Geochemical and Taphonomic Signatures of Freshwater Mussel Shells as Evidence of Mercury-Related Extirpations in the North Fork Holston River, Virginia

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Abstract

This study utilized freshwater mussel shells to assess the role of mercury contamination in the North Fork Holston River, an aquatic habitat affected by extensive extirpations of mussel populations starting in the early 1970's. Mussel shells (n=366) collected from 5 sites, upstream and downstream of Saltville (where mercury was used from 1950-1972) were analyzed to test if: (1) geochemical signatures of shells record variation in mercury levels relative to the contamination source; and (2) shell taphonomy could be used to differentiated affected and unaffected sites.

Analysis of 40 shells for geochemical signatures using atomic absorption spectroscopy indicated a strong longitudinal pattern. Mercury content was as follows: upstream sites had low Hg concentrations (<5 to 31ppb), shells directly below Saltville had high concentrations (23-4,637ppb), shells 18km downstream of Saltville displayed intermediate values (7-115ppb), and those 38.4km downstream were comparable to upstream sites (<10ppb). Two pre-industrial shells collected from Saltville in 1917 also yielded Hg estimates (5-6ppb) comparable with upstream estimates. The Hg content was not correlated with shell length ($r=-0.3$; $p=0.2$) or degree of taphonomic alteration ($r=0.18$; $p=0.28$). Analysis of 366 shells for taphonomic signatures indicated that shells are most heavily altered and fragmented directly downstream of Saltville. In contrast, upstream sites, inhabited by reproducing mussel populations, contain many fresh-dead shells. Taphonomic signatures can thus be used to differentiate sites with different extirpation histories. Relic mussel shells can provide useful spatial and temporal data on Hg concentrations in polluted ecosystems and offer a tool for delineating areas with unknown extirpation histories.

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Chapter 1: Introduction

There is increasing awareness that skeletal remains of benthic organisms (shells, tests, etc.) often provide important, and otherwise inaccessible historical insights into the anthropogenic contamination of aquatic ecosystems (Imlay, 1982; Carell et al., 1987; Bourgoin, 1990; Jeffree et al., 1995; Nystrom, 1996; Thomas and Bendell-Young, 1998; Amaral et al., 2000; Gundacker, 2000; Vander Putten et al., 2000; Giusti and Zhang, 2002; Markich et al., 2002; Yap et al., 2003). This utilization of remains may supplement and augment ongoing efforts of conservation biologists and environmental scientists. These non-invasive avenues of research, which do not require sampling from extant populations, have been recently applied to marine habitats. The multiple case studies have shown consistently that shelly remains of long dead mollusks can yield valuable and otherwise inaccessible insight into the recent history of the now degraded ecosystems (e.g., Bourgoin, 1990; Kowalewski et al., 2000; Vander Putten et al., 2000; Rodriguez et al., 2001; Giusti and Zhang, 2002; Yap et al., 2003). This approach deserves particular attention of freshwater ecologists because empty shells of freshwater mussels litter many streams, rivers, and lakes of the world and may provide data comparably valuable to those provided by remains left behind by the marine benthos.

The main goal of this study is to explore the utility of such non-invasive, shell-based techniques for studying freshwater ecosystems heavily affected by mercury contamination. Specifically, the study focuses on mussel shells collected from the North Fork Holston River of southwest Virginia. The study should have a wide applicability because (1) mercury pollution affects many habitats of the world (Amaral et al., 2000; Costa et al., 2000; Odzak et al., 2000) and (2) bivalves are among the key bio-monitoring tools used to assess the current state of aquatic ecosystems worldwide (Bourgoin, 1990; Avelar et al., 2000; Costa et al., 2000; Odzak et al., 2000; Sericano, 2000).

Mercury pollution affects many aquatic ecosystems worldwide (Amaral et al., 2000; Costa et al., 2000; Odzak et al., 2000). Much of the pollution is a result of the chlor-alkali industry that, for over 100 years, utilized a mercury cell in the electrolysis process (Kiefer, 2002). The process of producing chlorine and caustic soda first uses mercury to separate brine into chlorine gas and sodium. The sodium is dissolved in the mercury to produce sodium amalgam that passes out of the electrolytic cell and into

another reactor. The amalgam reacts with water to produce hydrogen gas, caustic soda, and regenerates the mercury (Dangwal, 1993). This process is not entirely efficient, and some mercury can be lost in the effluent. (A report prepared by the National Commission on Supplies and Shortages in 1976 estimated that chlorine plants in the United States required 463 metric tons of mercury each year to make up for what was being lost in the years 1964 to 1973 (Carter, 1977).) The chlor-alkali industry has been working toward preventing the discharge of mercury into aquatic ecosystems, but this pollution has already led to the degradation of many aquatic ecosystems and extirpation of species (Turner and Lindberg, 1978).

The catastrophic effect of the chlor-alkali industry in the North Fork Holston River of southwestern Virginia provides a suitable testing ground for demonstrating the utility of non-invasive shell-based approaches for studying the history of mercury pollution and its impact on local ecosystems. This area is ideal because there has been little independent documentation of the mercury pollution in the North Fork Holston River and the river was historically very diverse with mussel populations (Ortmann, 1918).

Freshwater mussels are important components of freshwater ecosystems. These filter-feeders purify the water and also play a significant role in the aquatic food chain. Mussels are often used as indicators of the health of freshwater systems because they are highly sensitive to increases in sediment load, dissolved oxygen content, impoundments, channelization and pollutants (Williams et al., 1993). These organisms may also be used as indicators of heavy metal pollution by providing a record of the contamination in the shell. Some species of freshwater mussels can live over 100 years and have been shown to document changes in pollutant levels over this long life-span (Nystrom et al., 1996). Freshwater mussel species are disappearing from historically abundant areas at alarming rates due to anthropogenic changes and stresses. For example, it has been estimated that 70% of North American mussels are extinct, endangered, or need special protection (Williams et al., 1993). Not surprisingly, deteriorating freshwater ecosystems, which were once dominated by mussels and other taxa indicative of habitat health, have been a subject of intense study by conservation biologists, ecotoxicologists, and other environmental researchers (e.g., Cherry et al., 1979; Balogh, 1988; Amaral et al., 2000;

Avelar et al., 2000; Costa et al., 2000; Odzak et al., 2000). Surprisingly, given this emphasis on studying mussels, the shell remains of freshwater mollusks have remained largely unexamined despite being a potential source of environmental information.

This project evaluates whether (1) the taphonomic signature of freshwater shells can provide detailed, independent assessments of the severity and timing of extirpations in a polluted system, and whether (2) the geochemical signatures of mercury extracted from shells can provide a separate line of evidence for documenting the contamination history of the polluted river system. If successful, these non-invasive techniques should become a functional research tool. The use of shells as monitoring tools is appealing because species are often so dangerously close to extinction that live sampling is no longer an option. Moreover, these strategies may yield useful information for the undocumented, pre-industrial history of the now polluted habitats. Specifically, the geochemical and taphonomic signatures extracted from shells of freshwater mussels will be used here to verify, and further explore, the mercury contamination and mussel extirpations in the North Fork Holston River. These techniques should yield new insights that are not accessible via routine techniques used in previous studies and may thus be invaluable in working towards understanding and documenting the pollution history of the North Fork Holston River. As importantly, this project may offer a model case example of an approach that can be transferred to other rivers affected by metal contamination.

The specific geochemical objectives of the study are to first analyze the mercury concentrations in shells collected at five locations along the North Fork Holston River. These concentrations will be compared to concentrations in shells collected in 1917, before the industry used mercury in the system. The mercury concentrations will also be compared to the shell length, a proxy for ontogenetic age, and to the total taphonomic grade to investigate the possibility of contamination by post-mortem absorption.

The specific taphonomic objectives of the study are to first determine the taphonomic grade of the shells based on a variety of different taphonomic features. The resulting taphonomic patterns will then be used to assess if such signatures can be reliably used to differentiate sites unaffected by mercury contamination from those sites that may have had their mussel populations wiped out entirely due to heavy mercury

contamination. Potentially confounding factors such as the biasing effects of the stream gradient or variation in relative abundance of species with different shell thickness will also be assessed.

Chapter 2: History of the Area

The Olin-Mathieson Chemical Company utilized natural, underground salt deposits located in Saltville, Virginia to produce chlorine and caustic soda. The plant used a mercury cell in the production process from 1950 until 1972. The plant was permanently closed in 1972 because water standards could not be met cost-effectively (Hill et al., 1974). Two unlined settling ponds that covered 44 hectares (Turner and Lindberg, 1978) and ran 4.16 km along the river were used to settle particulates from the waste slurry containing calcium chloride, sodium chloride, unreacted limestone particles, and mercury (Hill et al., 1974). The ponds drained directly into the river via pipes through the dike (Turner and Lindberg, 1978). By 1957, the elemental mercury and chloride salts led to extirpations of freshwater mussel populations as far as 112 km downstream of Saltville (Young-Morgan & Associates, 1990). The mercury contamination was also evident in fish samples more than 160 km downstream of Saltville by 1977 (Carter, 1977). It has been estimated that as much as 1,814 metric tons of salt and 34 kg of mercury were deposited per day into the plant's settling ponds during the final years of operation (Seivard et al., 1993). It is also estimated that after the closure of the plant, 100g of mercury seeped and eroded from these ponds into the river every day, while 99,773 kg of mercury were found on the grounds where the "cell building" once stood (Carter, 1977).

The North Fork Holston River in southwestern Virginia had an extremely diverse freshwater mussel fauna in the early 1900s. Ortmann (1918) found 42 species of freshwater mussels in the river, including 33 species downstream of Saltville. This originally high diversity is related to a favorable geological setting. In many places, the river flows directly over limestone bedrock, enriching the water with calcium, while numerous sandy pool areas provide habitats rich in nutrients. These conditions provide an ideal setting for nurturing a high diversity of mussel species (Starnes and Bogan, 1988).

The mussel diversity decreased dramatically in the North Fork Holston River following the mercury contamination. In 1998, only nine species of freshwater mussels were found living in the North Fork Holston River (Henley and Neves, 1999). Five of these nine species were only found below river mile 13.5, which is almost 107 km

downstream of Saltville (Henley and Neves, 1999). Transplanting efforts since 1975, which involved the re-introduction of mussels (relocated either from sites upstream of Saltville or from the nearby Clinch River) at multiple sites downstream of Saltville, have resulted in the increase in the number of living populations in the polluted portion of the river (Ahlstedt, 1979; Henley and Neves, 1999). Although Henley and Neves (1999) found that reproducing individuals could be found only at 4 out of 19 downstream sites at which live mussels were reintroduced.

Chapter 3: Materials and Methods

Study Site. The North Fork Holston River flows 216 km through rural southwest Virginia into the South Fork Holston River near Kingsport, Tennessee. Saltville is located about 76.8 km below the river's origin in Smyth County, Virginia (Turner and Lindberg, 1978). This medium-hardwater, high riffle-pool ratio stream has a substrate composed primarily of sand, gravel, and rubble with the shoal areas dominated by boulders (Hill et al., 1974).

Five collection sites were chosen on the North Fork Holston River (Fig. 1). These sites are easily accessible and were used in previous research on mussels of the region (Henley and Neves, 1999). Two sites are located upstream of Saltville at river miles 96 (site 1) and 85 (site 2). Site 1 is approximately 2 miles past Bradford, where Rt. 91 north meets the river at Sagewood Road, behind the Virginia Department of Transportation sign. Site 2 is approximately 50 meters downstream of the river gauging station on Rt. 91 South. Site 3 is located at river mile 79.9 and directly below the Olin-Mathieson settling ponds in Saltville. This site is approximately 0.8 km downstream from the last holding pond at the pullover with a yellow road gate on the left. The two downstream sites are located at river miles 68.6 (site 4) and 56 (site 5). Site 4 is approximately 2.4 km down Rt. 611 after it crosses Hortons Gap Road. This site is at the pullover just after the road becomes dirt. Continue down Rt. 611 until its junction with Rt. 687. Take a right on Rt. Porterfield Hwy and continue for 0.4 km until the next right, Rt. 876. Take the first right on Rt. 802 and Site 5 is at the end of Heinz Island off to the left of the road.

The stream gradient was calculated as a proxy for the energy of each site. The distance between topographic contour lines that encompassed each site was measured on U.S.G.S. topographic maps. The difference between the elevation of the two encompassing contour lines was divided by the distance between them. These calculations produced an estimate of stream gradient for each stretch of the river.

Figure 1. A schematic map of the study area showing the five collecting sites used in this study. The inset map shows the regional location of the study area (modified after Henley & Neves, 1999). The black arrow indicates the approximate position of the mercury contamination source at Saltville.

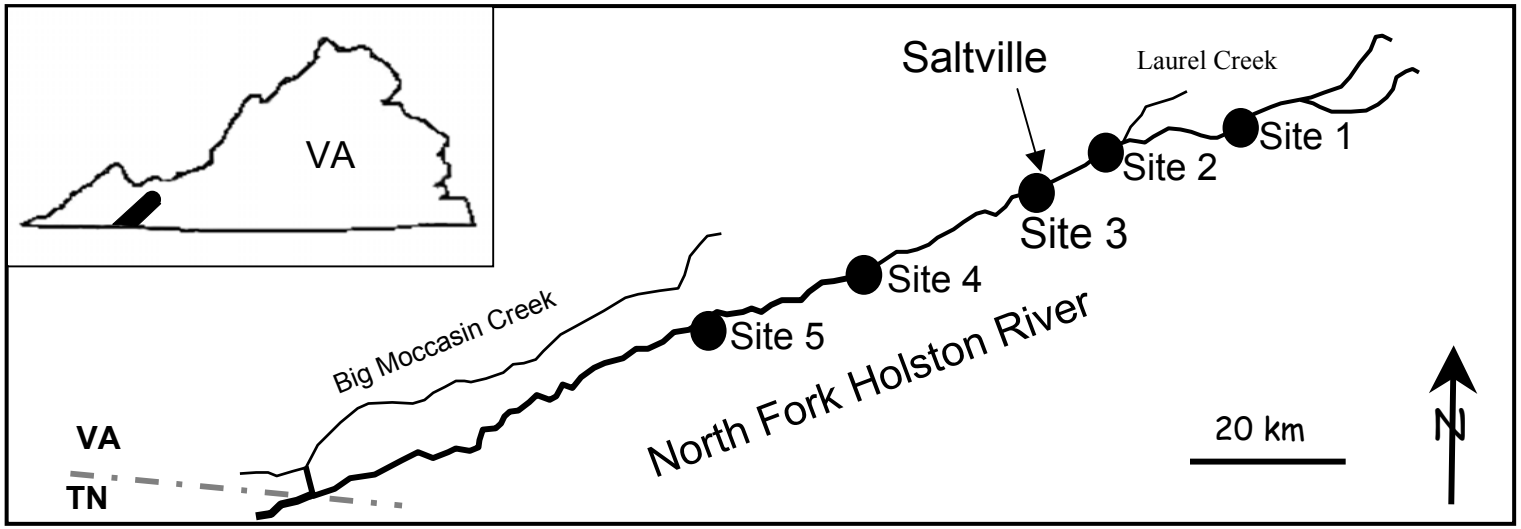
VA
Saltville
Laurel Creek
Site 1
Site 2
Site 3
Site 4
Site 5
Big Moccasin Creek
North Fork Holston River
VA
TN
20 km
N

Figure 1

Sampling of Mussel Shells. Freshwater mussel shells are so scarce in the North Fork Holston River that collection along transects (Henley and Neves, 1999) or in grids was not feasible. Consequently, collecting efforts consisted of systematic, exhaustive surveys with all shells and shell fragments collected by handpicking wherever they were found at the sampled site. The survey concentrated along the banks because the highest concentrations of shells were found in the soft sediment near the shoreline. To ensure comparable sampling intensity at all sites and for all sampling trip, ~45 minutes was spent collecting at each site during each of the three trips to each site. The surficial sampling strategy applied here may bias collecting efforts against larger specimens, which are more frequently found buried in sediments (R. Neves pers. comm., 2004) and against better preserved shells, which may likely be altered more heavily when exposed on the surface. However, because the same procedure was applied at all sites, the data are comparable across sites and should yield estimates that are meaningful in relative comparisons.

After collection, the shells were carefully placed into 35.5 cm X 66 cm plastic sampling bags so as not to cause damage to the shells. Each shell was labeled, and all specimens are housed in the Department of Geosciences at Virginia Tech, Blacksburg, Virginia.

Taphonomic Analysis of Shells. As used here, taphonomic analysis refers to study of post-mortem physical, chemical, and biological alterations of skeletal remains (bones, shells, etc.) left behind by dead organisms. Freshwater mussel shells consist of a high-organic, nacreous aragonitic shell that is poorly preserved in ephemeral and high-energy fluvial systems. Additionally, the weak hinge ligament allows the shells to be easily disarticulated (Cummins, 1994). Also, freshwater shells tend to dissolve easily because, unlike in marine systems, often freshwater is often undersaturated with respect to calcium carbonate (although the dissolution may be negligible in the specific case of North Fork Holston River, which is characterized by waters with pH below 7.0). All collected mussel shells and shell fragments were categorized as right or left valve, whole specimen or shell fragment. The maximum anterior-posterior length of each shell was measured with electronic calipers to the nearest 0.1mm. The genus and species were identified for most of the reasonably complete valves and also for many fragments by

freshwater malacologists, Dr. Richard Neves and Jess Jones (Virginia Tech Department of Fisheries and Wildlife).

A taphonomic scoring system was set up separately for each analyzed taphonomic variable. This taphonomic scale was adopted and modified from standard rank systems developed for marine mollusks (e.g. Kowalewski et al., 1994; Best & Kidwell, 2000; Kidwell et al., 2001; Henderson & Anderson, 2002). All shells and shell fragments were ranked for the following four taphonomic variables: (1) degree of fragmentation, (2) valve edge rounding, (3) shell exterior luster and (4) presence of articulation. For each taphonomic variable a score 0 was assigned for unaltered specimens, and increasingly higher ranks (up to 4 ranks in the case of some types of alterations that allow to distinguish multiple intermediate alteration stages) were assigned to remains displaying an increasing degree of alteration. The total taphonomic grade (TTG) was calculated as the arithmetic sum of individual scores for the articulation, fragmentation, edge preservation, and external luster.

Preparation of Shells for Mercury Analysis. Digital photographs of each specimen were taken prior to the chemical analyses. The shells were then soaked and scrubbed in bleach to remove any extraneous organic material or sediment that may have been attached. Shells with the periostracum were scrubbed with a nylon brush to remove the periostracum. The cleaned shells were allowed to air dry, and then small pieces were broken off the ventral edge. It should be noted that shells are not homogenous and consist of different layers. Consequently, chemical signals extracted from shells may vary notably depending on sample location (e.g., Goodfriend et al., 1997). Thus, all samples were consistently broken from the same area of the shell (i.e., the prismatic and nacreous layers, cleaned of the periostracum, were sampled along the ventral shell edge) to minimize the amount of variability in chemical signatures due to the heterogeneity of the shell. The pieces were placed in narrow mouth Nalgene bottles with two ceramic balls. The bottles were placed inside a ceramic ball-mill to powder the shell. The ceramic balls were cleaned with 12N hydrochloric acid and rinsed with distilled water between uses.

Mercury Analysis of Shells. Divalent metals have been shown to be absorbed into mussel tissue and incorporated into the calcareous shell as metabolic analogues to

calcium (Jeffree et al., 1995; Yap et al., 2003). Thus, the calcareous shell material can act as a potential archive of the metal contamination that occurred during the organism's lifetime (Gundacker, 2000; Markich et al., 2002). It should be noted that the shell chemistry is controlled by a variety of biological factors as well as water chemistry (Vander Putten et al., 2000). This is of little concern to this study because mercury levels resulting from such extensive contamination are expected to be very high relative to the background levels in the environment.

Analyzing the geochemical content of the shell has other advantages over methods targeting the soft tissues. Giusti and Zhang (2002) found that the soft tissue of mollusks is highly sensitive to short-term variations in water chemistry. Consequently, soft tissue primarily records the chemical conditions of the water at the time of sampling. In contrast, shells of mussels are the long-term recorders of the pollution history of an area (Bourgoin, 1990; Jeffree et al., 1995; Amaral et al., 2000; Vander Putten et al., 2000; Giusti and Zhang, 2002; Markich et al., 2002; Yap et al., 2003). For example, Yap et al. (2003) found that the shell of the mussel *Perna viridis* retained elevated concentrations of cadmium, lead, and zinc after exposure to the metals ended, while the tissue concentrations of Cd, Pb and Zn decreased (Fig. 2). Their study provides more evidence that examining shell material rather than tissue provides a long-term record of the pollution (Yap et al., 2003).

Five shells from various species at each site were selected initially to investigate whether mercury could be detected in the shells. The powdered samples were analyzed for mercury content at the Activation Laboratories Ltd. (Canada) by cold vapor FIMS (flow injection mercury system) using a Perkin Elmer Atomic Absorption Spectrometer with detection limits of 5 ppb. Negative values indicated concentrations below the detection limit. Prior to numerical analyses, all the negative values reflecting shells with mercury concentrations below the detection limits (<5 ppb) were given the value of 5 ppb (i.e., the highest possible value). Because most of <5 ppb specimens came from upstream sites, assigning the highest possible value made all analyses and tests more conservative. Repeat samples were tested of several specimens to investigate the variation due to laboratory error. This variation was found to be negligible, typically below the detection limit.

Figure 2. Illustration of changes in metal concentrations in the shell and tissue of mussels during exposure to contaminated water and after removal (depuration) from the contaminated environment (modified after Yap et al., 2003).

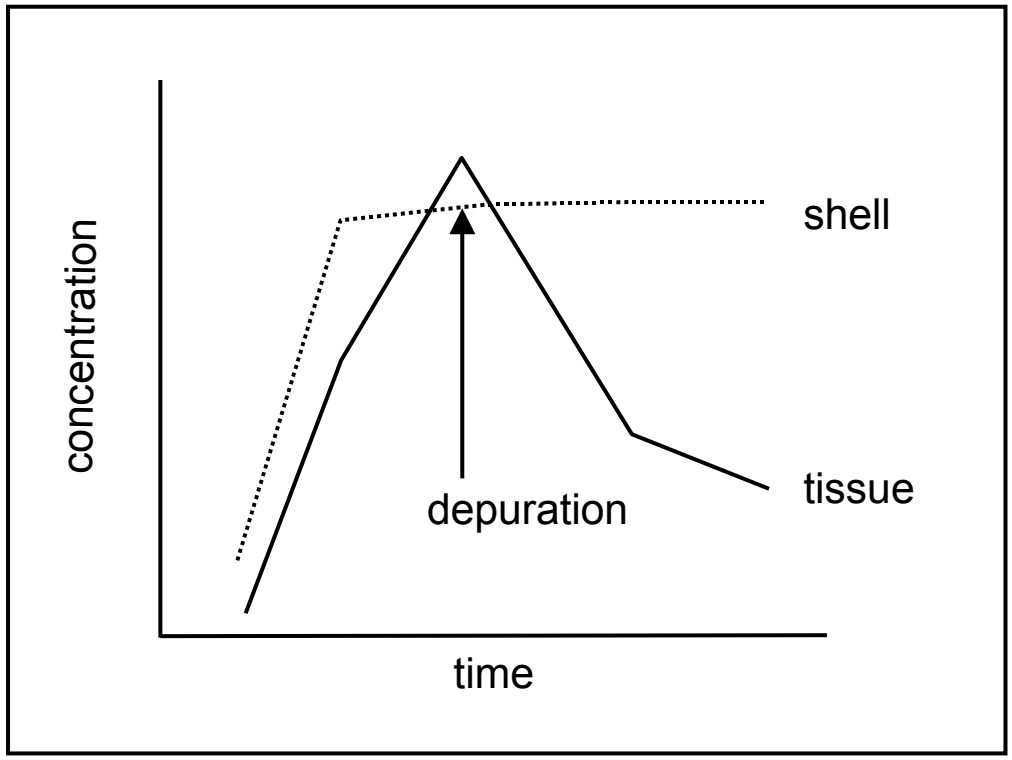

Figure 2

Following the initial analysis, all subsequent mercury analysis focused on one species, *Pluerobema oviforme,* the Tennessee clubshell. This species was selected for the following reasons: (1) it was found at all five sites; (2) it is native to the North Fork Holston River; (3) it has a robust shell that can last for many years after death; (4) it lives for many decades providing a suitable target for sclerochronological analyses in the future; (5) its shells from downstream sites analyzed in the initial analysis showed high levels of mercury; and (6) it occurs in many museum collections that include specimens live-collected in pre-industrial times.

Two shells of *Pluerobema oviforme* that were collected from Saltville in 1917 by C.C. Adams were also analyzed. These shells were obtained from the Carnegie Museum of Natural History to establish background mercury levels prior to the mercury pollution.

Statistical Analyses. Given the scarcity of the shells at some of the sampling sites and the time-consuming sampling processing for mercury analyses, the resulting sample sizes are relatively small. Consequently, in the analyses presented below, data are often pooled by groups of sites (e.g., "upstream" vs. "downstream") to maintain statistically reasonable sample sizes. It is worth stressing that, rather than invalidating the analyses, the pooling of data across sites makes in fact statistical tests more conservative. For example, the addition of specimens from the increasingly distant downstream sites (especially site 5) is expected to lower the estimates of the average mercury shell concentration within the polluted sample group, making it potentially more difficult to distinguish statistically the contaminated vs. uncontaminated sites.

Because of the small sample sizes and because the data include many rank and nominal variables, the statistical analyses presented below were based primarily on non-parametric rank techniques (Wilcoxon 2-Sample Test on medians and Spearman Rank Correlation Test) and standard contingency tests for enumeration data (Log-Likelihood G Ration) (e.g., Zar, 1998). Statistical decisions were based on the significance level of alpha=0.05 in all analyses presented below. Statistical tests were performed using Statistical Analysis Software (SAS Institute, 1989).

Chapter 4: Results

The shells in this study included four genera occurring in notable numbers at all of the five sites (Fig. 3). The sites vary significantly in terms of relative abundance of dominant genera ($G=67.3$; $p<0.0001$; $df=12$; Log-Likelihood Ratio Chi-Square Test). Rare genera were grouped to obtain sufficient sample sizes. Fourteen species were found in the death assemblage at the five sites (Table 1), including (1) Pheasantshell, *Actinonaias pectorosa* (Conrad, 1834); (2) Elktoe, *Alasmidonta marginata* (Say, 1818); (3) Spike, *Elliptio dilatata* (Rafinesque, 1820); (4) Tennessee Pigtoe, *Fusconaia barnesiana* (Lea, 1838); (5) Shiny Pigtoe, *F. cor* (Conrad, 1834); (6) Wavyrayed Lampmussel, *Lampsilis fasciola* (Rafinesque, 1820); (7) Pocketbook, *L. ovata* (Say, 1817); (8) Slabside Pearlymussel, *Lexingtonia dolabellodies* (Lea, 1840); (9) Cumberland Moccasinshell, *Medionidus conradicus* (Lea, 1834); (10) Tennessee Clubshell, *Pluerobema oviforme* (Conrad, 1834); (11) Kidneyshell, *Ptychobranchus fasciolaris* (Rafinesque, 1820); (12) Fluted Kidneyshell, *P. subtentum* (Say, 1825); (13) Rainbow Shell, *Villosa iris* (I. Lea, 1829); and (14) Mountain Creekshell, *V. vanuxemensis* (I. Lea, 1838).

Shell Mercury Concentrations Relative to the Pollution Source. The mercury content of the shells was analyzed by bulk shell analysis to investigate whether mussels incorporate mercury in the calcareous shell. The raw data are summarized in Tables 2 and 3.

The initial mercury analysis, based on specimens from 8 species belonging to 7 distinct genera, shows that shells collected upstream from Saltville have mercury concentration less than 10 ppb (Fig. 4A). In contrast, shells collected directly below the pollution source at Saltville contain substantially elevated mercury concentrations, with multiple specimens exceeding 100 ppb, the highest value of 176 ppb, and the mean value of 31.7 ppb. Shells collected further downstream have mercury concentrations that decrease with increasing distance from Saltville (Fig. 4A). The maximum mercury value at Site 4 is 115 ppb (mean value of 42.25 ppb) and the maximum observed value drops down to 14 ppb at Site 5 (mean value of 8.4 ppb), making it similar to estimates for the most upstream Site 1 (approximately 25.6 km upstream from Saltville).

Figure 3. The taxonomic (genus-level) composition of shell assemblages of freshwater mussels from the North Fork Holston River based on bulk sampling at the five targeted sites (n=266).

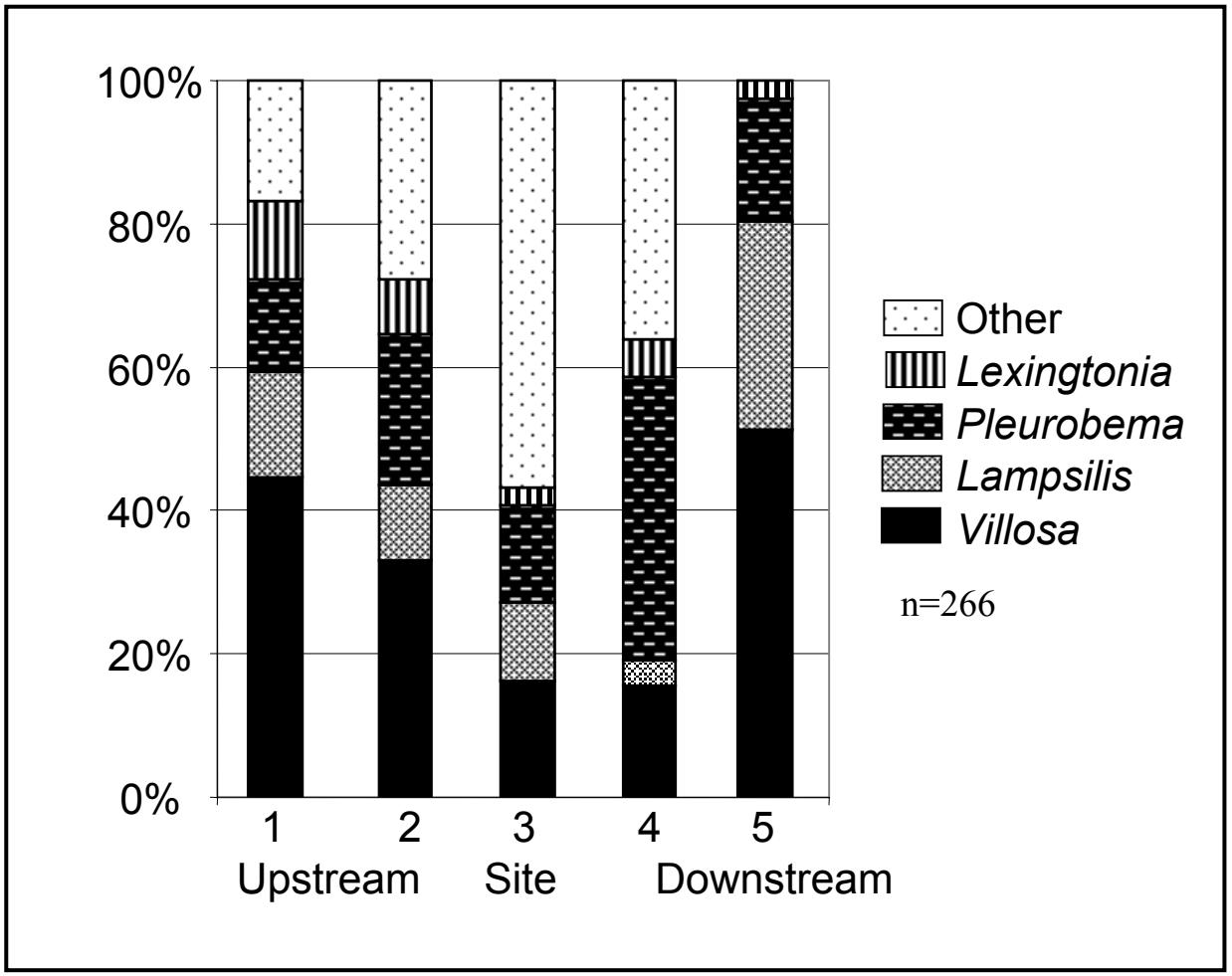
100%
80%
60%
40%
20%
0%
1
2
3
4
5
Upstream
Site
Downstream
Other
Lexingtonia
Pleurobema
Lampsilis
Villosa
n=266

Figure 3

Table 1. The species collected from the death assemblage at each of the five sites along the North Fork Holston River.

Site	Common Name	Genus	Species
1	Tennessee pigtoe	*Fusconaia*	*barnesiana*
	wavyrayed lampmussel	*Lampsilis*	*fasciola*
	slabside pearlymussel	*Lexingtonia*	*dolabelloides*
	cumberland moccasinshell	*Medionidus*	*condradicus*
	Tennessee clubshell	*Pleurobema*	*oviforme*
	fluted kidneyshell	*Ptychobranchus*	*subtentum*
	rainbow shell	*Villosa*	*iris*
	mountain creekshell	*Villosa*	*vanuxemensis*
2	Tennessee pigtoe	*Fusconaia*	*barnesiana*
	shiny pigtoe	*Fusconaia*	*cor*
	wavyrayed lampmussel	*Lampsilis*	*fasciola*
	slabside pearlymussel	*Lexingtonia*	*dolabelloides*
	cumberland moccasinshell	*Medionidus*	*conradicus*
	Tennessee clubshell	*Pleurobema*	*oviforme*
	fluted kidneyshell	*Ptychobranchus*	*subtentum*
	rainbow shell	*Villosa*	*iris*
	mountain creekshell	*Villosa*	*vanuxemensis*
3	pheasantshell	*Actinonaias*	*pectorosa*
	wavyrayed lampmussel	*Lampsilis*	*fasciola*
	pocketbook	*Lampsilis*	*ovata*
	slabside pearlymussel	*Lexingtonia*	*dolabelloides*
	cumberland moccasinshell	*Medionidus*	*conradicus*
	Tennessee clubshell	*Pleurobema*	*oviforme*
	fluted kidneyshell	*Ptychobranchus*	*subtentum*
	kidneyshell	*Ptychobranchus*	*fasciolaris*
	rainbow shell	*Villosa*	*iris*
	mountain creekshell	*Villosa*	*vanuxemensis*
4	pheasantshell	*Actinonaias*	*pectorosa*
	elktoe	*Alasmidonta*	*marginata*
	spike	*Elliptio*	*dilatata*
	shiny pigtoe	*Fusconaia*	*cor*
	Tennessee pigtoe	*Fusconaia*	*barnesiana*
	wavyrayed lampmussel	*Lampsilis*	*fasciola*
	slabside pearlymussel	*Lexingtonia*	*dolabelloides*

Site	Common Name	Genus	Species
4	Tennessee clubshell	*Pleurobema*	*oviforme*
	fluted kidneyshell	*Ptychobranchus*	*subtentum*
	kidneyshell	*Ptychobranchus*	*fasciolaris*
	rainbow shell	*Villosa*	*iris*
	mountain creekshell	*Villosa*	*vanuxemensis*
5	wavyrayed lampmussel	*Lampsilis*	*fasciola*
	slabside pearlymussel	*Lexingtonia*	*dolabelloides*
	Tennessee clubshell	*Pleurobema*	*oviforme*
	rainbow shell	*Villosa*	*iris*
	mountain creekshell	*Villosa*	*vanuxemensis*

Table 2. The mercury content of shells from various species at all five sites. Negative mercury values indicate concentrations below detection limits.

Site	River Mile	Mercury Content (ppb)	Length (mm)	Fragment Length (mm)	Genus	Species	Total Taphonomic Grade
Site 1	96	7	57.22		*Fusconaia*	*barnesiana*	2
Site 1	96	8	73.33		*Lampsilis*	*fasciola*	2
Site 1	96	6	65		*Lexingtonia*	*dolabelloides*	2
Site 1	96	9	54.03		*Lexingtonia*	*dolabelloides*	2
Site 1	96	6	68.39		*Pleurobema*	*oviforme*	2
Site 1	96	6	49.59		*Pleurobema*	*oviforme*	5
Site 1	96	-5	78.03		*Pleurobema*	*oviforme*	4
Site 1	96	-5	68.39		*Pleurobema*	*oviforme*	2
Site 1	96	-5		57	*Ptychobranchus*	*subtentum*	6
Site 2	85	7	42.04		*Fusconaia*	*barnesiana*	4
Site 2	85	9	56.28		*Fusconaia*	*barnesiana*	1
Site 2	85	5	70.04		*Fusconaia*	*barnesiana*	4
Site 2	85	6	66.55		*Pleurobema*	*oviforme*	2
Site 2	85	5	49.27		*Pleurobema*	*oviforme*	1
Site 2	85	-5	46.07		*Pleurobema*	*oviforme*	3
Site 2	85	8	72.25		*Pleurobema*	*oviforme*	3
Site 2	85	31	75.91		*Ptychobranchus*	*subtentum*	5
Site 3	79.9	162		72	*Actinonaias*	*pectorosa*	8
Site 3	79.9	41	73.21		*Lampsilis*	*fasciola*	3
Site 3	79.9	47	55.47		*Pleurobema*	*oviforme*	6
Site 3	79.9	4637	58.9		*Pleurobema*	*oviforme*	4
Site 3	79.9	23	78.27		*Ptychobranchus*	*subtentum*	5
Site 3	79.9	176	41.44		*Villosa*	*iris*	2
Site 4	68.6	25	114		*Actinonaias*	*pectorosa*	5
Site 4	68.6	7	55.82		*Lexingtonia*	*dolabelloides*	5
Site 4	68.6	115		54.19	*Pleurobema*	*oviforme*	6
Site 4	69	52	47.97		*Pleurobema*	*oviforme*	6
Site 4	69	20	45.3		*Pleurobema*	*oviforme*	5
Site 4	69	14	48.57		*Pleurobema*	*oviforme*	6
Site 4	68.6	39	39.09		*Villosa*	*iris*	2
Site 4	68.6	66	50.31		*Villosa*	*vanuxemensis*	1
Site 5	56	10	53.69		*Lampsilis*	*fasciola*	1
Site 5	56	14	63.64		*Lampsilis*	*fasciola*	3
Site 5	56	8	59.81		*Pleurobema*	*oviforme*	3
Site 5	56	6	59.06		*Pleurobema*	*oviforme*	5
Site 5	56	10	59.81		*Pleurobema*	*oviforme*	3
Site 5	56	7	59.06		*Pleurobema*	*oviforme*	5
Site 5	56	-5	55.89		*Pleurobema*	*oviforme*	6
Site 5	56	7	48		*Villosa*	*iris*	2

Table 3. The mercury content of shells collected at Saltville in 1917 by C. C. Adams.

Location	Year Collected	Mercury Content (ppb)	Length (mm)	Genus	Species
Saltville	1917	5	68	*Pleurobema*	*oviforme*
Saltville	1917	6	68	*Pleurobema*	*oviforme*

Figure 4. Results of the mercury analysis; each data point represents an estimate of the mercury content derived from different specimens of dead-collected shells. The hatched area represents expected background levels of mercury in uncontaminated habitats. The background levels are based on the highest concentrations (of 9 ppb) observed at Site 1, which is assumed unaffected by the mercury pollution (located over 20 km upstream of the contamination point). **A.** The initial analysis of specimens from several different genera. **B.** The second analysis (note log-scale y-axis) restricted to the species *Pleurobema oviforme* with the highest mercury values below Saltville two to four orders of magnitude higher than those observed at unaffected Site 1. Specimens collected in 1917, prior to the contamination event, have levels of mercury comparable to background levels observed upstream.

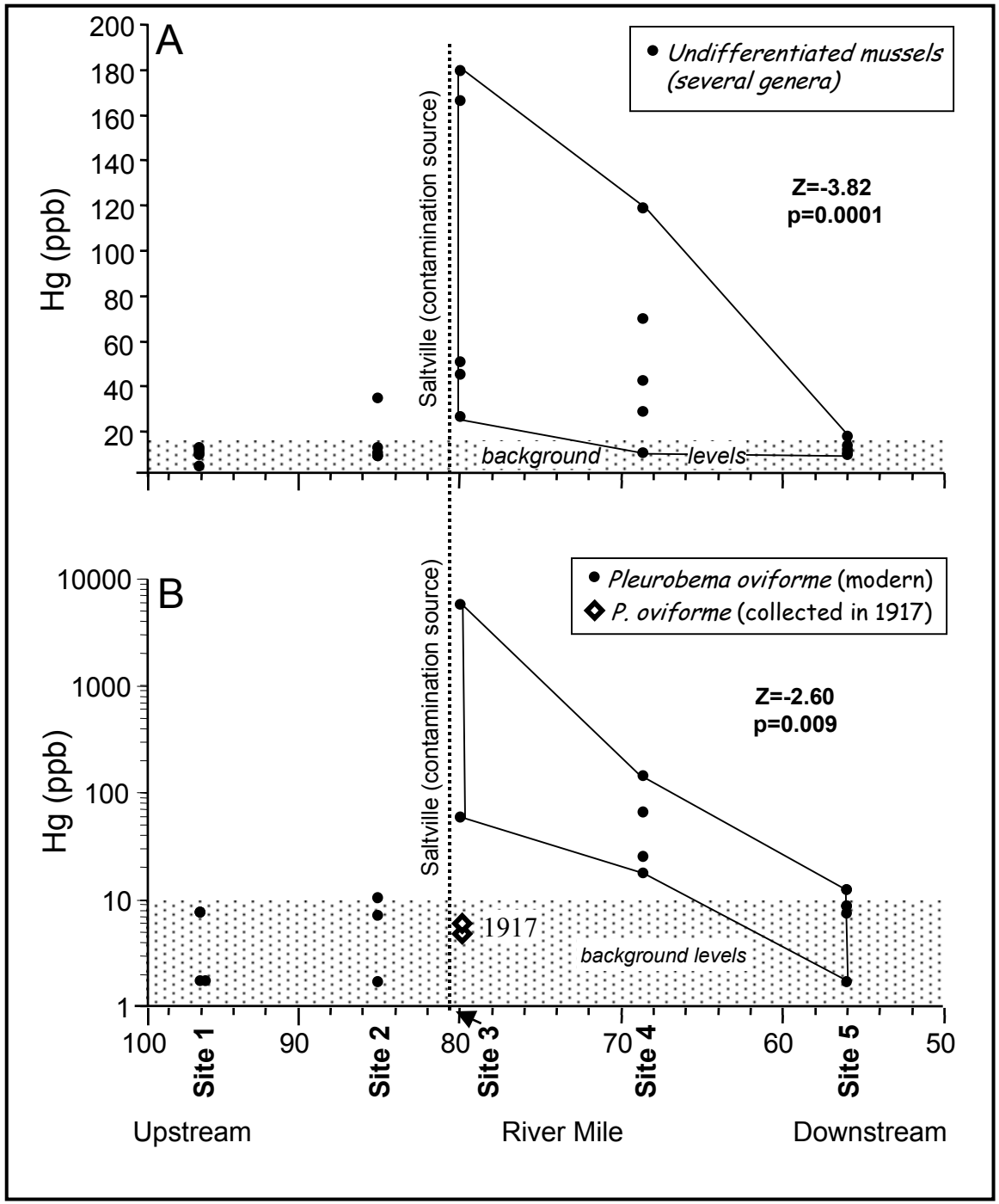
A
Hg (ppb)
200
180
160
140
120
100
80
60
40
20
Saltville (contamination source)
Undifferentiated mussels (several genera)
Z=-3.82
p=0.0001
background levels
B
Hg (ppb)
10000
1000
100
10
1
Saltville (contamination source)
Pleurobema oviforme (modern)
P. oviforme (collected in 1917)
Z=-2.60
p=0.009
1917
background levels
100
90
80
70
60
50
Site 1
Site 2
Site 3
Site 4
Site 5
Upstream
River Mile
Downstream

Figure 4

The mercury analysis of *P. oviforme* shells (Fig. 4B) is consistent with the mixed-genera analysis, but shows an even more dramatic spike in mercury concentration at and downstream of Saltville (note the y-axis is log-transformed). One specimen had mercury levels of 4637 ppb, several hundred times higher than recorded for any shell collected upstream. Also, both analyses suggested slightly elevated mercury levels at Site 2, the closest site above Saltville (Figure 4). As in the case of the mixed genera analysis, the mercury content in *P. oviforme* shells decreased downstream, with increasing distance from the contamination point at Saltville.

The statistical summary of mercury data is provided in Table 4. The mixed shells and *P. oviforme* shells do not differ significantly in their median Hg concentrations (Table 4). The lack of significant statistical differences between the two analyses and the striking similarity of mercury patterns across sites (Fig. 4A vs. 4B) both suggest that it should be acceptable to pool data across taxa (such pooling is needed to obtain sufficient sample sizes for statistical analyses across sites).

The pooled data were first grouped into "Upstream" (Sites 1 and 2) and "Downstream" (Sites 3-5) categories (Table 4). The comparison of these two groups shows that the shells collected from upstream sites had significantly lower mercury concentrations than the shells collected from the downstream sites (Table 4). Because the mercury levels at Site 5 are comparable to what is considered background level, the analysis was repeated with Site 5 excluded from "Downstream" sites. In that case, the resulting difference in mercury concentration becomes even more dramatic (Table 4). When the data are restricted to *P. oviforme* shells only, the significant pattern observed for pooled data persists; upstream sites had significantly lower mercury concentrations than the shells collected from downstream sites (Table 4).

The two shells collected from Saltville in 1917 show very low levels of mercury (6 ppb or less), comparable to the low levels observed at Site 1 (Table 3 and Fig. 4B). Because only two specimens were available for analysis, statistical tests cannot be applied to evaluate rigorously if these values are significantly lower than those observed in shells from the same site collected in post-industrial times. Nevertheless, it is remarkable that none of the post-industrial shells from Saltville have values lower than

Table 4. Summary of geochemical results for mercury analysis in mussel shells collected from the North Fork Holston River.

Sample groups	Sample size	Mercury concentrations in shells (ppb)						Wilcoxon 2-sample test	
		Min	Max	Mean	Median	Range	Standard deviation	Z	p
All data pooled	39*	5	4637	144.2	8	4632	739.4		
Mixed genera	21	5	176	31.7	9	171	48.3	-1.34	0.18
P. oviforme	18	5	4637	275.5	6.5	4632	1089		
Upstream Sites (1-2)	17	5	31	7.8	6	26	6.1	-3.83	0.0001
Downstream Sites (3-5)	22	5	4637	249.6	21.5	4632	981.2		
Upstream Sites (1-2)	17	5	31	7.8	6	26	6.1	4.33	<0.0001
Downstream Sites (3-4)	14	7	4637	387.4	44	4630	1224		
Site 1	9	5	9	6.3	6	4	1.4		
Site 2	8	5	31	9.5	6.5	26	8.8		
Site 3	6	23	4637	847.7	104.5	4614	1858	24**	<0.0001
Site 4	8	7	115	42.25	32	108	35.5		
Site 5	8	5	14	8.4	7.5	9	2.9		
Upstream Sites (1-2)	8***	5	8	5.75	5.5	3	1.0		
Downstream Sites (3-4)	10***	5	4637	491	17	4632	1457	-2.7	0.007

* – excludes two specimens collected in 1917

** – Chi-square parameter value for non-parametric Anova (Kruskal-Wallis test)

***– includes only *P. oviforme* data

23 ppb and most have mercury concentrations (ranging from 23 to 4637 ppb) 2 to 3 orders of magnitude higher than either of the two shells collected in pre-industrial times.

The mercury content also was compared to the length of the shell to determine whether mercury content was related to shell size (Fig. 5). No significant positive correlation was found between mercury content and shell length when including all genera, using either a rank test on raw data (r=-0.08; p=0.67; n=32; Spearman Rank Correlation Test) or a parametric test on log-transformed data (r=-0.01; p=0.93; n=32; Pearson Correlation Test). When data are restricted to *P. oviforme* data, and thus shell length is a somewhat more reasonable proxy for ontogenetic age of specimens than for data including various genera, there was also no correlation between shell length and mercury concentration (Fig. 6) (r=-0.3; p=0.2; n=19; Spearman Rank Correlation Test; r=-0.2; p=0.41; n=19; Pearson Correlation Test).

Taphonomic Signatures Relate Extirpation Patterns to the Pollution Source. The shells with the highest total taphonomic grade, i.e., those that are the most heavily altered and fragmented when all taphonomic variables are combined, dominate in areas directly downstream from the contamination point, at Site 3 (Fig. 7 and Fig. 8A). In contrast, upstream sites, unaffected directly by the mercury contamination, contain many fresh-dead shells with lowest total taphonomic grades. Site 4 has specimens that vary in the total taphonomic grade, many still exhibiting a higher degree of alteration. This total taphonomic grade decreases from Site 4 to Site 5, the farthest site downstream (Fig. 8A). Site 5 had similar numbers of shells with high and low degrees of total taphonomic alteration. This site is comparable to Sites 1 and 2 upstream. The shells collected from downstream sites are significantly more taphonomically altered than the shells collected from upstream sites (G=27.8; p=0.0005; df=8; Log-Likelihood Ratio Chi-Square Test). The median taphonomic grade is also significantly higher at sites downstream from Saltville when compared to upstream sites (Z=-3.66; p=0.0003; n=366; Wilcoxon Two-Sample Test with Normal Approximation).

The same pattern is also seen when each taphonomic variable is examined separately (Table 5 and Figure 8B). The taphonomic grade is low, between 0.5 and 1 for articulation and fragmentation and between 1.4 and 1.6 for edge preservation and external

Figure 5. Comparison of estimated shell mercury content and length of shells collected from the five targeted sites along the North Fork Holston River. Each data point represents a single specimen from various genera.

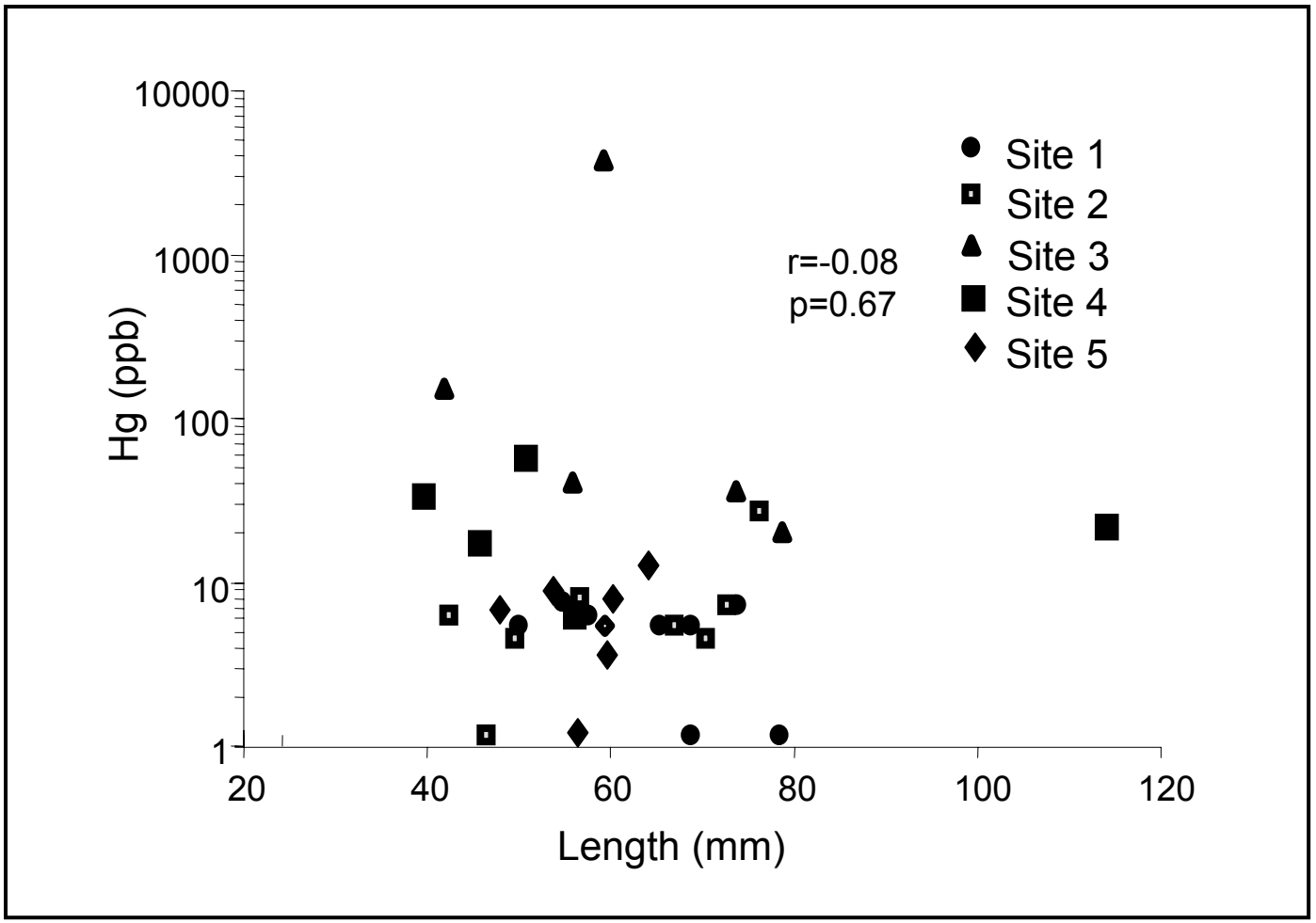
10000
1000
100
10
1
Hg (ppb)
20
40
60
80
100
120
Length (mm)
r=-0.08
p=0.67
Site 1
Site 2
Site 3
Site 4
Site 5

Figure 5

Figure 6. Comparison of estimated shell mercury content and length of *P. oviforme* collected from the five targeted sites along the North Fork Holston River. Each data point represents a single specimen.

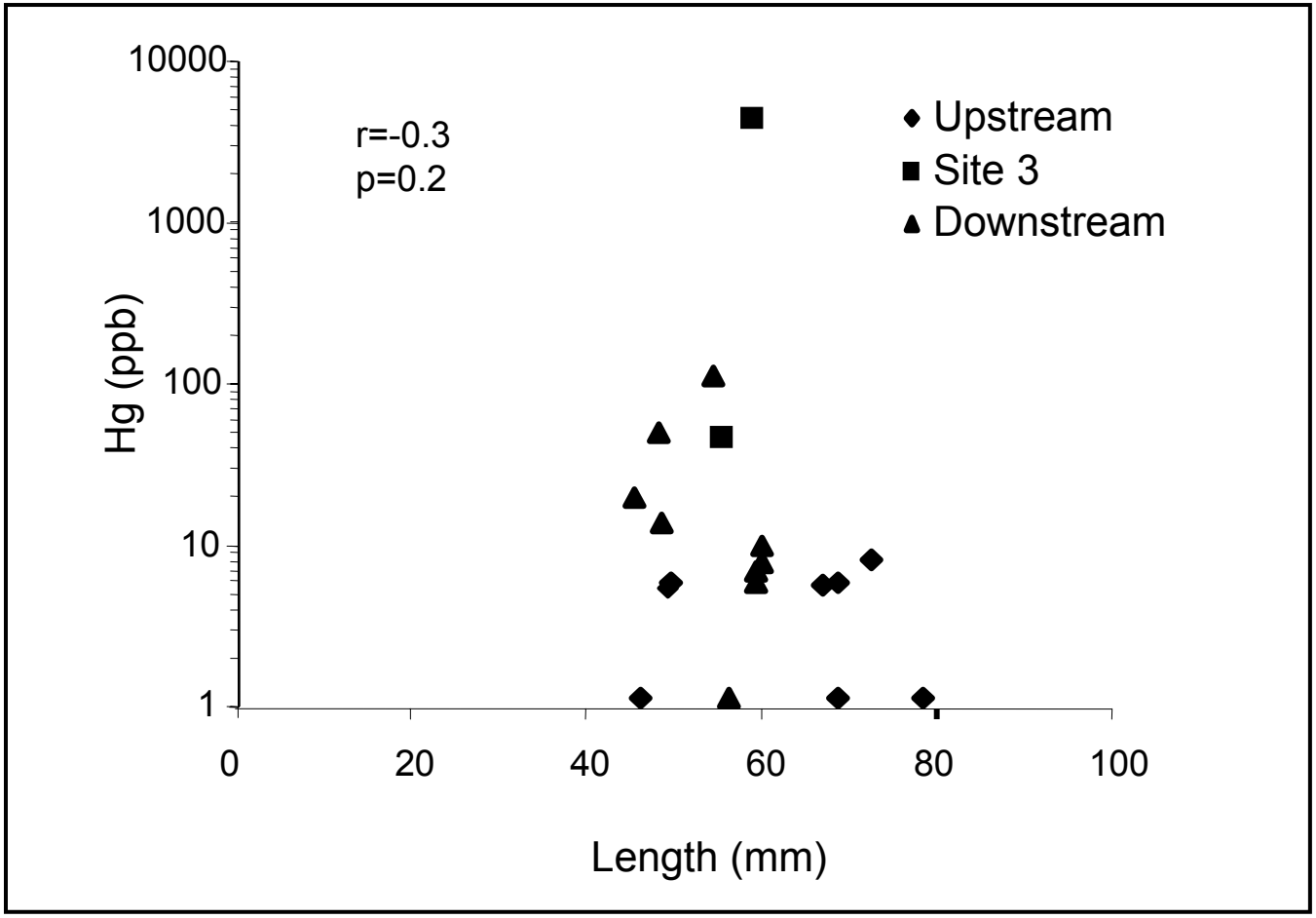
10000
1000
100
10
1
Hg (ppb)
r=-0.3
p=0.2
Upstream
Site 3
Downstream
0
20
40
60
80
100
Length (mm)

Figure 6

Figure 7. Taphonomic grade (TTG) of shells (i.e., the extent of physico-chemical shell alteration) along the North Fork Holston River, as observed in dead-collected shells from the five targeted sites, including sites upstream and downstream of the contamination.

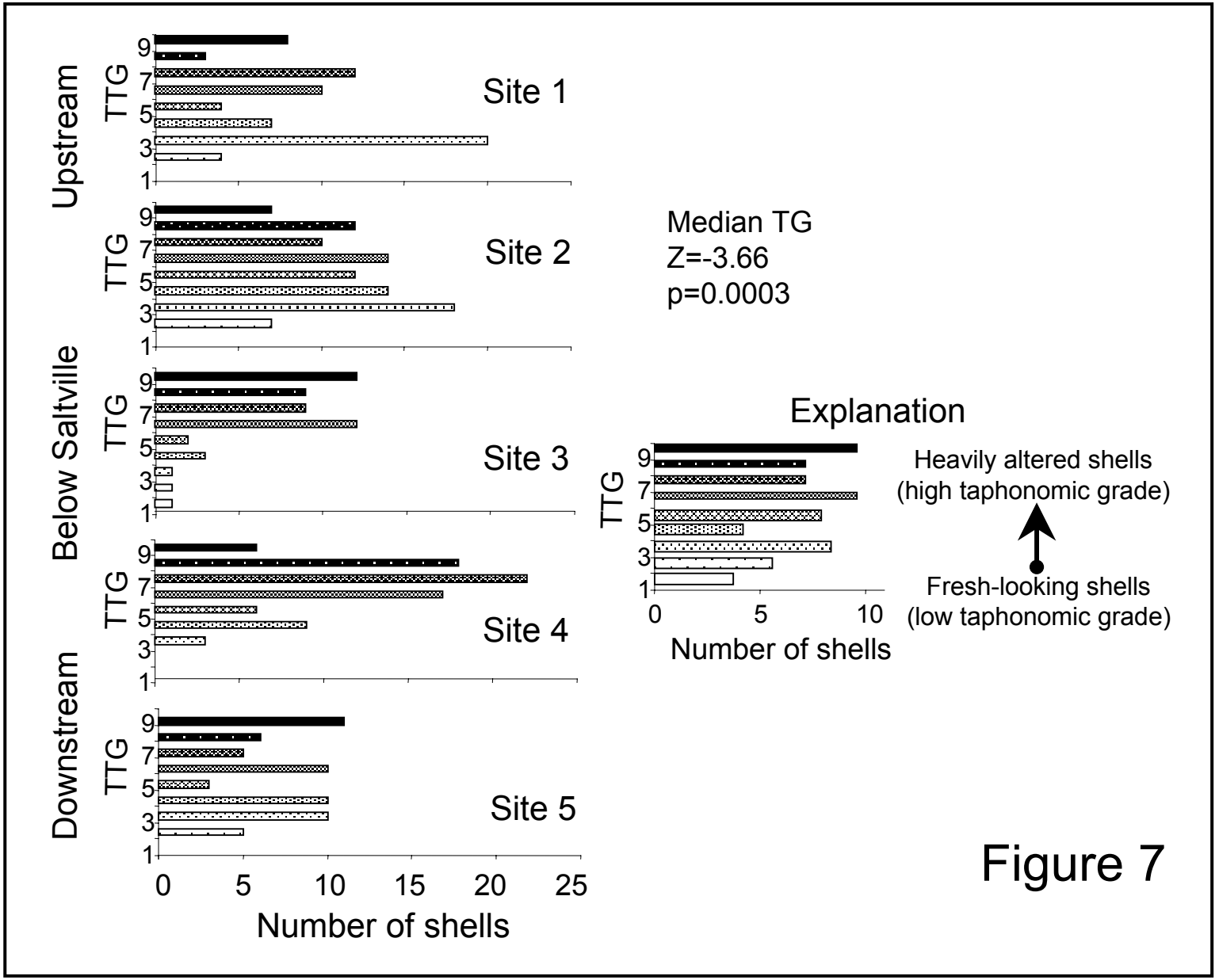
Upstream
Below Saltville
Downstream
TTG
Site 1
Site 2
Site 3
Site 4
Site 5
9
7
5
3
1
0
5
10
15
20
25
Number of shells
Median TG
Z=-3.66
p=0.0003
Explanation
Heavily altered shells
(high taphonomic grade)
Fresh-looking shells
(low taphonomic grade)

Figure 7

Figure 8. Changes in the overall taphonomic grade of shell assemblages (i.e., the extent of physico-chemical shell alteration) along the North Fork Holston River as observed in dead-collected shells from the five targeted sites. **A.** The change in the grade of taphonomic variables combined (the total taphonomic grade) as observed in dead-collected shells from five targeted sites. **B.** The change in average grade of each taphonomic variable graphed separately, as observed in dead-collected shells from the five targeted sites.

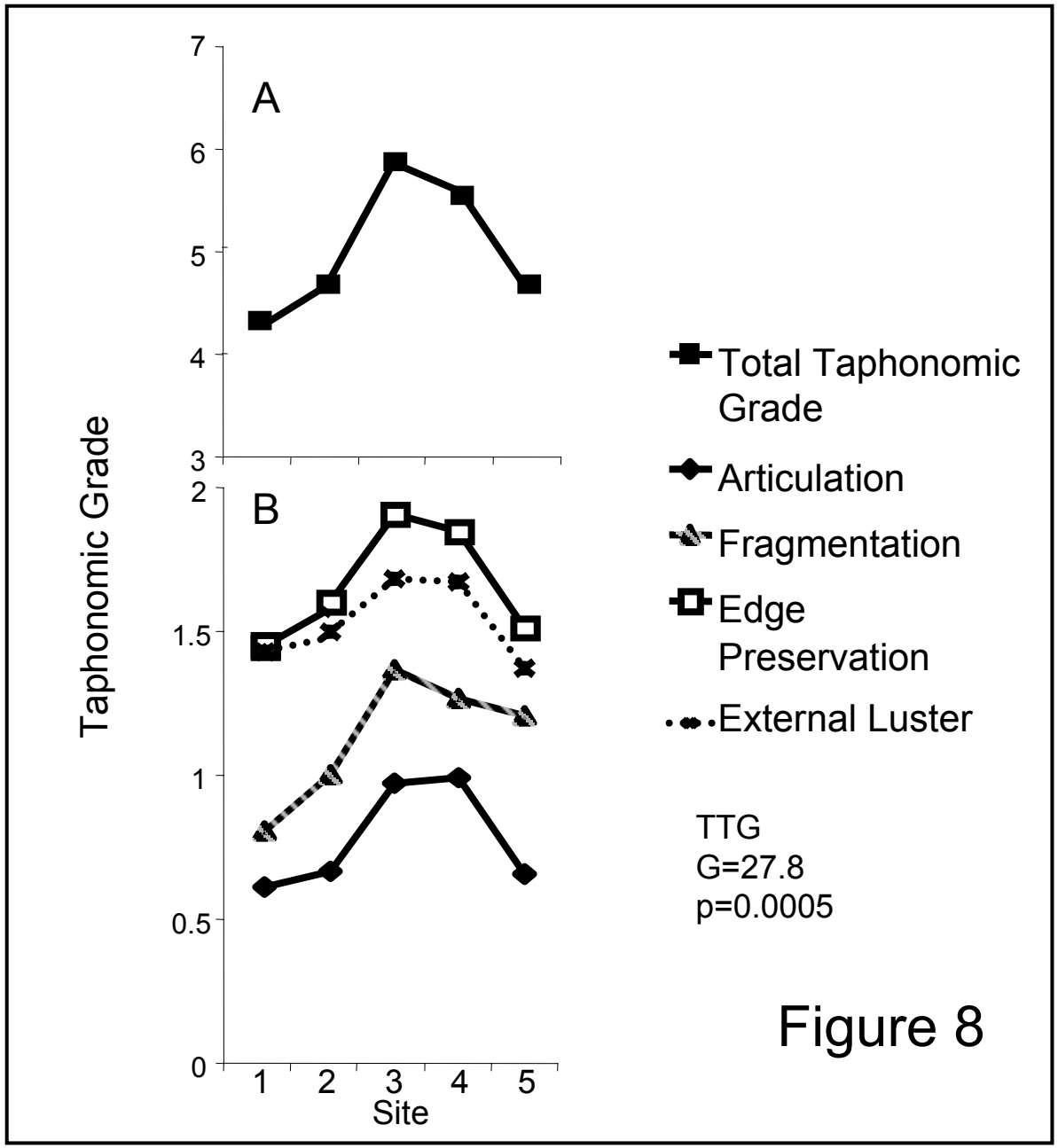
A
B
Taphonomic Grade
Site
7
6
5
4
3
2
1.5
1
0.5
0
1
2
3
4
5
Total Taphonomic Grade
Articulation
Fragmentation
Edge Preservation
External Luster
TTG
G=27.8
p=0.0005

Figure 8

Table 5. The summary of taphonomic data grouped by site and type of shell morphology.

Site	Number of shells	Articulation			Fragmentation			Edge preservation			External luster			Total taphonomic grade		
		mean	Median	Std. dev.	mean	Median	Std. dev.	mean	Median	Std. dev.	mean	Median	Std. dev.	mean	Median	Std. dev.
All data pooled																
1	68	0.59	1	0.50	0.78	0	1.05	1.43	1	0.61	1.44	1	0.50	4.24	4	2.23
2	104	0.64	1	0.48	0.97	0	1.17	1.56	2	0.65	1.47	1	0.54	4.64	5	2.29
3	53	0.94	1	0.23	1.36	1	1.26	1.87	2	0.39	1.66	2	0.59	5.83	6	1.92
4	81	0.96	1	0.19	1.12	1	0.98	1.80	2	0.46	1.64	2	0.48	5.53	6	1.56
5	60	0.63	1	0.49	1.17	1	1.25	1.47	1.5	0.57	1.35	1	0.63	4.62	5	2.37
Thick shells only																
1	30	0.53	1	0.51	0.70	0	0.99	1.57	2	0.50	1.50	1.5	0.51	4.30	5	2.17
2	60	0.68	1	0.47	0.92	0.5	1.11	1.68	2	0.60	1.55	2	0.53	4.83	5	2.10
3	30	0.97	1	0.18	1.07	1	1.14	1.97	2	0.18	1.73	2	0.45	5.73	6	1.34
4	61	0.95	1	0.22	1.07	1	0.87	1.88	2	0.32	1.75	2	0.43	5.66	6	1.39
5	20	0.40	0	0.50	0.60	0.5	0.68	1.40	1	0.60	1.20	1	0.62	3.60	3	1.67
Thin shells only																
1	31	0.58	1	0.50	0.58	0	0.85	1.20	1	0.65	1.29	1	0.46	3.64	3	1.96
2	34	0.47	0	0.51	0.62	0	1.04	1.20	1	0.69	1.20	1	0.48	3.50	3	2.15
3	16	0.88	1	0.34	1.31	1	1.35	1.62	2	0.62	1.38	2	0.81	5.19	5.5	2.66
4	12	1.00	1	0.00	0.58	0	0.90	1.25	1	0.75	1.17	1	0.39	4.00	3.5	1.60
5	21	0.57	1	0.51	0.52	0	0.98	1.10	1	0.44	0.95	1	0.50	3.14	3	1.68

luster at the upstream sites. The grades increase to the maximum grades for fragmentation and edge preservation at Site 3, while articulation and external luster have slightly higher taphonomic grades at Site 4. All of the taphonomic grades decrease downstream at Site 5. For each of those four taphonomic variables, the observed differences between the "Upstream" and "Downstream" sites are significant statistically (Table 6).

It should also be noted that thick-shelled and thin-shelled species vary in relative abundance among sites (Table 5 and Figure 9A). This variation is statistically significant ($G=23.3$; $p=0.0001$; $df=4$; Log-Likelihood Ratio Chi-Square Test). The thick-shelled and thin-shelled species also have different taphonomic signatures (Table 5 and Figure 9B), with thin-shelled taxa exhibiting significantly lower median taphonomic grades ($Z=5.15$, $p<0.0001$, thick: $n=201$, median=5, thin: $n=114$, median=3; Wilcoxon 2-sample Median Test). Yet, both thick-shelled and thin-shelled species reveal the same taphonomic trend of the highest taphonomic grade found at Site 3 and lower taphonomic grades upstream and downstream of the contamination point (Table 5 and Figure 9B). For the thick shells, this trend was shown to be statistically significant: thick shells collected from downstream sites are significantly more taphonomically altered than the thick shells collected from upstream sites ($G=19.6$; $p=0.007$; $df=7$; Log-Likelihood Ratio Chi Square Test). The taphonomic grade of thin shells was not compared between upstream and downstream sites because there were too few thin-shelled specimens in each taphonomic category to perform a contingency test.

The taphonomic pattern does not appear to relate with the stream gradient (Figure 9B). Sites 1 and 2 represent the highest-energy (steepest-gradient) hydrodynamic regimes, but contain the shells with a low degree of taphonomic alteration. Sites 3 and 4 have much lower stream gradients, yet contain shells with higher taphonomic grades than Sites 1 and 2. Although data consisting of 5 observations do not allow for a rigorous statistical treatment, it should be noted that the observed variation in stream gradient among the five studied sites is very minor (from the steepest being 1.84 m/km to the most gradual being 1.23 m/km), so it seems unlikely that these sites are exposed to notably different hydrological regimes.

Table 6. Summary of statistical tests for differences in taphonomic grades across sites. Site 5 was excluded from the analyses reported here. The analyses with Site 5 included are similar in most cases.

Compared data and variables	Downstream sites (3 and 4)		Upstream sites (1 and 2)		Wilcoxon 2-sample median test	
	n	median	n	median	|Z|	P
Articulation						
All**	172	1	134	1	6.84	<0.0001
Thick**	90	1	91	1	5.36	<0.0001
Thin**	65	1	28	1	3.72	0.0002
Fragmentation						
All**	172	0	134	1	2.96	0.003
Thick*	90	0	91	1	2.03	0.04
Thin	65	0	28	0.5	1.46	0.14
Edge preservation						
All**	172	2	134	2	5.08	<0.0001
Thick**	90	2	91	2	3.80	0.0001
Thin	65	1	28	2	1.85	0.06
External Luster						
All**	172	1	134	2	3.34	0.0008
Thick**	90	2	91	2	2.88	0.004
Thin	65	1	28	1	0.56	0.58
Total taphonomic grade						
All**	172	4.5	134	6	4.58	<0.0001
Thick**	90	5	91	6	3.40	0.0007
Thin**	65	3	28	5	2.30	0.02

* tests significant at alpha=0.05 level;
** tests significant at alpha=0.005 level.

Figure 9. Changes in the overall taphonomic grade of shell assemblages (i.e., the extent of physico-chemical shell alteration) along the North Fork Holston River as observed in dead-collected shells from the five targeted sites. **A.** The composition of dead shell assemblages across the five targeted sites with shells grouped into thick (more robust valves) or thin-shelled (less durable valves) species of freshwater mussels. **B.** The results are plotted separately for thick and thin-shelled species of freshwater mussels. The taphonomic grade is compared to the stream gradient at each site (a proxy for hydrodynamic regime of each site) estimated from topographic maps.

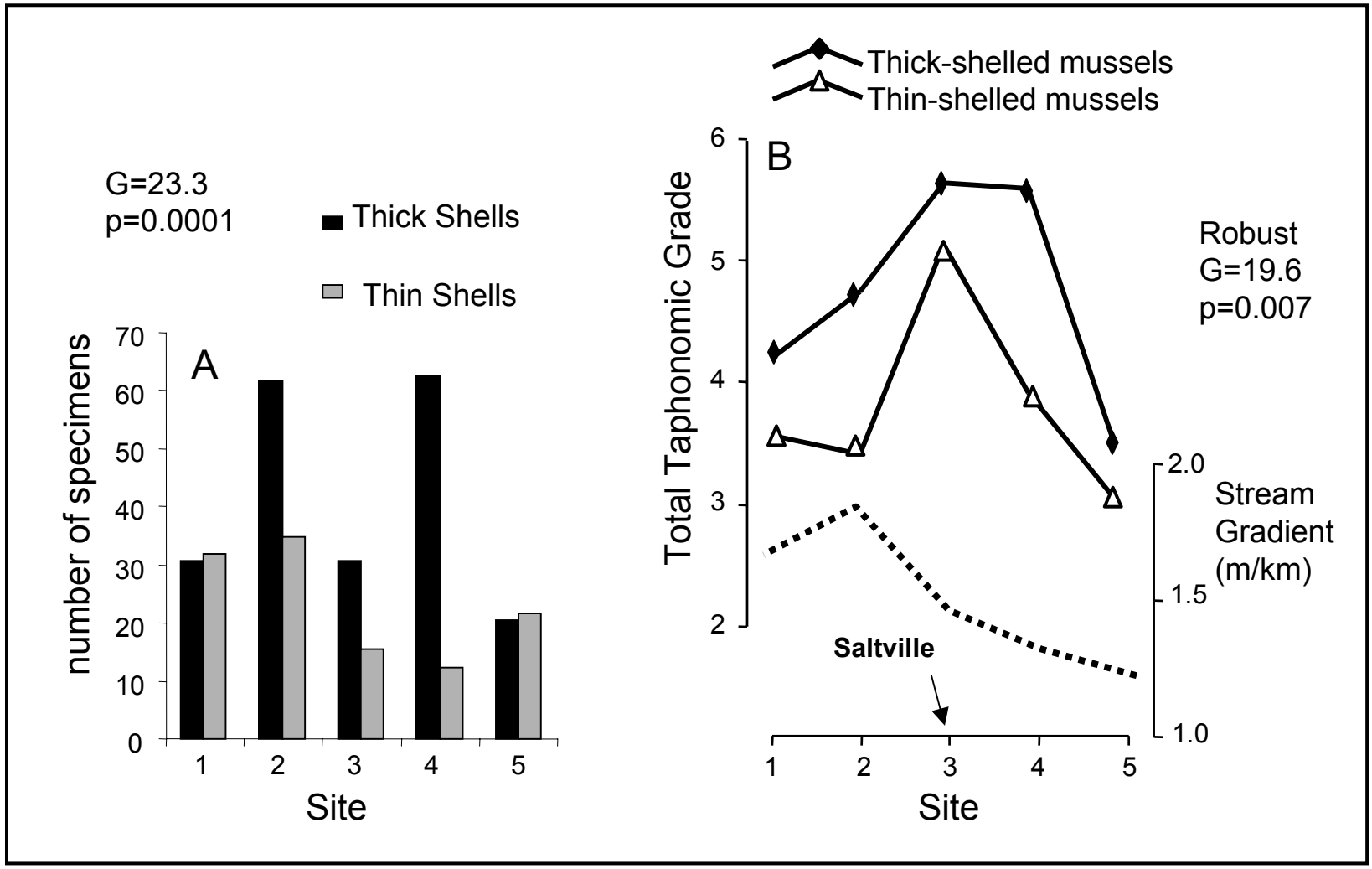
G=23.3
p=0.0001
Thick Shells
Thin Shells
A
number of specimens
70
60
50
40
30
20
10
0
1
2
3
4
5
Site
Thick-shelled mussels
Thin-shelled mussels
B
Total Taphonomic Grade
6
5
4
3
2
Robust
G=19.6
p=0.007
2.0
1.5
1.0
Stream
Gradient
(m/km)
Saltville
1
2
3
4
5
Site

Figure 9

The mercury content was compared to the total taphonomic grade to determine whether mercury was incorporated through post-mortem absorption (Fig. 10). Mercury levels would be higher in shells that have been in the river longer, subject to taphonomic processes for a long period of time, with higher taphonomic alteration if mercury was absorbed post-mortem. No correlation was found between the total taphonomic grade of a shell and its mercury concentration ($r=0.18$; $p=0.28$; $n=37$; Spearman Rank Correlation Test).

Figure 10. Comparison of the overall taphonomic state of the dead-collected shells (i.e., the extent of physico-chemical shell alteration) and the mercury content of shells collected from the five targeted sites along the North Fork Holston River. Each data point represents a single specimen.

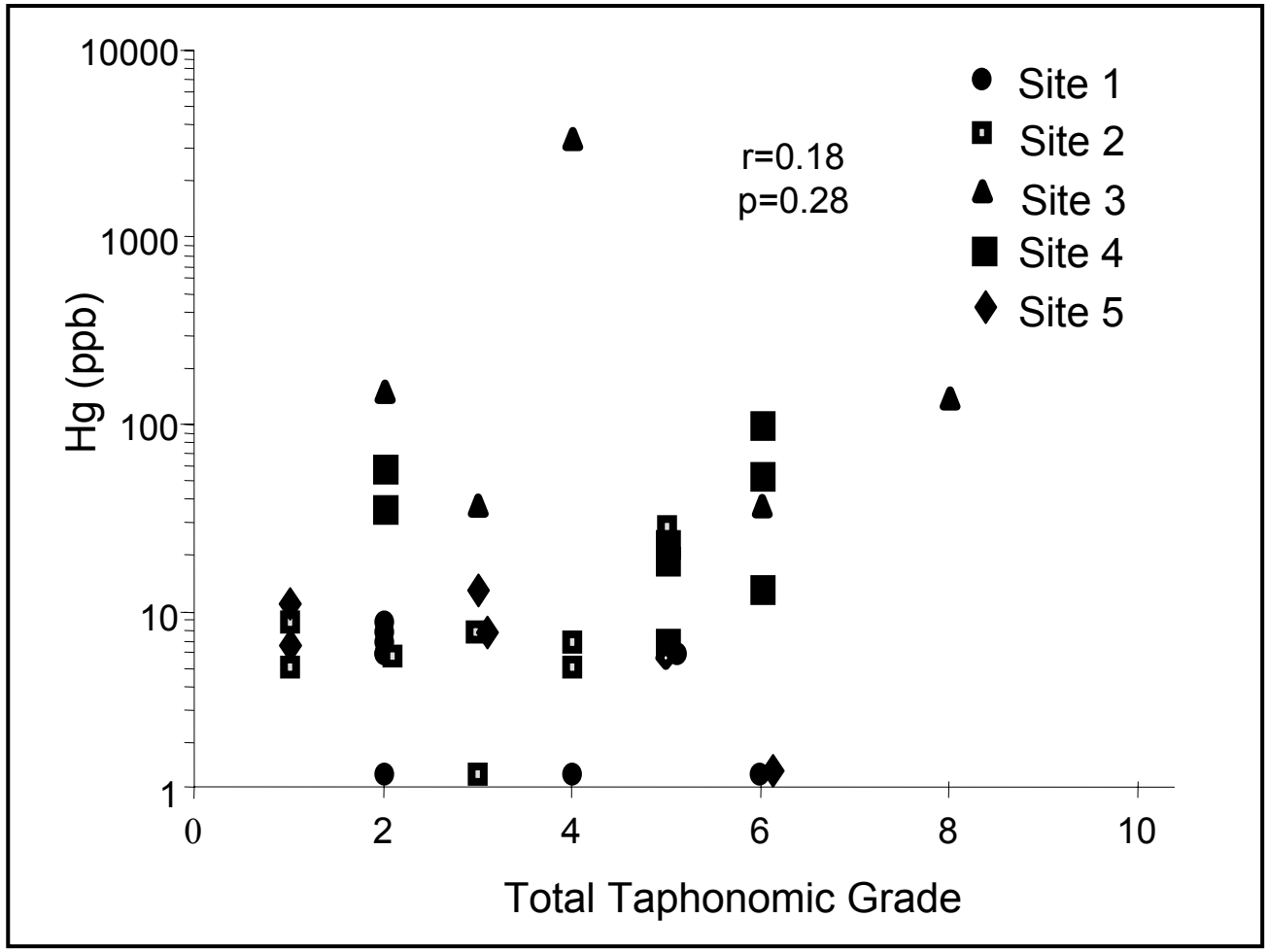
10000
1000
100
10
1
Hg (ppb)
r=0.18
p=0.28
Site 1
Site 2
Site 3
Site 4
Site 5
0
2
4
6
8
10
Total Taphonomic Grade

Figure 10

Chapter 5: Discussion

Shell Mercury Concentrations Relative to the Pollution Source. The geochemical analysis, based on empty shells of dead mussels, provided a record of the mercury contamination in the North Fork Holston River. The analyses showed that shells collected upstream from the pollution source, in areas unaffected by the contamination at Saltville, have invariably low mercury concentrations. The slightly elevated mercury levels at Site 2 provide tentative evidence supporting anecdotal reports of undocumented dumping sites upstream of Saltville (D. Cherry pers. comm., 2004). The shells collected directly below the pollution point contain statistically significantly higher mercury concentrations, reflecting their close downstream proximity to the plant. Shells collected further downstream have notable mercury concentrations that decrease with increasing distance from Saltville, which may be signatures of downstream dilution of the mercury that was discharging from the plant.

Results of this study corroborate recent research showing that freshwater mussel shells can provide a record of metal pollution. Markich et al. (2002) used the freshwater bivalve shell, *Velesunio angasi*, as an indicator of trace metal levels in a stream affected by acid mine drainage. In the Markich study, the whole shell was analyzed as a bulk sample and the annual laminations of the shell were analyzed using secondary ion mass spectrometry (SIMS). Similar to my study, Markich et al. (2002) found that copper, manganese, zinc, uranium, nickel, cobalt, and lead concentrations in the whole shell decreased with increasing distance from the pollution source. They also observed that there were no significant differences in the metal concentrations over the lifetime of the animals when examining annual laminations. Both studies illustrate that bivalve shells can be successfully used to document metal pollution.

The low levels of mercury in pre-industrial shells, collected from Saltville confirm that elevated levels of mercury were not present in the Saltville area in 1917. This result suggests that the high levels of mercury in shells collected recently at Site 3 do not record some natural (geological) sources of mercury eroded by the river over multiple centuries, but rather, reflect relatively recent (post 1917) anthropogenic release of mercury to the North Fork Holston river at and below Saltville.

This study sheds new light on the factors that affect incorporation of metals in mollusk shells, as debated in recent literature on shell geochemistry. First, metal incorporation is commonly reported to be related to the ontogenetic stage and physiological condition of the individual (Amaral et al., 2000). Markich et al. (2002) state that there is always inherent variability in metal concentrations due to the size, ontogenetic age and gender of the mussel. Yet, Wiesner et al. (2001) analyzed lead and cadmium levels in the shell of three different size classes of mussels and found no correlation between size and metal content. Wiesner's study corroborates claims of Thomas and Bendell-Young (1998) and Yap et al. (2003), who both showed that the variability in shell metal content is less variable than variability in the tissue because the incorporation of metals is not affected by physiological condition. When looking at all species studied, this study also found that larger, longer shells do not exhibit higher mercury concentrations, even when compared to other shells collected from the same site. Moreover, no correlation was seen when the data were restricted to a single species (*P. oviforme*), even when compared to other shells collected from the same site. Specifically, the results presented here suggest that mercury concentrations found in parts of shells secreted later in the ontogenetic age of individuals (ventral edge of large specimens) does not differ from mercury concentrations incorporated in more juvenile sectors of the shell (ventral edge of small specimens).

Taphonomic Signatures Relate Extirpation Patterns to the Pollution Source. This study shows that the taphonomic signatures of dead shells are a good proxy for distinguishing sites with active (reproducing) mussel populations from sites affected by local extirpation events. That is, upstream areas that have had a continuous input of fresh-dead material exhibited younger taphonomic signatures. The site directly below the contamination (Site 3), with no input of fresh-dead material from extant populations, exhibited the oldest taphonomic signature. This site has been devoid of viable populations for at least 30 years, and thus, is likely to be dominated by old specimens that have been altered by the prolonged action of various taphonomic factors operating in fluvial systems. The areas downstream, which have been partly re-colonized following the initial extirpation event, exhibited an intermediate taphonomic signature. The intermediate degree of alteration may reflect fresh-dead shell input from mussels re-

introduced in restoration efforts. In sum, the taphonomic index of shells proved to be a reliable predictor for distinguishing affected and unaffected sites along the North Fork Holston River. This approach may, therefore, be a useful tool for identifying freshwater communities with unknown extirpation histories, especially for aquatic systems, which are similar to the study area in terms of their faunal composition, climate, and local geomorphology (e.g., other comparably small rivers of the region). In addition, the spatial trends in taphonomic signatures across the five sampled sites are consistent with mercury analysis (Fig. 11) and provide additional, albeit more indirect evidence suggesting that the extirpation patterns relate to the pollution source in Saltville.

The outcome of this research is consistent with a prior study by Cummins (1994) who found a good agreement between the composition of death shell assemblages and life mussel populations and concluded that fresh-dead shells are continuously contributed to the death assemblage in habitats with active, reproducing populations. However, following a local extirpation event, shell input is either arrested or, at best, limited to few transport-battered shells (if more pristine sites still exist upstream). Consequently, shell taphonomy is expected to provide a metric for estimating the location and severity of extirpation events.

Transport of shells from upstream in the river is not a factor that could bias the taphonomic signatures of these death assemblages with different extirpation histories. Fresh-dead shell material may be washing from extant population to extirpated areas, but these shells will be physically altered in transport and the extirpated areas will still have death assemblages with the highest taphonomic alteration. Death assemblages in areas with extant populations downstream of extirpated areas will still exhibit a lower degree of taphonomic alteration as a result of the input of fresh-dead material.

The lack of relation between the stream gradient and taphonomic signatures observed here suggests that the extirpation history plays a primary role in controlling the taphonomic signatures of mussel shells in the North Fork Holston River. This pattern may not necessarily be valid for rivers with more variable stream gradients.

The differences in taphonomic signatures of thick-shelled and thin-shelled species most likely reflect the fact that more robust shells can withstand more damage before being completely destroyed and thus can obtain a higher taphonomic grade than thin-

Figure 11. Summary of the results of the mercury and taphonomic analyses. The mercury content in the shells document the pollution relative to the source, while the taphonomic data documents the location of die-off events.

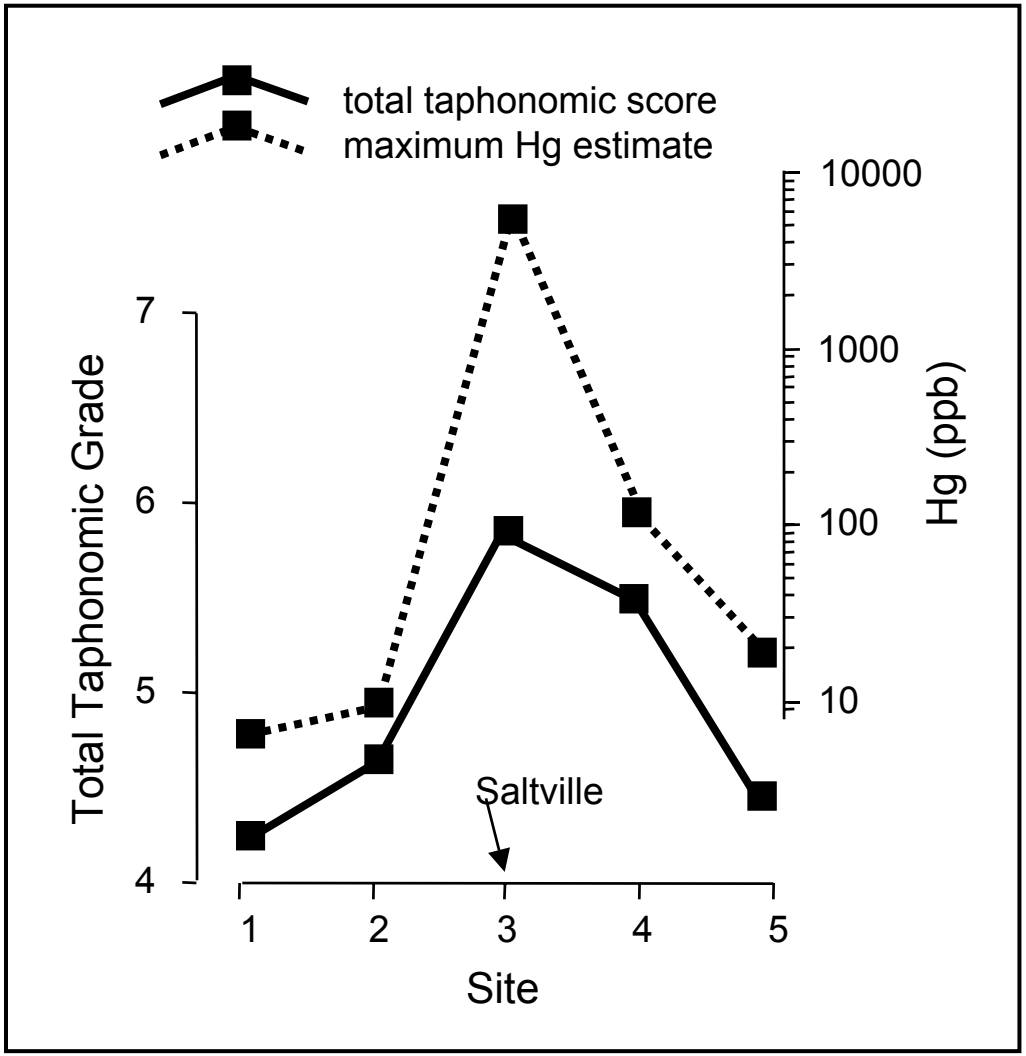
total taphonomic score
maximum Hg estimate
Total Taphonomic Grade
7
6
5
4
Hg (ppb)
10000
1000
100
10
Saltville
1
2
3
4
5
Site

Figure 11

shelled taxa. Yet, both thick-shelled and thin-shelled species reveal the same taphonomic trend related to the pollution source, again suggesting that taphonomic patterns are primarily driven by the location and severity of die-off events. In summary, the study suggests that the variation in taphonomic signatures of a death assemblage across sites is not a spurious artifact of changes in relative abundance of thick-shelled and thin-shelled species.

Thomas and Bendell-Young (1998) indicate that there may be potential contamination on the surface of the shell that is exposed to the contaminated water. The results of this study suggest that it is unlikely that the mercury signatures reflect post-mortem contamination of the shells. If post-mortem processes contaminated the shells, the highly deteriorated shells with higher taphonomic grades that typically represent specimens with longer post-mortem exposure should contain higher mercury levels. This was not seen because the total taphonomic grade of a shell and its mercury concentration were unrelated.

Chapter 6: Conclusions

This study investigated non-invasive techniques to obtain an assessment of the extirpation of freshwater mussels and the mercury pollution history of the North Fork Holston River. The major conclusions are as follows:

1. Shell chemistry provides an independent record of the presence and longitudinal variation of mercury contamination in the North Fork Holston River.
2. Shells collected in 1917 at Saltville contained only background levels of mercury, indicating that mercury pollution occurred sometime later in the pollution history of the North Fork Holston River.
3. Mercury levels in shells were similar in the outer margins of old mussels and young mussels after death, and do not support the hypothesis of post-mortem incorporation of mercury into shells.
4. Taphonomy of freshwater shell assemblages can be used to interpret the location and severity of the die-off event in the North Fork Holston River.
5. The taphonomic grade does not reflect a steeper stream gradient that represents the highest energy hydrodynamic regime, nor is it an artifact of the thickness of the shells even though more robust shells withstand more destruction than thinner shells.

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Appendix 1. Summary of taphonomic data for dead-collected shells from the five targeted sites along the North Fork Holston River.

Site	Shell	Articulation	Cracking	Holes	Fragmentation	Edge Preservation	External Peeling	External Luster
1	1	0	0	0	1	2	3	2
1	2	0	0	0	0	2	2	1
1	3	0	0	0	1	2	4	2
1	4	0	0	0	3	2	3	2
1	5	0	0	1	1	1	2	1
1	6	1	0	0	0	1	1	1
1	7	0	0	0	3	2	4	2
1	8	0	0	1	1	2	3	2
1	9	0	0	0	3	2	4	2
1	10	1	0	0	2	2	2	1
1	11	0	0	0	0	1	1	1
1	12	1	0	0	0	1	1	1
1	13	1	0	0	0	1	1	1
1	14	0	0	0	3	2	4	2
1	15	0	0	0	1	2	4	2
1	16	1	0	0	0	1	2	1
1	17	0	0	1	0	1	2	1
1	18	1	0	0	3	2	3	2
1	19	0	0	0	1	2	3	2
1	20	1	0	0	1	1	2	1
1	21	1	0	0	0	1	2	1
1	22	0	0	1	0	2	3	2
1	23	0	0	0	1	2	3	2
1	24	0	0	0	3	2	3	2
1	25	0	0	0	3	2	4	2
1	26	0	0	1	1	2	4	2
1	27	1	0	0	0	1	2	1
1	28	1	0	1	0	1	2	1
1	29	1	0	0	0	1	1	1
1	30	1	0	0	0	1	1	1
1	31	1	0	0	0	0	1	1
1	32	1	0	0	0	1	2	1
1	33	1	0	0	0	1	1	1
1	34	0	0	0	0	2	3	2
1	35	1	0	0	0	1	0	1
1	36	1	0	0	0	1	1	1
1	37	0	0	0	1	2	3	2
1	38	0	0	0	0	1	2	1
1	39	1	0	0	0	1	2	1
1	40	0	0	0	1	2	1	1
1	41	1	0	0	0	1	1	1
1	42	0	0	0	1	2	3	2
1	43	1	0	0	0	0	2	1
1	44	1	0	0	0	2	2	2
1	45	0	0	0	0	2	3	2
1	46	1	0	0	0	0	2	1

Site	Shell	Articulation	Cracking	Holes	Fragmentation	Edge Preservation	External Peeling	External Luster
1	47	1	0	0	0	1	2	1
1	48	0	0	0	3	2	4	2
1	49	0	0	0	1	2	3	2
1	50	1	0	0	0	0	2	1
1	51	0	0	0	0	2	2	2
1	52	0	0	0	1	2	3	2
1	53	1	0	0	0	1	2	1
1	54	0	0	0	1	2	2	2
1	55	0	0	0	0	1	1	1
1	56	0	0	0	2	1	2	1
1	57	0	1	0	0	1	2	1
1	58	1	0	0	0	1	2	1
1	59	0	0	0	1	1	3	2
1	60	0	1	0	0	1	2	1
1	61	0	0	0	1	2	2	1
1	62	0	0	0	0	2	3	2
1	63	0	0	0	3	2	4	2
1	64	0	0	1	1	1	2	1
1	65	0	0	0	2	2	3	2
1	66	1	0	0	0	1	2	1
1	67	0	0	0	2	2	3	2
1	68	1	0	0	0	1	2	1
2	1	0	0	0	0	2	3	2
2	2	0	0	0	0	2	1	1
2	3	0	0	0	2	2	2	1
2	4	1	0	1	0	0	2	1
2	5	1	0	1	1	0	2	1
2	6	0	0	1	1	2	3	2
2	7	1	0	0	0	0	3	1
2	8	1	0	0	0	0	2	1
2	9	0	0	1	1	1	2	1
2	10	1	0	0	0	1	2	1
2	11	0	0	0	2	2	3	2
2	12	0	0	0	3	2	3	2
2	13	1	0	1	0	0	2	1
2	14	0	0	0	3	2	3	2
2	15	1	0	1	0	1	2	1
2	16	1	1	1	0	2	3	2
2	17	0	1	0	1	2	2	1
2	18	0	0	0	3	2	4	2
2	19	0	0	0	3	2	3	2
2	20	0	0	0	2	2	3	2
2	21	0	0	0	2	2	3	2
2	22	0	0	0	0	2	3	2
2	23	0	0	0	3	2	3	2
2	24	0	0	0	1	2	4	2
2	25	0	0	0	0	1	1	1
2	26	0	1	1	1	1	2	1
2	27	1	0	1	0	2	3	1
2	28	1	0	0	0	1	2	1

Site	Shell	Articulation	Cracking	Holes	Fragmentation	Edge Preservation	External Peeling	External Luster
2	29	0	0	0	1	1	2	1
2	30	0	0	0	1	2	4	2
2	31	0	0	0	3	2	3	2
2	32	0	0	0	0	1	1	1
2	33	0	0	1	0	2	2	1
2	34	0	0	0	1	2	4	2
2	35	0	0	1	0	2	2	1
2	36	0	0	0	3	2	4	2
2	37	0	0	0	3	2	4	2
2	38	1	0	1	0	2	3	1
2	39	0	0	0	3	2	4	2
2	40	0	0	0	3	2	4	2
2	41	0	0	0	3	2	4	2
2	42	1	0	0	0	1	2	1
2	43	1	0	1	0	1	2	1
2	44	0	0	0	2	2	3	2
2	45	1	1	0	0	2	2	1
2	46	1	0	0	0	1	2	1
2	47	1	0	0	0	1	2	1
2	48	0	0	1	0	2	3	2
2	49	0	0	1	0	2	4	2
2	50	0	0	0	0	1	2	1
2	51	0	0	0	1	2	4	2
2	52	1	0	1	0	1	2	1
2	53	1	0	1	0	1	2	1
2	54	1	0	1	0	2	2	1
2	55	1	1	1	1	2	2	1
2	56	0	0	0	1	2	3	2
2	57	0	0	1	0	1	2	1
2	58	0	1	0	0	1	2	1
2	59	0	1	1	0	1	1	0
2	60	1	0	1	1	1	2	2
2	61	0	1	1	1	2	3	2
2	62	1	0	0	1	2	2	1
2	63	0	0	0	1	2	1	1
2	64	0	0	0	3	2	4	2
2	65	0	0	1	0	2	3	2
2	66	1	0	1	1	2	3	2
2	67	1	0	0	0	0	2	1
2	68	1	0	1	0	1	2	1
2	69	1	0	0	0	1	2	1
2	70	1	0	1	0	0	1	1
2	71	0	1	1	0	2	3	2
2	72	0	0	0	3	2	3	2
2	73	0	0	1	2	2	4	2
2	74	1	0	0	0	1	2	1
2	75	1	0	1	0	2	2	2
2	76	0	0	0	2	2	4	2
2	77	0	0	1	0	2	3	2
2	78	0	0	0	1	2	3	2

Site	Shell	Articulation	Cracking	Holes	Fragmentation	Edge Preservation	External Peeling	External Luster
2	79	0	1	0	0	2	2	0
2	80	1	0	1	0	1	2	1
2	81	0	1	1	1	2	2	1
2	82	0	0	0	1	2	3	1
2	83	0	0	0	0	2	3	2
2	84	1	0	0	0	1	2	1
2	85	0	0	0	0	1	2	1
2	86	1	1	1	0	2	2	1
2	87	0	0	0	2	2	4	2
2	88	0	0	0	2	2	4	2
2	89	0	0	0	0	1	2	1
2	90	0	0	1	0	2	2	2
2	91	1	0	1	0	0	3	2
2	92	1	0	1	0	1	2	1
2	93	0	0	0	2	2	4	2
2	94	0	0	0	3	2	3	2
2	95	0	1	0	3	2	2	1
2	96	0	0	0	2	2	3	2
2	97	0	0	0	3	2	3	2
2	98	0	1	1	0	1	1	1
2	99	0	0	0	3	2	3	2
2	100	0	0	0	3	2	2	1
2	101	0	0	0	3	2	4	2
2	102	1	0	1	0	2	3	2
2	103	1	0	0	0	0	2	1
2	104	1	0	0	2	2	3	2
3	1	0	0	0	1	2	4	2
3	2	0	0	0	0	1	1	0
3	3	0	0	0	0	1	2	1
3	4	0	0	0	0	2	3	2
3	5	0	0	0	3	2	4	2
3	6	0	0	0	3	2	1	2
3	7	0	0	0	0	2	1	1
3	8	0	0	0	0	2	1	1
3	9	1	0	0	0	1	1	0
3	10	1	0	0	0	0	0	0
3	11	0	0	0	3	2	4	2
3	12	0	0	0	3	2	4	2
3	13	0	0	0	1	2	3	2
3	14	0	1	0	1	2	2	1
3	15	0	0	0	0	2	2	2
3	16	1	0	0	1	1	1	1
3	17	0	0	0	2	2	4	2
3	18	0	0	0	2	2	4	2
3	19	0	0	0	2	2	3	2
3	20	0	0	0	3	2	3	2
3	21	0	0	0	3	2	2	2
3	22	0	0	0	3	2	4	2
3	23	0	0	0	3	2	3	2
3	24	0	0	0	2	2	3	2

Site	Shell	Articulation	Cracking	Holes	Fragmentation	Edge Preservation	External Peeling	External Luster
3	25	0	0	0	3	2	4	2
3	26	0	0	0	3	2	4	2
3	27	0	0	0	3	2	4	2
3	28	0	0	0	3	2	4	2
3	29	0	0	0	0	1	2	1
3	30	0	0	0	3	2	2	1
3	31	0	0	1	3	2	2	1
3	32	0	0	0	0	2	3	2
3	33	0	0	1	0	2	4	2
3	34	0	0	0	1	2	4	2
3	35	0	0	0	0	2	3	2
3	36	0	0	0	0	2	4	2
3	37	0	0	0	1	2	4	2
3	38	0	0	0	0	2	3	2
3	39	1	0	0	0	1	2	1
3	40	1	0	0	0	1	2	1
3	41	1	0	0	0	0	0	1
3	42	0	0	0	1	2	4	2
3	43	0	0	0	2	2	3	2
3	44	0	0	0	1	2	2	1
3	45	0	0	0	3	2	4	2
3	46	0	0	0	1	2	3	2
3	47	0	0	0	0	2	2	1
3	48	0	0	0	0	2	2	2
3	49	0	0	0	1	2	3	2
3	50	0	0	0	0	2	2	1
3	51	0	0	0	2	2	2	2
3	52	0	0	0	0	2	2	2
3	53	0	0	0	1	2	2	2
3	54	0	0	0	3	2	2	1
3	55	0	0	0	0	2	2	2
3	56	0	0	0	1	2	4	2
4	1	0	1	1	0	2	4	2
4	2	0	0	0	0	2	4	2
4	3	0	0	0	1	2	4	2
4	4	0	0	1	2	2	3	2
4	5	0	0	0	0	0	1	1
4	6	1	0	0	0	1	2	1
4	7	0	0	0	0	2	3	2
4	8	0	0	0	2	2	4	2
4	9	0	0	0	2	2	4	2
4	10	0	0	0	0	2	4	2
4	11	0	0	0	0	1	2	1
4	12	0	0	1	0	2	3	1
4	13	0	0	0	1	2	2	1
4	14	0	0	0	2	2	4	2
4	15	0	0	1	1	2	4	2
4	16	0	0	0	2	2	4	2
4	17	0	0	0	0	2	3	1
4	18	0	0	0	3	2	3	1

Site	Shell	Articulation	Cracking	Holes	Fragmentation	Edge Preservation	External Peeling	External Luster
4	19	0	0	0	1	2	3	1
4	20	0	0	0	1	1	2	1
4	21	0	0	0	3	2	3	1
4	22	0	0	0	1	2	3	2
4	23	0	0	0	0	0	2	1
4	24	0	0	0	0	1	2	1
4	25	0	0	0	1	2	2	1
4	26	0	0	0	1	2	3	2
4	27	0	0	0	1	2	3	2
4	28	0	0	0	3	2	4	2
4	29	0	0	0	2	2	3	2
4	30	0	0	0	0	2	2	1
4	31	0	0	0	1	2	3	2
4	32	0	0	0	3	2	4	2
4	33	0	1	0	0	1	1	1
4	34	0	1	0	1	2	2	1
4	35	0	0	0	1	2	3	2
4	36	0	0	0	3	2	2	1
4	37	0	0	0	0	1	2	1
4	38	0	0	0	1	2	4	2
4	39	0	0	0	3	2	4	2
4	40	0	0	0	0	2	1	1
4	41	0	0	0	1	2	3	1
4	42	0	0	0	2	2	4	2
4	43	0	0	1	0	2	4	2
4	44	0	0	0	2	2	4	2
4	45	0	0	0	1	2	3	1
4	46	0	0	0	1	2	3	2
4	47	0	0	0	3	2	4	2
4	48	0	0	0	3	2	2	1
4	49	0	0	0	2	2	4	2
4	50	0	0	0	2	2	4	2
4	51	0	0	0	1	2	4	2
4	52	0	0	0	2	2	3	2
4	53	0	0	0	3	2	4	2
4	54	0	0	0	1	2	3	2
4	55	0	0	0	1	2	3	1
4	56	0	0	0	3	2	3	2
4	57	1	0	0	1	1	2	1
4	58	0	0	0	0	1	2	1
4	59	0	0	0	0	2	3	2
4	60	0	0	0	2	2	4	2
4	61	0	0	0	1	2	4	2
4	62	0	0	0	1	2	4	2
4	63	0	0	0	1	2	3	2
4	64	0	0	0	1	2	2	1
4	65	0	0	0	1	2	4	2
4	66	0	0	0	0	1	1	1
4	67	0	0	1	1	2	3	2
4	68	0	0	0	0	2	3	2

Site	Shell	Articulation	Cracking	Holes	Fragmentation	Edge Preservation	External Peeling	External Luster
4	69	0	0	0	1	2	4	2
4	70	0	0	0	1	2	4	2
4	71	0	0	0	0	2	3	2
4	72	0	0	0	1	2	4	2
4	73	0	0	0	0	1	2	1
4	74	1	0	0	1	2	3	2
4	75	0	0	1	1	2	4	2
4	76	0	0	0	1	2	3	2
4	77	0	0	1	2	2	3	2
4	78	0	0	0	2	2	3	2
4	79	0	0	0	0	1	1	1
4	80	0	0	0	0	1	2	2
4	81	0	0	0	1	2	2	2
5	1	0	0	0	0	1	3	1
5	2	1	0	0	0	2	3	1
5	3	1	1	1	0	1	1	0
5	4	0	0	0	1	2	2	1
5	5	0	0	0	2	2	2	1
5	6	1	0	0	0	1	2	1
5	7	0	0	0	3	2	4	2
5	8	1	0	0	1	1	2	1
5	9	0	0	0	3	2	4	2
5	10	0	1	0	1	1	1	1
5	11	1	0	0	1	0	1	0
5	12	1	0	1	0	1	2	1
5	13	0	0	0	2	2	4	2
5	14	0	0	0	0	1	3	2
5	15	0	0	0	0	2	4	2
5	16	0	0	0	0	2	4	2
5	17	0	0	0	3	2	4	2
5	18	0	0	0	3	2	4	2
5	19	0	0	0	3	2	2	1
5	20	0	0	0	3	2	4	2
5	21	0	0	0	3	2	4	2
5	22	0	0	0	3	2	4	2
5	23	1	0	0	0	1	1	1
5	24	1	1	0	0	1	1	0
5	25	1	0	0	0	1	1	0
5	26	1	0	0	0	1	2	1
5	27	1	0	1	0	1	2	1
5	28	1	0	1	1	2	3	2
5	29	0	0	0	3	2	4	2
5	30	1	0	0	0	1	2	2
5	31	1	0	0	0	1	2	1
5	32	1	1	0	1	1	1	1
5	33	0	0	0	3	2	4	2
5	34	0	0	0	3	2	2	1
5	35	1	0	0	1	1	2	1
5	36	0	0	0	3	2	3	2
5	37	0	0	0	2	2	4	2

Site	Shell	Articulation	Cracking	Holes	Fragmentation	Edge Preservation	External Peeling	External Luster
5	38	0	0	0	0	1	2	1
5	39	0	0	1	0	0	1	0
5	40	0	0	0	1	2	4	2
5	41	0	0	0	2	2	4	2
5	42	0	0	0	3	2	4	2
5	43	1	0	0	0	1	2	1
5	44	1	1	0	0	1	1	1
5	45	1	0	0	0	1	2	1
5	46	0	0	0	1	2	3	2
5	47	0	0	0	3	1	2	1
5	48	0	0	0	3	1	4	2
5	49	0	1	0	2	1	2	1
5	50	0	0	0	0	1	2	1
5	51	1	0	0	0	1	3	1
5	52	0	1	0	0	1	3	1
5	53	1	1	0	1	2	2	1
5	54	0	0	0	0	1	2	1
5	55	0	0	0	0	2	4	2
5	56	0	0	0	0	2	3	2
5	57	1	0	0	2	2	4	2
5	58	0	0	1	0	2	3	2
5	59	0	0	0	2	1	1	1
5	60	0	0	0	1	2	2	1

Site	Shell	Length (mm)	Fragment Size (mm)	Genus	Species	Valve	Thickness
1	1	45.97		*Villosa*	*iris*	L	Thin
1	2	46.84		*Villosa*	*iris*	L	Thin
1	3	58.95		*Lexingtonia*	*dolabelloides*	R	Thick
1	4		15.02	not identified		F	
1	5		37.43	*Villosa*	*iris*	R	Thin
1	6	40.46		*Villosa*	*vanuxemensis*	B	Thin
1	7		35.56	not identified		F	Thick
1	8		44.82	*Villosa*	*iris*	L	Thin
1	9		25.98	not identified		F	
1	10		46.06	*Lampsilis*	*fasciola*	B	Thick
1	11	31.62		*Lexingtonia*	*dolabelloides*	L	Thick
1	12	64.92		*Lexingtonia*	*dolabelloides*	B	Thick
1	13	54.03		*Lexingtonia*	*dolabelloides*	B	Thick
1	14		25.32	not identified		F	Thick
1	15		57.25	*Ptychobranchus*	*subtentum*	R	Thick
1	16	46.67		*Lampsilis*	*fasciola*	B	Thick
1	17	49.14		*Villosa*	*iris*	L	Thin
1	18		32.07	not identified		B	Thin
1	19	40.33		*Villosa*	*iris*	L	Thin
1	20	38.79		*Medionidus*	*condradicus*	B	Thin
1	21	77.77		*Lampsilis*	*fasciola*	B	Thick
1	22	44.54		*Villosa*	*vanuxemensis*	R	Thin
1	23		70.18	*Ptychobranchus*	*subtentum*	R	Thick

Site	Shell	Length (mm)	Fragment Size (mm)	Genus	Species	Valve	Thickness
1	24		21.93	not identified		F	
1	25		22.06	not identified		F	
1	26		63.73	not identified		L	Thick
1	27	37.73		not identified		B	
1	28	42.24		*Villosa*	*iris*	B	Thin
1	29	57.22		*Fusconaia*	*barnesiana*	B	Thick
1	30	59.64		*Lampsilis*	*fasciola*	B	Thick
1	31	51.17		*Villosa*	*iris*	B	Thin
1	32	68.39		*Pleurobema*	*oviforme*	B	Thick
1	33	47.2		*Villosa*	*vanuxemensis*	B	Thin
1	34	42.92		not identified		R	
1	35	48.54		*Lexingtonia*	*dolabelloides*	B	Thick
1	36	47.47		*Villosa*	*iris*	B	Thin
1	37		48.47	*Pleurobema*	*oviforme*	L	Thick
1	38	44.62		*Villosa*	*iris*	R	Thin
1	39	73.33		*Lampsilis*	*fasciola*	B	Thick
1	40	49.59		*Pleurobema*	*oviforme*	R	Thick
1	41	48.36		*Pleurobema*	*oviforme*	B	Thick
1	42		40.01	*Lampsilis*	*fasciola*	R	Thick
1	43	44.13		*Medionidus*	*conradicus*	B	Thin
1	44	78.03		*Pleurobema*	*oviforme*	B	Thick
1	45	62.12		*Lampsilis*	*fasciola*	L	Thick
1	46	65.8		*Villosa*	*iris*	B	Thin
1	47	68.39		*Pleurobema*	*oviforme*	B	Thick
1	48		44.05	not identified		F	Thick
1	49		27.51	*Medionidus*	*conradicus*	F	Thin
1	50	26.71		*Villosa*	*vanuxemensis*	B	Thin
1	51	61.76		*Lampsilis*	*fasciola*	L	Thick
1	52		44.39	*Ptychobranchus*	*subtentum*	R	Thick
1	53	32.58		*Villosa*	*vanuxemensis*	B	Thin
1	54		26.55	not identified		L	Thin
1	55	40.09		*Villosa*	*vanuxemensis*	R	Thin
1	56		32.9	*Villosa*	*iris*	L	Thin
1	57	37.32		*Villosa*	*iris*	R	Thin
1	58	42.36		*Villosa*	*iris*	B	Thin
1	59	26.84		*Villosa*	*vanuxemensis*	L	Thin
1	60	33.93		*Villosa*	*iris*	R	Thin
1	61		34.45	*Medionidus*	*conradicus*	R	Thin
1	62	27.88		*Lexingtonia*	*dolabelloides*	L	Thick
1	63		42.94	*Medionidus*	*conradicus*	F	Thin
1	64		25.31	*Villosa*	*iris*	R	Thin
1	65		37.39	not identified		F	
1	66	37.51		*Pleurobema*	*oviforme*	B	Thick
1	67		59.23	not identified		L	Thick
1	68	54.9		*Villosa*	*iris*	B	Thin
2	1	75.91		*Ptychobranchus*	*subtentum*	L	Thick
2	2	42.04		*Fusconaia*	*barnesiana*	L	Thick
2	3		53.35	not identified		L	Thin
2	4	48.1		*Villosa*	*iris*	B	Thin
2	5	48.56		*Villosa*	*iris*	B	Thin

Site	Shell	Length (mm)	Fragment Size (mm)	Genus	Species	Valve	Thickness
2	6		58.51	not identified		L	Thick
2	7	49.27		*Pleurobema*	*oviforme*	B	Thick
2	8	56.28		*Fusconaia*	*barnesiana*	B	Thick
2	9	35.08		*Lexingtonia*	*dolabelloides*	L	Thick
2	10	32.65		*Medionidus*	*conradicus*	B	Thin
2	11		43.46	not identified		L	Thick
2	12		38.73	not identified		L	Thin
2	13	28.91		*Villosa*	*iris*	B	Thin
2	14		35.81	not identified		F	
2	15	50.58		*Villosa*	*iris*	B	Thin
2	16	60.08		*Lampsilis*	*fasciola*	B	Thick
2	17		37.45	*Villosa*	*vanuxemensis*	L	Thin
2	18		34.31	not identified		F	Thick
2	19		34.44	not identified		F	
2	20		38.59	not identified		L	
2	21		39.84	not identified		L	
2	22	34.79		*Villosa*	*iris*	L	Thin
2	23		44.74	*Lampsilis*	*fasciola*	F	Thick
2	24	52.62		*Pleurobema*	*oviforme*	R	Thick
2	25	35.6		*Fusconaia*	*cor*	L	Thick
2	26	44.12		*Villosa*	*iris*	L	Thin
2	27	54.57		*Villosa*	*iris*	B	Thin
2	28	49.84		*Lexingtonia*	*dolabelloides*	B	Thick
2	29		34.73	*Pleurobema*	*oviforme*	R	Thick
2	30		44.73	*Villosa*	*iris*	L	Thin
2	31		35.27	not identified		F	Thick
2	32	45.56		*Villosa*	*iris*	L	Thin
2	33	55.98		*Villosa*	*iris*	R	Thin
2	34		49.62	*Pleurobema*	*oviforme*	R	Thick
2	35	45.12		*Lexingtonia*	*dolabelloides*	L	Thick
2	36		50.33	not identified		F	Thick
2	37		51.07	*Ptychobranchus*	*subtentum*	L	Thick
2	38	46.07		*Pleurobema*	*oviforme*	B	Thick
2	39		30.3	not identified		R	Thin
2	40		39.19	not identified		F	
2	41		30.75	not identified		F	
2	42	56.89		*Villosa*	*iris*	B	Thin
2	43	48.23		*Villosa*	*iris*	B	Thin
2	44		73.76	*Ptychobranchus*	*subtentum*	R	Thick
2	45	79.26		*Lampsilis*	*fasciola*	B	Thick
2	46	49.55		*Villosa*	*iris*	B	Thin
2	47	57.39		*Villosa*	*iris*	B	Thin
2	48	82.84		*Ptychobranchus*	*subtentum*	L	Thick
2	49	63.29		*Pleurobema*	*oviforme*	L	Thick
2	50	52.16		*Villosa*	*iris*	L	Thin
2	51		58.79	*Pleurobema*	*oviforme*	R	Thick
2	52	66.55		*Pleurobema*	*oviforme*	B	Thick
2	53	48.69		*Villosa*	*iris*	B	Thin
2	54	72.25		*Pleurobema*	*oviforme*	B	Thick
2	55	51.66		*Lampsilis*	*fasciola*	B	Thick

Site	Shell	Length (mm)	Fragment Size (mm)	Genus	Species	Valve	Thickness
2	56		34.95	not identified		L	
2	57	46.52		*Villosa*	*iris*	L	Thin
2	58	47.82		*Lampsilis*	*fasciola*	L	Thick
2	59	36.66		*Medionidus*	*conradicus*	L	Thin
2	60		24.2	*Villosa*	*iris*	B	Thin
2	61		58.12	*Lampsilis*	*fasciola*	L	Thick
2	62	63.77		*Ptychobranchus*	*fasciolaris*	B	Thick
2	63	48.15		*Fusconaia*	*cor*	R	Thick
2	64		42.84	not identified		F	Thick
2	65	65.22		*Pleurobema*	*oviforme*	L	Thick
2	66		54.92	*Pleurobema*	*oviforme*	B	Thick
2	67	52.07		*Lampsilis*	*fasciola*	B	Thick
2	68	50.3		*Villosa*	*iris*	B	Thin
2	69	45.89		*Fusconaia*	*cor*	B	Thick
2	70	34.56		*Lexingtonia*	*dolabelloides*	B	Thick
2	71	67.63		*Pleurobema*	*oviforme*	R	Thick
2	72		33.66	not identified		F	
2	73		62.07	*Pleurobema*	*oviforme*	L	Thick
2	74	51.41		*Lexingtonia*	*dolabelloides*	B	Thick
2	75	54.33		*Ptychobranchus*	*fasciolaris*	B	Thick
2	76		46.13	*Pleurobema*	*oviforme*	F	Thick
2	77	49.63		*Lexingtonia*	*dolabelloides*	L	Thick
2	78		46.02	*Lampsilis*	*fasciola*	L	Thick
2	79	55.04		*Fusconaia*	*cor*	L	Thick
2	80	51.61		*Pleurobema*	*oviforme*	B	Thick
2	81		54.06	*Ptychobranchus*	*subtentum*	L	Thick
2	82		49.17	*Ptychobranchus*	*subtentum*	R	Thick
2	83	66.55		*Pleurobema*	*oviforme*	L	Thick
2	84	57.18		*Villosa*	*iris*	B	Thin
2	85	34.21		*Fusconaia*	*cor*	R	Thick
2	86		25.88	*Medionidus*	*conradicus*	B	Thin
2	87		37.24	not identified		L	Thick
2	88		29.31	not identified		L	
2	89	37.67		*Fusconaia*	*barnesiana*	L	Thick
2	90	63.48		not identified		L	Thick
2	91	51.51		*Villosa*	*iris*	B	Thin
2	92	42.83		*Villosa*	*vanuxemensis*	B	Thin
2	93		49.88	not identified		R	Thick
2	94		41.05	not identified		F	Thick
2	95		34.51	not identified		F	
2	96		39.94	not identified		L	Thin
2	97		42.47	not identified		F	Thick
2	98	29.96		*Villosa*	*iris*	L	Thin
2	99		22.35	not identified		R	Thin
2	100		32.28	not identified		F	Thin
2	101		43.77	not identified		R	Thick
2	102	70.04		*Fusconaia*	*barnesiana*	B	Thick
2	103	48.97		*Villosa*	*iris*	B	Thin
2	104		75.72	*Ptychobranchus*	*subtentum*	B	Thick
3	1	55.47		*Pleurobema*	*oviforme*	L	Thick

Site	Shell	Length (mm)	Fragment Size (mm)	Genus	Species	Valve	Thickness
3	2	41.44		*Villosa*	*iris*	L	Thin
3	3	47.24		*Villosa*	*iris*	L	Thin
3	4	78.27		*Ptychobranchus*	*subtentum*	R	Thick
3	5		41.54	not identified		F	
3	6		25.93	not identified		L	
3	7	58.9		*Pleurobema*	*oviforme*	R	Thick
3	8	77.96		*Ptychobranchus*	*fasciolaris*	L	Thick
3	9	45.99		*Villosa*	*iris*	B	Thin
3	10	40.45		*Villosa*	*iris*	B	Thin
3	11		41.11	not identified		F	Thick
3	12		36.18	not identified		F	Thick
3	13		38.72	*Pleurobema*	*oviforme*	R	Thick
3	14	69.35		*Actinonaias*	*pectorosa*	L	Thick
3	15	57.26		*Lexingtonia*	*dolabelloides*	R	Thick
3	16	73.21		*Lampsilis*	*fasciola*	B	Thick
3	17		47.89	not identified		L	
3	18		35.07	not identified		L	
3	19		38.83	*Lampsilis*	*fasciola*	L	Thick
3	20		17.17	not identified		F	Thin
3	21		26.67	*Villosa*	*iris*	R	Thin
3	22		15.37	not identified		F	Thin
3	23		71.54	*Actinonaias*	*pectorosa*	F	Thick
3	24		34.03	*Medionidus*	*conradicus*	R	Thin
3	25		38.18	not identified		F	
3	26		22.85	not identified		F	
3	27		20.53	not identified		F	Thick
3	28		21.42	not identified		F	Thin
3	29	49.82		not identified		R	Thin
3	30		28.53	*Ptychobranchus*	*subtentum*	F	Thick
3	31		43.38	*Ptychobranchus*	*subtentum*	R	Thick
3	32	70.63		not identified		L	Thick
3	33	56.53		not identified		L	Thick
3	34		52.24	not identified		L	Thick
3	35	60.26		not identified		L	Thick
3	36	34.12		not identified		L	Thin
3	37		67.64	not identified		R	Thick
3	38	59.94		not identified		L	Thick
3	39	72.51		not identified		B	Thick
3	40	58.36		not identified		B	Thick
3	41	60.07		not identified		B	
3	42		45.12	not identified		R	Thick
3	43		31.24	not identified		R	Thin
3	44		51.97	not identified		R	Thin
3	45		23.44	not identified		F	
3	46		53.23	not identified		R	Thick
3	47	72.61		not identified		L	Thick
3	48	48.99		not identified		R	Thick
3	49		71.32	not identified		L	Thick
3	50	57.9		not identified		L	Thick
3	51		56.83	not identified		R	Thick

Site	Shell	Length (mm)	Fragment Size (mm)	Genus	Species	Valve	Thickness
3	52	64.53		not identified		L	Thick
3	53		57.67	not identified		R	Thick
3	54		23.87	not identified		F	Thin
3	55	30.46		not identified		L	Thin
3	56		50.92	not identified		L	Thick
4	1	113.6		*Actinonaias*	*pectorosa*	L	Thick
4	2	55.82		*Lexingtonia*	*dolabelloides*	L	Thick
4	3		50.89	not identified		R	Thick
4	4		48.09	*Ptychobranchus*	*subtentum*	L	Thick
4	5	39.09		*Villosa*	*iris*	L	Thin
4	6	57.01		*Pleurobema*	*oviforme*	B	Thick
4	7		48.29	*Ptychobranchus*	*subtentum*	R	Thick
4	8		53.77	not identified		F	Thick
4	9		50.02	not identified		F	Thick
4	10		60.67	*Ptychobranchus*	*subtentum*	L	Thick
4	11	38.81		*Pleurobema*	*oviforme*	R	Thick
4	12		41.39	*Pleurobema*	*oviforme*	L	Thick
4	13		35.39	*Pleurobema*	*oviforme*	R	Thick
4	14		43.65	not identified		F	
4	15		69.3	*Ptychobranchus*	*fasciolaris*	L	Thick
4	16		52.83	not identified		R	Thick
4	17	51.59		*Pleurobema*	*oviforme*	L	Thick
4	18		24.14	not identified		F	
4	19	40.21		*Fusconaia*	*cor*	L	Thick
4	20		40.92	*Pleurobema*	*oviforme*	L	Thick
4	21		30.08	not identified		F	
4	22		36.85	not identified		L	
4	23	50.31		*Villosa*	*vanuxemensis*	L	Thin
4	24	57.5		*Lampsilis*	*fasciola*	R	Thick
4	25		37.59	*Pleurobema*	*oviforme*	L	Thick
4	26		47.97	*Pleurobema*	*oviforme*	R	Thick
4	27		47.89	*Pleurobema*	*oviforme*	L	Thick
4	28		23.5	not identified		F	
4	29		40.09	not identified		R	Thick
4	30	45.91		*Fusconaia*	*barnesiana*	L	Thick
4	31	37.73		*Fusconaia*	*cor*	R	Thick
4	32		44.12	*Ptychobranchus*	*subtentum*	R	Thick
4	33	57.65		*Elliptio*	*dilatata*	R	Thin
4	34	45.07		*Villosa*	*vanuxemensis*	L	Thin
4	35		54.19	*Pleurobema*	*oviforme*	L	Thick
4	36		22.84	not identified		F	
4	37	43.51		*Villosa*	*iris*	R	Thin
4	38		43.74	*Ptychobranchus*	*subtentum*	L	Thick
4	39		29.06	not identified		F	
4	40	40.53		*Pleurobema*	*oviforme*	R	Thick
4	41	45.3		*Pleurobema*	*oviforme*	L	Thick
4	42		37.11	not identified		L	Thick
4	43	53.23		*Ptychobranchus*	*subtentum*	L	Thick
4	44		47.29	*Lexingtonia*	*dolabelloides*	R	Thick
4	45		34.38	not identified		L	Thin

Site	Shell	Length (mm)	Fragment Size (mm)	Genus	Species	Valve	Thickness
4	46		52.55	*Villosa*	*iris*	R	Thin
4	47		36.06	*Ptychobranchus*	*subtentum*	L	Thick
4	48		25.78	not identified		F	Thin
4	49		34.91	*Fusconaia*	*cor*	R	Thick
4	50		33.09	not identified		L	Thick
4	51		48.57	*Pleurobema*	*oviforme*	R	Thick
4	52		56.83	*Ptychobranchus*	*subtentum*	R	Thick
4	53		40.78	not identified		L	Thick
4	54	51.62		*Pleurobema*	*oviforme*	R	Thick
4	55		46.67	not identified		L	
4	56		31.35	*Ptychobranchus*	*subtentum*	F	Thick
4	57		48.83	not identified		B	Thick
4	58	59.1		not identified		R	Thick
4	59	76.2		not identified		L	Thick
4	60		36.76	not identified		R	Thick
4	61		41.27	not identified		L	Thick
4	62		61.06	not identified		L	Thick
4	63		53.5	not identified		L	Thick
4	64		53.17	not identified		L	Thin
4	65	52.23		not identified		L	Thick
4	66	56.13		not identified		R	Thick
4	67	41.72		not identified		L	Thick
4	68	55.22		not identified		L	Thick
4	69		28.07	not identified		L	Thick
4	70		37.44	not identified		L	Thick
4	71	47.29		not identified		L	Thick
4	72		47.53	not identified		L	Thick
4	73	55.51		not identified		L	Thin
4	74		68.85	not identified		B	Thick
4	75		41.04	not identified		R	Thick
4	76		39.79	not identified		L	Thick
4	77		38.01	not identified		R	Thick
4	78		27.29	not identified		L	Thick
4	79	43.6		not identified		L	Thin
4	80	39.28		not identified		L	Thin
4	81		36.84	not identified		L	Thick
5	1	44.41		*Villosa*	*vanuxemensis*	R	Thin
5	2	59.81		*Pleurobema*	*oviforme*	B	Thick
5	3	47.53		*Villosa*	*iris*	B	Thin
5	4	60.35		*Lexingtonia*	*dolabelloides*	R	Thick
5	5	45.23		*Lampsilis*	*fasciola*	L	Thick
5	6	55.8		*Villosa*	*vanuxemensis*	B	Thin
5	7		38.25	not identified		F	
5	8	59.18		*Lampsilis*	*fasciola*	B	Thick
5	9		37.95	not identified		L	
5	10		35.26	*Villosa*	*iris*	R	Thin
5	11	53.69		*Lampsilis*	*fasciola*	B	Thick
5	12	42.77		*Villosa*	*vanuxemensis*	B	Thin
5	13		46.53	not identified		F	
5	14	42.14		*Villosa*	*vanuxemensis*	R	Thin

Site	Shell	Length (mm)	Fragment Size (mm)	Genus	Species	Valve	Thickness
5	15		47.22	not identified		F	
5	16		44.26	not identified		L	
5	17		22.05	not identified		F	
5	18		31.15	not identified		F	
5	19		27.69	*Villosa*	*iris*	F	Thin
5	20		35.17	not identified		F	
5	21		28.55	not identified		F	
5	22		20.89	not identified		L	
5	23	40.71		*Lampsilis*	*fasciola*	B	Thick
5	24	47.63		*Villosa*	*iris*	B	Thin
5	25	54.46		*Lampsilis*	*fasciola*	B	Thick
5	26	56.15		*Villosa*	*iris*	B	Thin
5	27	41.76		*Villosa*	*iris*	B	Thin
5	28	59.06		*Pleurobema*	*oviforme*	B	Thick
5	29		32.95	not identified		L	
5	30	63.64		*Lampsilis*	*fasciola*	B	Thick
5	31	51.82		*Villosa*	*iris*	B	Thin
5	32	50.64		*Villosa*	*iris*	B	Thin
5	33		18.88	not identified		F	
5	34		28.69	not identified		F	
5	35	45.13		*Lampsilis*	*fasciola*	B	Thick
5	36		31.37	not identified		F	
5	37		58.7	not identified		L	
5	38	47.45		*Villosa*	*vanuxemensis*	R	Thin
5	39	42.32		*Villosa*	*iris*	R	Thin
5	40	45.49		*Pleurobema*	*oviforme*	R	Thick
5	41		40.38	not identified		R	
5	42		32.66	not identified		R	
5	43	52.33		*Villosa*	*vanuxemensis*	B	Thin
5	44	67.15		*Lampsilis*	*fasciola*	B	Thick
5	45	56.98		*Lampsilis*	*fasciola*	B	Thick
5	46	55.89		*Pleurobema*	*oviforme*	L	Thick
5	47		38.09	*Villosa*	*vanuxemensis*	F	Thin
5	48		30.68	not identified		F	
5	49		41.16	*Villosa*	*iris*	R	Thin
5	50	50.07		*Lampsilis*	*fasciola*	L	Thick
5	51	44.37		*Lampsilis*	*fasciola*	B	Thick
5	52	61.01		*Villosa*	*iris*	L	Thin
5	53	66.8		*Lampsilis*	*fasciola*	B	Thick
5	54	55.48		*Villosa*	*iris*	L	Thin
5	55	49.64		*Pleurobema*	*oviforme*	L	Thick
5	56	52.22		*Pleurobema*	*oviforme*	R	Thick
5	57		42.12	not identified		B	
5	58	42.75		*Villosa*	*vanuxemensis*	L	Thin
5	59		31.19	*Pleurobema*	*oviforme*	L	Thick
5	60	53.77		*Villosa*	*vanuxemensis*	L	Thin

Appendix B: Curriculum Vitae

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EDUCATION

M. S. in Geosciences (expected spring 2004), Department of Geosciences, Virginia Tech, Blacksburg, VA
ADVISOR: Michal Kowalewski
THESIS: Geochemical and Taphonomic Signatures of Freshwater Mussel Shells as Evidence of Mercury-Related Extirpations in the North Fork Holston River, Virginia

B. A. in Biology, 2002, Department of Biology, University of Virginia, Charlottesville, Virginia

PROFESSIONAL EXPERIENCE

Spring, 2004	Teaching Assistant, Dept. of Geosciences, Virginia Tech
Summer, 2002	Research Assistant, Dept. of Soil, Water, and Environmental Science, University of Arizona
Fall, 1999-Spring, 2000	Office Assistant, Dept. of Environmental Science, University of Virginia
Summer, 1999	Teaching Assistant for high school summer biology program, Dept. of Biology, Virginia State University
1994-1998	Laboratory Assistant, BioTech Laboratory

GRANTS, AWARDS, AND HONORS

Spring, 2004	Geological Society of America Travel Grant, $100
Fall, 2003	Graduate Student Assembly Travel Grant, $170
Spring, 2003	The Theodore Roosevelt Memorial Fund, American Museum of Natural History, $1000
Spring, 2003	David R. Wones Geosciences Fund, Virginia Tech, $1000
2002-2003	Powell Fellowship Award, Multicultural Academic Opportunities Program, Virginia Tech
1999-2002	Dean's Honor Roll, University of Virginia

ABSTRACT PUBLICATIONS

Brown, M.E., Kowalewski, M., Cherry, D., Neves, R. and Schreiber, M., 2004, Using Geochemical and Taphonomic Signatures of Freshwater Mussel Shells to Explore Industry-Related Extirpations in the North Fork Holston River, VA. *Geological Society of America, Abstracts with Programs*, v. 36 no. 2

Brown, M. E. and Kowalewski, M., 2003, Do Local Extinctions Correlate with Taphonomic Signatures of Freshwater Mussel Shells in the North Fork Holston River, VA? *Geological Society of America, Abstracts with Programs*, v. 35 no. 6

PROFESSIONAL AFFILIATIONS

2003-2004	Geological Society of America (GSA)
2003-2004	Virginia Academy of Science
2003-2004	The Paleontological Society