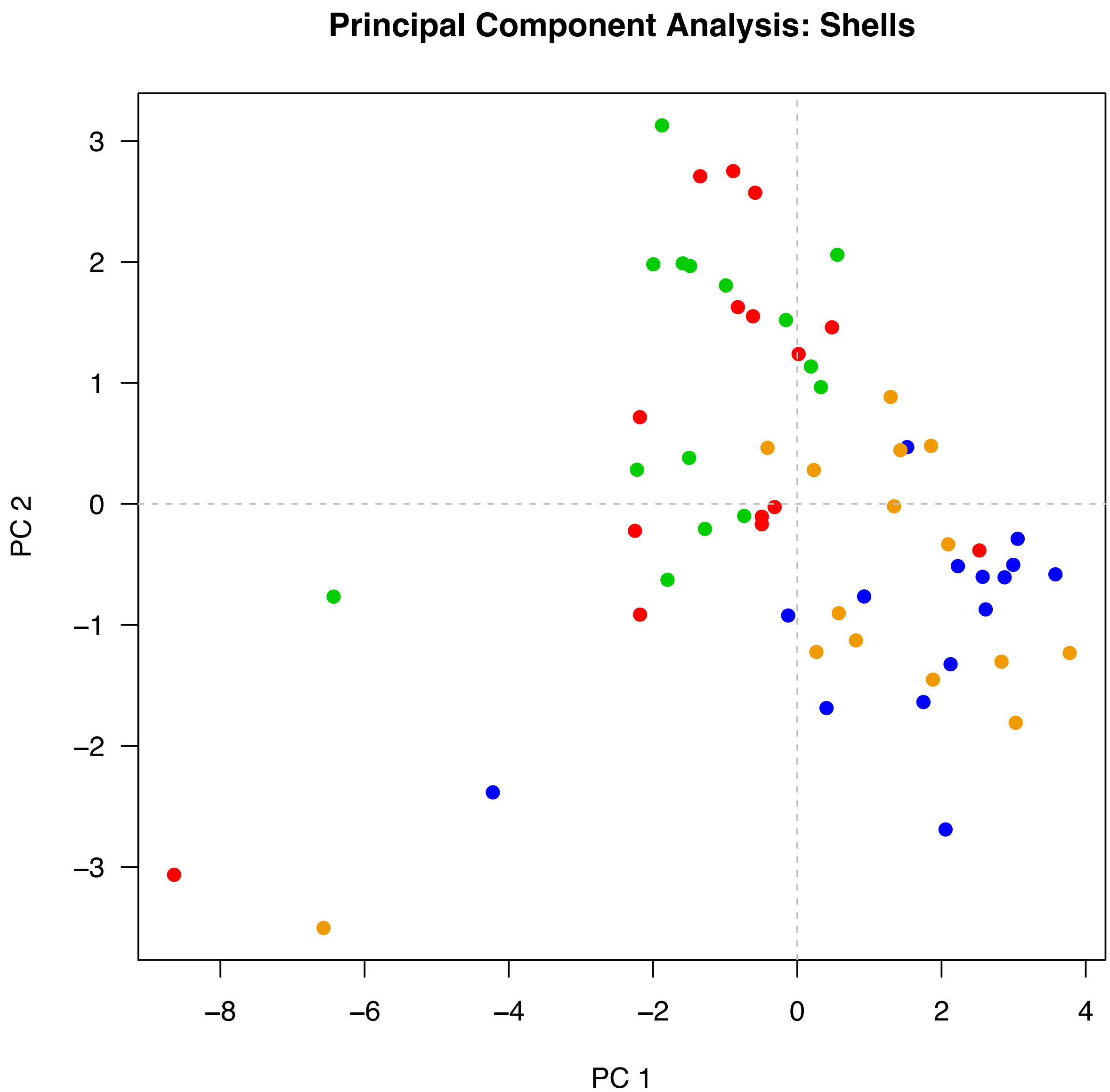


FIGURE 12. Principal component analysis (PCA) of log-normalized *Corbicula fluminea* shell data from Sites A (green), B (blue), C (red), and D (orange).

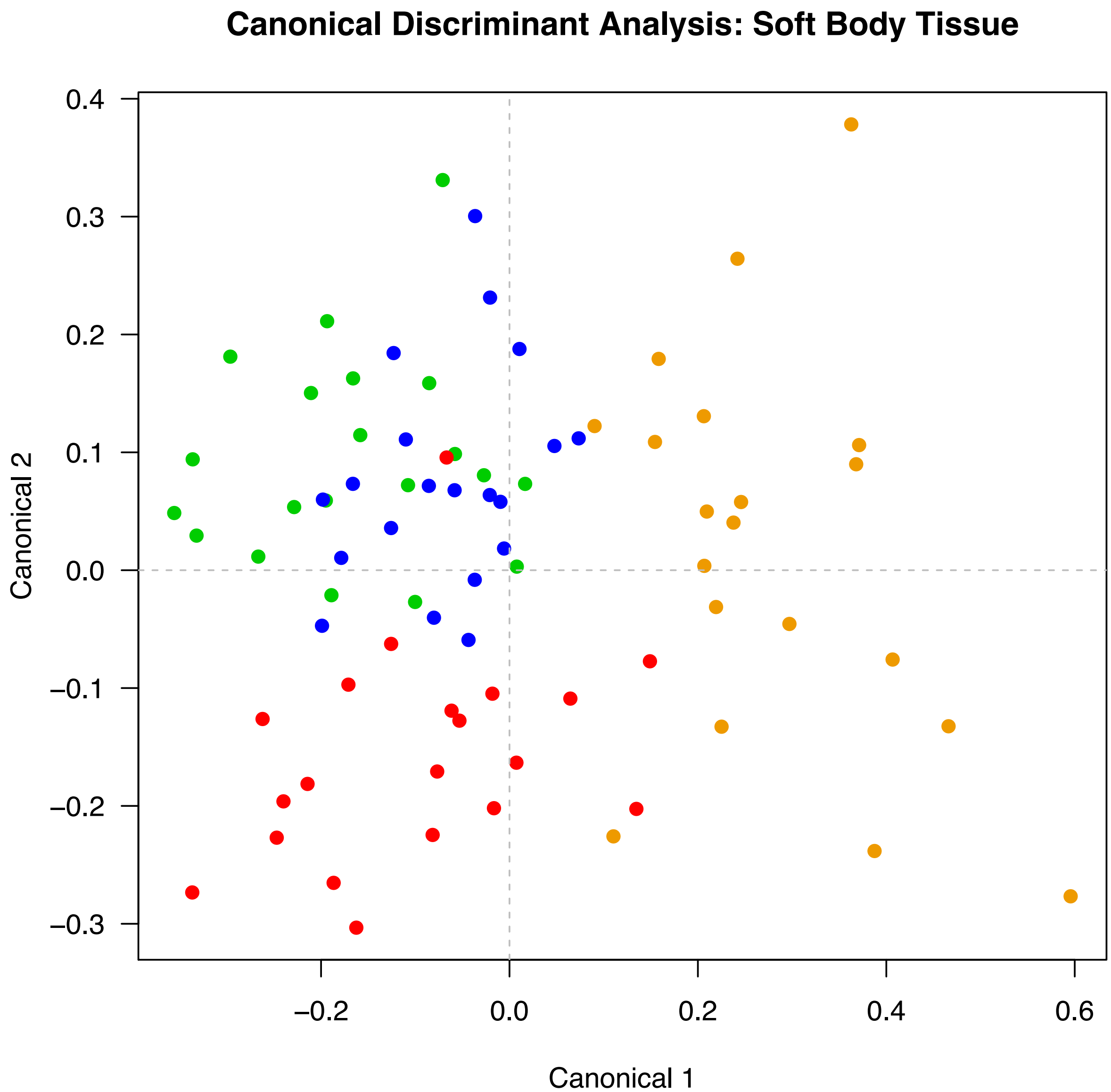


FIGURE 13. Canonical discriminant analysis (CDA) of log-normalized *Corbicula fluminea* soft body tissue data from Sites A (green), B (blue), C (red), and D (orange).

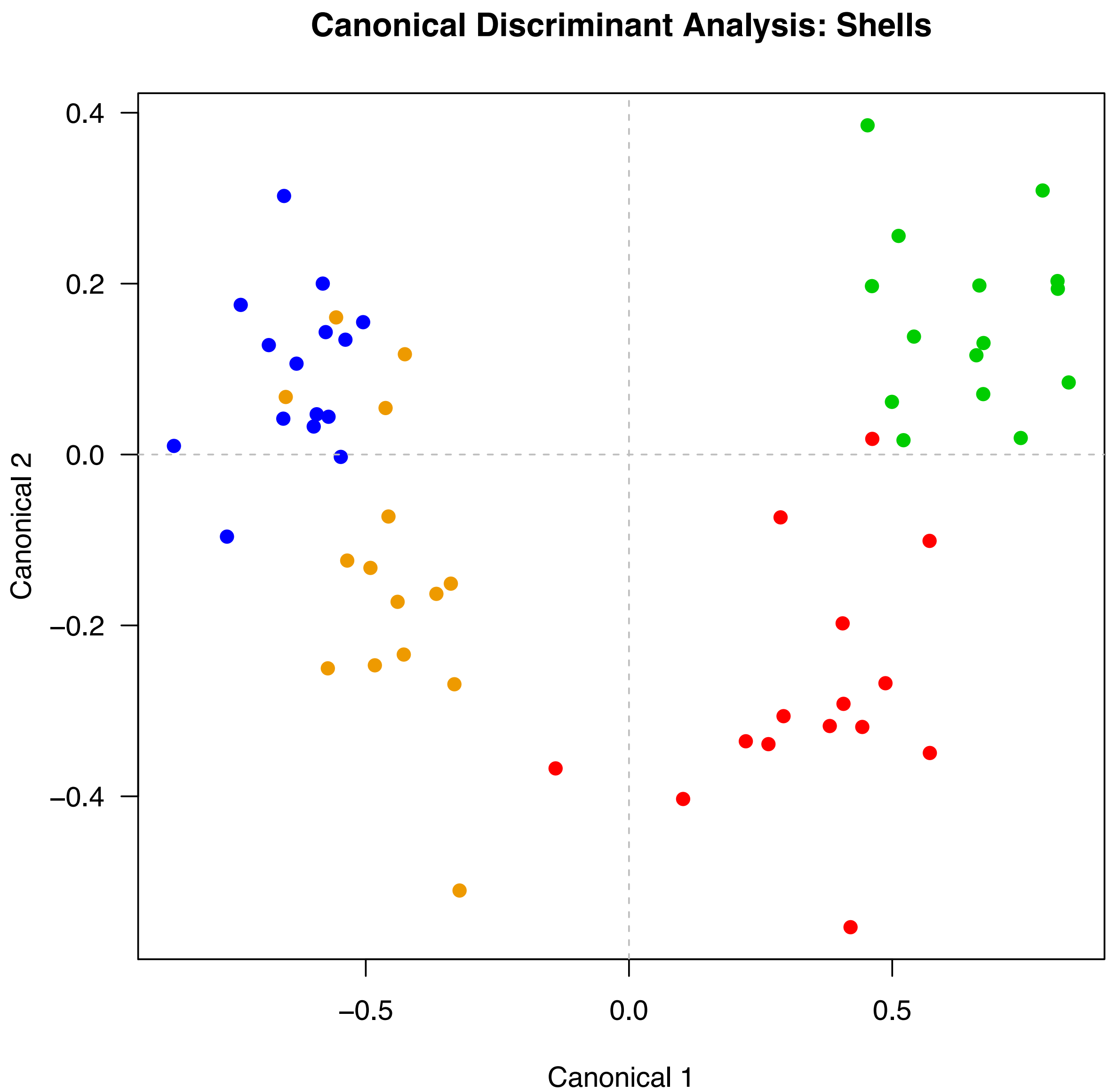


FIGURE 14. Canonical discriminant analysis (CDA) of log-normalized *Corbicula fluminea* shell data from Sites A (green), B (blue), C (red), and D (orange).

3.8 ANOVA: Soft Body Tissue

The univariate ANOVAs failed to detect an effect among sites ($p>0.05$) for the concentrations of Ag, As, Ba, Cd, Li, Mg, Ni, Pb, Rb, Se, and Sr in the soft body tissue (Table A-20). There were significant differences ($p<0.05$), however, among sites for Al, Cu, Fe, and Mn, and the results of the post-hoc Tukey's HSD tests are indicated in each element's box plot (Figure 7).

The mean concentration of Al was highest in those clams collected from Site C and lowest from Site B. According to the post- hoc Tukey's HSD test, the concentration of Al at Site C was significantly higher than Sites B and D ($p=0.0103$). The soft body tissues of clams collected from Site D were significantly enriched in Cu compared to the three other sites ($p<0.0001$), while the concentration of Fe from the same site was significantly lower ($p=0.0021$). Clams from Site A were significantly enriched in Mn compared to Sites C and D ($p=0.0002$), but did not differ significantly from Site B.

3.9 ANOVA: Shells

The univariate ANOVAs failed to detect an effect among sites ($p>0.05$) for the concentrations of As, Cd, Cu, and Mg in the *Corbicula* shells (Table A-21). However, there were significant differences ($p<0.05$) among sites for Al, Ba, Fe, Mn, Ni, Pb and Se, and the results of the post-hoc Tukey's HSD tests are indicated in each element's box plot (Figure 8).

Corbicula shells from Site A were significantly lower in Al than shells from Sites B and D, but did not differ significantly from Site C ($p=0.001$). Samples from Sites B and D were significantly enriched in Ba ($p<0.0001$). Site B clams were significantly enriched in

Fe compared to Sites A and C, while Site A clams were significantly lower than Sites B and D ($p<0.0001$). Site B clams were also significantly enriched in Mn, while those from Site C were significantly lower ($p<0.0001$). Concentrations of Ni and Pb were significantly higher in Sites B and D ($p<0.0001$). Site B showed significantly higher levels of selenium than Site A, but did not differ from the other two sites ($p=0.0191$).

CHAPTER 4 - DISCUSSION

4.1 Shell and soft tissue correlation

A laboratory study involving trace metal accumulation in *Perna perna* mussel soft tissue and shells using tanks spiked with varying levels of Cd, Cr, Cu, Ni, Pb, and Zn, showed that an increase in soft tissue concentration was consistently coupled with an increase in the shell concentration for all elements ($n=15$; $p<0.05$; $r=0.89$, 0.84, 0.72, and 0.84, respectively) (Bellotto and Miekeley 2007). Thus, the relatively weak, albeit often significant, correlations between the soft body tissue and shell concentrations I found for *Corbicula fluminea* were initially surprising (Table A-10). However, Bellotto and Miekeley's method involved analysis of only the newest growth ring to account for recent bioaccumulation (Bellotto and Miekeley 2007), which explains why the shell concentrations are so highly correlated with the soft tissue.

Soft tissue concentrations vary drastically over time, and, as such, largely represent only the water quality near the time of sampling (Brown et al. 2005). However, analysis of an entire shell, as my method entails, offers an average of bioaccumulation over a bivalve's lifetime (Brown et al. 2005). Thus, whole shell analysis should only correlate strongly with the soft body tissue if the average of water quality conditions over the organism's lifetime roughly matched the conditions at the time of sampling. My results show that for a number of elements including Al, Ba, Fe, Mn, Ni, Pb and Se, shell concentration data better differentiates among sites than does soft tissue data. As stated previously, the majority of work done using *Corbicula* as a biomonitor involves analysis of body tissue alone, but these results highlight the efficacy of using shells as records of lifelong exposure to contaminants.

4.2 Differences among sampling sites and legacy pollution

As previously mentioned, the remediation efforts undertaken by TVA between March 2009 and June 2010, removed over 2.6 million cubic meters of contaminated sediment from the Emory River (Bednar et al. 2013). Hydraulic dredges were used to pump ash and sediment out of the river and ultimately into a stilling pond (Figure 15). The water from the stilling pond was then used as cooling water for the power plant and released into the Clinch River. Site D is just downstream of the cooling water release site (Figure 15). Given these remediation efforts, I expected *Corbicula fluminea* from Site C, the site immediately downstream of the spill, to be lower in coal ash associated elements than those collected from Site D, but higher in concentration than those collected from Sites A, B and E.

Based on the coal ash to soil ratios reported by Ruhl et al. 2009 (Table 1), I expected the concentration of Al, As, Ba, Be, Cd, Cr, Co, Cu, Pb, Mg, Mo, Ni, Sr, V and Zn to be higher in *Corbicula* from those sites affected by the ash spill, Sites C and D. However, I expected that the *Corbicula* from these same sites would have similar or slightly lower concentrations of Fe, Mn, and Se, as these elements had an ash/soil ratio of less than one (Table 1) (Ruhl et al. 2009). Therefore, of the elements that were significantly different among sites in the soft tissue and shell data, I expected the concentrations of Al, Ba, Cu, Ni and Pb to be higher in samples from Sites C and D and the concentrations of Fe, Mn and Se in Sites C and D *Corbicula* to be similar or lower than those collected from the other sites. Figures 16 and 17 show the concentration by site of those elements that differed significantly among sampling sites for the soft tissue

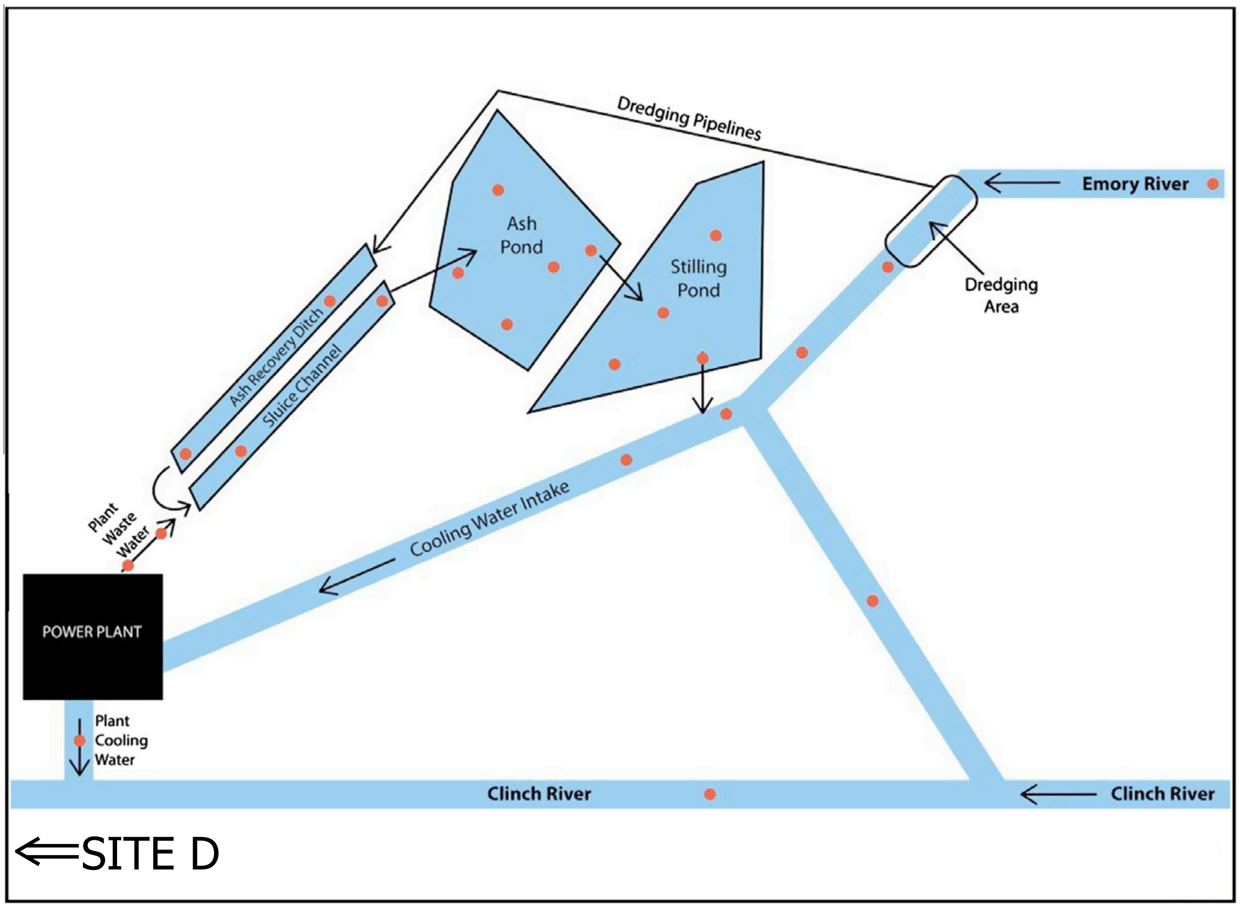


FIGURE 15. Diagram of TVA's hydraulic dredging remediation efforts taken from Bednar et al. 2013. The red dots indicate sampling locations of the Bednar et al. study, while the location of Site D in the present study is indicated with an arrow.

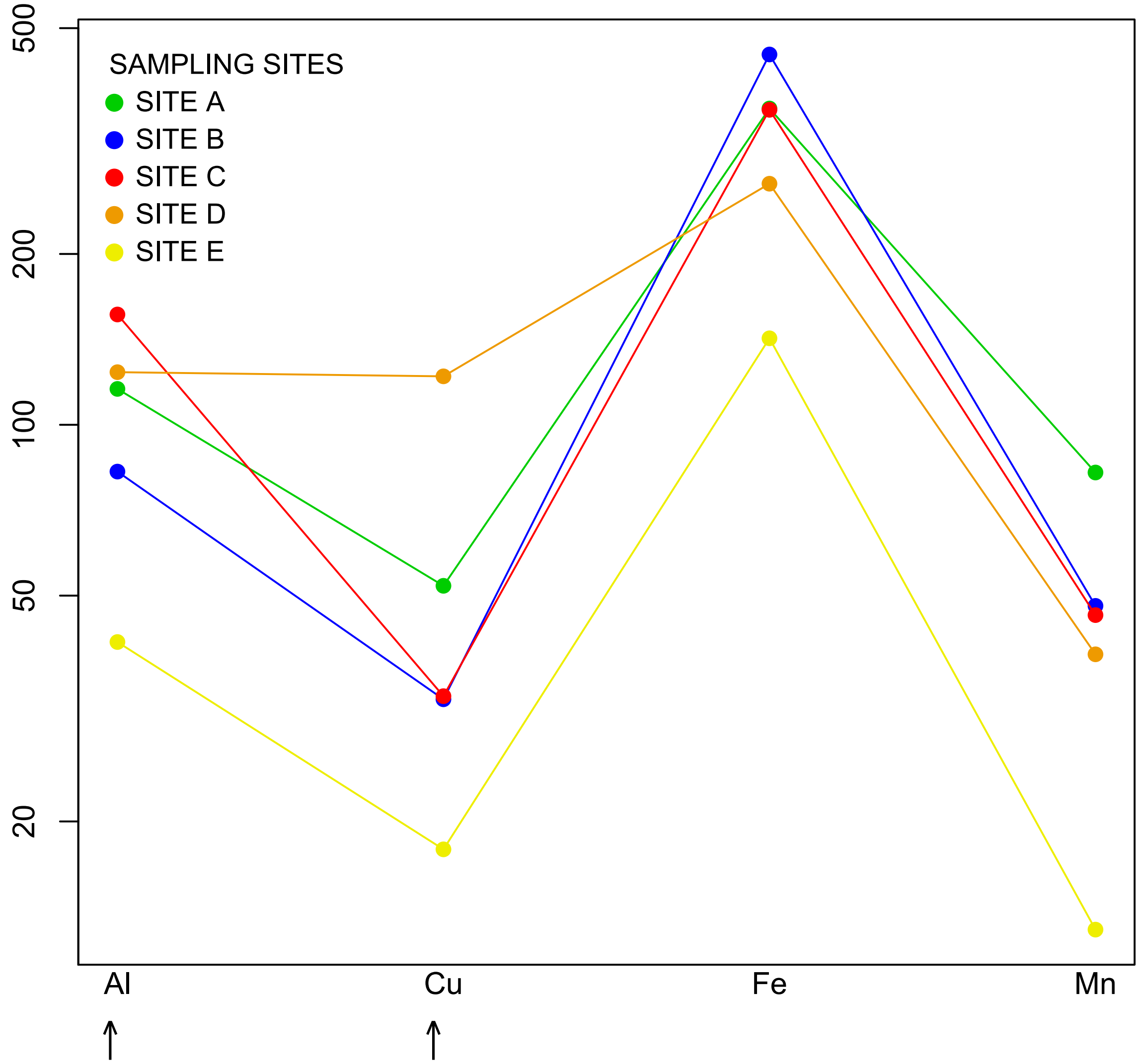


FIGURE 16. Average soft body tissue concentration (μg/g) of those elements that differed significantly among sampling sites based on analyses of variance (ANOVA). Arrows indicate a coal ash/background soil ratio of greater than one and, thus, the expectation that the concentration would increase in *Corbicula* from ash-affected sites.

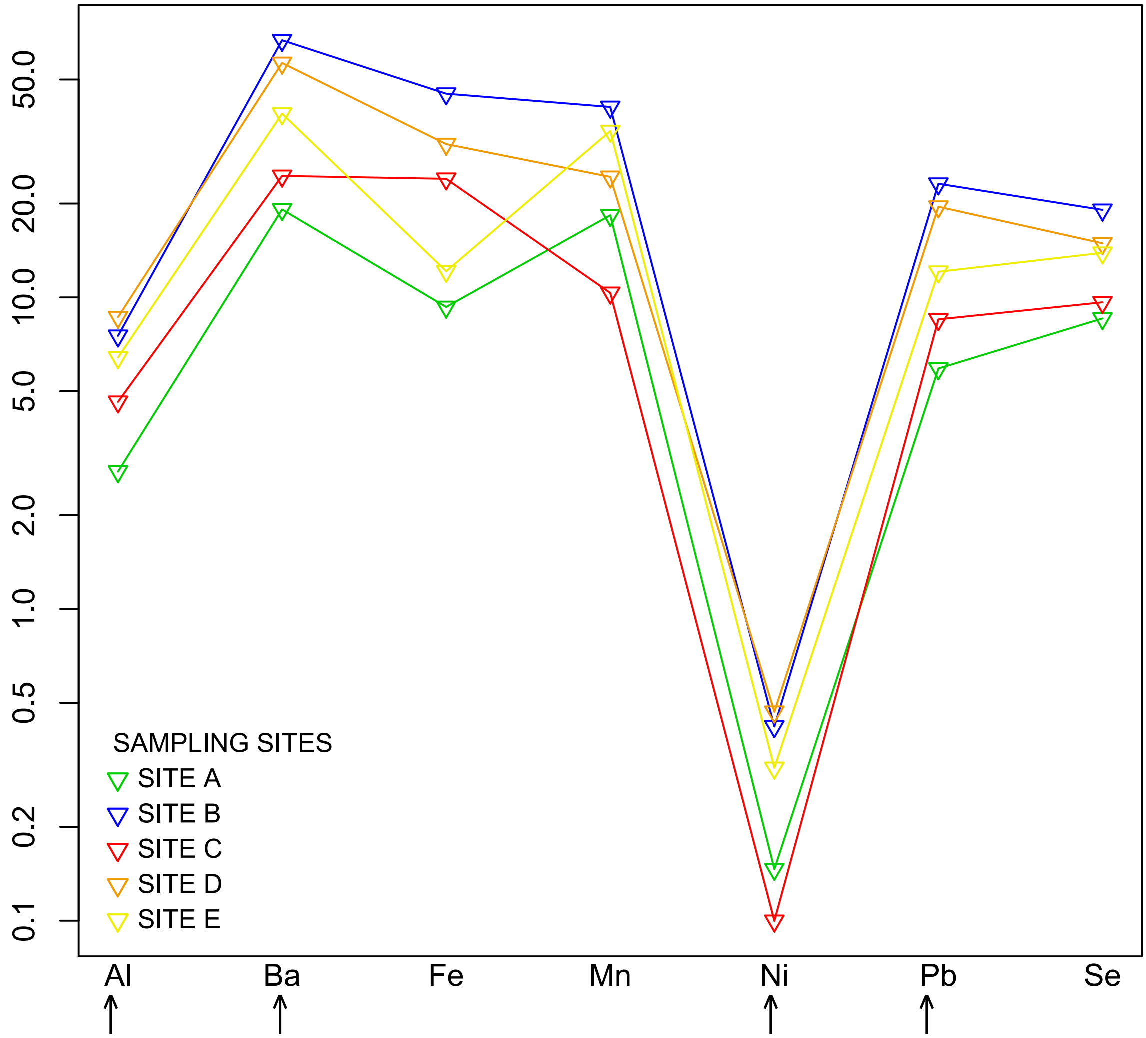


FIGURE 17. Average shell concentration (μg/g) of those elements that differed significantly among sampling sites based on analyses of variance (ANOVA). Arrows indicate a coal ash/background soil ratio of greater than one and, thus, the expectation that the concentration would increase in *Corbicula* from ash-affected sites.

and shell data sets. Arrows below the x-axis indicate elements with a coal ash/background soil ratio of greater than one (Figures 16 & 17).

While there were significant differences among sites for some of the elements in both the shell and soft tissue data, the elevated levels were not consistently associated with the Kingston Fossil Plant ash spill-affected sites (Figures 7 & 8, Tables A-20 & A-21, Figures 16 & 17). The soft bodies from ash-affected Site D, for example, were significantly enriched in Cu, but lower in Al and Fe compared to other sites. However, the coal ash composition reported by Ruhl et al. sheds light on this, as the ash/background soil ratios for Al and Fe were 1.5 and 0.9, respectively, while the concentration of Cu in the ash was 2.9 times that of the background soil (Table 1) (Ruhl et al. 2009). In the shell data, Sites B and D are consistently higher in concentration than Sites A and C. The data show that both Site B and Site D are significantly enriched in Al, Ba, Fe, Ni and Pb (ash/soil ratios: Al=1.5, Ba=4.8, Fe=0.9, Ni=5.2 & Pb=1.1 (Ruhl et al. 2009)). Site B shells were also highest in Mn and Se (ash/soil ratios: Mn=0.1 & Se=0.2 (Ruhl et al. 2009)), although only differing significantly from Site A in Se.

The Emory and Clinch Rivers are part of the Cumberland Plateau drainage basin of East Tennessee (Figures 18 & 19) (Etnier and Starnes 1993). This region has been extensively mined for its coal reserves, and early mining practices did not include runoff prevention or reclamation following strip-mining (Etnier and Starnes 1993). Thus, many tributaries of the Emory River system and, to a lesser extent, the lower Clinch were in the past and continue to be adversely affected by acid mine drainage (AMD) and mining-related siltation, which, along with other anthropogenic point and non-point

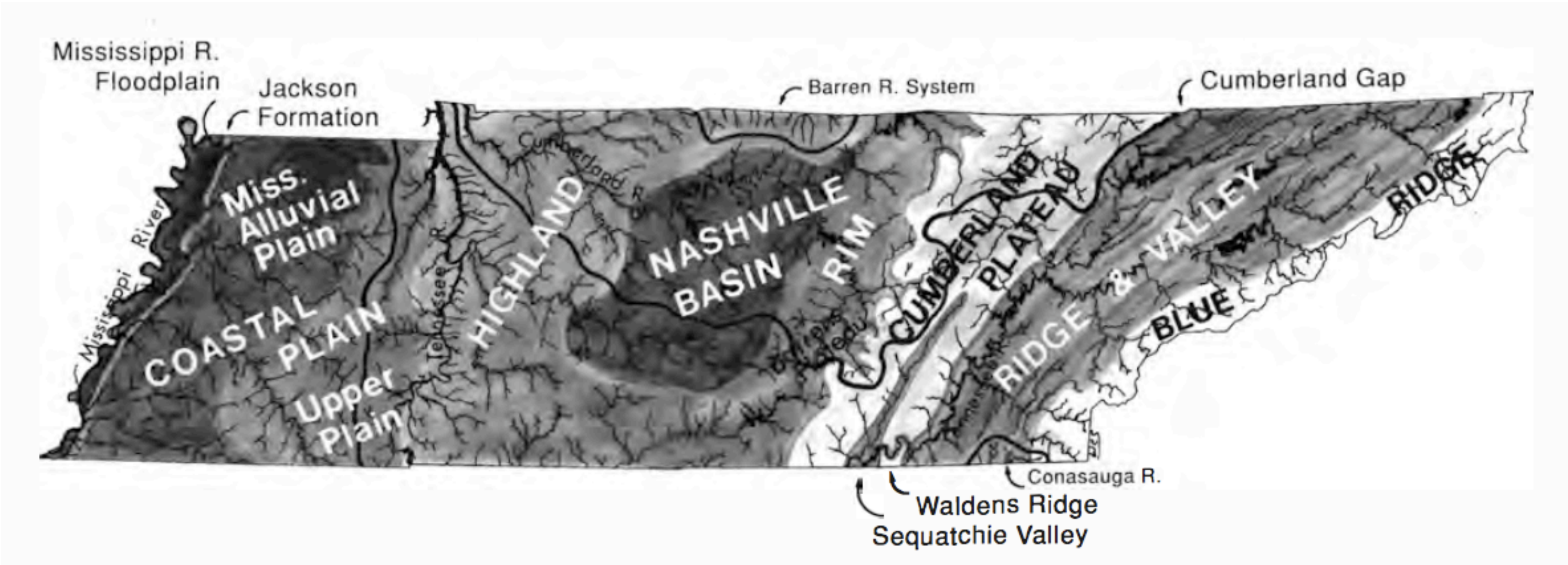


FIGURE 18. Physiographic provinces, selected geologic features, and major drainage basins of Tennessee (Etnier and Starnes 1993).

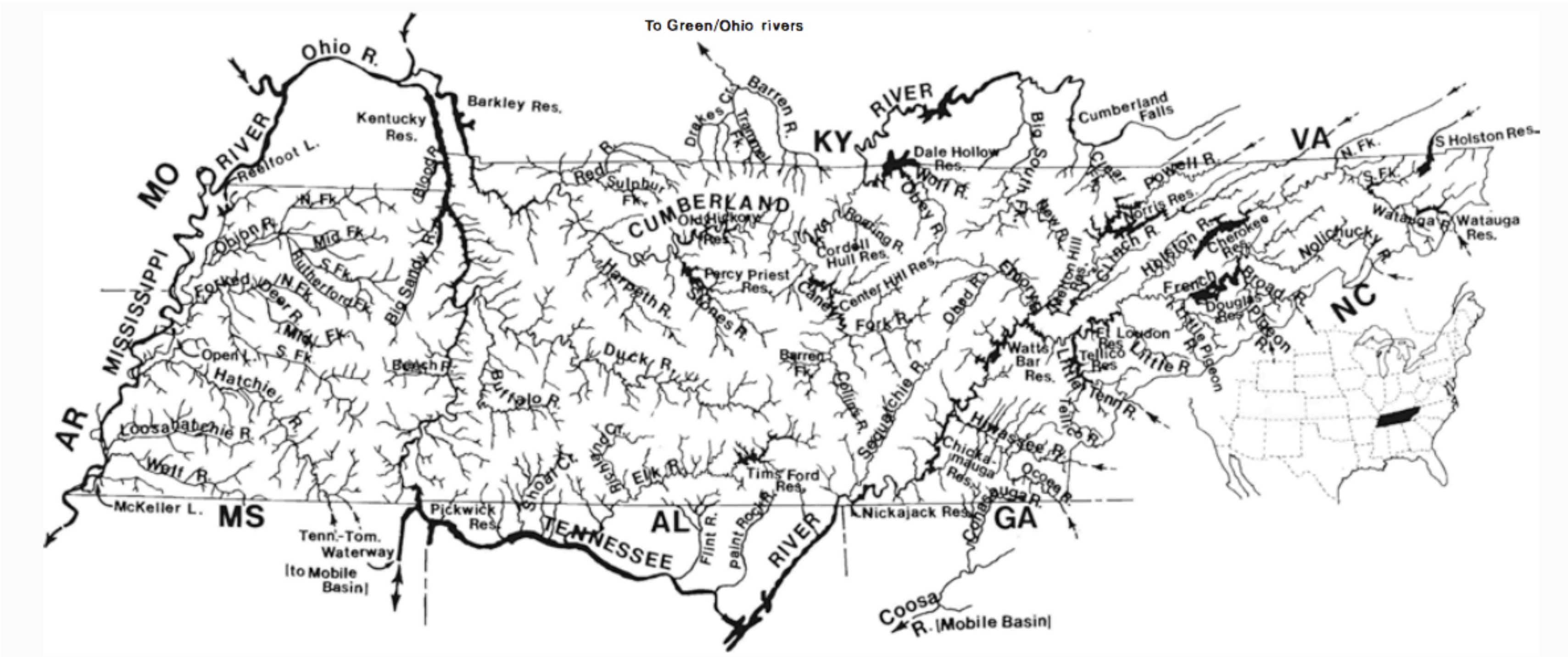


FIGURE 19. Drainages of Tennessee denoting major streams, lakes, and reservoirs (Etnier and Starnes 1993).

sources of pollution, has led to the extirpation and decline of countless aquatic organisms in the region and the accumulation of coal-related trace elements in the rivers' substrate (Etnier and Starnes 1993). Therefore, even ignoring the largest coal ash spill in United States history, the watersheds in this region of Tennessee are far from pristine.

4.3 Comparisons to past work

It is important to note that all of the elements included in the analysis are, of course, naturally occurring and would, to some extent, be found in the sediment, water, and organisms of the rivers even in the absence of human activity. Therefore, without a historical record of what a normal amount of bioaccumulation would be for *Corbicula* in these water bodies, it is impossible to say that these elements are necessarily bioaccumulating to a greater degree due to human activity. However, given the documented history of pollution in the Emory, Clinch and Tennessee Rivers, I also compared my results to the reported elemental concentrations of several previous studies including Peltier et al. 2008, Shoults-Wilson et al. 2010, Lewbart et al. 2010, and Graney et al. 1983. Comparison with reference sites of other studies may offer a better idea of the concentrations of some elements that might be expected to occur naturally in *Corbicula,* but it must be noted that natural levels would differ between watersheds due to the varying geology of the regions.

Peltier et al. collected *Corbicula fluminea* from 15 sites in the Chattahoochee and Broad River Basins in Georgia and report the soft body tissue concentrations (μg/g dry mass) of As, Cd, Cu, Hg, Se, and Zn (2008). I compared the mean concentrations of these elements from six of their sites, including three reference sites and three sites near coal fired power plants, to those concentrations in the *Corbicula fluminea* from the Kingston Fossil Plant sites (Figure 20). The levels of Cu they report from their reference sites are comparable to the levels in Sites B and C and roughly double the average of Site E. Site A levels fall in the range of the coal fired power plant affected sites, while Site D levels are double that of the average concentration of Cu in the three coal-affected sites. The Zn concentration of Sites B and C are similar to those from the coal-affected sites of Peltier et al., while the Zn concentration of Sites A and D are considerably higher. The Zn concentration of Site E is lower than the average for the Peltier et al. reference sites. Hg levels cannot be compared, unfortunately, as the level of Hg in my samples was below the detection limit of the ICP-OES.

Shoults-Wilson et al. report the average *Corbicula* soft tissue concentration of As, Cd, Cu, Hg, Mn, Ni, Pb, and Zn from 13 sites in the Altamaha River system of Georgia (Shoults-Wilson et al. 2010). The Kingston Fossil Plant river system sites show elevated levels of Cd, Ni, Pb and Zn compared to the Shoults-Wilson et al. average (Figure 21). The reported average level of Mn was similar to the level of Sites B, C and D, while Site A shows elevated levels, and Site E shows lower levels. The level of Cu in soft tissue from Site D is nearly double the average reported by Shoults-Wilson et al., while Sites A, B, C, and especially E are lower than the average. Hg levels could not be compared, as my diluted samples were below the detection limit.

As previously stated, the majority of work done using *Corbicula fluminea* as a biomonitor has been done using soft tissue concentrations alone, but Lewbart et al. looked at accumulation in both soft tissue and shells (2010). They report the soft tissue

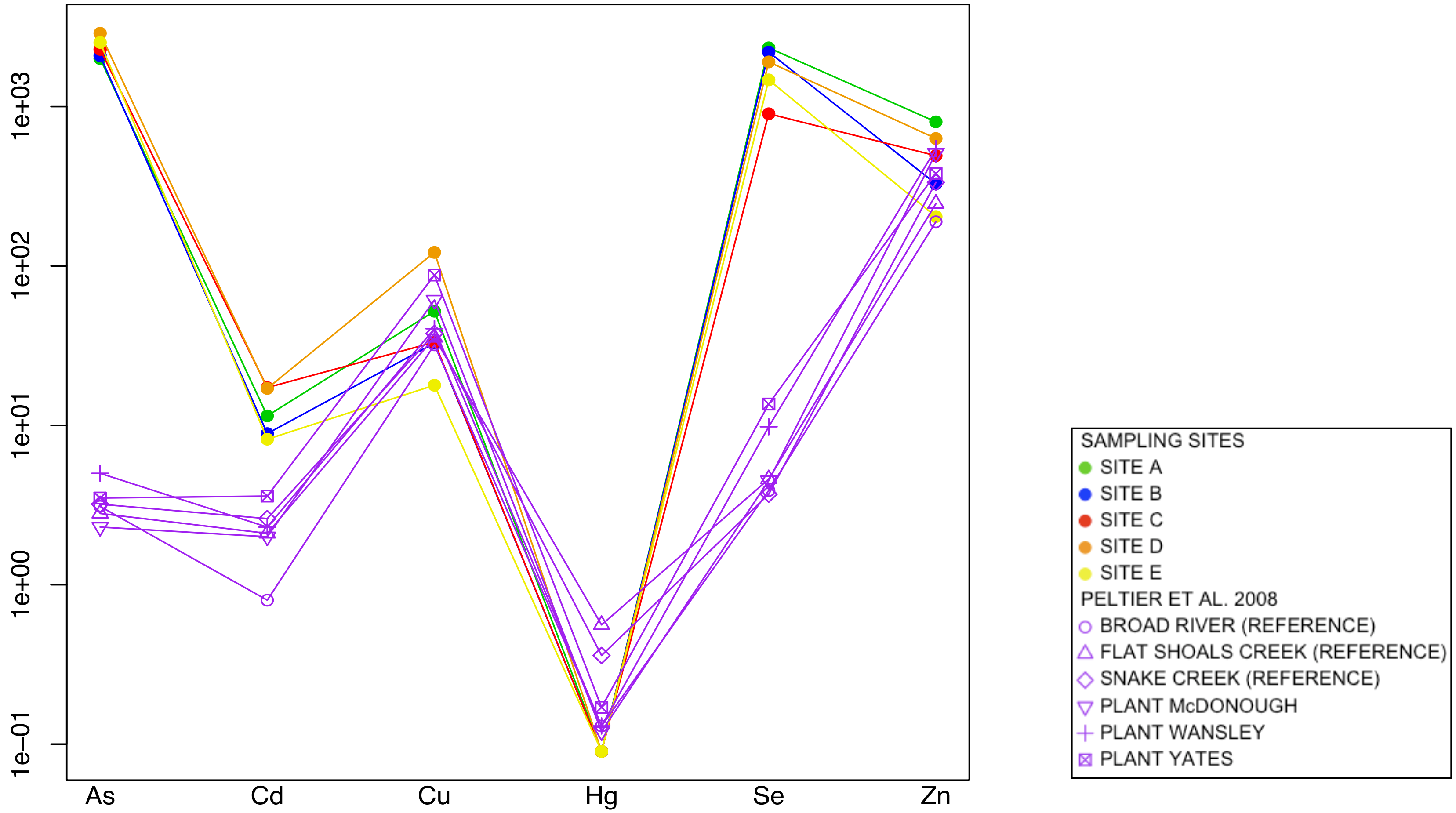


FIGURE 20. Soft tissue concentration (μg/g) comparison with data from Peltier et al. 2008.

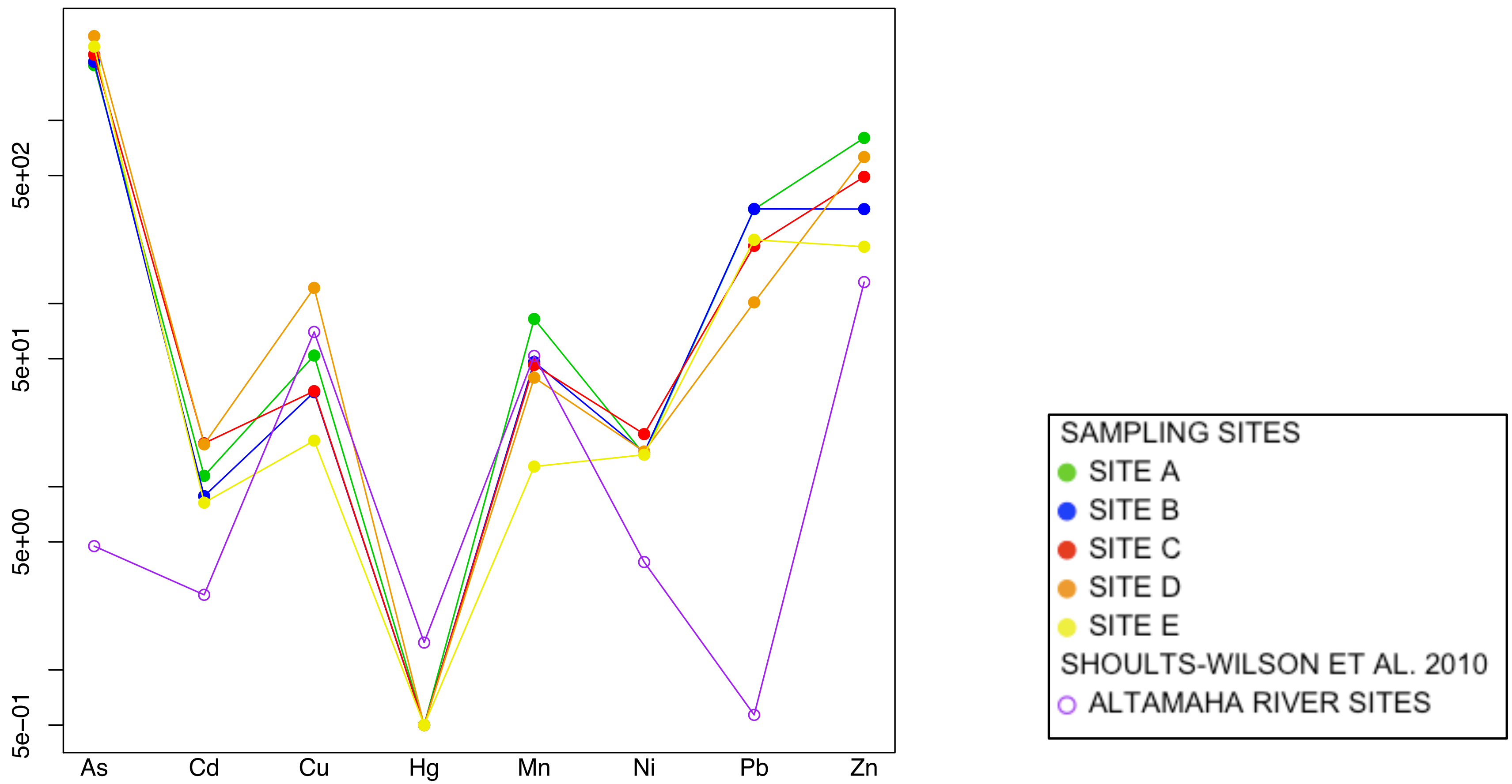


FIGURE 21. Soft tissue concentration (μg/g) comparison with Shoults-Wilson et al. 2010 data.

and shell concentration of As, Cd, Cu, Fe, Mn, Pb and Zn in Corbicula fluminea collected from the Choctawhatchee and Dead Rivers in Florida and Crabtree Creek and Lake Wheeler in North Carolina (2010). The soft body tissue data show that levels of Cd, Cu, Fe, Mn, Pb and Zn were, for the most part, considerably higher in the Kingston sites than those reported by Lewbart et alii (Figure 22). Levels of Cu, Fe, and Mn from Site E, however, were similar to those from the Lewbart et al. study. Levels of As were below the detection limit in the soft body tissue.

In the Lewbart et al. study, levels of As, Cd and Pb in the *Corbicula fluminea* shells were below the detection limit for their method, and cannot, therefore, be compared to levels of those elements in the present study (Lewbart et al. 2010). Zn concentrations were similar for all sites, with slightly elevated levels at Sites C and E (Figure 23). Levels of Mn were considerably higher in the Lewbart et al. shells across all sites. Cu levels were roughly four times higher in Kingston *Corbicula* than the average of those reported by Lewbart et al.

Graney et al. used in-field artificial stream systems spiked with varying levels of Cd, Cu and Zn to study the effectiveness of using *Corbicula* as a biomonitor (1983). They found that, of the three metals, Cu was concentrated to the greatest degree in the soft tissue relative to water concentration, while Zn showed the lowest potential for concentration (Graney Jr et al. 1983). The Cd concentration of the Graney et al. control group (average Cd concentration in water: 0.003 mg/L) is similar to concentrations in *Corbicula* soft tissues from Sites A, C and D, and slightly higher than that of Sites B and E (Figure 24) (1983). The average concentration of Cu in the Graney et al. artificial

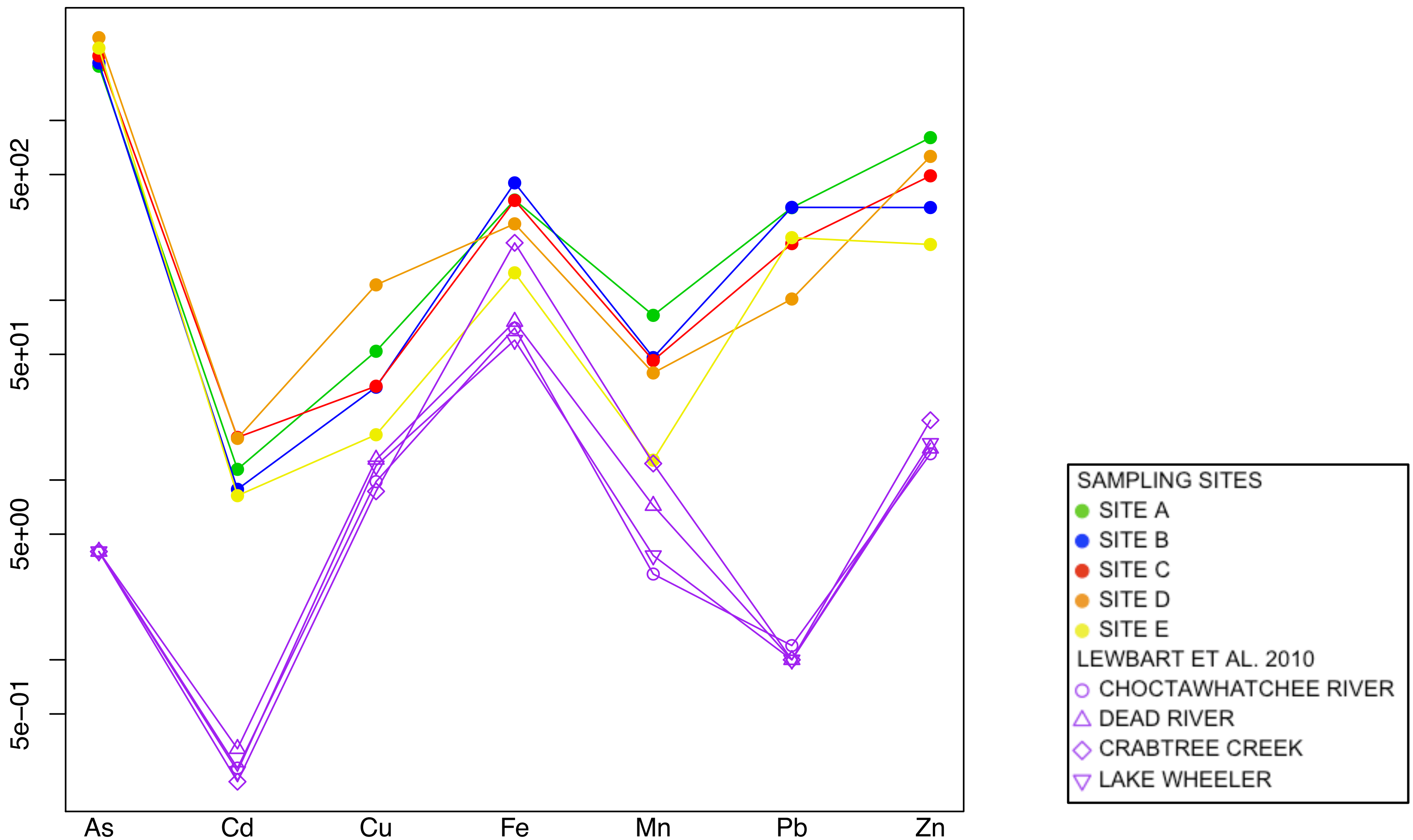


FIGURE 22. Soft tissue concentration (μg/g) comparison with Lewbart et al. 2010 data.

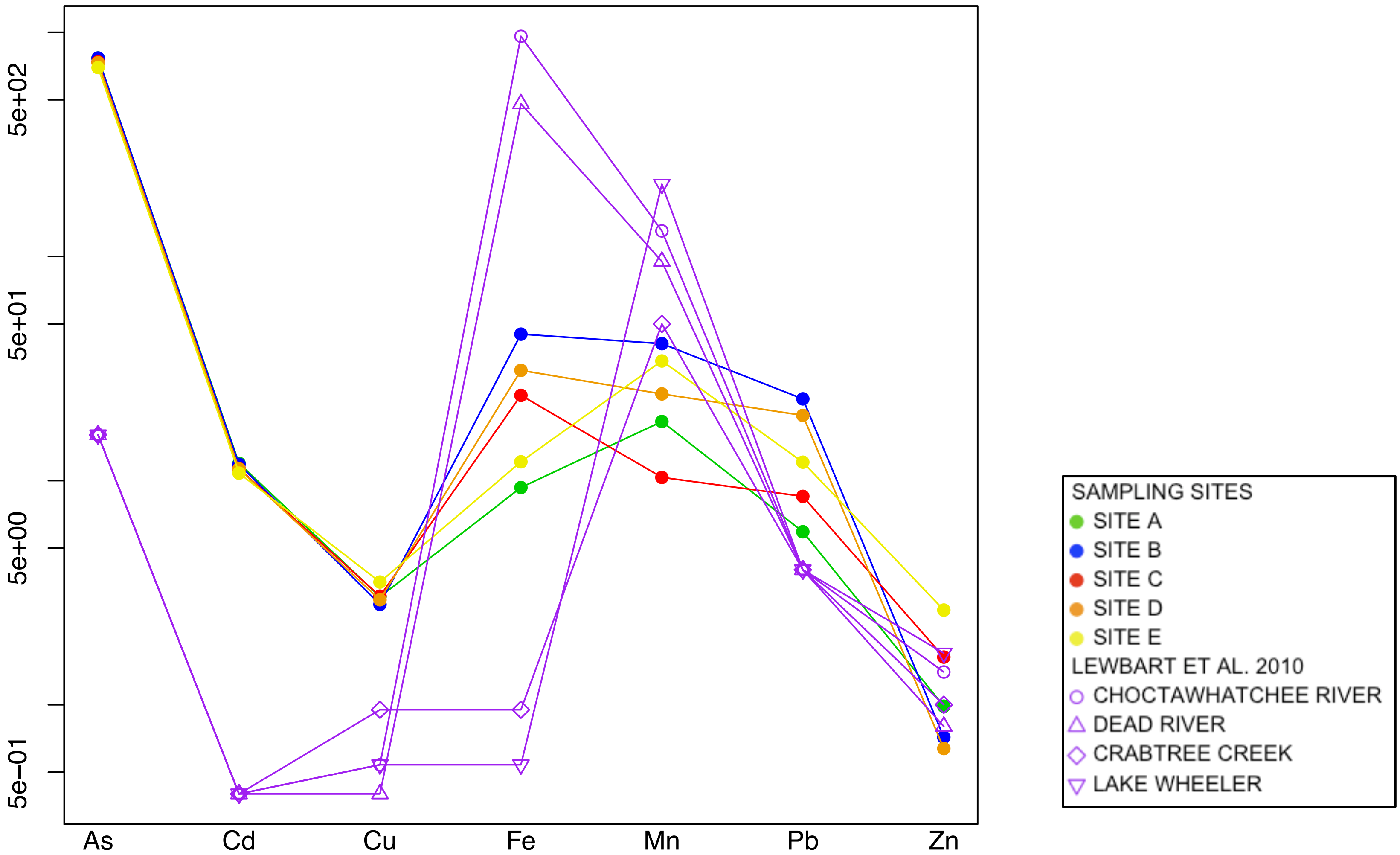


FIGURE 23. Shell concentration (μg/g) comparison with Lewbart et al. 2010 data.

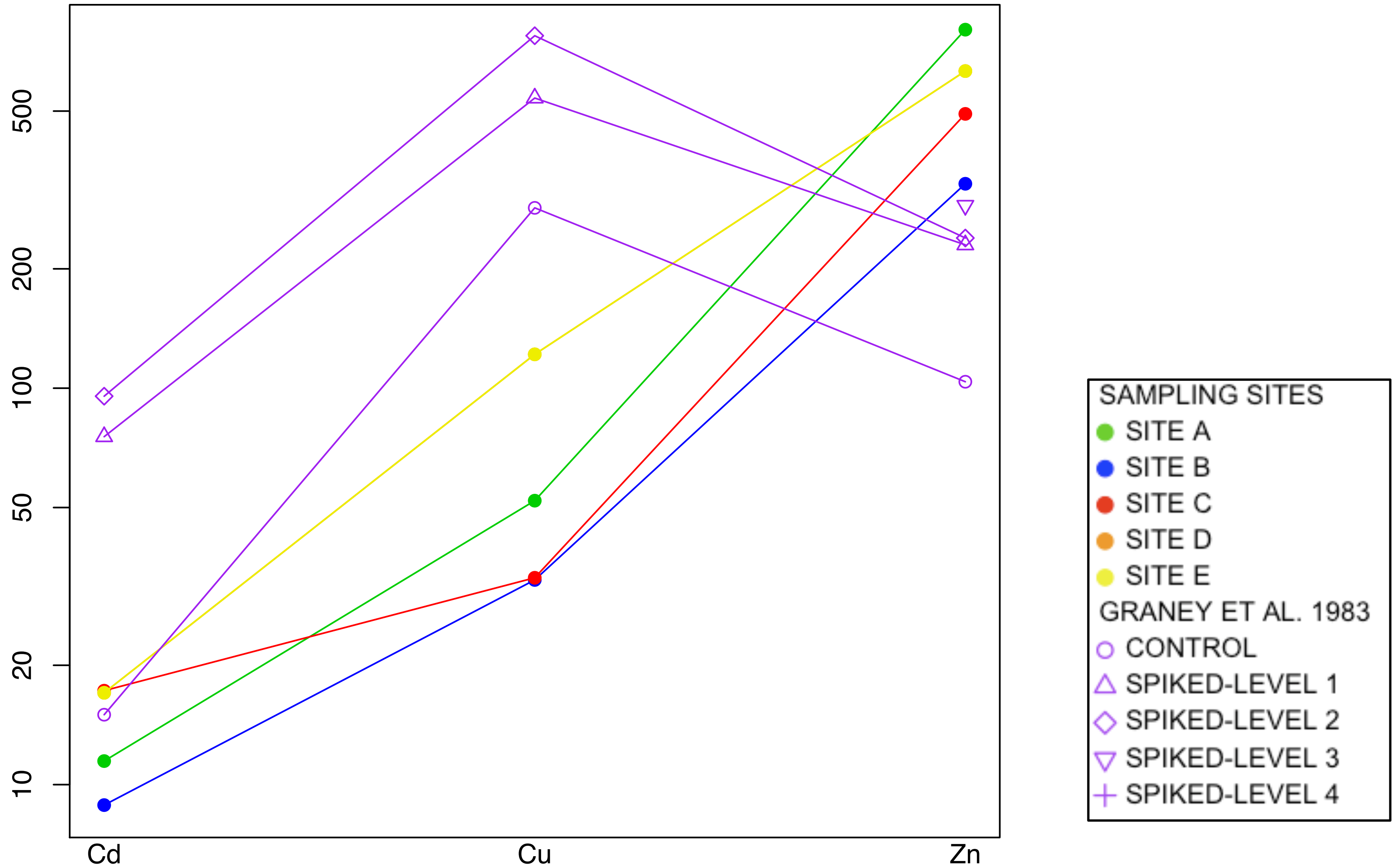


FIGURE 24. Soft tissue concentration (μg/g) comparison with Graney et al. 1983 data.

stream control group (average Cu concentration in water: 0.001 mg/L) is 285.01 μg/g (1983), which is more than double that of Cu enriched Site D and considerably higher than levels from Sites A, B, C and E. The levels of Zn in *Corbicula* soft tissue from Sites A, C, D, and, to a lesser degree, B exceeded that of even the highest dosed clams in the Graney et al. study, suggesting that the average water concentration of Zn at those sites exceeds Graney's reported average dosage of 0.835 mg/L (Graney Jr et al. 1983). The soft tissue concentration of Site E is similar to clams in water dosed with an average Zn concentration of 0.218 mg/L (Graney Jr et al. 1983).

The levels of Se and As in my samples are orders of magnitude higher than those reported by others (Peltier et al. 2008, Lewbart et al. 2010, Shoults-Wilson et al. 2010), which makes me question the validity of my results for those two elements from the ICP-OES. It is possible that the levels of As and Se in the standards used to calibrate the machine were lower than reported, which would skew the calibration curve and cause higher levels to be reported. If this is the case, the relative levels between sampling sites would still be valid (e.g. As levels at Site D are higher than at the other four sampling sites), but the reported μg/g would not be correct.

4.4 Bioaccumulation in the food web & human health implications

The majority of the work on the effects of coal combustion wastes on organisms has been done in lentic environments, in which stunted growth, population declines and extirpations, and increased offspring mortality in fish and amphibians have all been reported (Rowe et al. 2002, Peltier 2006, Peltier et al. 2009). While dilution in lotic environments may lead to decreased body burdens compared to those observed in lentic systems, even relatively small increases in the trace element concentrations in lower levels of a food web can lead to detrimental accumulation in higher trophic levels (Peltier et al. 2009). Many coal-related elements are not readily soluble in water, which can lead to concentration of the elements in sediment that far exceeds that of the river water (Strayer et al. 2004). Because they are not extremely soluble in water, these contaminants may persist indefinitely in the river system (Strayer et al. 2004). Areas of higher deposition in rivers and streams may accumulate nontrivial amounts of coal-related sediments over time, and invertebrates living in these hot spots may then cycle these elements throughout the food web (Peltier 2006, Peltier et al. 2009).

The concentration of trace elements in benthic macroinvertebrates, as this study exhibits in *Corbicula fluminea*, provides insight on inevitable bioaccumulation in higher trophic levels. Many molluscivorous fish are known to consume *Corbicula* based on gut content analysis (Robinson and Wellborn 1988). Robinson and Wellborn present a by no means exhaustive list of a number of documented *Corbicula* feeders including carp (*Cyprinus carpio*), smallmouth buffalo (*Ictiobus bubalus*), black buffalo (*Ictiobus niger*), warmouth (*Lepomis gulosis*), bluegill (*Lepomis macrochirus*), freshwater drum (*Aplodinotus frunniens*), American shad (*Alosa sapidissima*), striped bass (*Roccus saxatilis*), river redhorse (*Moxostoma carinatum*), channel catfish (*Ictalurus punctatus*), blue catfish (*Ictalurus furcatus*), and sturgeon (*Acipenser)* (Robinson and Wellborn 1988). All of these species are present in the large reservoirs of East Tennessee and are consumed by both wildlife and humans (TWRA 2008). Exposure to coal ash-related toxins is associated with an increased risk of a number of different types of cancer, lung disease, and heart disease (Ruhl et al. 2009). Thus, the accumulation of these toxins in our food supply is far from ideal.

CHAPTER 5 - CONCLUSION

This study demonstrates that trace elements associated with coal mining and coal combustion are bioavailable to *Corbicula fluminea* in the river systems in the vicinity of the Kingston Fossil Plant. The presence of *Corbicula fluminea* in both lentic and lotic freshwater habitats worldwide coupled with the multitude of traits that make the species an exceptional biological monitor make the invasive clam an excellent model organism for use in ecotoxicology (Sousa et al. 2008). Given the species' ubiquitous presence in the aquatic systems surrounding the Kingston Fossil Plant, *Corbicula fluminea* could be used as a component of the long term monitoring of the 2008 ash spill. The use of a globally widespread and abundant organism such as *Corbicula* to assess the amount of bioavailable metals in polluted systems decreases the need to use threatened native mussels and fish as biomonitors.

The global demand for energy will continue to rise with the pressures of increased population and economic development, and the combustion of coal will continue to be used to meet that growing need (Tuberty et al. 2009). Currently, the long-term effects of coal-related pollution on ecosystems is understudied and poorly understood, and the majority of the knowledge we do have is based on laboratory tests or closed-system field studies (Tuberty et al. 2009). The Kingston Fossil Plant ash spill offers an opportunity to boost our understanding of the manner in which trace elements move and cycle in open aquatic systems and how these contaminants bioaccumulate in the food chain after a coal-related spill (Tuberty et al. 2009). The methods of remediation following an ash spill are far from perfect, and understanding the way these methods affect aquatic systems may lead to improved techniques for future spills.

As Etnier and Starnes so eloquently put it, “the extremely involved geologic history of Tennessee and the correspondingly complex physiographic makeup of the state…have resulted in a theater of evolution for aquatic organisms unmatched elsewhere in North America; and many groups, including fishes, have responded wonderfully, giving us our richest of all faunal heritages” (Etnier and Starnes 1993). Hopefully, an increased understanding of the long-term effects of coal combustion on aquatic ecosystems will influence energy policy and the regulation of coal waste storage in the future (Tuberty et al. 2009), and perhaps we will be able to salvage what remains of the rich aquatic diversity of Tennessee.

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TWRA. 2008. TWRA's Region 4-East Tennessee Reservoir Fisheries Management Program. Photo Gallery of Tennessee Reservoir/Big River Fish.

APPENDICES

APPENDIX A

TABLE A-1. GPS coordinates for the five *Corbicula fluminea* collection sites.

	N	**W**
SITE A	35^{o} 54.955'	084^{o} 26.199'
SITE B	35^{o} 56.999	084^{o} 34.555'
SITE C	35^{o} 47.437'	084^{o} 39.256'
SITE D	35^{o} 52.910	084^{o} 31.931'
SITE E	35^{o} 53.361	084^{o} 29.379'

TABLE A-2. *Corbicula fluminea* morphometric data and the total volume of the stock solutions following acid digestion.

CLAM ID	SHELL LENGTH (mm)	SHELL WIDTH (mm)	SHELL HEIGHT (mm)	SOFT BODY DRY WEIGHT (g)	SHELL DRY WEIGHT (g)	ACID DIGESTION VOLUME (ml)
A1	14.33	13.18	10.21	0.01751	1.03789	5.8
A2	17.35	15.91	11.8	0.05468	1.55317	8.2
A3	14.59	13.66	10.44	0.02561	1.05653	5.5
A4	13.2	11.87	9.46	0.01501	0.77643	4.9
A5	11.99	10.23	8.04	0.00936	0.45765	5.1
A6	12.33	11.06	8.81	0.01434	0.60876	5.1
A7	17.05	15.75	11.82	0.04369	1.5827	9.7
A8	13.99	12.98	10.21	0.02049	0.96331	6
A9	11.9	10.5	8.06	0.01306	0.47452	5
A10	14.39	12.74	9.69	0.02148	0.89365	5.9
A11	17.87	16.84	12.68	0.05673	1.87684	10.1
A12	19.44	18.08	13.08	0.07417	2.36616	12.4
A13	14.95	13.82	10.46	0.02493	1.1676	7.3
A14	11.51	10.07	7.98	0.00942	0.46515	5.2
A15	13.29	11.91	9.45	0.01472	0.77529	5.8
A16	14.22	13.16	10.2	0.02343	1.01676	6
A17	16.63	15.08	11.43	0.04156	1.44175	9.8
A18	13.47	11.88	9.21	0.01442	0.73214	5.8
A19	18.65	17.92	13.58	0.0743	2.3849	10.1
A20	14.67	13.39	10.18	0.02773	1.01385	5.6
B1	15.19	13.47	9.93	0.02773	0.9114	6
B2	13.69	11.96	8.58	0.01968	0.63221	5
B3	13.73	12.16	8.71	0.01812	0.58816	5.1
B4	12.63	11.07	8.01	0.01159	0.41517	5.1
B5	17.58	15.69	10.89	0.04277	1.30282	8.6
B6	16.72	15.34	11.27	0.04206	1.26947	6.1
B7	17.42	15.54	11.01	0.03566	1.35179	7.5
B8	14.42	12.81	9.39	0.02008	0.74521	5
B9	12.63	10.96	8.53	0.01556	0.49309	5
B10	13.81	12.09	8.59	0.01876	0.57411	4.9
B11	16.58	14.6	10.46	0.03656	1.11817	7.5
B12	16.17	14.62	10.67	0.03244	1.21241	6.1
B13	14.46	12.89	9.38	0.02026	0.72162	5
B14	15.06	13.2	9.89	0.02793	0.90162	5.9
B15	11.97	10.72	8.26	0.01388	0.47342	5
B16	13.96	12.3	8.95	0.02025	0.65432	4.9
B17	18.84	16.92	12.25	0.05113	1.88387	10
B18	15.98	14.44	10.72	0.03547	1.08961	6.2
B19	16.32	14.14	10.04	0.03517	1.03404	5.8

TABLE A-2. Continued.

CLAM ID	SHELL LENGTH (mm)	SHELL WIDTH (mm)	SHELL HEIGHT (mm)	SOFT BODY DRY WEIGHT (g)	SHELL DRY WEIGHT (g)	ACID DIGESTION VOLUME (ml)
B20	15.12	13.52	9.65	0.03156	0.86331	5.9
C1	16.63	14.64	10.9	0.04412	1.10293	6
C2	11.59	10.74	8.47	0.01764	0.53551	5
C3	15.27	13.71	10.7	0.03456	1.13698	6
C4	14.27	13.14	9.92	0.03198	0.91622	6
C5	13.14	12.58	9.42	0.02283	0.79691	6
C6	18.71	16.53	12.26	0.07109	1.62876	9.9
C7	16.4	14.36	11.24	0.04207	1.11539	6.1
C8	14.58	13.02	10.03	0.03146	0.83021	5.9
C9	14.02	12.86	9.83	0.02326	0.88217	6
C10	12.56	10.62	8.68	0.01967	0.50648	4.9
C11	18.23	15.76	12.13	0.06087	1.57655	10
C12	14.26	12.98	9.96	0.02564	0.87871	5.9
C13	15.67	13.63	10.55	0.03797	1.01413	5.8
C14	15.83	13.55	10.76	0.04126	0.93314	5.8
C15	14.02	12.02	9.59	0.02126	0.64739	4.9
C16	19.13	18.14	13.46	0.13957	2.00416	9.9
C17	14.01	12.27	9.5	0.03134	0.80388	5.8
C18	15.05	13.89	10.54	0.03888	1.12497	5.9
C19	13.11	11.62	9.25	0.02563	0.63911	4.9
C20	13.96	11.95	9.14	0.02354	0.64298	5.2
D1	12.82	12.48	9.45	0.01811	0.69169	5
D2	16.33	14.93	10.49	0.04198	1.10974	6
D3	13.41	13.96	10.53	0.02932	1.21892	6.6
D4	13.83	14.5	10.95	0.02973	1.36318	8
D5	12.03	11.53	8.67	0.01546	0.55598	5.1
D6	16.68	15.76	12.09	0.02781	1.38749	7.1
D7	14.54	15.89	12.28	0.04786	1.75883	9.9
D8	12.2	12.59	9.41	0.02064	0.86397	6
D9	11.9	12.73	9.61	0.02332	0.92249	5.9
D10	15.23	13.86	10.31	0.04145	0.94278	5.8
D11	14.44	15.09	11.62	0.02136	1.53502	9.5
D12	16.95	15.76	11.91	0.05121	1.4996	6.9
D13	17.97	18.91	14.48	0.05964	2.91324	12.6
D14	15.54	14.08	10.11	0.03459	1.00095	5.6
D15	12.67	12	9.07	0.01777	0.65382	5.7
D16	13.69	14.69	10.97	0.03044	1.36864	10.1
D17	15.26	16.31	12.67	0.05579	1.94871	7.5
D18	14.05	14.55	11	0.02227	1.37702	8.4
D19	15.86	16.55	12.53	0.04674	1.99851	12.2
D20	13	13.95	10.28	0.03226	1.15446	8.4
E1	21.74	18.91	13.76	0.16376	2.39397	12.1

TABLE A-2. Continued.

CLAM ID	LENGTH (mm)	WIDTH (mm)	HEIGHT (mm)	SOFT BODY DRY WEIGHT (g)	SHELL DRY WEIGHT (g)	ACID DIGESTION VOLUME (ml)
E2	17.33	15.3	10.91	0.08236	1.20584	6
E3	17.7	16.02	11.25	0.02857	1.22496	6
E4	15.72	13.39	9.16	0.05454	0.74503	5.9
E5	16.22	14.59	10.2	0.05914	0.98297	5.9
E6	20.65	17.85	12.5	0.10332	1.72697	10.1
E7	14.26	12.57	9.23	0.02936	0.71229	5
E8	13.11	10.9	7.72	0.03144	0.38761	5.1
E9	14.86	12.66	9	0.04228	0.62275	5.9
E10	15.92	13.97	10.05	0.05296	0.91465	5.8
E11	23.01	20.47	14.53	0.17096	3.00917	13
E12	21.58	18.53	13.33	0.13694	2.28692	11.9
E13	21.6	19.45	13.25	0.14729	2.3821	12
E14	10.93	9.18	6.53	0.00839	0.23729	4.9
E15	11.38	9.24	6.63	0.01322	0.25233	5.1
E16	27.18	23.68	16.61	0.25688	4.03611	14.5
E17	23.72	20.77	14.92	0.19659	3.04545	14.6
E18	22.91	21.11	15.98	0.08874	3.39206	13.6
E19	22.98	20.34	14.03	0.15514	2.90973	13.5
E20	21.34	18.76	13.18	0.14447	2.32223	13.9

TABLE A-3. Pearson's correlations (r) between log-transformed shell length, width and height across all sites. Bold values indicate significance at $p<0.05$.

	LENGTH	**WIDTH**	**HEIGHT**
LENGTH	1	**0.9416**	**0.8952**
WIDTH	**0.9416**	1	**0.977**
HEIGHT	**0.8952**	**0.977**	1

TABLE A-4. Principal component analysis (PCA) on log-transformed length, width, and height of *Corbicula fluminea* shells across all sites.

PC NUMBER	**EIGENVALUE**	**PERCENT**	**CUMULATIVE PERCENT**
1	2.8763	95.875	95.875
2	0.1093	3.642	99.518
3	0.0145	0.482	100
LOADING MATRIX			
	PC1	**PC2**	**PC3**
LENGTH	0.96541	0.25902	0.02995
WIDTH	0.99379	-0.05761	-0.0952
HEIGHT	0.97807	-0.19713	0.06717

TABLE A-5. ANOVA and Tukey's HSD results for shell length, shell width, shell height, body dry weight, and shell dry weight. Sites not connected by the same letter are significantly different.

	F RATIO	p VALUE	SITE A	SITE B	SITE C	SITE D	SITE E
SHELL LENGTH	8.383	<0.0001	B	b	b	b	a
SHELL WIDTH	4.9768	0.0011	B	b	b	ab	a
SHELL HEIGHT	3.0475	0.0207	Ab	b	ab	ab	a
BODY DRY WEIGHT	13.9945	<0.0001	B	b	b	ab	a
SHELL DRY WEIGHT	4.8117	0.0014	B	b	b	b	a

TABLE A-6. Micrograms of element per gram (μg/g) dry weight of soft body tissue. Values of zero indicate concentrations in the samples below the detection limits of the ICP-OES.

SAMPLE	Ag	Al	As	Ba	Be	Cd	Co	Cr	Cu	Fe	Ga	Hg	K
A1B	94.031	151.275	2253.847	70.484	0.000	56.039	0.000	37.846	255.955	456.684	6.398	0.000	4425.344
A2B	30.120	86.780	1333.964	29.761	0.000	41.643	0.000	42.839	90.525	387.385	0.000	0.000	2561.449
A3B	18.624	163.325	4065.189	50.465	0.000	20.240	0.000	3.630	69.761	435.598	0.000	0.000	3356.056
A4B	5.711	113.233	0.000	35.803	0.000	0.000	0.000	0.000	28.117	350.717	0.000	0.000	2428.609
A5B	9.337	185.929	2944.971	0.000	0.000	0.000	0.000	0.000	7.799	431.137	0.000	0.000	2930.447
A6B	15.767	157.314	4231.816	2.650	0.000	0.000	0.000	0.000	2.102	435.031	0.000	0.000	2717.308
A7B	1.320	101.361	2781.270	4.318	0.000	19.753	0.000	0.319	21.468	306.215	0.000	0.000	3290.294
A8B	3.695	177.625	263.354	28.453	0.000	46.091	0.000	0.000	0.000	475.442	0.000	0.000	2957.392
A9B	0.000	205.807	4051.949	0.000	0.000	0.000	0.000	0.000	0.000	431.647	0.000	0.000	2865.287
A10B	0.000	126.346	2805.074	0.000	0.000	0.000	0.000	0.000	0.000	380.777	0.000	0.000	3034.071
A11B	37.570	117.407	1150.467	0.000	0.000	34.765	0.000	0.000	261.322	197.257	0.000	0.000	2302.764
A12B	3.135	77.945	0.000	7.202	0.000	0.000	0.000	0.000	46.068	201.957	0.000	0.000	2466.700
A13B	0.375	94.302	367.966	5.284	0.000	0.000	0.000	0.000	0.010	302.524	0.000	0.000	2118.081
A14B	0.000	172.168	897.792	0.000	0.000	0.000	0.000	0.000	0.000	388.404	0.000	0.000	2361.093
A15B	1.298	59.736	3418.405	0.000	0.000	0.000	0.000	0.000	0.000	263.641	0.000	0.000	2166.520
A16B	14.387	49.196	0.000	0.000	0.000	10.817	13.036	0.000	203.189	239.046	0.000	0.000	1929.295
A17B	0.000	55.487	464.790	0.000	0.000	0.000	0.000	0.000	36.200	221.386	0.000	0.000	2082.937
A18B	0.000	41.640	6040.405	0.000	0.000	0.000	0.000	0.000	0.000	329.702	0.000	0.000	2252.131
A19B	0.000	66.042	2320.072	0.000	0.000	0.000	0.000	0.000	18.074	221.515	0.000	0.000	2162.514
A20B	0.000	110.541	656.699	0.000	0.000	0.000	0.000	111.705	0.000	761.574	0.000	0.000	2254.674
B1B	86.468	159.012	4350.324	68.637	0.000	59.431	0.000	21.096	168.829	444.361	0.000	0.000	3313.965
B2B	39.644	124.006	535.592	108.940	0.000	34.975	0.000	6.587	111.415	459.221	0.000	0.000	3987.054
B3B	21.418	123.920	2610.983	56.989	0.000	0.000	0.000	0.000	41.452	402.030	0.000	0.000	2744.889
B4B	11.629	189.722	2850.083	10.386	0.000	16.011	0.000	205.962	17.393	1415.442	0.000	0.000	2612.963
B5B	1.849	36.768	179.117	18.616	0.000	25.162	0.000	1.937	10.113	259.796	0.000	0.000	3074.132
B6B	10.167	66.448	2407.207	13.543	0.000	0.000	0.000	0.000	15.123	254.025	0.000	0.000	2516.562
B7B	1.994	79.630	1073.795	32.550	0.000	0.000	0.000	0.000	10.450	355.039	0.000	0.000	3417.317
B8B	0.000	121.606	3151.875	32.438	0.000	0.000	0.000	0.000	2.626	498.659	0.000	0.000	3626.222
B9B	0.000	110.452	2253.479	4.111	0.000	28.319	0.000	0.000	0.000	344.587	0.000	0.000	2782.545
B10B	0.000	105.639	605.173	0.000	0.000	0.000	0.000	0.000	0.000	370.941	0.000	0.000	2871.748
B11B	3.983	26.080	396.679	0.000	0.000	0.000	0.000	0.000	107.960	311.370	0.000	0.000	2782.006
B12B	0.691	38.998	1504.441	0.000	0.000	0.000	0.000	0.000	26.121	274.351	0.000	0.000	3066.326
B13B	0.000	84.548	1064.391	0.000	0.000	0.000	0.000	73.188	0.000	689.374	0.000	0.000	2960.426

TABLE A-6. Continued.

SAMPLE	Ag	Al	As	Ba	Be	Cd	Co	Cr	Cu	Fe	Ga	Hg	K
B14B	0.000	63.696	957.873	0.000	0.000	0.000	0.000	0.000	0.000	354.134	0.000	0.000	2373.563
B15B	0.000	34.783	5450.344	0.000	0.000	0.000	0.000	59.377	0.000	1066.160	0.000	0.000	2712.418
B16B	5.301	106.165	3921.427	0.000	0.000	0.000	11.276	15.377	104.815	443.722	0.000	0.000	2358.307
B17B	0.000	38.050	3334.971	0.000	0.000	0.000	0.000	0.000	24.258	230.864	0.000	0.000	2288.965
B18B	0.000	68.383	931.112	0.000	0.000	0.000	0.000	0.000	11.285	307.271	0.000	0.000	2422.257
B19B	0.000	46.294	1088.395	0.000	0.000	13.709	0.000	0.000	4.821	298.327	0.000	0.000	2555.045
B20B	0.000	29.876	3077.213	0.000	0.000	0.000	0.000	0.000	0.000	206.912	0.000	0.000	2275.717
C1B	145.319	165.457	52.807	103.140	0.000	96.455	0.000	17.981	177.991	373.870	0.000	0.000	4285.349
C2B	36.328	177.655	124.407	77.961	0.000	66.235	0.000	13.377	122.869	342.490	0.000	0.000	3399.951
C3B	22.295	143.571	4487.040	40.684	0.000	36.112	0.000	10.001	90.737	330.888	31.320	0.000	3735.706
C4B	1.951	166.761	1523.696	27.634	0.000	0.000	0.000	0.000	33.156	363.060	0.000	0.000	2525.501
C5B	0.000	150.203	0.000	24.277	0.000	61.981	0.000	0.000	4.014	356.731	0.000	0.000	2263.996
C6B	12.263	134.434	1513.052	31.981	0.000	5.504	0.000	4.388	18.088	279.766	0.000	0.000	2805.624
C7B	0.262	132.128	2038.073	32.140	0.000	0.000	0.000	0.000	19.043	346.110	0.000	0.000	3853.084
C8B	0.000	272.407	2577.977	9.072	0.000	0.000	0.000	0.000	0.000	405.650	0.000	0.000	3271.775
C9B	0.000	185.968	3688.792	0.000	0.000	44.586	0.000	0.000	0.000	425.774	0.000	0.000	3260.240
C10B	0.000	108.920	4376.960	0.000	0.000	0.000	0.000	0.000	0.000	420.218	0.000	0.000	2413.432
C11B	3.192	225.151	3427.524	18.310	0.000	0.000	0.000	0.000	77.110	445.181	0.000	0.000	2790.189
C12B	0.678	123.048	1796.280	0.000	0.000	0.000	0.000	0.000	17.846	342.671	0.000	0.000	3049.566
C13B	0.000	198.005	1295.693	0.000	0.000	0.000	0.000	0.000	16.533	417.001	0.000	0.000	2699.557
C14B	0.000	81.237	2875.298	0.000	0.000	0.000	0.000	0.000	0.209	275.293	0.000	0.000	2316.742
C15B	0.000	57.850	3083.029	0.000	0.000	18.527	0.000	0.000	0.000	327.591	0.000	0.000	3094.313
C16B	0.000	33.240	2079.026	0.000	0.000	12.765	0.000	0.000	57.566	164.410	0.000	0.000	2557.446
C17B	0.000	130.921	3346.640	0.000	0.000	0.000	0.000	0.000	11.970	315.437	0.000	0.000	2165.776
C18B	0.000	84.264	3354.278	0.000	0.000	2.989	5.509	0.000	17.862	270.614	0.000	0.000	2091.916
C19B	0.000	409.035	2062.968	0.000	0.000	0.000	0.000	0.000	0.000	590.905	0.000	0.000	2323.058
C20B	0.000	148.752	2006.433	0.000	0.000	0.000	0.000	0.000	0.000	385.570	0.000	0.000	2620.394
D1B	66.731	510.177	3313.575	93.167	0.000	23.329	0.000	18.851	325.710	276.350	0.000	0.000	3198.329
D2B	20.195	74.087	3959.262	55.286	0.000	36.423	0.000	4.797	145.196	244.766	0.000	0.000	3421.870
D3B	11.842	157.069	3081.771	29.814	0.000	67.565	0.000	4.303	123.950	281.949	0.000	0.000	2798.927
D4B	5.114	89.093	1637.912	51.302	0.000	0.000	0.000	0.000	100.892	257.905	0.000	0.000	2434.632
D5B	0.000	539.583	4448.954	61.793	0.000	89.300	0.000	0.000	124.871	384.830	0.000	0.000	2510.600
D6B	8.452	42.917	1846.967	101.430	0.000	24.424	0.000	5.283	230.673	273.058	0.000	0.000	2872.256
D7B	0.937	73.565	1134.419	46.588	0.000	0.000	0.000	0.000	93.273	225.696	0.000	0.000	2690.713

TABLE A-6. Continued.

SAMPLE	Ag	Al	As	Ba	Be	Cd	Co	Cr	Cu	Fe	Ga	Hg	K
D8B	0.000	183.276	2831.792	32.974	0.000	0.000	0.000	0.000	38.580	204.323	0.000	0.000	3262.514
D9B	0.000	77.327	2414.066	0.000	0.000	26.749	0.000	0.000	30.783	255.392	0.000	0.000	2501.919
D10B	0.000	74.763	763.557	13.130	0.000	21.176	0.000	0.000	72.209	225.479	0.000	0.000	2729.631
D11B	6.200	72.340	4003.025	37.296	0.000	0.000	0.000	0.000	242.384	270.035	0.000	0.000	2929.080
D12B	0.000	36.181	4263.948	0.000	0.000	21.405	0.000	0.000	164.322	230.985	0.000	0.000	2989.744
D13B	0.000	18.980	2338.872	15.873	0.000	5.574	0.000	0.000	94.724	175.880	0.000	0.000	3263.589
D14B	0.000	32.561	1452.538	0.000	0.000	20.281	4.107	0.000	79.172	199.210	0.000	0.000	2544.234
D15B	0.000	305.315	4818.716	2.914	0.000	0.000	7.793	0.000	113.692	542.125	0.000	0.000	1729.978
D16B	0.000	39.061	0.000	0.000	0.000	0.000	0.000	0.000	120.055	195.728	0.000	0.000	2125.248
D17B	0.000	2.954	1286.186	0.000	0.000	0.000	16.263	0.000	73.325	106.782	0.000	0.000	1975.371
D18B	0.000	90.590	5379.287	30.099	0.000	0.000	0.000	0.000	150.224	377.639	0.000	0.000	2345.385
D19B	0.000	36.194	4212.924	10.130	0.000	4.753	0.000	0.000	74.050	174.012	0.000	0.000	2314.205
D20B	0.000	19.998	4522.022	0.000	0.000	0.000	0.000	37.352	36.487	417.168	0.000	0.000	1879.422
E1B	24.959	34.343	1572.824	10.752	0.000	17.589	0.000	8.097	70.429	108.902	0.000	0.000	2639.638
E2B	19.914	122.400	2172.241	51.851	0.000	29.361	0.000	39.535	62.151	281.505	0.000	0.000	2854.269
E3B	12.610	104.452	3249.845	15.286	0.000	0.000	0.000	17.351	44.261	248.915	0.000	0.000	3002.221
E4B	4.205	43.407	1231.554	0.000	0.000	42.617	0.000	0.272	15.511	89.342	0.000	0.000	2269.559
E5B	0.391	73.358	3516.773	0.000	0.000	28.159	0.000	0.000	2.076	162.114	0.000	0.000	1904.350
E6B	6.155	60.521	627.897	27.980	0.000	0.000	0.000	5.585	7.298	147.906	0.000	0.000	2777.080
E7B	0.000	72.143	4729.236	6.673	0.000	0.000	0.000	0.000	0.000	145.181	0.000	0.000	2856.477
E8B	1.000	54.438	273.238	0.000	0.000	0.000	0.000	0.000	0.000	174.503	0.000	0.000	2635.942
E9B	0.000	41.796	2543.348	0.000	0.000	0.000	0.000	0.000	0.000	200.358	0.000	0.000	2920.750
E10B	0.000	41.660	640.300	0.000	0.000	0.000	0.000	0.000	0.000	134.656	0.000	0.000	2200.906
E11B	3.083	19.332	2712.680	0.000	0.000	36.407	0.000	0.000	40.989	98.953	0.000	0.000	2461.409
E12B	0.000	20.892	4167.664	0.000	0.000	2.681	0.000	0.000	14.367	130.406	0.000	0.000	2473.540
E13B	0.000	8.326	748.486	0.000	0.000	0.000	0.000	0.000	12.779	109.281	0.000	0.000	2437.406
E14B	0.000	42.576	9525.009	0.000	0.000	0.000	39.576	0.000	0.000	101.329	0.000	0.000	2489.362
E15B	0.000	15.298	7228.763	0.000	0.000	0.000	0.000	0.000	0.000	93.008	0.000	0.000	2165.938
E16B	0.000	3.515	1198.802	0.000	0.000	0.000	0.000	0.000	47.481	86.403	0.000	0.000	2113.175
E17B	0.000	1.577	2293.216	0.000	0.000	6.868	0.000	0.000	23.128	87.041	0.000	0.000	2090.540
E18B	0.000	50.550	17.542	0.000	0.000	0.000	0.000	0.000	5.083	170.597	0.000	0.000	2792.904
E19B	0.000	13.052	295.373	0.000	0.000	0.000	0.000	1.852	8.125	152.741	0.000	0.000	1825.357
E20B	0.000	4.748	1786.363	0.000	0.000	0.000	0.000	0.000	3.500	117.680	0.000	0.000	2601.569

TABLE A-6. Continued.

SAMPLE	Li	Mg	Mn	Na	Ni	Pb	Rb	Se	Sr	U	V	Zn
A1B	175.875	2069.674	76.788	26576.577	0.000	228.735	238.271	5927.522	200.685	15.771	28.133	0.000
A2B	88.461	1406.153	73.171	11136.160	0.000	1087.511	127.195	587.181	138.646	1.647	0.000	0.000
A3B	87.558	1659.102	103.911	9754.382	0.000	0.000	136.049	0.000	192.935	18.115	0.000	0.000
A4B	92.575	1652.323	60.763	10576.469	24.641	0.000	134.041	0.000	175.598	0.000	0.000	0.000
A5B	131.259	1757.328	99.902	9066.052	0.000	0.000	139.451	6140.827	128.456	12.428	0.000	0.000
A6B	96.255	1592.784	87.587	7029.345	0.000	501.200	139.687	3314.205	190.451	0.000	0.000	0.000
A7B	44.167	1730.974	66.685	5952.931	12.753	0.000	70.622	2978.912	82.367	3.616	0.000	0.000
A8B	59.274	1949.553	130.246	6400.168	35.660	0.000	85.093	4867.837	103.008	4.064	0.000	0.000
A9B	85.702	1587.451	120.374	5947.135	0.000	2556.089	91.119	12498.683	106.295	0.000	0.000	0.000
A10B	52.019	1605.567	84.621	6167.085	22.079	472.583	55.344	61.258	53.128	8.701	0.000	0.000
A11B	62.880	1442.400	55.116	1624.606	0.000	208.351	28.223	735.138	22.367	14.978	0.000	0.000
A12B	36.706	1210.121	62.638	3057.114	0.000	299.143	23.050	4986.516	13.170	0.000	0.000	0.000
A13B	46.150	1386.322	131.017	3531.609	0.000	0.000	36.599	0.000	16.761	0.000	0.000	0.000
A14B	119.419	1292.825	171.993	2362.766	108.436	0.000	68.046	1492.529	25.814	0.000	0.000	5822.606
A15B	68.602	1256.082	44.316	2781.877	0.000	712.736	41.055	3165.232	26.034	0.000	0.000	6349.895
A16B	61.449	1194.869	35.960	2100.316	17.058	0.000	35.958	0.000	13.886	3.700	0.000	0.000
A17B	39.459	1295.223	44.758	2096.852	0.000	0.000	19.393	0.000	0.829	0.000	0.000	0.000
A18B	70.355	1556.602	38.742	2372.940	1.003	245.482	53.363	0.000	18.349	7.848	0.000	0.000
A19B	30.377	1212.017	56.056	2029.440	36.543	0.000	20.292	0.000	15.926	0.000	0.000	2491.033
A20B	42.712	1324.494	103.314	1459.409	41.850	230.767	26.270	0.000	13.998	0.000	0.000	1383.320
B1B	164.326	1565.268	63.291	18129.033	26.626	0.000	209.148	0.000	170.395	0.000	0.000	0.000
B2B	139.267	1977.111	82.445	17848.492	0.000	0.000	209.915	5013.079	215.100	4.862	0.000	0.000
B3B	89.800	1585.402	53.856	8501.790	53.134	0.000	134.605	4077.204	193.222	3.496	0.000	0.000
B4B	118.827	1537.334	78.454	7938.726	10.717	1094.444	173.823	0.000	138.434	0.000	0.000	0.000
B5B	45.239	1410.481	16.076	6428.917	48.336	1959.172	64.250	919.470	59.271	0.000	0.000	0.000
B6B	48.377	1317.326	38.724	5895.097	5.352	707.159	67.479	0.000	99.793	1.053	0.000	0.000
B7B	56.802	1852.459	36.531	8907.917	0.000	1076.222	66.114	0.000	84.508	0.000	0.000	0.000
B8B	56.714	1932.563	74.637	7651.779	0.000	0.000	68.007	2578.883	82.830	0.000	0.000	3559.464
B9B	74.103	1517.706	38.253	6550.993	0.000	0.000	89.549	1961.237	83.508	0.000	0.000	441.614
B10B	61.478	1610.867	60.361	8518.786	54.228	0.000	56.078	721.161	45.317	0.000	0.000	0.000
B11B	52.980	1538.518	32.749	4188.794	25.681	751.056	35.362	4170.162	21.761	0.000	0.000	0.000
B12B	55.627	1580.660	22.373	4124.862	0.000	0.000	58.426	0.000	17.213	6.670	0.000	2473.475
B13B	72.383	1624.967	47.537	4023.152	0.000	0.000	74.553	7892.635	21.615	0.000	0.000	0.000
B14B	57.345	1369.967	45.610	2327.329	0.000	0.000	34.635	4349.515	16.623	0.000	0.000	0.000

TABLE A-6. Continued.

SAMPLE	Li	Mg	Mn	Na	Ni	Pb	Rb	Se	Sr	U	V	Zn
B15B	73.286	1256.355	35.840	1328.981	34.910	0.000	69.729	2403.099	26.231	0.000	0.000	0.000
B16B	58.304	1240.813	80.257	3371.540	46.765	0.000	36.883	0.000	12.976	25.098	0.000	0.000
B17B	34.236	1231.180	37.445	3384.827	0.000	0.000	26.146	0.000	5.265	0.000	0.000	0.000
B18B	44.696	1174.725	62.461	2322.499	0.000	0.000	40.379	2272.771	4.591	0.000	0.000	81.634
B19B	44.936	1258.467	35.269	2697.960	1.152	798.110	33.126	2782.171	7.921	0.000	0.000	0.000
B20B	49.017	1096.214	17.122	1814.890	0.000	181.920	32.254	4844.087	8.585	5.587	0.000	0.000
C1B	200.848	1796.150	63.320	31631.337	56.363	0.000	307.810	2055.179	360.456	11.991	0.000	0.000
C2B	112.479	1746.476	72.631	14780.904	112.408	279.333	179.159	0.000	264.816	0.000	0.000	1594.804
C3B	103.003	2088.724	44.019	10616.071	0.000	0.000	195.208	0.000	188.276	0.000	0.000	0.000
C4B	65.641	1395.344	52.978	6069.777	18.298	76.610	104.954	0.000	105.534	0.000	0.000	0.000
C5B	56.321	1430.665	34.019	5209.777	95.664	0.000	69.246	0.000	68.672	0.000	0.000	3827.803
C6B	44.307	1464.720	65.735	5985.208	0.000	466.759	83.491	0.000	137.380	6.300	0.000	0.000
C7B	46.671	2030.494	50.313	6286.331	25.783	957.704	54.040	0.000	61.668	5.329	0.000	195.476
C8B	59.122	1789.686	53.337	7327.770	0.000	0.000	70.109	5340.530	75.203	0.000	0.000	0.000
C9B	58.663	1830.646	51.558	5585.623	0.947	0.000	53.505	0.000	60.608	2.754	0.000	0.000
C10B	56.523	1490.994	12.613	4362.391	0.000	909.390	55.724	0.000	68.715	10.331	0.000	0.000
C11B	48.289	1792.743	90.294	3852.971	0.000	422.342	31.015	2323.131	21.077	0.000	0.000	0.000
C12B	66.099	1732.066	36.396	3573.133	0.000	113.282	43.300	0.000	32.208	0.000	0.000	1633.387
C13B	43.305	1633.020	71.649	2975.291	49.096	124.723	40.119	0.000	15.312	0.000	0.000	0.000
C14B	44.136	1388.012	25.066	3413.161	6.605	206.501	23.210	585.337	11.943	0.000	0.000	0.000
C15B	48.546	1924.642	14.200	4287.052	0.000	0.000	27.758	0.000	9.299	0.000	0.000	0.000
C16B	29.158	790.705	8.059	1352.931	0.000	227.885	19.414	0.000	4.608	0.000	0.000	0.000
C17B	51.440	1163.597	30.275	794.328	0.000	0.000	33.024	0.000	6.333	0.000	0.000	1939.722
C18B	40.163	1252.511	22.402	1251.206	5.681	0.000	20.207	3958.183	11.693	0.000	0.000	0.000
C19B	61.960	1258.229	89.619	402.429	16.716	345.636	36.106	3480.835	14.656	0.000	0.000	648.088
C20B	55.572	1570.940	35.544	2542.564	0.000	0.000	30.748	291.943	1.538	0.000	0.000	0.000
D1B	173.053	1690.009	51.956	20616.086	52.354	0.000	175.574	0.000	234.535	7.224	0.000	2110.194
D2B	74.029	1778.432	54.773	10266.251	58.492	0.000	123.533	479.568	164.451	0.000	0.000	144.523
D3B	117.639	1507.282	70.597	9845.667	0.000	0.000	171.179	3741.734	207.698	0.000	0.000	0.000
D4B	68.245	1432.707	49.656	6419.295	0.000	219.562	77.931	0.000	130.017	3.790	0.000	0.000
D5B	83.504	1468.252	101.385	7353.088	7.484	0.000	91.202	5694.412	87.078	0.000	0.000	4653.320
D6B	48.362	1638.944	50.742	10891.470	0.000	0.000	70.038	4277.644	134.468	0.000	0.000	0.000
D7B	37.559	1429.890	33.009	4783.736	0.000	0.000	48.934	0.000	56.625	0.000	0.000	0.000
D8B	59.683	1490.523	18.452	6063.718	14.961	0.000	61.222	345.587	70.061	0.000	0.000	0.000

TABLE A-6. Continued.

SAMPLE	Li	Mg	Mn	Na	Ni	Pb	Rb	Se	Sr	U	V	Zn
D9B	62.413	1301.026	13.960	4578.489	6.981	1139.051	51.638	1830.624	54.889	17.364	0.000	0.000
D10B	41.668	1485.643	29.368	4158.910	23.831	0.000	53.517	952.545	57.912	0.000	0.000	0.000
D11B	77.127	1837.148	26.303	4937.646	0.000	0.000	61.071	0.000	37.552	0.000	0.000	1227.827
D12B	50.681	1302.234	19.086	3791.121	0.000	0.000	42.123	3920.413	23.971	0.000	0.000	848.141
D13B	67.002	1717.981	0.048	3280.855	0.000	0.000	37.811	3644.795	19.897	0.113	0.000	0.000
D14B	51.310	1376.682	16.471	1935.140	0.000	0.000	37.191	2261.887	12.225	0.000	0.000	0.000
D15B	56.372	1177.275	149.564	1811.821	0.000	71.525	35.898	1905.475	20.382	0.000	0.000	0.000
D16B	53.380	1254.205	29.404	1324.173	56.471	0.000	28.073	942.623	13.181	0.000	0.000	464.216
D17B	42.201	1028.832	3.843	1083.213	0.000	80.526	27.712	2910.725	8.841	0.000	0.000	1923.302
D18B	57.222	1506.698	54.135	3801.780	27.449	0.000	35.014	0.000	15.481	0.000	0.000	0.000
D19B	48.949	1398.172	6.658	1319.401	63.367	520.614	16.902	0.000	18.024	0.000	0.000	241.783
D20B	47.354	1039.595	8.678	431.240	0.000	0.000	36.638	5230.474	3.725	0.000	0.000	1004.849
E1B	68.218	950.804	22.287	7624.348	0.000	498.934	90.414	767.132	73.837	2.630	0.000	0.000
E2B	82.814	1146.829	30.662	9405.527	67.710	0.000	133.372	3433.519	183.221	0.000	0.000	0.000
E3B	61.699	1403.003	46.173	11408.905	0.000	0.000	119.592	0.000	119.354	2.171	0.000	0.000
E4B	37.594	978.054	2.512	3108.947	25.037	0.000	43.477	114.498	46.930	0.000	0.000	0.000
E5B	57.494	988.049	30.042	3944.906	0.000	963.505	75.206	0.000	86.149	8.075	0.000	0.000
E6B	46.580	1290.833	23.437	5525.267	105.624	0.000	65.229	52.548	85.285	4.402	0.000	0.000
E7B	65.286	1134.133	22.834	7688.228	10.803	0.000	91.391	5488.874	133.858	36.254	0.000	0.000
E8B	54.049	1137.296	0.000	2831.031	49.197	0.000	58.626	1074.170	63.979	0.000	0.000	0.000
E9B	40.453	1154.779	7.983	2324.773	0.000	0.000	33.036	467.061	32.870	0.000	0.000	0.000
E10B	46.469	1045.081	7.564	3308.131	12.194	0.000	53.808	1381.936	54.485	0.000	0.000	0.000
E11B	37.838	1055.111	10.155	2226.122	0.000	410.726	22.149	2528.338	8.333	2.801	0.000	0.000
E12B	37.126	907.243	10.337	1183.632	0.000	351.886	35.270	0.000	14.818	0.000	0.000	0.000
E13B	30.570	944.716	6.503	1329.522	17.698	0.000	20.724	546.160	1.089	3.401	0.000	0.000
E14B	130.078	1334.632	0.000	225.321	0.000	0.000	80.181	0.000	27.843	13.257	0.000	0.000
E15B	78.577	926.305	0.000	123.192	0.000	1424.111	73.101	2058.261	4.354	0.000	0.000	0.000
E16B	25.766	852.266	5.429	1919.106	0.000	0.000	20.768	1212.746	2.407	0.000	0.000	469.402
E17B	32.129	1024.967	8.663	1098.576	0.000	0.000	16.980	4553.615	3.430	0.000	0.000	0.000
E18B	41.514	1288.785	14.197	4477.442	0.000	202.805	45.703	1136.236	20.727	14.467	0.000	3614.665
E19B	28.929	854.197	0.929	1708.909	3.467	0.000	12.930	1142.498	6.852	0.000	0.000	0.000
E20B	30.785	1037.196	8.177	1624.212	7.362	605.998	22.858	3468.074	4.459	0.000	0.000	0.000

TABLE A-7. Micrograms of element per gram ($\mu g/g$) of dry weight of shell. Values of zero indicate concentrations in the samples below the detection limits of the ICP-OES.

SAMPLE	Ag	Al	As	Ba	Be	Cd	Co	Cr	Cu	Fe	Ga	Hg	K
A6S	0.000	3.391	254.215	18.014	0.000	8.461	0.000	0.000	2.579	3.388	0.000	0.000	205.222
A7S	0.000	1.904	407.449	21.267	0.000	8.582	0.000	0.000	3.245	14.285	0.000	0.000	156.261
A8S	0.000	2.406	415.504	17.314	0.000	8.357	0.000	0.000	2.920	5.034	0.000	0.000	171.182
A9S	0.000	4.646	384.518	17.870	0.000	8.505	0.000	0.000	1.558	6.628	0.000	0.000	229.719
A10S	0.000	3.299	415.418	17.148	0.000	8.746	0.000	0.000	2.960	5.233	0.000	0.000	229.088
A11S	0.000	4.651	984.068	24.361	0.000	14.279	0.000	0.000	3.319	11.640	0.000	0.000	141.835
A12S	0.000	3.633	1029.360	25.339	0.000	14.694	0.000	0.000	3.845	9.409	0.000	0.000	121.752
A13S	0.000	5.879	1085.985	22.203	0.000	15.222	0.000	0.179	4.020	46.422	0.000	0.000	181.547
A14S	0.000	4.678	1148.767	21.291	0.000	16.627	0.000	0.000	3.596	7.552	0.000	0.000	198.308
A15S	0.000	2.083	1023.342	18.200	0.000	15.393	0.000	0.000	3.990	2.415	0.000	0.000	171.749
A16S	0.000	0.554	988.496	17.618	0.000	14.264	0.000	0.000	3.449	3.393	0.000	0.000	145.324
A17S	0.000	1.117	1067.318	20.865	0.000	14.926	0.000	0.000	3.288	7.813	0.000	0.000	126.002
A18S	0.000	1.661	1030.629	17.224	0.000	14.655	0.000	0.000	3.822	5.560	0.000	0.000	161.694
A19S	0.000	1.321	961.841	21.838	0.000	12.582	0.000	0.103	2.308	8.256	0.000	0.000	99.332
A20S	0.000	0.192	320.776	6.677	0.000	3.898	0.138	0.000	0.575	2.694	0.000	0.000	26.157
B6S	0.000	9.933	400.325	80.791	0.000	8.620	0.000	0.000	4.111	48.225	0.000	0.000	148.124
B7S	0.000	9.085	413.891	77.039	0.000	8.795	0.000	0.000	3.234	44.099	0.000	0.000	157.872
B8S	0.000	2.477	421.920	71.899	0.000	8.862	0.000	0.000	1.537	21.128	0.000	0.000	172.846
B9S	0.000	10.230	413.940	86.615	0.000	8.276	0.000	0.000	1.581	61.766	0.000	0.000	190.410
B10S	0.000	3.806	419.501	70.927	0.000	8.630	0.000	0.000	1.077	26.674	0.000	0.000	197.019
B11S	0.000	8.749	978.165	68.876	0.000	14.390	0.000	0.000	3.923	42.598	0.000	0.000	116.134
B12S	0.000	9.913	1053.570	69.403	0.000	14.822	0.448	0.000	3.828	69.101	0.000	0.000	126.417
B13S	0.000	3.507	1084.159	68.194	0.000	15.442	0.041	0.000	3.115	18.407	0.000	0.000	169.998
B14S	0.000	6.626	1104.448	71.958	0.000	15.766	0.000	0.000	4.031	43.010	0.000	0.000	149.608
B15S	0.000	14.875	1156.883	66.357	0.000	16.068	0.020	0.000	3.504	96.880	0.000	0.000	137.651
B16S	0.000	9.784	1001.699	61.401	0.000	14.513	0.247	0.000	2.610	53.786	0.000	0.000	132.653
B17S	0.000	9.830	996.484	64.240	0.000	14.193	0.018	0.000	3.225	42.238	0.000	0.000	89.083
B18S	0.000	7.859	986.970	64.750	0.000	13.311	0.279	0.000	3.308	58.641	0.000	0.000	98.761
B19S	0.000	4.719	830.905	64.250	0.000	11.281	0.173	0.083	2.405	38.616	0.000	0.000	71.717
B20S	0.000	1.520	256.352	15.982	0.000	4.287	0.490	0.000	0.551	10.274	0.000	0.000	19.583

TABLE A-7. Continued.

SAMPLE	Ag	Al	As	Ba	Be	Cd	Co	Cr	Cu	Fe	Ga	Hg	K
C6S	0.000	3.970	413.169	29.615	0.000	8.597	0.000	0.000	3.853	20.136	0.000	0.000	211.637
C7S	0.000	5.361	418.141	34.693	0.000	8.727	0.000	0.000	3.916	15.201	0.000	0.000	265.230
C8S	0.000	8.871	417.552	24.177	0.000	8.248	0.000	0.000	3.183	24.376	0.000	0.000	244.951
C9S	0.000	3.303	355.496	20.140	0.000	7.521	0.000	0.000	2.135	13.984	0.000	0.000	161.106
C10S	0.000	3.650	388.442	24.250	0.000	7.679	0.000	0.000	0.930	5.210	0.000	0.000	259.739
C11S	0.000	2.909	930.854	22.722	0.000	13.817	0.000	0.000	3.408	12.047	0.000	0.000	130.653
C12S	0.000	4.375	1090.776	24.281	0.000	15.620	0.000	0.000	4.115	15.815	0.000	0.000	171.449
C13S	0.000	2.874	1015.151	23.538	0.000	14.980	0.000	0.000	3.905	10.243	0.000	0.000	174.703
C14S	0.000	2.367	1041.796	27.670	0.000	15.330	0.000	0.000	4.176	13.152	0.000	0.000	230.714
C15S	0.000	2.513	1061.771	23.138	0.000	15.041	0.000	0.000	3.314	7.723	0.000	0.000	257.270
C16S	0.000	15.922	1018.472	44.690	0.000	14.128	0.442	0.000	3.239	155.719	0.000	0.000	155.042
C17S	0.000	7.934	1009.444	23.915	0.000	15.261	0.028	0.000	4.060	27.711	0.000	0.000	157.985
C18S	0.000	4.299	995.623	25.492	0.000	13.791	0.222	0.000	3.371	23.291	0.000	0.000	145.171
C19S	0.000	0.994	708.746	16.355	0.000	9.697	0.410	0.000	2.172	15.209	0.000	0.000	110.695
C20S	0.000	0.000	200.516	3.240	0.000	1.166	0.288	0.000	0.000	0.334	0.000	0.000	11.936
D6S	0.000	31.353	407.957	77.078	0.000	8.775	0.000	0.000	4.051	59.392	0.000	0.000	108.307
D7S	0.000	11.364	429.582	76.264	0.000	9.251	0.000	0.000	3.125	47.306	0.000	0.000	147.424
D8S	0.000	5.184	420.042	62.034	0.000	9.010	0.000	0.000	2.959	18.922	0.000	0.000	167.133
D9S	0.000	6.135	395.022	59.823	0.000	8.399	0.000	0.000	2.275	28.823	0.000	0.000	160.914
D10S	0.000	7.908	408.506	59.558	0.000	8.310	0.000	0.000	2.673	32.510	0.000	0.000	209.030
D11S	0.000	7.291	1032.766	55.806	0.000	15.418	0.000	0.000	3.448	31.656	0.000	0.000	96.188
D12S	0.000	16.165	992.680	78.842	0.000	14.087	0.000	0.000	3.721	44.615	0.000	0.000	95.279
D13S	0.000	11.565	947.013	58.546	0.000	13.331	0.000	0.000	2.805	54.761	0.000	0.000	97.546
D14S	0.000	9.140	1004.825	51.338	0.000	14.273	0.023	0.000	3.385	21.864	0.000	0.000	161.727
D15S	0.000	5.760	1106.382	58.316	0.000	15.629	0.000	0.000	3.818	18.921	0.000	0.000	100.581
D16S	0.000	5.171	1082.445	56.735	0.000	15.657	0.000	0.000	3.359	34.075	0.000	0.000	121.066
D17S	0.000	0.832	928.391	53.153	0.000	12.672	0.000	0.000	3.132	7.302	0.000	0.000	93.715
D18S	0.000	6.400	972.745	47.709	0.000	13.880	0.000	0.000	2.582	38.976	0.000	0.000	78.913
D19S	0.000	4.711	701.656	41.213	0.000	9.312	0.000	0.000	2.335	23.563	0.000	0.000	60.437
D20S	0.000	0.641	150.278	11.248	0.000	0.989	0.300	0.000	0.436	2.805	0.000	0.000	22.666
E6S	0.000	10.961	399.262	43.026	0.000	8.584	0.000	0.000	3.801	31.258	0.000	0.000	134.276

TABLE A-7. Continued.

SAMPLE	Ag	Al	As	Ba	Be	Cd	Co	Cr	Cu	Fe	Ga	Hg	K
E7S	0.000	13.004	412.707	54.653	0.000	8.889	0.000	0.000	4.208	17.312	0.000	0.000	162.132
E8S	0.000	4.201	392.473	32.302	0.000	8.165	0.000	0.000	1.921	5.746	0.000	0.000	286.160
E9S	0.000	7.245	386.835	40.330	0.000	7.975	0.000	0.000	3.114	19.601	0.000	0.000	205.182
E10S	0.000	5.006	368.256	37.618	0.000	8.251	0.000	0.000	2.488	11.039	0.000	0.000	197.432
E11S	0.000	2.626	903.612	39.397	0.000	13.705	0.000	0.000	4.176	7.143	0.000	0.000	101.706
E12S	0.000	4.889	998.491	38.210	0.000	14.346	0.000	0.062	4.438	17.326	0.000	0.000	127.495
E13S	0.000	3.741	1025.508	58.407	0.000	14.471	0.000	0.000	4.279	11.982	0.000	0.000	111.795
E14S	0.000	8.003	1013.555	31.478	0.000	16.472	0.000	0.000	5.762	5.068	0.000	0.000	222.069
E15S	0.000	10.223	1126.219	36.711	0.000	16.243	0.000	0.000	5.194	7.025	0.000	0.000	244.242
E16S	0.000	7.929	974.535	55.913	0.000	12.706	0.008	0.000	4.004	17.283	0.000	0.000	96.008
E17S	0.000	3.042	958.919	41.697	0.000	12.949	0.000	0.000	3.720	9.171	0.000	0.000	98.814
E18S	0.000	13.782	945.427	44.811	0.000	13.035	0.000	0.000	4.678	17.026	0.000	0.000	119.380
E19S	0.000	1.392	494.855	22.814	0.000	5.755	0.000	0.000	1.067	3.996	0.000	0.000	16.068
E20S	0.000	0.180	46.648	5.404	0.000	0.368	0.012	0.000	0.066	1.002	0.000	0.000	3.622

TABLE A-7. Continued.

SAMPLE	Li	Mg	Mn	Na	Ni	Pb	Rb	Se	Sr	U	V	Zn
A6S	0.000	32.837	15.132	SATURATED	0.398	3.560	0.000	13.507	177.807	0.000	0.000	0.000
A7S	0.000	34.959	19.818	SATURATED	0.680	4.923	0.000	3.345	167.525	0.000	0.000	0.000
A8S	0.000	33.417	24.496	SATURATED	0.277	5.340	0.000	17.183	163.747	0.000	0.000	0.000
A9S	0.000	37.030	21.770	SATURATED	0.015	1.796	0.831	2.860	186.892	0.000	0.000	0.000
A10S	0.000	35.160	20.448	SATURATED	0.161	4.618	0.000	6.173	166.727	0.000	0.000	0.000
A11S	0.536	27.486	38.048	SATURATED	0.172	9.941	0.000	7.926	SATURATED	0.670	0.000	2.430
A12S	0.523	31.216	23.382	SATURATED	0.212	7.773	0.000	15.959	SATURATED	1.520	0.000	1.748
A13S	0.682	35.593	33.572	SATURATED	0.245	3.919	0.000	0.000	SATURATED	0.000	0.000	4.527
A14S	0.975	27.049	14.019	SATURATED	0.000	11.814	0.000	20.961	156.464	0.330	0.000	0.000
A15S	0.757	25.490	8.861	SATURATED	0.000	5.636	0.000	0.000	SATURATED	0.432	0.000	0.000
A16S	0.626	23.894	8.175	SATURATED	0.000	5.417	0.000	14.461	SATURATED	0.984	0.000	2.106
A17S	0.631	25.929	15.000	SATURATED	0.000	9.827	0.000	8.476	SATURATED	0.000	0.000	0.000
A18S	0.728	24.251	7.980	SATURATED	0.000	4.164	0.000	15.386	134.732	0.933	0.000	0.000
A19S	0.594	25.263	19.683	SATURATED	0.034	8.371	0.000	0.000	SATURATED	0.433	0.000	2.505
A20S	0.362	4.259	4.774	1468.183	0.000	1.588	0.000	2.157	20.286	0.000	0.000	1.457
B6S	0.000	33.560	49.281	SATURATED	0.668	14.552	0.000	6.354	SATURATED	0.000	0.000	0.000
B7S	0.000	33.265	36.921	SATURATED	0.562	12.514	0.000	19.707	SATURATED	0.000	0.000	0.000
B8S	0.000	30.301	32.293	SATURATED	0.190	13.721	0.000	4.645	SATURATED	0.000	0.000	0.000
B9S	0.000	39.420	41.628	SATURATED	0.677	19.508	1.943	31.606	SATURATED	0.000	0.000	0.000
B10S	0.000	33.891	53.667	SATURATED	0.298	13.401	0.000	8.508	SATURATED	0.000	0.000	0.000
B11S	0.483	27.568	35.664	SATURATED	0.743	29.583	0.000	19.458	SATURATED	1.325	0.000	0.089
B12S	0.402	27.255	65.553	SATURATED	0.405	30.348	0.000	12.485	SATURATED	0.000	0.000	0.000
B13S	0.508	29.709	26.820	SATURATED	0.395	25.509	0.000	7.323	SATURATED	0.078	0.000	3.235
B14S	0.504	27.354	38.162	SATURATED	0.649	34.478	0.000	33.620	SATURATED	0.000	0.000	0.365
B15S	0.752	28.860	77.545	SATURATED	0.355	34.066	0.000	26.586	SATURATED	0.000	0.000	0.000
B16S	0.555	26.585	39.276	SATURATED	0.202	28.629	0.000	27.240	SATURATED	0.000	0.000	0.000
B17S	0.417	26.912	38.592	SATURATED	0.454	30.957	0.000	24.047	SATURATED	0.000	0.000	0.000
B18S	0.461	24.530	39.443	SATURATED	0.528	29.260	0.000	31.689	SATURATED	0.000	0.000	3.574
B19S	0.457	16.496	24.783	SATURATED	0.171	22.142	0.000	25.405	SATURATED	0.000	0.000	1.750
B20S	0.397	3.198	12.770	1400.959	0.000	8.624	0.000	7.619	50.750	0.000	0.000	1.732
C6S	0.000	33.560	10.450	SATURATED	0.200	7.403	0.000	15.989	186.751	0.000	0.000	0.000

TABLE A-7. Continued.

SAMPLE	Li	Mg	Mn	Na	Ni	Pb	Rb	Se	Sr	U	V	Zn
C7S	0.000	35.323	8.791	SATURATED	0.243	9.083	0.000	11.108	SATURATED	0.000	0.000	0.000
C8S	0.000	33.621	9.209	SATURATED	0.147	7.787	0.000	9.729	179.731	0.000	0.000	0.000
C9S	0.000	27.691	11.545	SATURATED	0.000	1.700	0.000	14.916	162.595	0.000	0.000	0.000
C10S	0.000	31.620	6.598	SATURATED	0.334	5.342	0.943	9.991	181.740	0.000	0.000	0.000
C11S	0.551	25.511	7.851	SATURATED	0.051	10.361	0.000	5.214	SATURATED	0.000	0.000	0.000
C12S	0.625	26.219	7.617	SATURATED	0.141	6.745	0.000	0.000	SATURATED	1.711	0.000	1.961
C13S	0.580	27.947	6.417	SATURATED	0.162	6.999	0.000	14.912	SATURATED	0.396	0.000	0.000
C14S	0.648	24.844	6.793	SATURATED	0.000	11.125	0.000	0.000	SATURATED	0.468	0.000	1.730
C15S	0.720	25.847	6.070	SATURATED	0.000	10.423	0.000	0.000	SATURATED	0.490	0.000	0.000
C16S	0.494	27.548	30.210	SATURATED	0.219	19.468	0.000	25.459	SATURATED	0.000	0.000	3.604
C17S	0.673	34.192	12.918	SATURATED	0.000	10.023	0.000	11.369	SATURATED	0.000	0.000	6.569
C18S	0.524	29.080	15.022	SATURATED	0.000	11.149	0.000	9.965	SATURATED	0.000	0.000	0.000
C19S	1.691	15.124	13.227	SATURATED	0.000	5.999	0.000	5.564	113.043	0.017	0.000	3.718
C20S	0.445	0.897	2.332	1458.004	0.000	3.940	0.000	10.435	18.495	0.000	0.000	6.894
D6S	0.000	38.766	77.869	SATURATED	0.589	14.254	0.000	14.314	SATURATED	0.000	0.000	0.000
D7S	0.000	41.505	30.292	SATURATED	0.543	12.277	0.000	17.884	SATURATED	0.000	0.000	0.000
D8S	0.000	35.022	25.742	SATURATED	0.467	12.790	0.000	16.923	218.553	0.000	0.000	0.000
D9S	0.000	28.650	15.519	SATURATED	0.359	11.732	0.000	11.020	208.688	0.000	0.000	0.000
D10S	0.000	32.775	19.193	SATURATED	0.299	12.492	0.000	6.564	207.577	0.000	0.000	0.000
D11S	0.555	30.245	20.365	SATURATED	0.496	24.175	0.000	4.371	SATURATED	0.702	0.000	2.265
D12S	0.486	31.668	45.687	SATURATED	1.172	33.811	0.000	20.712	SATURATED	0.000	0.000	0.052
D13S	0.439	29.376	24.629	SATURATED	0.999	28.057	0.000	29.463	SATURATED	0.000	0.000	0.000
D14S	0.546	30.074	15.073	SATURATED	0.260	24.155	0.000	10.795	SATURATED	0.143	0.000	0.810
D15S	0.722	26.481	15.126	SATURATED	0.262	23.142	0.000	7.436	SATURATED	0.000	0.000	0.000
D16S	0.616	25.140	20.858	SATURATED	0.670	25.626	0.000	27.865	SATURATED	0.000	0.000	1.473
D17S	0.382	27.813	15.275	SATURATED	0.279	26.209	0.000	30.702	SATURATED	0.051	0.000	0.272
D18S	0.577	24.114	21.064	SATURATED	0.445	20.406	0.000	6.509	SATURATED	0.000	0.000	3.560
D19S	1.252	17.605	16.501	SATURATED	0.178	16.875	0.000	0.000	113.928	0.195	0.000	0.000
D20S	0.457	3.507	2.146	1335.309	0.000	7.075	0.000	19.129	23.275	0.000	0.000	1.135
E6S	0.000	45.015	67.048	SATURATED	0.415	9.660	0.000	9.162	SATURATED	0.000	0.000	0.000
E7S	0.000	55.426	78.439	SATURATED	0.309	7.238	0.000	10.949	206.020	0.000	0.000	0.000

TABLE A-7. Continued.

SAMPLE	Li	Mg	Mn	Na	Ni	Pb	Rb	Se	Sr	U	V	Zn
E8S	0.000	52.357	20.789	SATURATED	0.705	5.614	6.174	46.884	284.832	0.000	0.000	0.000
E9S	0.000	41.546	21.413	SATURATED	0.438	10.086	0.000	20.188	267.011	0.000	0.000	0.000
E10S	0.000	40.847	52.877	SATURATED	0.264	6.941	0.000	4.012	SATURATED	0.000	0.000	0.000
E11S	0.381	38.863	22.632	SATURATED	0.230	16.817	0.000	36.987	SATURATED	1.952	0.000	2.273
E12S	0.443	47.776	21.164	SATURATED	0.485	17.237	0.000	3.624	SATURATED	0.661	0.000	0.000
E13S	0.439	42.697	24.954	SATURATED	0.609	23.238	0.000	21.973	SATURATED	0.677	0.000	4.608
E14S	1.399	49.073	13.161	SATURATED	0.000	2.050	0.000	0.000	219.050	0.000	0.000	15.972
E15S	1.362	46.470	16.591	SATURATED	0.094	0.000	0.000	0.000	245.061	0.000	0.000	9.586
E16S	0.347	34.664	71.835	SATURATED	0.470	25.591	0.000	20.879	SATURATED	0.805	0.000	1.258
E17S	0.407	45.538	21.527	SATURATED	0.116	17.338	0.000	11.689	SATURATED	0.571	0.000	0.000
E18S	0.478	48.290	64.851	SATURATED	0.400	20.038	0.000	9.891	SATURATED	1.651	0.000	3.149
E19S	0.338	9.016	14.295	SATURATED	0.102	8.813	0.000	9.410	54.739	0.000	0.000	2.834
E20S	0.325	1.309	0.980	136.685	0.000	10.567	0.000	2.845	90.789	0.000	0.000	0.000

TABLE A-8. Pearson's correlations (r) between trace element concentrations in log-transformed soft body tissue data across Sites A, B, C & D. Bold values indicate significance at $p<0.05$.

	Ag	**Al**	**As**	**Ba**	**Cd**	**Cu**	**Fe**	**Li**	**Mg**	**Mn**	**Ni**	**Pb**	**Rb**	**Se**	**Sr**
Ag	1	**0.2836**	-0.1044	**0.6098**	**0.4807**	**0.5274**	0.1171	**0.697**	**0.3925**	**0.3254**	0.091	0.0323	**0.7205**	-0.0625	**0.6656**
Al	**0.2836**	1	0.0776	**0.348**	0.1579	-0.0885	**0.5315**	**0.4628**	**0.4402**	**0.7079**	0.1857	-0.0261	**0.4695**	-0.0742	**0.4834**
As	-0.1044	0.0776	1	-0.0693	-0.0748	-0.1118	0.1651	0.0309	0.0928	-0.0553	**-0.3013**	0.0818	0.0253	0.0486	0.005
Ba	**0.6098**	**0.348**	-0.0693	1	**0.4273**	**0.4901**	0.0537	**0.4535**	**0.5209**	0.1946	0.1496	-0.0936	**0.6603**	-0.1066	**0.7365**
Cd	**0.4807**	0.1579	-0.0748	**0.4273**	1	**0.3705**	-0.042	**0.3918**	**0.2599**	0.0029	0.1124	-0.1292	**0.4622**	0.079	**0.4543**
Cu	**0.5274**	-0.0885	-0.1118	**0.4901**	**0.3705**	1	**-0.3611**	0.1919	0.0122	-0.1329	0.0624	-0.115	0.1934	-0.1085	0.2149
Fe	0.1171	**0.5315**	0.1651	0.0537	-0.042	**-0.3611**	1	**0.4015**	**0.3218**	**0.5619**	0.1409	0.0316	**0.4071**	0.027	**0.2682**
Li	**0.697**	**0.4628**	0.0309	**0.4535**	**0.3918**	0.1919	**0.4015**	1	**0.4912**	**0.337**	0.1315	-0.1108	**0.8499**	0.1163	**0.6764**
Mg	**0.3925**	**0.4402**	0.0928	**0.5209**	**0.2599**	0.0122	**0.3218**	**0.4912**	1	**0.2825**	0.0369	-0.1158	**0.5816**	-0.037	**0.6151**
Mn	0.3254	0.7079	-0.0553	0.1946	0.0029	-0.1329	0.5619	0.337	0.2825	1	0.159	-0.0108	**0.3992**	0.01	**0.3657**
Ni	0.091	0.1857	**-0.3013**	0.1496	0.1124	0.0624	0.1409	0.1315	0.0369	0.159	1	-0.0419	0.1204	-0.1451	0.1941
Pb	0.0323	-0.0261	0.0818	-0.0936	-0.1292	-0.115	0.0316	-0.1108	-0.1158	-0.0108	-0.0419	1	-0.0946	-0.0469	0.0493
Rb	**0.7205**	**0.4695**	0.0253	**0.6603**	**0.4622**	0.1934	**0.4071**	**0.8499**	**0.5816**	**0.3992**	0.1204	-0.0946	1	0.0548	**0.886**
Se	-0.0625	-0.0742	0.0486	-0.1066	0.079	-0.1085	0.027	0.1163	-0.037	0.01	-0.1451	-0.0469	0.0548	1	0.0151
Sr	**0.6656**	**0.4834**	0.005	**0.7365**	**0.4543**	0.2149	**0.2682**	**0.6764**	**0.6151**	**0.3657**	0.1941	0.0493	**0.886**	0.0151	1

TABLE A-9. Pearson's correlations (r) between trace element concentrations in log-transformed shell data across Sites A, B, C & D. Bold values indicate significance at $p<0.05$.

	Al	**As**	**Ba**	**Cd**	**Cu**	**Fe**	**Mg**	**Mn**	**Ni**	**Pb**	**Se**
Al	1	0.2404	**0.7596**	**0.4079**	**0.485**	**0.8521**	**0.5819**	**0.7126**	**0.645**	**0.5932**	**0.3258**
As	0.2404	1	**0.3026**	**0.9035**	**0.6844**	**0.3378**	**0.3831**	0.2483	0.0533	**0.4996**	-0.0712
Ba	**0.7596**	**0.3026**	1	**0.4599**	**0.4341**	**0.8298**	**0.5827**	**0.7636**	**0.7214**	**0.8146**	**0.3565**
Cd	**0.4079**	**0.9035**	**0.4599**	1	**0.8327**	**0.4335**	**0.6994**	**0.4132**	0.1864	**0.4462**	-0.0444
Cu	**0.485**	**0.6844**	**0.4341**	**0.8327**	1	**0.4267**	**0.7455**	**0.3615**	**0.2669**	**0.3457**	0.0061
Fe	**0.8521**	**0.3378**	**0.8298**	**0.4335**	**0.4267**	1	**0.4718**	**0.7533**	**0.6116**	**0.7067**	**0.3087**
Mg	**0.5819**	**0.3831**	**0.5827**	**0.6994**	**0.7455**	**0.4718**	1	**0.5324**	**0.3963**	0.2375	0.066
Mn	**0.7126**	0.2483	**0.7636**	**0.4132**	**0.3615**	**0.7533**	**0.5324**	1	**0.6428**	**0.5772**	**0.3148**
Ni	**0.645**	0.0533	**0.7214**	0.1864	**0.2669**	**0.6116**	**0.3963**	**0.6428**	1	**0.5938**	**0.4135**
Pb	**0.5932**	**0.4996**	**0.8146**	**0.4462**	**0.3457**	**0.7067**	0.2375	**0.5772**	**0.5938**	**1**	**0.4004**
Se	**0.3258**	-0.0712	**0.3565**	-0.0444	0.0061	**0.3087**	0.066	**0.3148**	**0.4135**	**0.4004**	1

TABLE A-10. Pearson's correlations (r) and Spearman's rank correlations between trace element concentrations in log-transformed soft body tissue data and shell data across Sites A, B, C & D. Bold values indicate significance at $p<0.05$.

	Correlation	**P Value**	**Spearman's Rank Correlation**	**Spearman's Rank P-Value**
Ag	NA	NA	NA	NA
Al	-0.1041276	0.42850218	-0.2277299	0.0802218
As	-0.1975795	0.13022619	-0.1275981	0.33127051
Ba	**0.29106212**	0.02405667	**0.29025506**	0.02446911
Be	NA	NA	NA	NA
Cd	-0.0149836	0.90953349	-0.2525871	0.05152595
Co	0.05783265	0.66072389	0.13779902	0.2937402
Cr	-0.0754525	0.56666134	-0.089648	0.49575995
Cu	**0.33647379**	0.00857333	0.2416462	0.06287436
Fe	-0.1536149	0.24126294	-0.2270075	0.08120487
Ga	NA	NA	NA	NA
Hg	NA	NA	NA	NA
K	**0.35428814**	0.0054816	**0.39799944**	0.00176955
Li	0.00427905	0.97411455	0.05688198	0.66596879
Mg	**0.26136492**	0.04368015	**0.40400111**	0.00148824
Mn	0.06569128	0.6180065	0.04879133	0.71056317
Na	**0.38297728**	0.00252621	**0.2912643**	0.02395427
Ni	-0.0631554	0.63166258	-0.0786398	0.5503382
Pb	**-0.2994766**	0.02009509	**-0.2915515**	0.02380945
Rb	**0.29790946**	0.02078775	**0.29779791**	0.02083782
Se	0.22288872	0.08693775	0.15899332	0.22498224
Sr	0.20427662	0.11744385	-0.1640107	0.21049837
U	-5.67E-05	0.99965673	-0.0972664	0.45970734
V	NA	NA	NA	NA
Zn	0.169021	0.19670323	0.20815173	0.11049894

TABLE A-11. Principal component analysis (PCA) on log-transformed soft body tissue data across Sites A, B, C & D.

Number	Eigenvalue	Percent	Percent	Cum Percent
1	5.2967	35.311	+++	35.311
2	2.2471	14.98	+	50.292
3	1.3961	9.307		59.599
4	1.12	7.467		67.066
5	0.9851	6.568		73.633
6	0.7742	5.161		78.795
7	0.6964	4.642		83.437
8	0.6145	4.097		87.533
9	0.568	3.787		91.32
10	0.361	2.406		93.727
11	0.3483	2.322		96.049
12	0.2858	1.905		97.954
13	0.1533	1.022		98.976
14	0.1104	0.736		99.712
15	0.0432	0.288		100

TABLE A-12. Loading matrix for the principal component analysis (PCA) of log-transformed soft body tissue data across Sites A, B, C & D.

	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8	PC9	PC10	PC11	PC12	PC13	PC14	PC15
Ag	0.79143	-0.3275	0.01487	0.12942	0.17144	-0.24817	0.03113	-0.21536	-0.00981	0.111	-0.08443	0.16956	-0.19054	-0.16265	0.00572
Al	0.63731	0.50732	-0.16816	0.05074	-0.12148	-0.19554	0.06628	0.32486	-0.0803	-0.06057	-0.17694	-0.29803	-0.03181	-0.10306	0.02848
As	-0.01739	0.26627	0.68011	0.30271	-0.27353	0.07755	0.48394	0.11691	0.16598	-0.02721	-0.05967	0.11477	-0.02805	0.01037	0.0005
Ba	0.75532	-0.34985	-0.02916	0.14684	-0.14567	0.04846	-0.16337	0.23253	0.20347	-0.13192	0.26734	-0.06259	-0.19179	0.09608	-0.01231
Cd	0.52837	-0.44159	0.08988	-0.19987	0.07884	0.1426	0.26079	0.238	-0.56326	0.04098	0.0613	0.04883	-0.00251	0.03804	-0.00699
Cu	0.29529	-0.76393	-0.05639	0.11559	-0.05781	-0.3475	0.20021	0.07012	0.20833	0.22342	0.07273	-0.11401	0.17848	0.00861	0.00598
Fe	0.42679	0.73499	-0.02511	-0.03984	0.0144	0.0282	0.16229	-0.20891	-0.04841	0.18128	0.39993	-0.0739	0.01685	-0.05576	-0.02056
Li	0.82985	0.03263	0.14735	-0.14619	0.10723	-0.00305	0.11787	-0.36797	0.01872	-0.00481	-0.20175	-0.18801	-0.03427	0.15364	-0.07208
Mg	0.68082	0.13534	0.16062	0.03809	-0.29521	0.31785	-0.36079	0.0996	0.01645	0.37708	-0.12739	0.05144	0.02813	0.01876	0.01307
Mn	0.52687	0.56851	-0.25854	-0.03242	0.05652	-0.44979	-0.02877	0.17023	-0.00373	0.00248	-0.02645	0.27514	0.04639	0.13119	-0.01486
Ni	0.22225	0.00688	-0.73564	-0.1189	0.11931	0.40766	0.36563	0.06671	0.24709	0.0547	-0.08172	0.07092	-0.02707	0.00248	0.01422
Pb	-0.08863	0.13367	0.06241	0.68477	0.67969	0.10871	-0.04731	0.09841	-0.04618	0.08228	-0.00824	-0.06163	0.00888	0.05182	0.01482
Rb	0.92654	-0.00275	0.13842	-0.05129	0.05511	0.07897	-0.0231	-0.19651	0.01721	-0.18377	0.03512	0.02841	0.09757	0.02558	0.15819
Se	-0.01027	0.07773	0.45756	-0.6428	0.49477	-0.0208	-0.0337	0.24134	0.24423	0.07134	0.01534	-0.01731	-0.02505	-0.02693	0.00789
Sr	0.89402	-0.06164	0.06621	0.09908	0.09647	0.18989	-0.11484	0.0658	0.0757	-0.22703	0.00583	0.08136	0.17658	-0.11874	-0.10319

TABLE A-13. Principal component analysis (PCA) on log-transformed shell data across Sites A, B, C & D.

Number	Eigenvalue	Percent	Percent	Cumulative Percent
1	5.96	54.182	+++++	54.182
2	2.1424	19.476	+	73.658
3	0.9073	8.248		81.906
4	0.6807	6.188		88.094
5	0.4063	3.694		91.788
6	0.3349	3.045		94.833
7	0.2531	2.301		97.134
8	0.1435	1.305		98.439
9	0.115	1.045		99.484
10	0.0393	0.357		99.841
11	0.0175	0.159		100

TABLE A-14. Loading matrix for the principal component analysis (PCA) of log-transformed shell data across Sites A, B, C & D.

	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8	PC9	PC10	PC11
Al	0.84729	-0.22329	-0.20868	-0.06823	-0.24803	-0.24968	0.11407	0.09469	-0.19287	-0.03142	0.00483
As	0.55677	0.67699	0.4422	-0.03795	-0.00531	0.05394	0.11898	0.10454	0.0112	-0.0002	0.08097
Ba	0.89904	-0.25293	0.00771	-0.12413	0.09069	0.01991	-0.2929	-0.03214	0.02868	-0.12283	0.03046
Cd	0.7087	0.66896	0.10787	0.06899	0.0111	0.09826	0.01384	0.10961	0.01012	-0.05597	-0.09288
Cu	0.69157	0.58037	-0.12629	0.21872	0.04449	-0.1898	0.0798	-0.27657	0.01235	-0.00846	0.00423
Fe	0.86591	-0.22949	0.00069	-0.2271	-0.27106	-0.11032	0.03109	0.01122	0.23932	0.04264	-0.01147
Mg	0.71924	0.32774	-0.50164	0.23343	0.04855	0.06247	-0.20606	0.10738	-0.00351	0.0919	0.02092
Mn	0.80981	-0.24865	-0.15517	-0.12294	-0.09737	0.44377	0.14465	-0.11157	-0.05761	0.00871	0.00387
Ni	0.70206	-0.46332	-0.11577	0.01785	0.4667	-0.07088	0.21815	0.07438	0.05266	-0.0006	-0.00131
Pb	0.78084	-0.19867	0.50869	-0.1618	0.11525	-0.06169	-0.14722	-0.06469	-0.11225	0.09779	-0.02766
Se	0.34321	-0.55667	0.30444	0.67742	-0.1342	0.04196	0.00408	0.01674	0.02423	-0.00967	0.00177

TABLE A-15. Results of the multivariate analysis of variance (MANOVA) for log-transformed soft body tissue concentrations among Sites A, B, C & D.

TEST	VALUE	F	DF	DenDF	p VALUE
WILKS' LAMBDA	0.1244717	4.1785	45	184.97	<.0001
PILLAI'S TRACE	1.392589	3.6964	45	192	<.0001
HOTELLING-LAWLEY	3.4971503	4.7285	45	144.09	<.0001
ROY'S MAX ROOT	2.3789735	10.1503	15	64	<.0001

TABLE A-16. Eigenvectors of the multivariate analysis of variance (MANOVA) for log-transformed soft body tissue concentrations among Sites A, B, C & D.

EIGENVECTOR	1	2	3	4	5	6	7	8
Ag	-0.184361	-0.0278563	0.04890114	0.00075857	-0.0963919	0.04676855	-0.02865	0.06618517
Al	-0.0329844	-0.259253	-0.0374868	0.052721	-0.0519974	-0.0577207	-0.0115407	0.01348082
As	0.04596613	0.01008855	0.03110445	0.02238894	-0.0018372	0.06944698	0.05034523	-0.0394549
Ba	0.07454312	0.02108157	0.03020436	-0.0218537	0.02984671	0.01829032	0.00160665	0.02295676
Cd	-0.0023158	-0.0164469	-0.0281606	0.0214865	0.02669087	-0.0104953	0.06828374	0.02194277
Cu	0.15778282	0.00181948	-0.0222077	0.03217418	-0.0126767	-0.0402075	0.03245959	0.01102466
Fe	-0.0072819	-0.000403	0.13266556	-0.0663492	-0.0031613	-0.103279	0.04119101	0.05451161
Li	0.10500067	0.1240133	-0.1276806	-0.0002625	0.14139996	-0.0262117	-0.0789128	-0.0505807
Mg	-0.0488395	-0.0400878	-0.0123925	0.03060621	0.01694294	0.03585384	-0.0334679	-0.0294419
Mn	-0.0491226	0.23565769	-0.1240642	0.02118497	-0.0005788	0.04376949	0.09523515	-0.0495014
Ni	0.00505136	-0.0051223	0.03618821	0.06479371	-0.0300651	0.03853014	-0.0113632	-0.0657102
Pb	-0.0334973	-0.0217989	-0.0140936	0.02980006	0.08788847	0.0125314	0.03521315	-0.0015341
Rb	-0.0927864	-0.1064736	0.18537177	0.19475931	-0.0271714	0.03736168	0.01050632	0.03114188
Se	0.03032467	0.02572996	0.02018788	0.03096924	-0.0087755	-0.0466274	0.01104176	-0.0036475
Sr	0.10872051	0.097201	-0.1139593	-0.2043755	-0.0042521	-0.0069242	0.00275143	0.01043478

TABLE A-16. Continued.

EIGENVECTOR	9	10	11	12	13	14	15
Ag	-0.0116664	0.0137946	-0.0386782	-0.0860933	0.07985073	-0.0693132	-0.0641329
Al	0.05690176	0.01095082	0.03471178	-0.0556287	-0.0250788	-0.0671814	-0.0334013
As	0.02406962	0.01246795	-0.0393154	-0.0140668	0.0006455	-0.028486	-0.004666
Ba	-0.0178411	0.01625185	-0.0709213	-0.0432396	-0.0770131	0.11383355	-0.1101799
Cd	-0.0765871	-0.0200425	-0.0544674	0.05642212	0.0014811	-0.0003888	0.00367466
Cu	0.04328636	0.01035923	0.06746994	0.00913065	0.05579275	0.03086782	0.09001588
Fe	-0.0286821	0.10643912	-0.0256063	-0.0120219	0.05777664	0.0315387	0.05857938
Li	-0.0268972	0.06781834	-0.1334046	-0.0377989	-0.0238106	0.03539079	0.03588505
Mg	0.05023573	-0.0058287	0.01739616	0.10468618	0.11535839	0.06191747	-0.0155123
Mn	0.01359586	-0.0174112	-0.0232181	0.05418804	-0.0433458	0.09637681	0.02218694
Ni	-0.0510601	0.02179571	0.03140564	-0.0078171	0.00859337	0.00728724	-0.0189399
Pb	-0.0058965	-0.022547	0.05447061	-0.0277153	0.01531268	0.04230905	-0.0084508
Rb	0.0936951	-0.2110102	0.1912211	0.06522211	-0.1066783	0.0073301	0.02923971
Se	0.02026947	0.00701171	0.02828036	-0.0035983	0.03124298	-0.0291622	-0.0819472
Sr	-0.0530784	0.17873391	0.01898469	0.02391146	0.02231215	-0.1387235	0.0604097

TABLE A-17. Results of the multivariate analysis of variance (MANOVA) for log-transformed shell concentrations among Sites A, B, C & D.

TEST	VALUE	F	DF	DenDF	p VALUE
WILKS' LAMBDA	0.0148599	13.099	33	136.23	<.0001
PILLAI'S TRACE	1.8267558	6.7942	33	144	<.0001
HOTELLING-LAWLEY	19.276024	26.2303	33	99.18	<.0001
ROY'S MAX ROOT	17.124739	74.7261	11	48	<.0001

TABLE A-18. Eigenvectors of the multivariate analysis of variance (MANOVA) for log-transformed shell concentrations among Sites A, B, C & D.

EIGENVECTOR	1	2	3	4	5	6	7	8	9	10	11
Al	-0.015	-0.071	-0.116	-0.099	0.051	0.178	0.005	-0.073	-0.059	0.033	-0.145
As	0.089	-0.008	-0.322	0.423	-0.199	0.054	0.201	-0.072	-0.346	0.011	-0.110
Ba	-0.629	0.067	-0.148	0.167	-0.164	-0.013	-0.028	-0.056	0.101	-0.077	-0.331
Cd	-0.003	0.103	0.365	-0.451	0.373	-0.140	-0.119	0.120	0.334	-0.032	-0.154
Cu	0.043	0.020	-0.048	-0.078	-0.246	0.042	0.032	0.070	0.084	-0.045	0.057
Fe	0.032	-0.193	0.242	0.067	0.025	-0.058	0.022	0.109	-0.019	-0.112	0.127
Mg	0.349	-0.143	-0.062	0.248	0.077	0.041	0.022	-0.050	-0.075	0.158	0.255
Mn	0.032	0.359	0.030	-0.011	-0.053	0.067	0.017	-0.042	-0.045	0.002	0.065
Ni	0.015	0.014	-0.115	-0.015	0.082	-0.073	0.006	0.107	-0.038	-0.082	0.020
Pb	-0.084	-0.106	0.086	-0.181	0.095	-0.036	0.014	-0.086	0.033	0.178	0.328
Se	0.004	0.013	0.040	0.024	-0.033	-0.003	-0.039	0.093	-0.035	0.082	-0.064

TABLE A-19. Canonical scores of the canonical discriminant analysis (CDA) across Sites A, B, C & D.

CLAM ID	BODY CANON 1	BODY CANON 2	BODY CANON 3	SHELL CANON 1	SHELL CANON 2	SHELL CANON 3
A1	-0.0579	0.0986	-0.0348			
A2	-0.0852	0.1587	-0.0218			
A3	-0.0271	0.0806	-0.0493			
A4	-0.1077	0.0722	-0.0910			
A5	-0.1660	0.1628	-0.1091			
A6	-0.3362	0.0941	0.0040	0.6656	0.1978	0.0097
A7	0.0077	0.0031	0.0223	0.6729	0.0707	-0.0208
A8	-0.3322	0.0294	0.0340	0.7858	0.3090	-0.0214
A9	-0.1584	0.1146	-0.1822	0.8140	0.2030	0.0212
A10	-0.2666	0.0116	-0.0392	0.8146	0.1940	-0.0247
A11	-0.1890	-0.0212	-0.3669	0.4532	0.3853	0.0332
A12	-0.1949	0.0591	-0.2635	0.5121	0.2558	-0.0634
A13	-0.2963	0.1812	-0.1480	0.8352	0.0844	0.0917
A14	-0.0709	0.3310	-0.1842	0.5215	0.0168	0.0917
A15	-0.1935	0.2113	-0.1322	0.6599	0.1162	-0.2199
A16	-0.2286	0.0536	-0.1906	0.6734	0.1305	0.0500
A17	-0.1002	-0.0269	-0.0789	0.5413	0.1380	0.1943
A18	-0.2107	0.1504	-0.0106	0.7442	0.0194	0.0291
A19	0.0166	0.0732	-0.1093	0.4614	0.1971	0.0377
A20	-0.3558	0.0485	0.0766	0.4994	0.0617	0.0886
B1	-0.0096	0.0580	0.0528			
B2	0.0106	0.1877	-0.0049			
B3	0.0477	0.1054	0.1907			
B4	-0.1990	-0.0471	0.2710			
B5	-0.0435	-0.0592	0.1566			
B6	-0.1786	0.0105	0.0462	-0.6842	0.1281	-0.0765
B7	-0.0802	-0.0403	-0.0106	-0.6566	0.0420	-0.0030
B8	0.0735	0.1119	0.0539	-0.7375	0.1751	0.1941
B9	-0.0057	0.0184	-0.0372	-0.7636	-0.0961	0.0715
B10	-0.1662	0.0733	0.0008	-0.6552	0.3026	0.1852
B11	-0.0206	0.2313	0.0247	-0.5932	0.0471	-0.0681
B12	-0.0369	-0.0081	0.0743	-0.5817	0.2001	0.1583
B13	-0.1980	0.0599	0.2214	-0.5761	0.1433	-0.0641
B14	-0.1230	0.1842	-0.0389	-0.6315	0.1064	0.0372
B15	-0.0364	0.3005	0.4663	-0.5050	0.1549	0.2721
B16	-0.0582	0.0678	0.0454	-0.5474	-0.0028	0.2708
B17	-0.0209	0.0638	0.0133	-0.5707	0.0442	0.0544
B18	-0.0856	0.0716	0.0847	-0.5989	0.0328	0.1100
B19	-0.1257	0.0358	0.0051	-0.8644	0.0101	0.2232
B20	-0.1100	0.1109	0.0519	-0.5386	0.1344	0.4981

TABLE A-19. Continued.

CLAM ID	BODY CANON 1	BODY CANON 2	BODY CANON 3	SHELL CANON 1	SHELL CANON 2	SHELL CANON 3
C1	-0.0667	0.0956	-0.0219			
C2	-0.1256	-0.0625	-0.0853			
C3	-0.0531	-0.1276	0.0109			
C4	0.0646	-0.1089	0.0678			
C5	-0.0816	-0.2246	-0.0670			
C6	-0.2621	-0.1262	-0.0230	0.2649	-0.3389	0.1512
C7	0.0074	-0.1633	-0.0012	0.1028	-0.4031	-0.0160
C8	-0.0767	-0.1708	0.0113	0.4208	-0.5531	0.1237
C9	-0.2143	-0.1813	-0.1416	0.5710	-0.1010	0.2110
C10	-0.1626	-0.3033	0.0707	0.2936	-0.3062	-0.1600
C11	-0.0181	-0.1048	-0.0772	0.4075	-0.2918	0.1267
C12	-0.0615	-0.1192	-0.1740	0.4431	-0.3188	-0.0605
C13	-0.1866	-0.2653	0.0164	0.4872	-0.2676	-0.0019
C14	-0.1710	-0.0971	-0.0241	0.2219	-0.3355	0.0872
C15	-0.2471	-0.2269	0.0148	0.3814	-0.3177	-0.0203
C16	0.1347	-0.2025	0.0122	-0.1394	-0.3673	0.4229
C17	-0.0165	-0.2019	0.0040	0.5715	-0.3493	0.2332
C18	0.1491	-0.0773	-0.0384	0.4058	-0.1976	0.2448
C19	-0.2399	-0.1961	-0.0086	0.4621	0.0183	0.3493
C20	-0.3367	-0.2734	0.1209	0.2879	-0.0735	-0.1162
D1	0.1104	-0.2258	-0.1731			
D2	0.0903	0.1223	0.0465			
D3	0.2063	0.1307	-0.1534			
D4	0.1545	0.1088	-0.1415			
D5	0.4661	-0.1324	-0.1804			
D6	0.2420	0.2643	-0.0029	-0.5563	0.1605	-0.1323
D7	0.2193	-0.0312	-0.0167	-0.5352	-0.1241	-0.0260
D8	0.3875	-0.2382	-0.0299	-0.4624	0.0544	-0.0495
D9	0.2251	-0.1328	-0.0973	-0.5720	-0.2502	0.0691
D10	0.2972	-0.0456	-0.0128	-0.4827	-0.2467	0.0344
D11	0.2068	0.0037	-0.0288	-0.3659	-0.1630	-0.0475
D12	0.3711	0.1062	-0.0578	-0.6520	0.0674	-0.3175
D13	0.5957	-0.2767	0.1807	-0.4278	-0.2341	-0.1072
D14	0.2458	0.0579	-0.0689	-0.3317	-0.2688	-0.0709
D15	0.2378	0.0404	-0.1464	-0.4911	-0.1327	-0.0728
D16	0.1584	0.1793	-0.2202	-0.4569	-0.0725	0.0288
D17	0.3628	0.3783	0.0567	-0.4257	0.1174	-0.0422
D18	0.3680	0.0899	0.0044	-0.3384	-0.1511	0.0813
D19	0.4067	-0.0758	-0.1437	-0.4395	-0.1724	-0.0195
D20	0.2095	0.0498	0.4135	-0.3218	-0.5102	-0.1392

TABLE A-20. Results of univariate analyses of variance (ANOVAs) for soft body tissue concentrations testing the effect of site across Sites A, B, C & D. Sites not connected by the same letter are significantly different based on post-hoc Tukey's HSD.

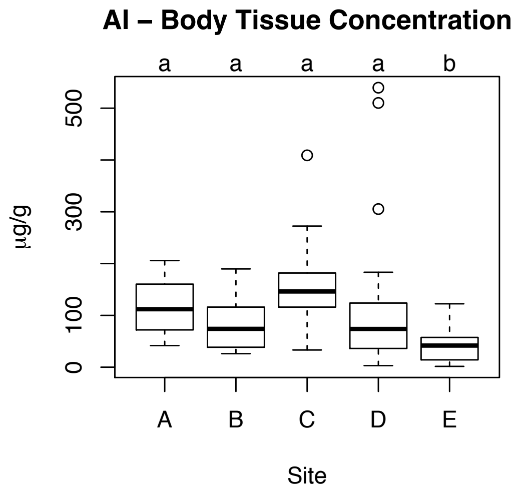
ELEMENT	F RATIO	P VALUE	SITE A	SITE B	SITE C	SITE D
Ag	0.8314	0.4807				
Al	4.0257	0.0103	ab	b	a	b
As	1.3249	0.2725				
Ba	1.6273	0.19				
Cd	0.7489	0.5264				
Cu	9.401	<.0001	b	b	b	a
Fe	5.3635	0.0021	a	a	a	b
Li	0.4316	0.731				
Mg	0.6679	0.5744				
Mn	7.5504	0.0002	a	ab	b	b
Ni	0.0266	0.9941				
Pb	1.4746	0.2282				
Rb	0.3703	0.7747				
Se	2.1176	0.105				
Sr	0.1063	0.9562				

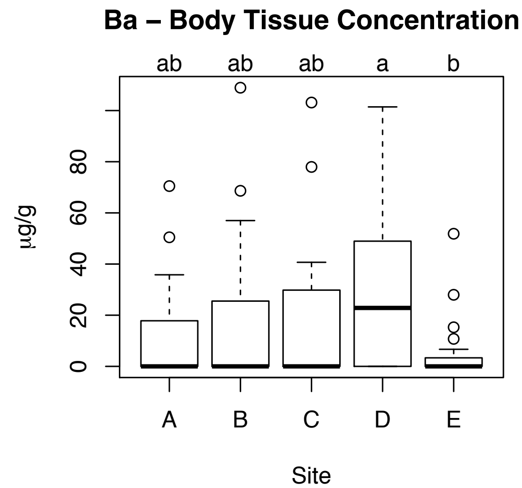
TABLE A-21. Results of univariate analyses of variance (ANOVAs) for shell concentrations testing the effect of site across Sites A, B, C & D. Sites not connected by the same letter are significantly different based on post-hoc Tukey's HSD.

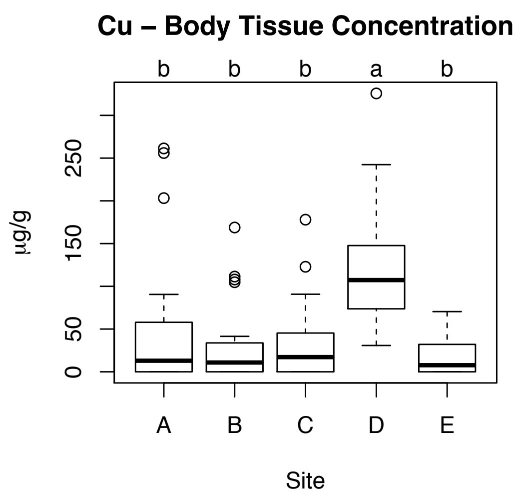
ELEMENT	F RATIO	P VALUE	SITE A	SITE B	SITE C	SITE D
Al	6.2659	0.001	B	a	ab	A
As	0.0504	0.9849				
Ba	30.6437	<.0001	B	a	b	A
Cd	0.2091	0.8897				
Cu	0.1167	0.95				
Fe	13.3574	<.0001	C	a	bc	Ab
Mg	0.1286	0.9427				
Mn	16.0372	<.0001	B	a	c	B
Ni	12.1672	<.0001	B	a	b	A
Pb	29.2201	<.0001	B	a	b	A
Se	3.5901	0.0191	B	a	ab	Ab

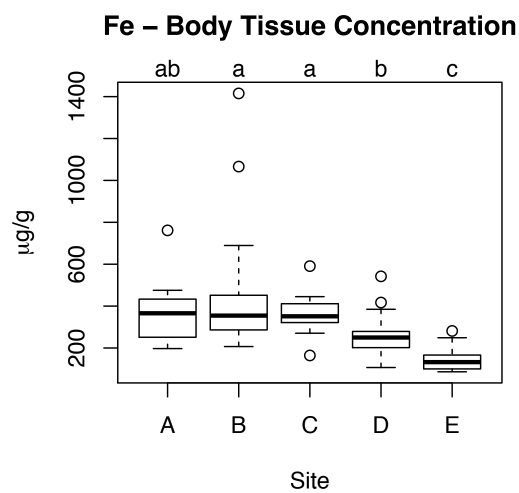
APPENDIX B

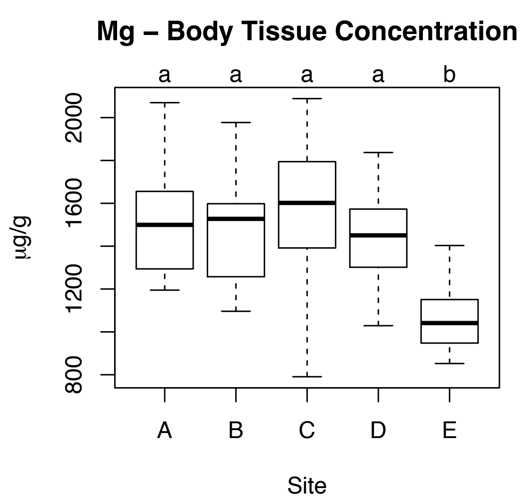
FIGURE B-1. Body tissue concentration box plots including results from Tukey's HSD post-hoc tests for Sites A, B, C, D & E. Sites not connected by the same letter are significantly different. Note that only those elements included in the MANOVA were subjected to Tukey's HSD

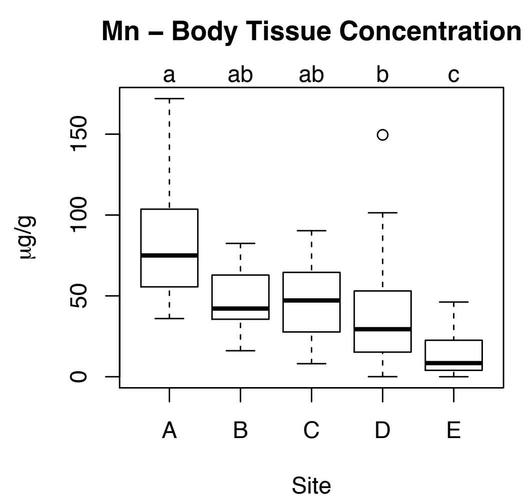

Al – Body Tissue Concentration
a a a a b
µg/g
0 100 300 500
A B C D E
Site

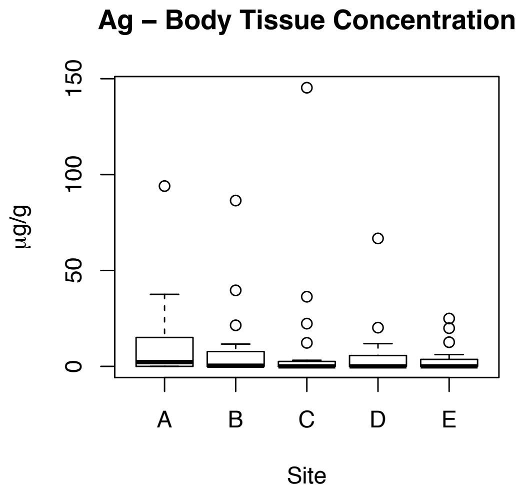

Ba – Body Tissue Concentration
ab ab ab a b
µg/g
0 20 40 60 80
A B C D E
Site

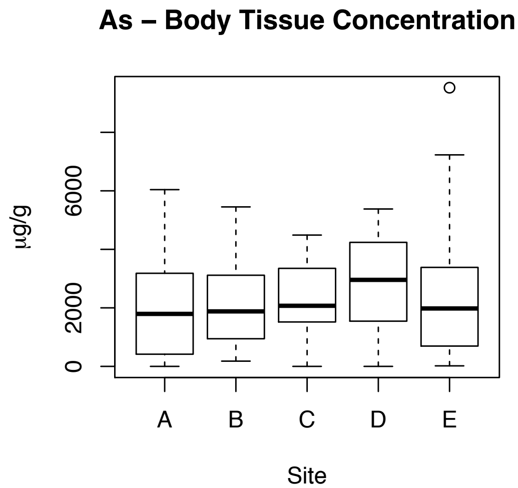

Cu – Body Tissue Concentration
b b b a b
µg/g
0 50 150 250
A B C D E
Site

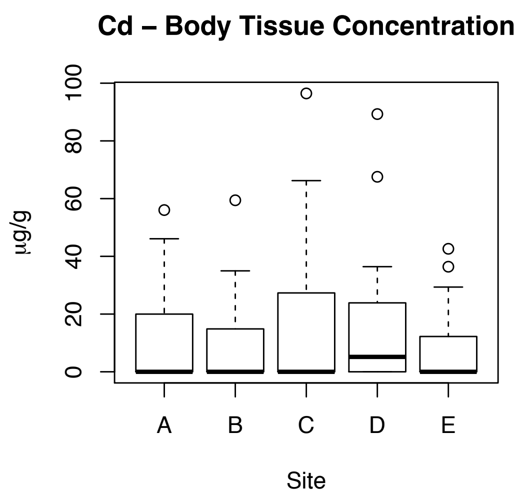

Fe – Body Tissue Concentration
ab a a b c
µg/g
200 600 1000 1400
A B C D E
Site

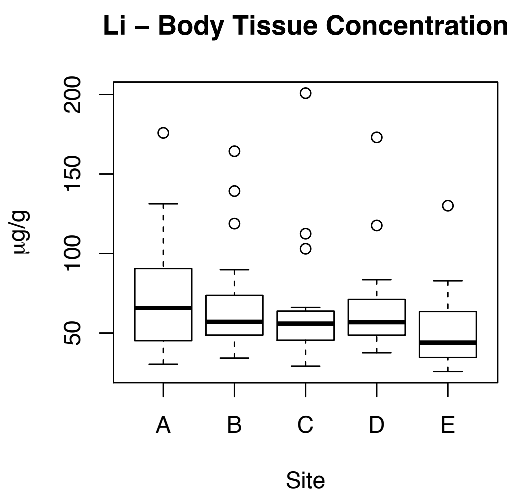

Mg – Body Tissue Concentration
a a a a b
µg/g
800 1200 1600 2000
A B C D E
Site

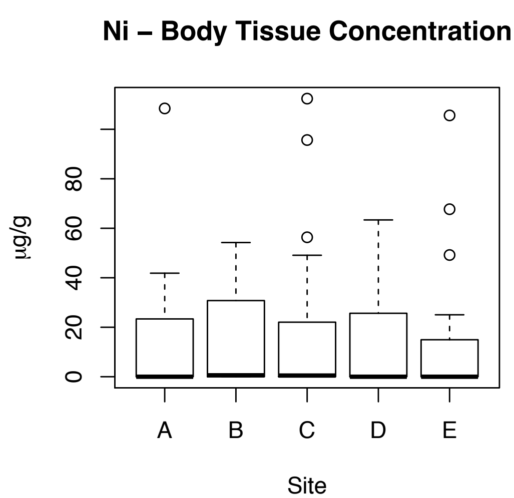

Mn – Body Tissue Concentration
a ab ab b c
µg/g
0 50 100 150
A B C D E
Site

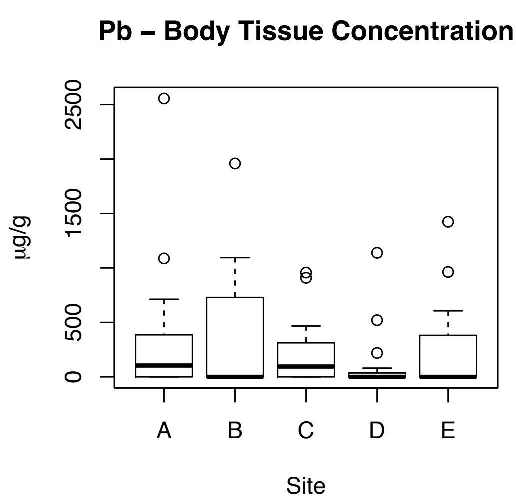

Ag – Body Tissue Concentration
µg/g
0
50
100
150
A
B
C
D
E
Site

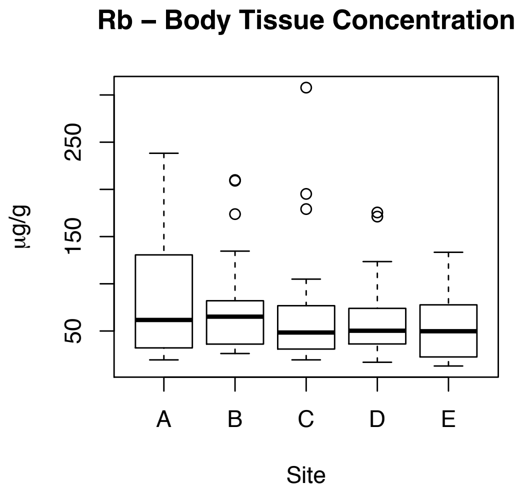

As – Body Tissue Concentration
µg/g
0
2000
6000
A
B
C
D
E
Site

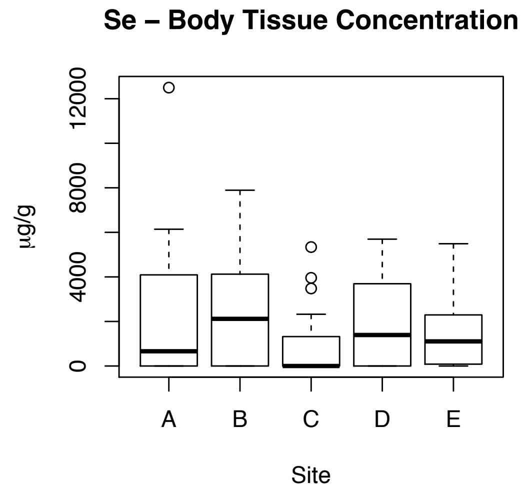

Cd – Body Tissue Concentration
µg/g
0
20
40
60
80
100
A
B
C
D
E
Site


Li – Body Tissue Concentration
µg/g
50
100
150
200
A
B
C
D
E
Site


Ni – Body Tissue Concentration
µg/g
0
20
40
60
80
A
B
C
D
E
Site


Pb – Body Tissue Concentration
µg/g
0
500
1500
2500
A
B
C
D
E
Site


Rb – Body Tissue Concentration
µg/g
50
150
250
A
B
C
D
E
Site


Se – Body Tissue Concentration
µg/g
0
4000
8000
12000
A
B
C
D
E
Site

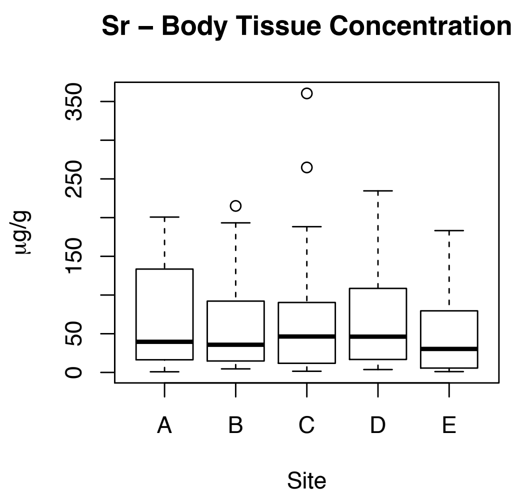
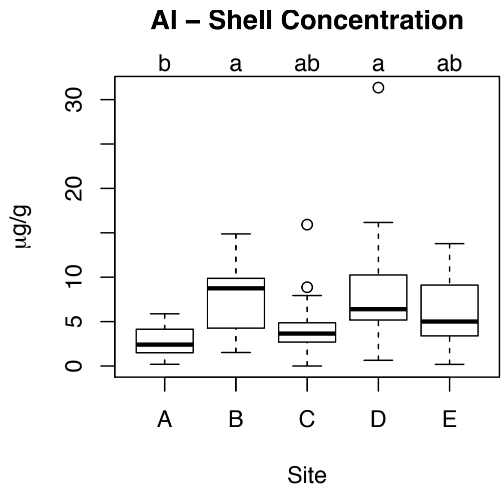
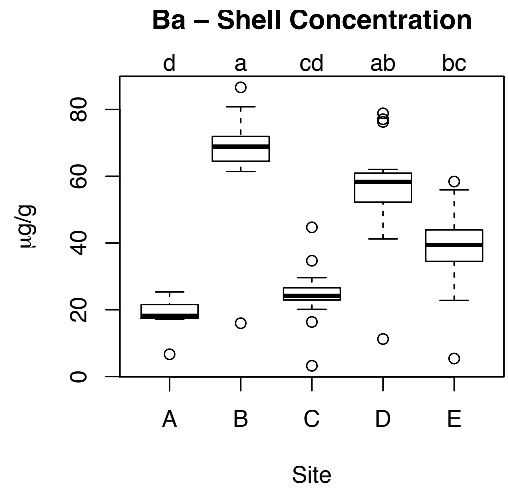
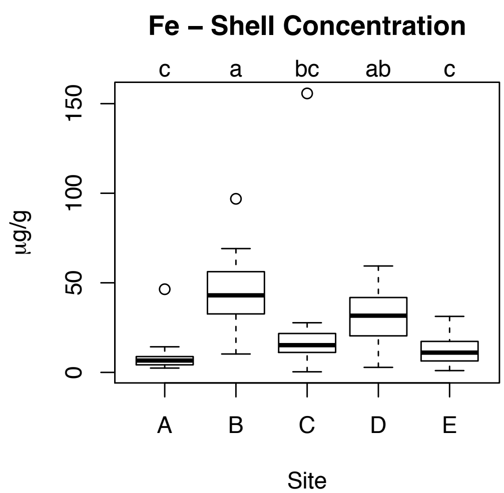
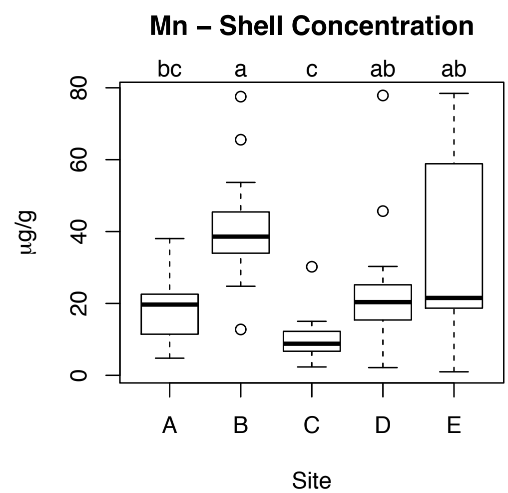

Sr – Body Tissue Concentration
µg/g
0
50
150
250
350
A
B
C
D
E
Site

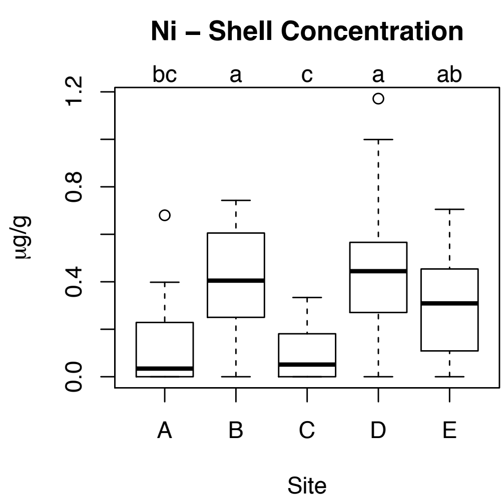
FIGURE B-2. Shell concentration box plots including results from Tukey's HSD post-hoc tests for Sites A, B, C, D & E. Sites not connected by the same letter are significantly different. Note that only those elements included in the MANOVA were subjected to Tukey's HSD.

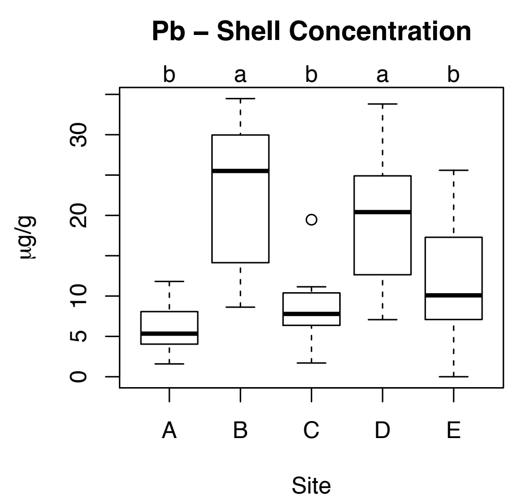

Al – Shell Concentration
b a ab a ab
µg/g
0 5 10 20 30
A B C D E
Site

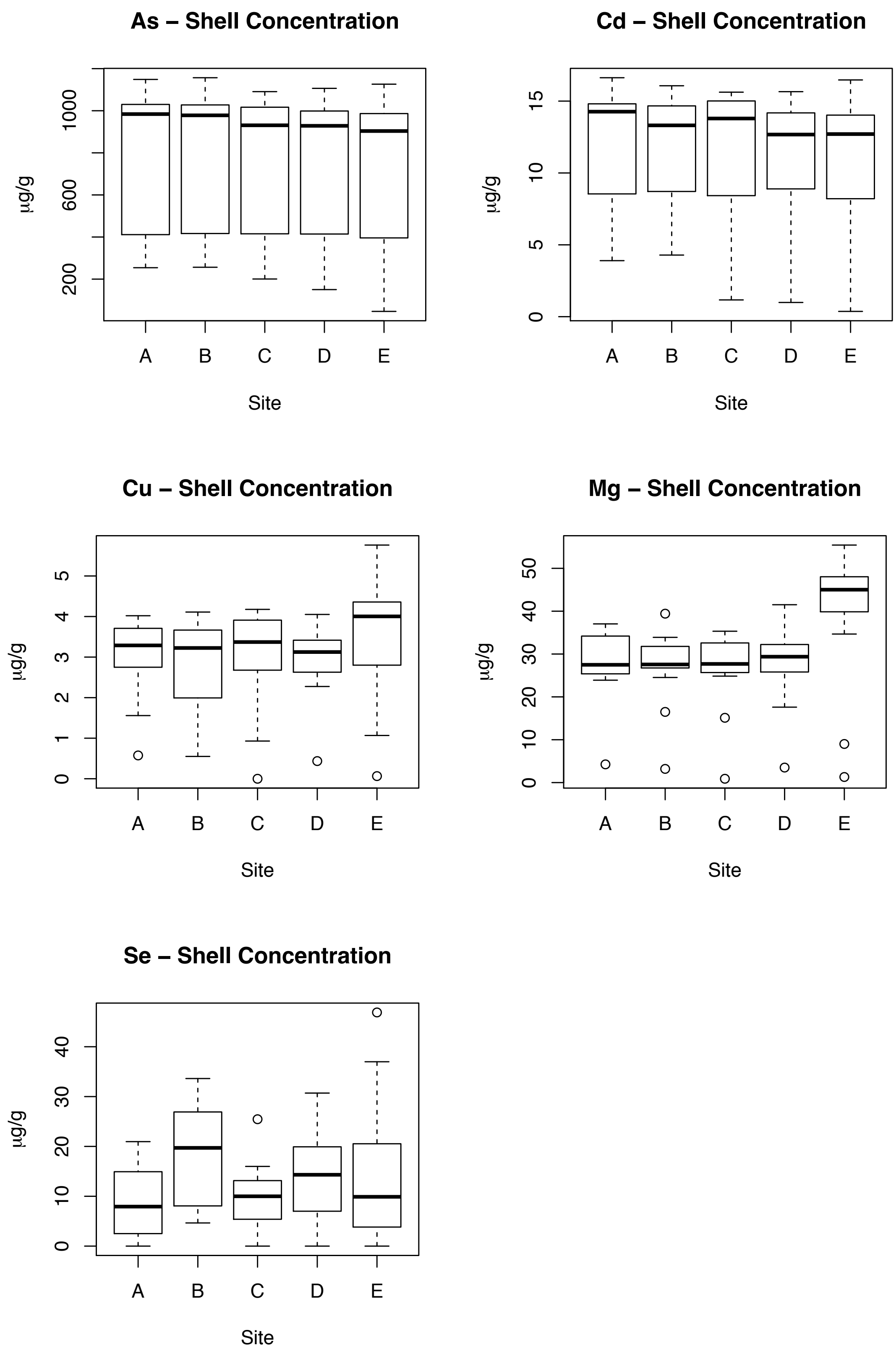

Ba – Shell Concentration
d a cd ab bc
µg/g
0 20 40 60 80
A B C D E
Site


Fe – Shell Concentration
c a bc ab c
µg/g
0 50 100 150
A B C D E
Site


Mn – Shell Concentration
bc a c ab ab
µg/g
0 20 40 60 80
A B C D E
Site


Ni – Shell Concentration
bc a c a ab
µg/g
0.0 0.4 0.8 1.2
A B C D E
Site


Pb – Shell Concentration
b a b a b
µg/g
0 5 10 20 30
A B C D E
Site


As – Shell Concentration
µg/g
200
600
1000
A
B
C
D
E
Site
Cd – Shell Concentration
µg/g
0
5
10
15
A
B
C
D
E
Site
Cu – Shell Concentration
µg/g
0
1
2
3
4
5
A
B
C
D
E
Site
Mg – Shell Concentration
µg/g
0
10
20
30
40
50
A
B
C
D
E
Site
Se – Shell Concentration
µg/g
0
10
20
30
40
A
B
C
D
E
Site

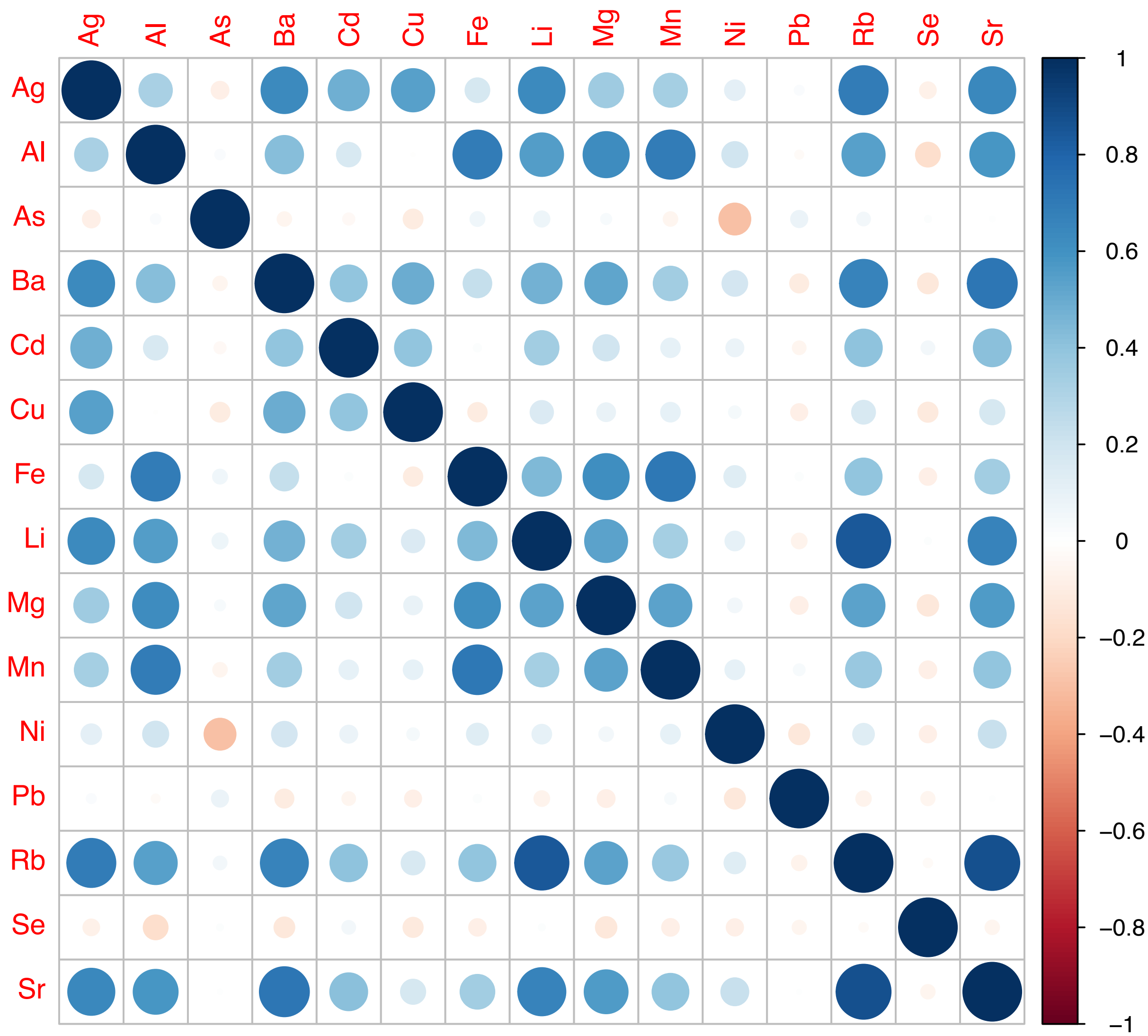


FIGURE B-3. Correlation between elements in the *Corbicula fluminea* soft body tissue across all sites.

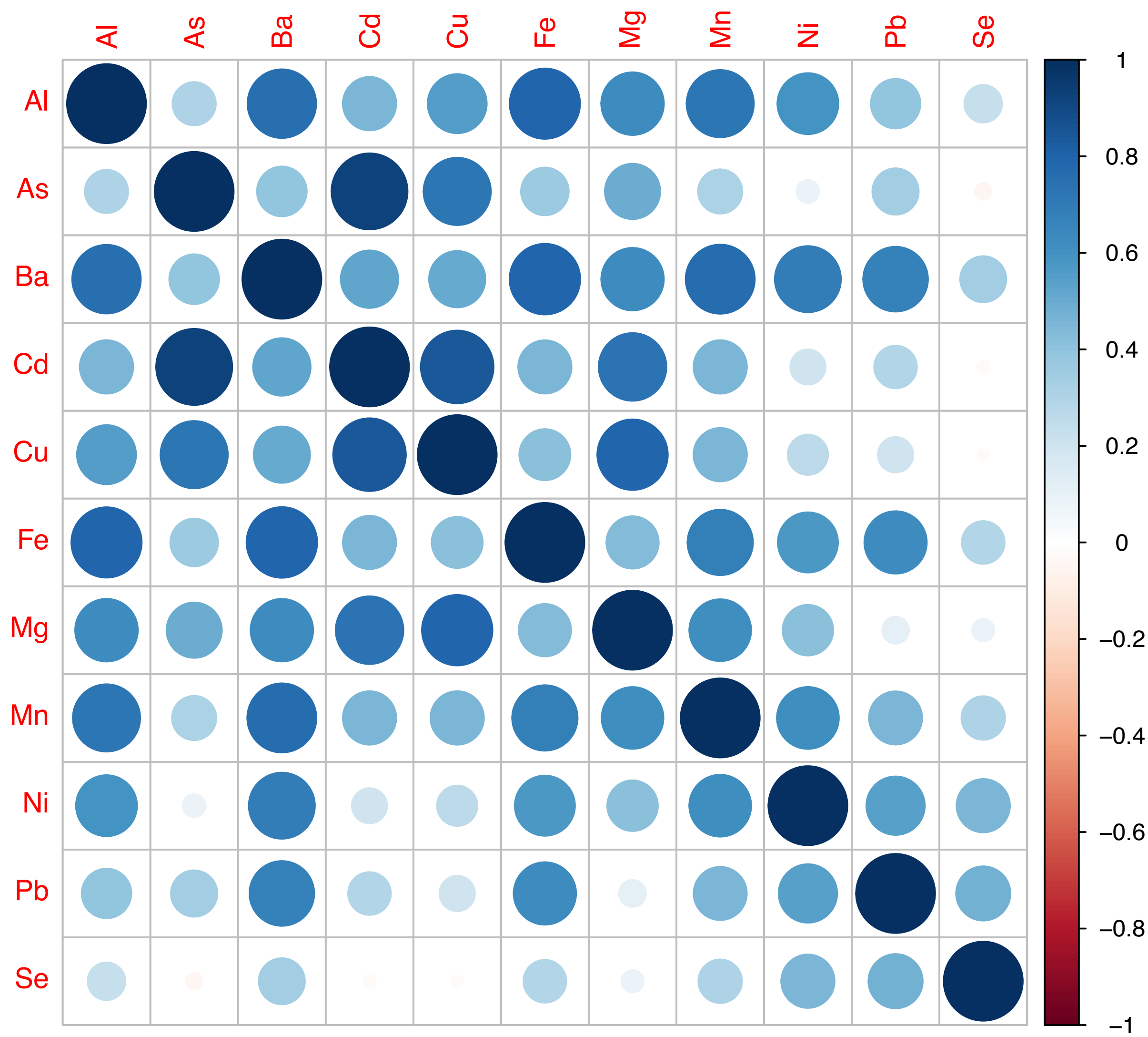


FIGURE B-4. Correlation between elements in the *Corbicula fluminea* shells across all sites.

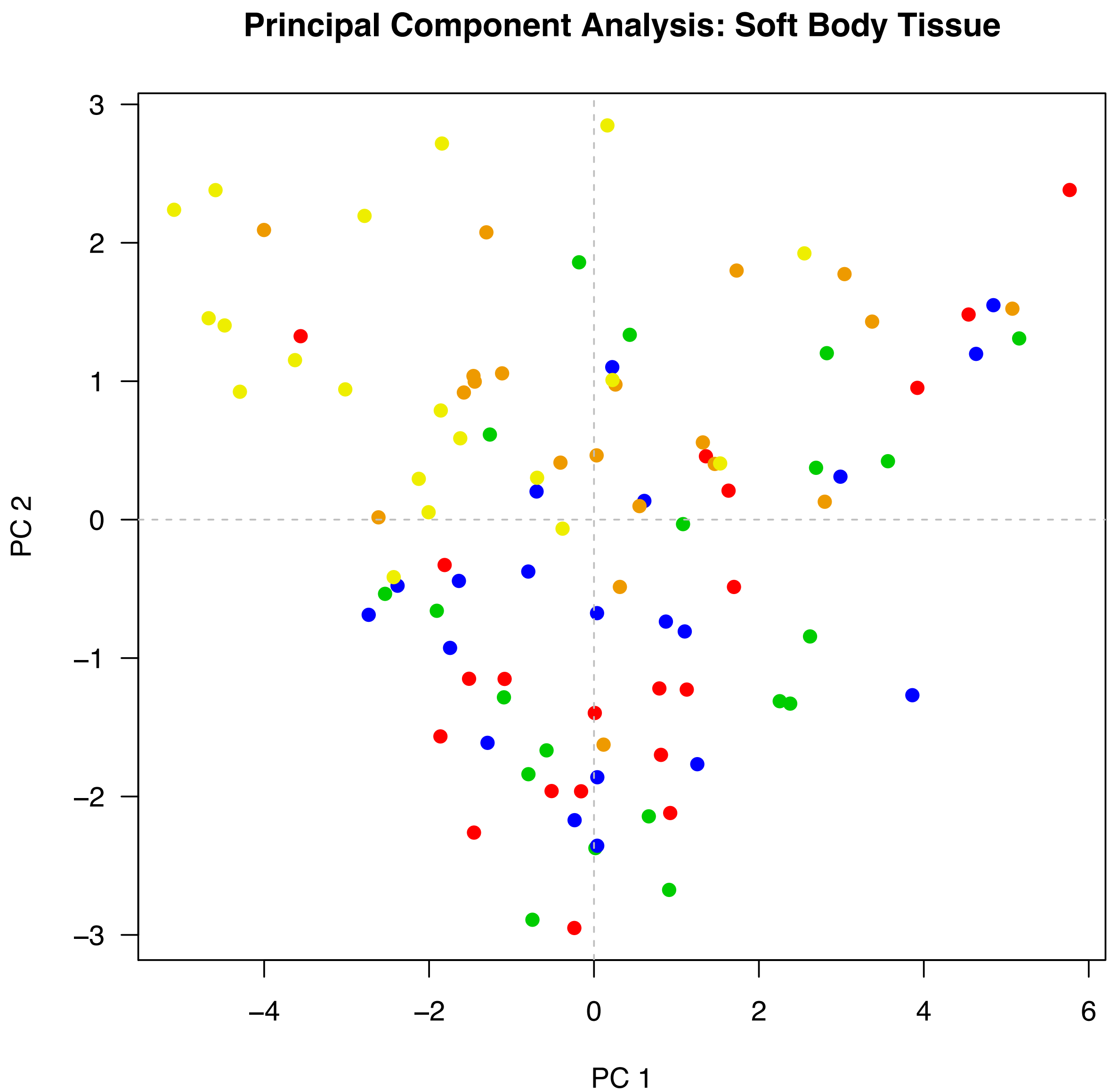


FIGURE B-5. Principal component analysis (PCA) of log-normalized *Corbicula fluminea* soft body tissue data across Sites A (green), B (blue), C (red), D (orange), and E (red).

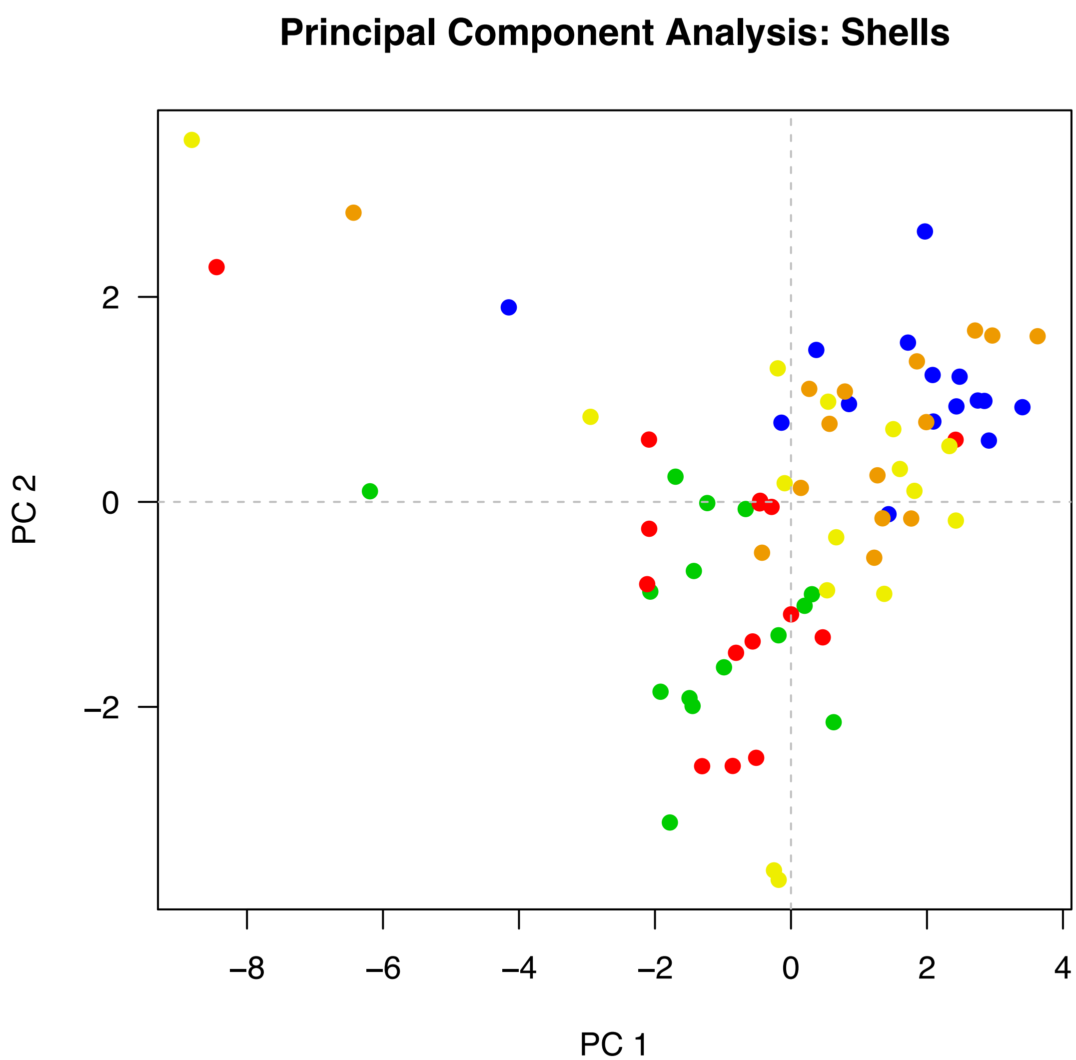


FIGURE B-6. Principal component analysis (PCA) of log-normalized *Corbicula fluminea* shell data across Sites A (green), B (blue), C (red), D (orange), and E (red).

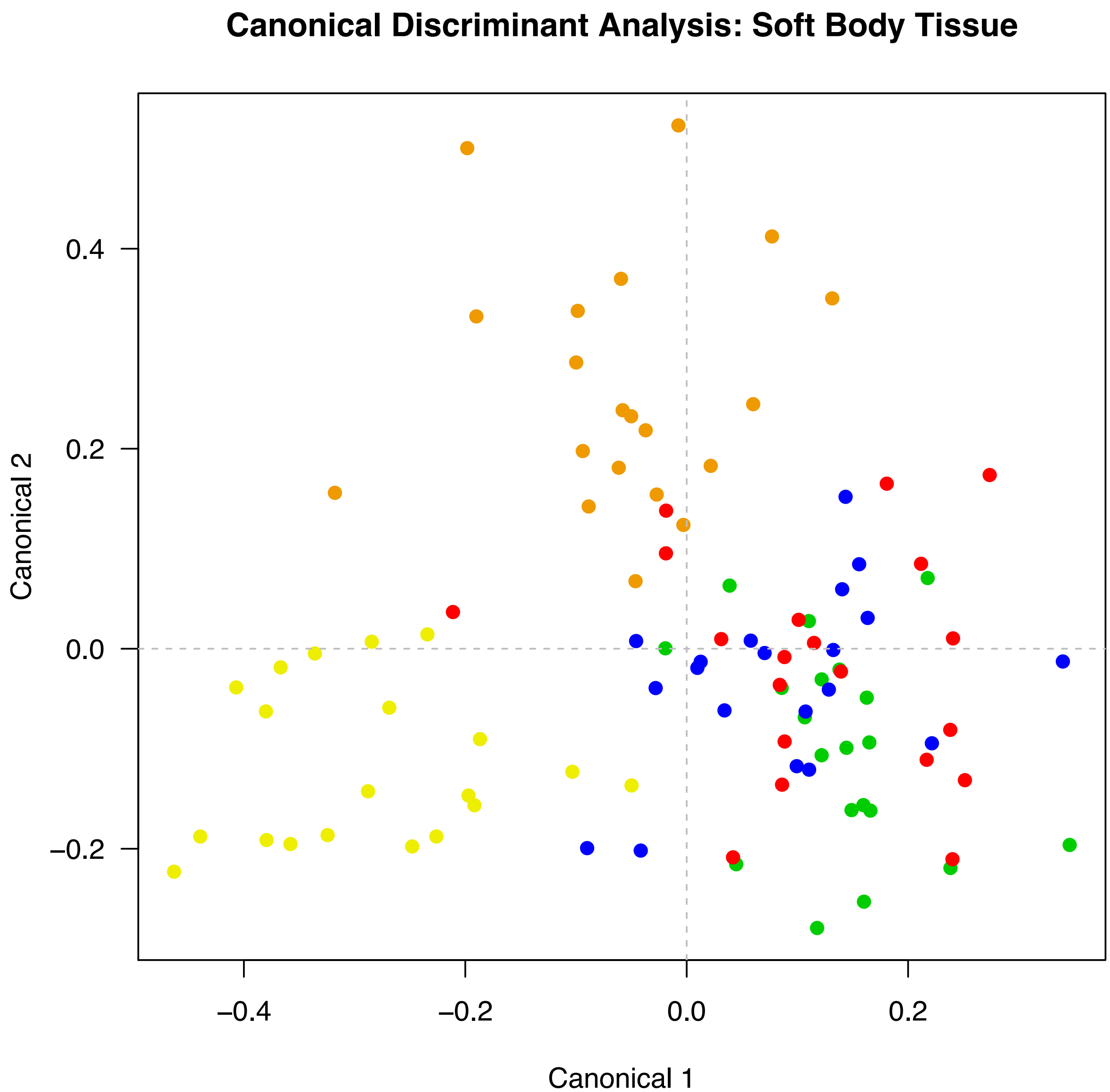


FIGURE B-7. Canonical discriminant analysis (CDA) of log-normalized *Corbicula fluminea* soft body tissue data across Sites A (green), B (blue), C (red), D (orange), and E (red).

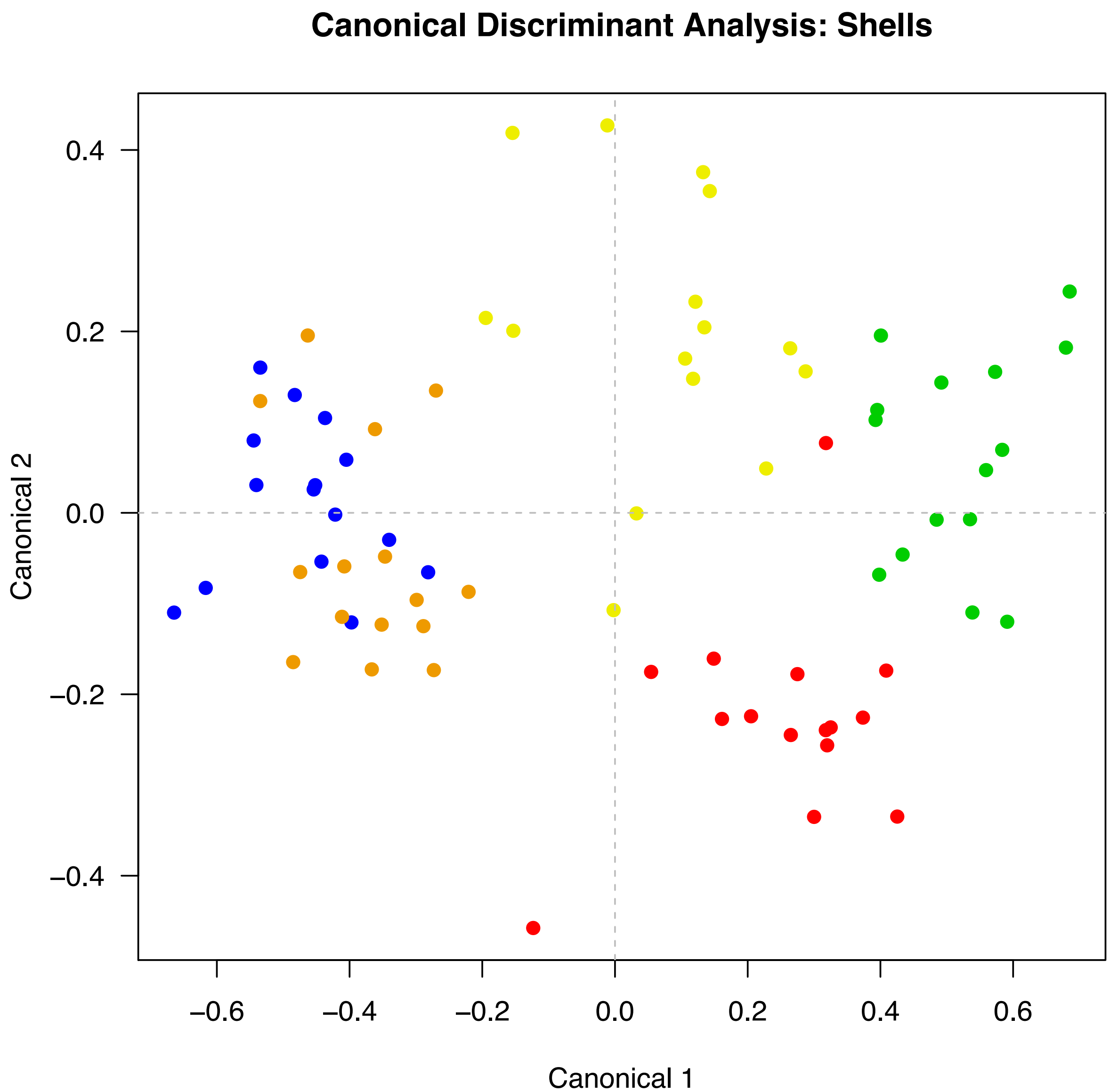


FIGURE B-8. Canonical discriminant analysis (CDA) of log-normalized *Corbicula fluminea* shell data across Sites A (green), B (blue), C (red), D (orange), and E (red).

VITA

Hannah Ruth Johnson was born in Chattanooga, Tennessee, and grew up playing outside with her brother and sister on Signal Mountain with parents who instilled in her a respect for the environment. She attended Girls Preparatory School of Chattanooga, where she was honored to receive the Woman of the Wilderness award her senior year. After spending her freshman year of college attending UNCA in Asheville, North Carolina, she transferred to the University of Tennessee and has yet to escape the charms of Knoxville. After two years of architecture school, Hannah changed her major to Environmental Studies and has ever since been working with Dr. Mike McKinney. After finishing her BA in Environmental Studies, she went on to complete a BS in Ecology and Evolutionary Biology. She then began her graduate work with Dr. McKinney in the spring of 2011.