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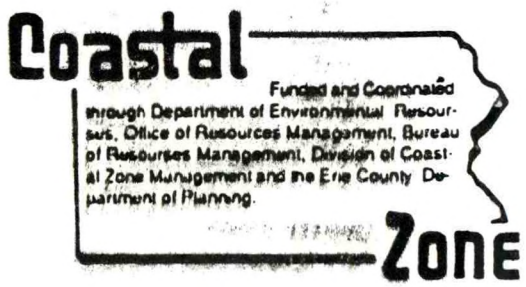

E.C. Masteller

BASELINE ANALYSIS OF BIOACCUMULATION OF HEAVY METALS IN ZEBRA MUSSELS IN LAKE ERIE AND PRESQUE ISLE BAY, ERIE, PENNSYLVANIA

This study was undertaken to determine the extent of bioaccumulation of heavy metals by the zebra mussel (*Dreissena polymorpha*). Samples were taken from nine different locations along the Pennsylvania shoreline of Lake Erie (Figure 1). Samples were collected for analysis from June to October with two samples from six sites to determine if bioaccumulation was increasing with size and time of exposure. Samples were also obtained from western Lake Erie (Detroit Edison Power Company) and Lake St. Clair for comparison.

A total of fifteen samples were analyzed over the summer of 1990 for cadmium, lead, and chromium (Table 1). Background water data for the sample sites for these samples was too low to be detectable for these heavy metals. Calcium levels were determined for all sites as well as recording the physical parameters of temperature, pH, dissolved oxygen, conductivity, and oxidation reduction potential.

It has been observed that mollusks will accumulate heavy metals in both the periostracum and the soft body parts (Czarnezki, 1987; Karbe et al., 1975; Zadory, 1983; Hinch and Bailey, 1988; Hinch and Green, 1989). The latter reference found *Elliptio complanata* a good monitor of acidification and heavy metal concentration. These unionids are long-lived individuals; *Elliptio complanata* and *E. dilatata* live 15 years and *Lampsilis radiata* 12 years in St. Lawrence River channels, but others have been estimated to live 0-40 years (Imlay, 1982). Hinch and Stephenson (1987) found that concentrations of metals in tissues is related to ambient metal concentration, pH, and alkalinity.

Bivalves are well suited for in-situ monitoring as they bioaccumulate environmental pollutants, they are sedentary, numerous, and large enough to provide tissue analysis. Anderson (1977) found that in six species of clams the body tissue generally reflected the concentrations of Cd, Cu, Pb, and Zn found in the sediment. The six species used for analysis were *Lampsilis siliquoidea*, *L. ventricosa*, *Strophitis rugosus*, *Anodonta marginata*, *Lasmigona complanata*, and *Sphaerium sp*. The order of metal concentrations he found were Cd < Cu < Pb < Zn. Pugsley et al. (1988) found levels of 7.1 mg/kg for lead in clams which was one-half that found in sediments, whereas cadmium concentrations were 30 times higher in clam tissues than in the surrounding sediments (6.4 mg/kg). Bias and Karbe (1985) used radioactive cadmium and found accumulation factors of more than 70,000 in the periostracum and up to 3,000 for the whole mussel. Koide et al. (1982) suggested that byssal threads, because of their enrichment of transuranic actinide elements (those elements with an atomic number greater than or equal to 92) and their ease in handling, may be useful in monitoring heavy metals. They reported the level of eight heavy metals in *Mytilus edulis* with Ni the highest. Karbe et al. (1975) reported that the accumulation of metals from water was strongly influenced by the water quality of the sampling site. Tessier et al. (1984) found that metal accumulation was influenced by the protective or competitive effect of sediment constituents, notably amorphous iron oxyhydroxides. Slooff et al. (1983) reported on the biological activity of mussels exposed to different heavy metal levels. Green et al. (1989) have shown that bioconcentration of metals by mussels is influenced by their growth rate.

Nine sites were sampled for bioaccumulation of three heavy metals: Cd, Pb, and Cr. The first samples in June required development of our technique, therefore, these data are not reported. Six of the sites were sampled a second

time to determine if there might be any effect of size or season. The results of all sites are in Table 1. Lampe Marina had the highest levels of cadmium with 10.1 ppm, but no increase in the cadmium level appeared with size increase (1.7 cm to 2.1 cm average length from August 24, 1990 to September 18, 1990). Data are not available from the June collection at this site when the average length of the zebra mussel shells was less than 1 cm. The range of cadmium for all sites was 2.8 to 10.1 ppm with an average for all sites of 5.3 ppm. Chromium was excessively high in Horseshoe Pond at Presque Isle, but at present we attribute this to the paint that was removed with the mussels collected from houseboats. A chromium level excluding Horseshoe Pond of 6.0 - 6.4 ppm was present at Raccoon Creek, Thompson Bay at Presque Isle, and Freeport Beach at Northeast with a range from all sites of 2.6 to 6.4 ppm with an average of 4.2 ppm. Most of these high readings did occur in late summer. Lead showed the same problem at Horseshoe Pond but excluding this a level of 8.0 - 9.4 ppm was present at four sites: Raccoon Creek, Thompson Bay-Presque Isle, Lampe Marina, and Freeport Beach. The range for lead at all sites was 2.6 to 9.4 ppm with an average of 7.4 ppm (Figure 2).

Samples acquired from two sources to the west of Erie, Pennsylvania were analyzed in our laboratory at Penn State-Behrend. The values from the Erie, Pennsylvania region all tended to be higher than those found in zebra mussels collected in western Lake Erie (Detroit Edison) and Lake St. Clair (Table 2). The levels we found were also higher than background water which gave no detectable amount for any of the metals tested (Table 2). Calcium levels were recorded from all collection sites (Table 4). These values could be of importance as baseline data as the zebra mussel population increases.

Physical parameters for temperature, dissolved oxygen, pH, conductivity, and oxidation reduction potential were also collected when sampling (Table 3). The pH readings are of interest as all but two were slightly acid.

This data should provide a beginning set of baseline data plus identify potential areas for future study. Appendix A has the collection and handling procedures while Appendix B the spectrophotometer analysis procedures. No action levels have been established for zebra mussels. If we use levels for fish, it would appear there is presently no reason for concern. Zebra mussels could be put in landfills with present levels but this should be reevaluated during 1991. It may be of interest to monitor several other heavy metals and selected organic compounds.

From this initial effort it is felt that there is the need to establish a statistical sampling format from the data present in Table 1. Close surveillance should be made of the zebra mussel population in Presque Isle Bay. The zebra mussels were not large enough to provide an adequate sample for analysis during the summer of 1990. This area could have a high concentration of heavy metals, and it may be important to check several other heavy metals. Also, it may be important to monitor some of the organic compounds.

We wish to thank those groups and agencies that funded this research. We developed our sampling and analysis techniques and obtained some baseline data. This would not have been possible without the quick response which was given by various individuals. Also, thanks specifically to James Samuels who conducted all of the laboratory work and most of the collections, the many students that assisted us with little or no remuneration, Penn State-Behrend for the use of their facilities and equipment, and especially Dr. A. Pulsifer for his support.

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FIGURE 1

Heavy Metal Sampling

of Zebra Mussels

Erie County Pennsylvania

Lake Erie

1 Thompson Bay
2 Sommerheim water intake
3 Lampe Marina
4 Raccoon Creek
5 Walnut Creek
6 McCords Point (Shipwreck)
7 Freeport Beach
8 Presque Isle stone jetty
9 Horseshoe Pond

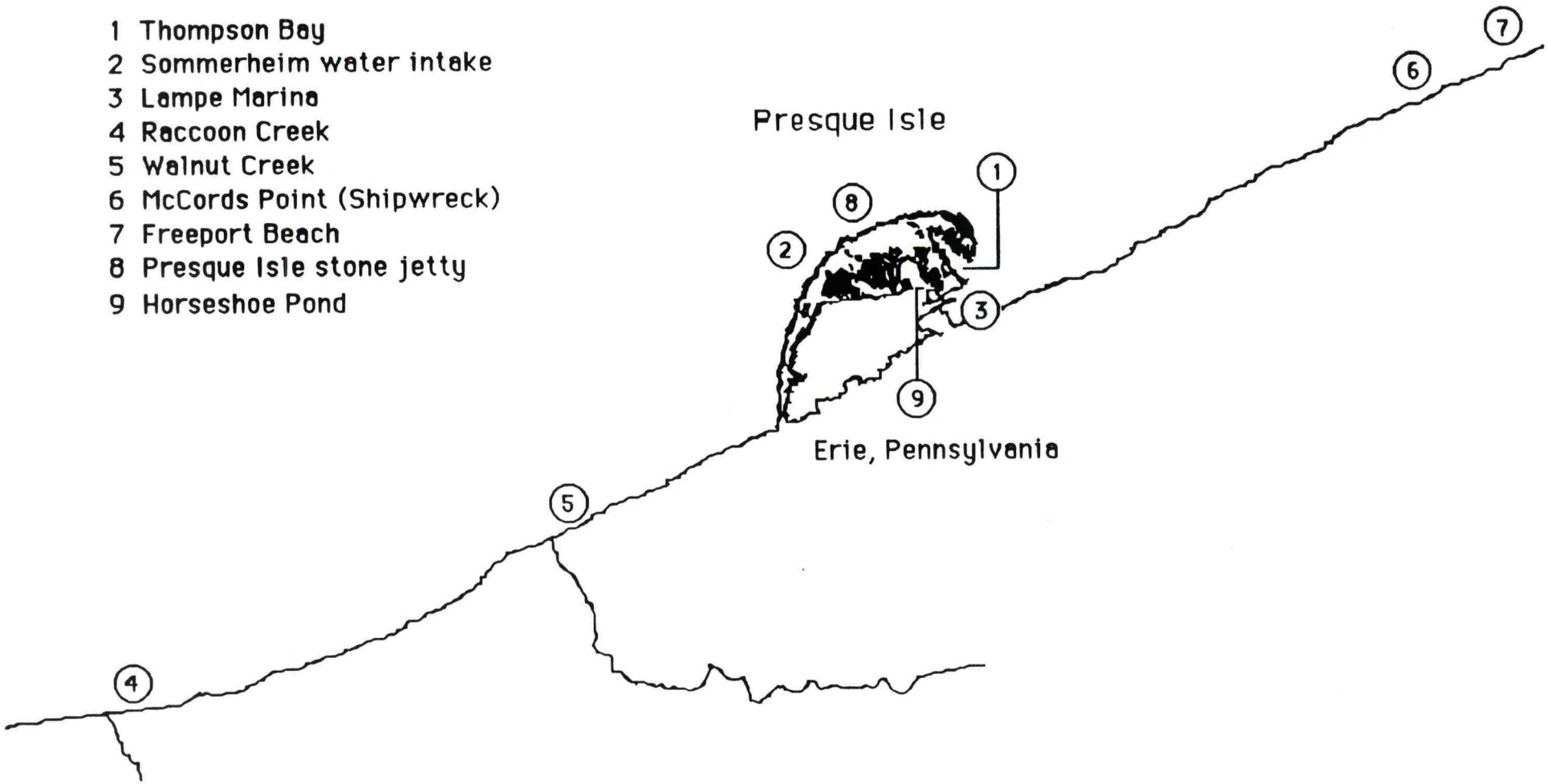
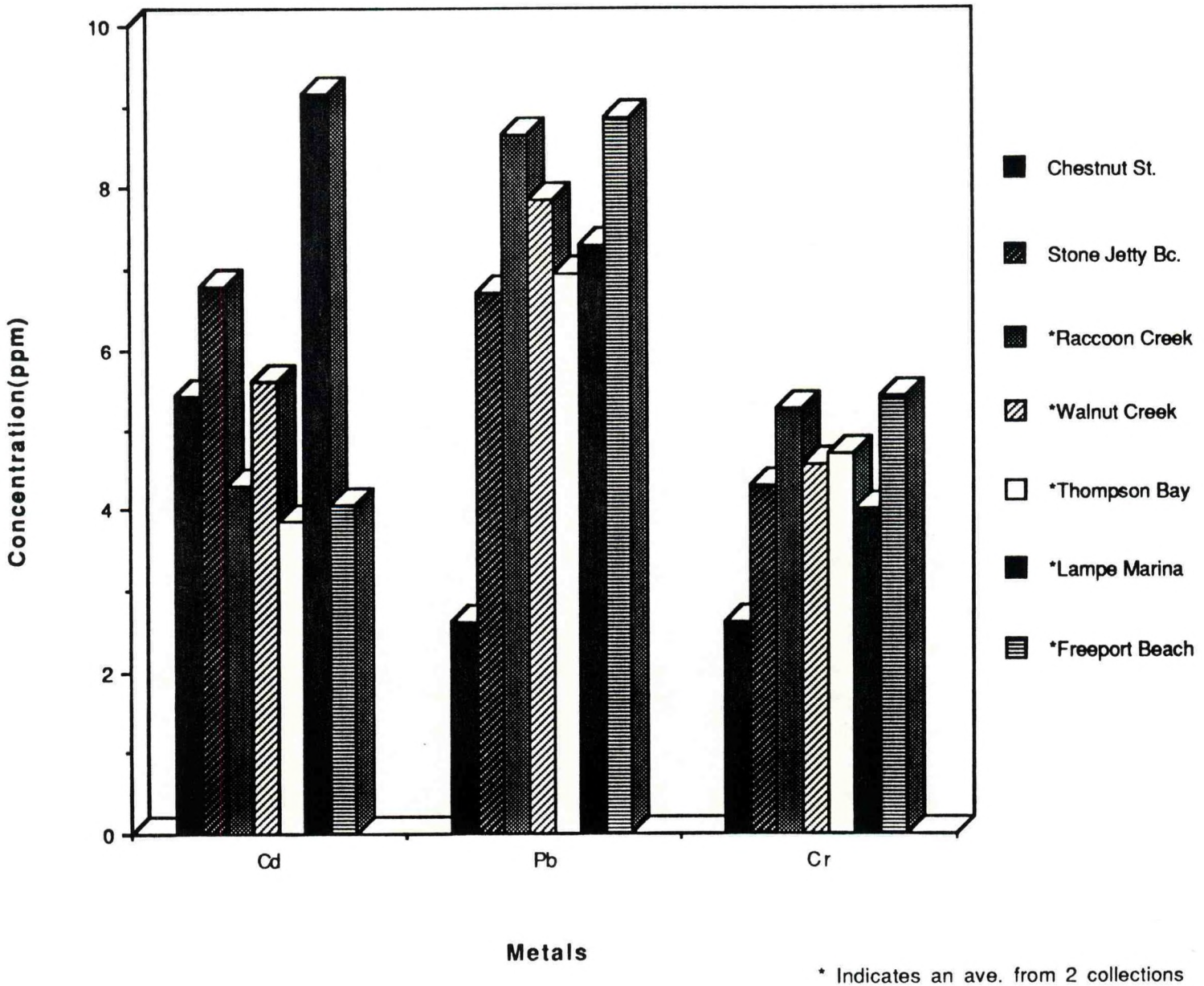


Figure 2

Comparison of Heavy Metal Concentrations in Zebra Mussels From Different Locations in Lake Erie



* Indicates an ave. from 2 collections

Table 1.

Total Metals Analysis of Zebra Mussels From Erie Lakeshore Sites From West to East

Concentrations (in ppm) are based on dried tissue only, shells and byssal threads are not included.

Sample Site	Date (1990)	Size (cm)	# of Batches	Cd (+/-)	Pb (+/-)	Cr (+/-)	Zn (+/-)
Mouth of Raccoon Ck.	14 August	1.2	6 X 1g	4.9(0.3)	8.0(2.0)	4.1(0.5)	.....
	3 October	1.2	2 X 1g	3.7(0.3)	9.3(2.0)	6.4(0.5)	104(1.0)
Mouth of Walnut Ck.	14 August	1.2	7 X 1g	5.9(0.3)	7.5(2.0)	3.8(0.5)	.....
	3 October	1.3	6 X 1g	5.3(0.3)	8.2(2.0)	5.3(0.5)	108(1.0)
Chestnut St. Water Intake	10 July	1.4	5 X 1g	5.4(0.3)	2.6(2.0)	2.6(0.5)	.....
*Stone Jetty Beach	12 Sept.	1.5	8 X 1g	6.8(0.3)	6.7(2.0)	4.3(0.5)	.....
*Thompson Bay	13 June	0.8	6 X 1g	3.6(0.3)	4.5(2.0)	3.4(0.5)	.....
	8 Sept.	1.4	6 X 1g	4.1(0.3)	9.4(2.0)	6.0(0.5)	.....
*Horseshoe Pd. Houseboat	18 Sept	1.3	3 X 1g	2.8(0.3)	192.0(2.0)	47.7(0.5)	.....
(Note: Houseboat hull was coated with paint, possibly lead based)							
Lampe Marina, Wayne St.	24 August	1.7	7 x 1g	10.1(0.3)	6.6(2.0)	3.8(0.5)	.....
	18 Sept	2.1	3 X 1g	8.2(0.3)	8.0(2.0)	4.2(0.5)	.....
Freeport Beach, N.E.	23 August	1.0	3 x 1g	4.6(0.3)	9.4(2.0)	6.3(0.5)	112(1.0)
	3 Oct	1.1	2 X 1g	3.5(0.3	8.3(2.0)	4.5(0.5)	.....

*Located at Presque Isle State Park

Table 2

Other Areas

Sample site	Date Collected	# Tested	Cd (+/-)	Pb (+/-)	Cr (+/-)
Detroit Edison Pwr., MI	26 June 1990	5 X 1gram	1.9 (0.3)	6.6 (2.0)	3.6 (0.5)
Lake St. Clair, MI	18 April 1990	2 X 1gram	4.2 (0.3)	6.1 (2.0)	4.6 (0.5)

Water Quality Network 601 Analysis From Erie County Health Dept.
For Water Taken From Chestnut Street Water Intake In Lake Erie

Date Collected	Pb	Ca
5 February 1990	<4ppb	.....
7 March 1990	<4ppb	.....
3 April 1990	<4ppb	34.7 ppm
3 May 1990	<4ppb	32.0 ppm
6 June 1990	<4ppb	34.7 ppm

Table 3

Hydrolab Readings

Sample	Date	DO*	Temp	pH	Cond$^{\Delta}$	ORP†	Depth
Raccoon Creek	8/14/90	10.7	21.35	6.60	0.294	0.218	0.6m
Walnut Creek	8/14/90	7.96	23.45	7.38	0.309	0.158	1.1m
Fisher Drive	9/08/90	10.10	21.71	6.65	0.343	0.283	0.0m
		9.50	21.73	6.79	0.341	0.279	1.0m
Thompson Bay	9/08/90	10.65	22.63	6.50	0.347	0.257	0.0m
		10.32	21.97	6.67	0.345	0.250	0.8m
W. of Perry Mon.	9/11/90	12.72	23.57	7.20	0.349	0.286	0.0m
Horseshoe Pond	9/18/90	10.05	16.87	6.65	0.361	0.360	0.9m
		10.08	17.12	6.73	0.362	0.353	0.2m
Lampe Marina	9/18/90	9.33	17.30	6.14	0.405	0.349	1.5m
		10.17	17.52	6.27	0.406	0.339	0.0m

Key

*Dissolved Oxygen units in ppm or mg/l

Δ Conductivity units in μS/cm

† Oxidation Reduction Potential in mv

Table 4

Calcium Analysis of Local Bodies of Water (in ppm)

Sample site	Date Collected	Ca	(+/-)
Mouth Raccoon Ck., Springfield Township	14 August 1990	41.6	(0.4)
Mouth Walnut Ck., Millcreek Township	14 August 1990	51.4	(0.4)
Thompson Bay, Presque Isle St. Park	20 July 1990	50.6	(0.4)
	8 Sept. 1990	48.6	(0.4)
Marina Entrance, Presque Isle St. Park	20 July 1990	47.6	(0.4)
Leo's Landing, Presque Isle St. Park	20 July 1990	58.4	(0.4)
Long Pond, Presque Isle St. Park	5 Ocotober 1989	42.8	(0.4)
Kanty Pond, City of Erie	2 November 1989	80.4	(0.4)
Lake Pleasant, Erie County	15 June 1990	50.8	(0.4)
	11 Sept. 1990	39.12	(0.4)
Stone Jetty Beach, Presque Isle St. Park	12 Sept. 1990	53.2	(0.4)
Freeport Beach, North East, PA	23 Aug 1990	60.6	(0.4)

Appendix A. Zebra Mussel Handling and Analysis

Collection:

1. Sediment samples were collected in acid washed polyethylene bottles whenever mussels were collected from clams or rocks near the sea floor. Collect 200 grams.
2. Water samples were collected in polyethylene bottles and fixed with 2ml of concentrated Nitric Acid/ 100ml. collect 500 ml.
3. Zebra mussels were collected from rocks and other surfaces by scraping with stainless steel spatulas or knives, taking care not to crush the shells. The mussels were transported to the lab in plastic bags from the site. 300 grams were collected.
4. The hydrolab readings were taken wherever mussels were obtained.
5. All samples were transported in coolers containing ice packs.

Processing:

1.Enough mussels were selected to fill a 600 ml beaker. This is enough to obtain at least 7 grams of dried flesh.
2. Mussels to be processed were rinsed with deionized water to remove mud and loose particles clinging to shells.
3. Mussels were heated in DI water on a hot plate without boiling until they popped open.
4. Excess water was drained off and mussels were shucked placing the wet flesh into cleaned, acid washed, petri dishes. Shells were put into labeled beakers for measuring.
5. Flesh and shells were dried in an oven at 105 Celsius overnight.
6. Dried flesh was ground with a mortar and pestle and placed in oven to dry completely.

Digestion:

1. At least Six, one-gram portions of powdered flesh, when available, were weighed out to two decimal places but weights were recorded to three places.
2. One digestion spike was prepared for every collection site, as well as a digestion blank. Samples were spiked as follows: .8ppm Pb, Cd, Cr .
3.10 ml of 1:1 HNO_3(Analytical grade or best available) was added to each sample. These were allowed to react at room temperature for one hour.
4.Beakers were placed on hot plates and refluxed for 15 min. at 95 Celsius.
5.Samples were taken off the burners and Allowed to cool for 10 min., 5 ml of concentrated HNO_3 was added and the watch glasses were replaced and the samples were refluxed for 30 minutes. Samples were taken off the burners and allowed to cool for10 min. 5 more ml HNO_3 were added and the samples were refluxed 30 min.. Using the watch glass, the solutions were evaporated on the hot plate to about 5 ml.
6. The samples were removed from the hot plate and allowed to cool. 2 ml of DI water was added and 3 ml of 30% H_2O_2. The samples were placed on the hot plate at the same setting as in step 4 with the watch glasses covering the beakers. The beakers were cooled and 2 more ml of peroxide were added and The samples were heated again until effervescence subsided. This process was repeated until a maximum of 10ml was used.
7. After the final aliquot of peroxide was added and effervescence subsided, the beakers were removed from the hot plates and allowed to cool. 5ml of concentrated HCl and 10ml of DI were then added. The beakers were returned to the hot plate and the contents were evaporated down below 20 ml without boiling.
8. Samples were filtered into 25ml volumetric flasks and filters and beakers were rinsed with DI into the flasks. Flasks were diluted up to the mark with DI.

Appendix B. AA Analysis Using the Perkin Elmer 2380

Cadmium:

1. One 2ppm standard was used to calibrate the instrument. Standards were remade every month by diluting 1000 ppm stock solutions from Fischer scientific.
2. A separate .2ppm (AA readings of .18- .22 acceptable) check solution was prepared using different glassware.
3. A Fischer multi-element lamp was used as the source.
4. All standards contained 1% HNO_3(purest grade available) to maintain ions in solution.

Chromium:

1. The Burner head was lowered an additional 1/4 turn to compensate for the change in the flame mixture for this anylate.
2. While the instrument was running in the continuous mode, the signal was on absorbance, and the flame was lit on normal settings, a 1 ppm standard was aspirated and the fuel flow was adjusted until there was a hint of yellow in the flame and the absorbance reading was .056.
3. Two standards were used: 1ppm, 2 ppm, and a .5 ppm check solution was prepared (readings of .48-.52 acceptable). The flame was checked periodically to see if the 1ppm standard was giving an absorbance of .056.

Lead:

1. A 2 ppm standard was used to calibrate and a 1 ppm check solution was read after every fifth sample (readings of .95-1.05 ppm are acceptable).
2. The same multi-element lamp was used as for Cadmium.
3. Lead has a small signal to noise ratio and therefore we recalibrated more frequently because small changes in operating conditions over time lead to significant changes in absorbance. A distilled water blank was read after every sample reading and the instrument was auto-zeroed whenever the blank read higher or lower than +/- .02ppm.

Calcium:

1. Standard additions method was used.
2. All Standards, blanks, and samples contained LaO_3 as well as 1% HNO_3.
2. To make up the LaO_3 solution 12.7 grams of LaO_3 and 250 ml of Conc. HCl were added to about 100 ml of distilled water in a 500 ml volumetric flask. Thus was diluted to the mark with distilled water.
3. 50 ml of the water sample were pipetted into a 100 ml volumetric flask. Included in this flask were 10 ml of LaO_3 solution and 1 ml of concentrated HNO_3. The flask was diluted to the mark and inverted 20 times.
4. 1ml of 100 ppm Calcium standard was pipetted into a 10ml volumetric flask and diluted to the mark with the sample prepared in step 3.
5. a 10% solution of LaO_3 in distilled water was prepared to be used as a blank.
6. After setting up the AA and allowing burner head to heat up for 3 min. or more, we aspirated distilled water and then Auto Zeroed on the sample prepared in step 3.
7. Next, the 10 ppm standard prepared in step 4 was aspirated and assigned a concentration of 10 ppm by the instrument.
8. Finally, distilled water prepared in step 5 was read .
9. This reading is the negative of the concentration of your diluted sample.