RELATIONSHIPS BETWEEN THE PARTITIONING OF LEAD IN SEDIMENTS AND ITS ACCUMULATION IN THE MARINE MUSSEL, *MYTILUS EDULIS* NEAR A LEAD SMELTER

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ABSTRACT. Surficial sediment samples and specimens of the filter-feeding marine mussel, *Mytilus edulis*, were collected at thirteen sites near a Pb/Zn ore stockpile and smelting complex (Canada). Aside from measuring the total S content in the sediment samples, each of these samples were subjected to a sequential extraction procedure designed to determine the partitioning of Fe and Pb among various geochemical phases and compared with the Pb levels measured in the mussel tissues. About one third of the Fe and more than 90% of the Pb were extracted from the sediments. Sediments collected within Belledune Harbour generally yielded the greatest amount of Pb whereas the highest levels of S were measured in Dalhousie Harbour sediments. Regression analysis indicated that the total S content in the sediments played an important role in defining the Pb accumulation in the mussel tissues.

1. INTRODUCTION

Realistic prediction of the impact of trace metals in sediments upon aquatic organisms requires an understanding on how physical and chemical factors affect metal uptake by organisms. These factors are not completely understood, and simple relationships are seldom found in natural systems between trace metal levels in the organisms and total metal concentrations in either the sediment or the water to which the organisms are exposed (Luoma, 1989).

A previous study (Bourgoin, 1990) has shown that the tissue Pb levels in blue mussels collected near a Pb smelter on New Brunswick's north shore (Canada), were significantly correlated with the Pb levels in the suspended particulate material in the water column. The objective of this study was to identify factors influencing Pb bioavailability to the common blue mussel, *Mytilus edulis*, collected near Pb/Zn ore stockpile and smelting facilities.

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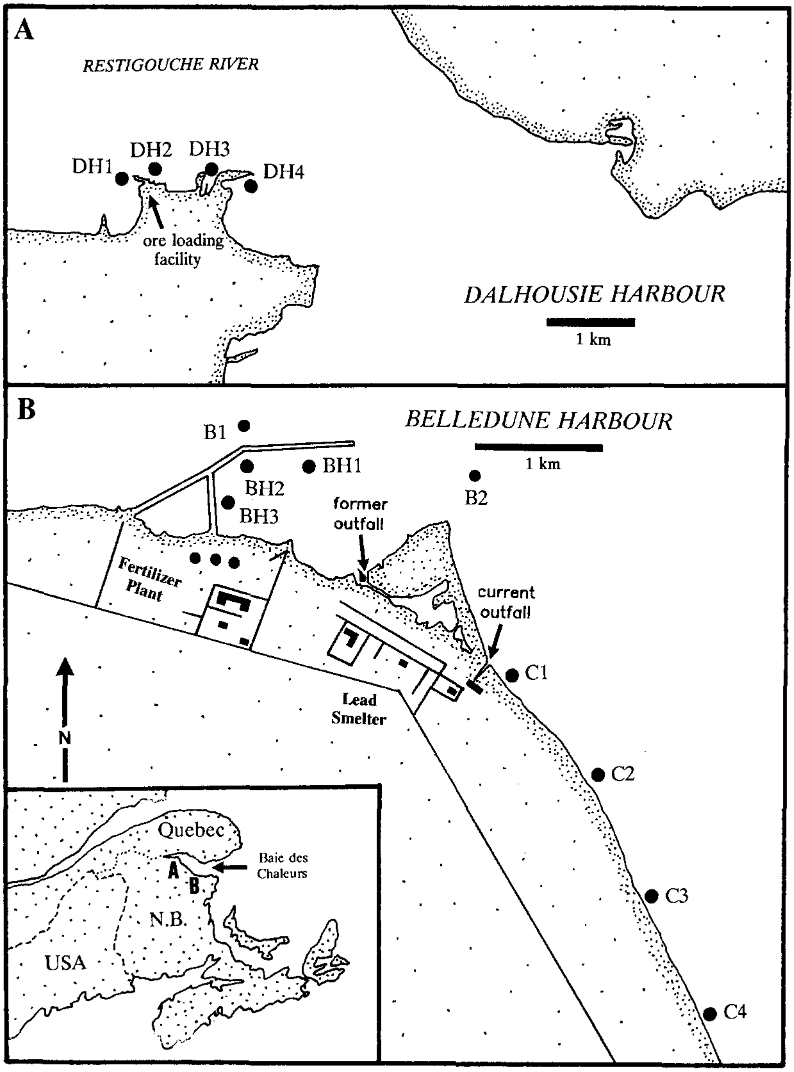

Figure 1. Location of the study area (inset). A) Sampling sites in Dalhousie Harbour; ore stockpiling and loading facilities are located on the western portion of the harbour. B) Sampling sites in the Belledune Harbour area.

2. Materials and Methods

2.1 SAMPLE COLLECTION

A total of 13 mussel and sediment samples were collected in the Baie des Chaleurs on the New Brunswick north shore (Figure 1, inset). Four sampling stations were located at Dalhousie Harbour (DH1 - DH4; Figure 1A) and 3 were in Belledune Harbour (BH1 - BH3; Fig 1B). The remaining 5 stations were located near Belledune; 2 exterior of the harbour (B1 & B2) and 4 coastal stations south of Belledune Harbour at 1000 m intervals (C1 - C4).

The sediment samples were collected by a diver using a clean acrylic boxcore (0.1 m^2). The boxcore was brought to the surface and the water within the boxcore was slowly drained out. The superficial sediment layer (top 5 mm) was sampled with a Teflon spatula, transferred in acid-cleaned polyethylene containers and frozen at -20 °C until further processing in the laboratory 14 days later.

Mussels greater than 50 mm (shell length) were collected in nylon bags by divers who avoided any specimens anchored to metallic structures. Special attention was taken to select mussels from a single clump to ensure environmental homogeneity (Lobel et al., 1982). The mussels were allowed to depurate their gut content in ambient seawater for 48 hr and stored frozen at -20 °C until required for analysis.

2.2 SEDIMENT ANALYSES

The sediments samples were wet sieved through 100 μm polyethylene mesh with dilute seawater, the slurry centrifuged at 1200 g for 20 min, after which the supernatant was discarded and the sediments were immediately sequentially extracted for Fe and Pb or dried at 60 °C.

2.2.1 *Sequential Extraction.* The sequential extraction procedure was designed to partition the metals into the following 5 fractions:

Fraction 1: exchangeable metal; sediment sample was extracted for 30 min with 0.5 M $MgCl_2$ at pH 7.

Fraction 2: metals bound to carbonates or specifically adsorbed; residue from fraction 1 was leached for 5 hr with 1 M NaOAc adjusted to pH 5 with HOAc.

Fraction 3: metals bound to Fe-Mn oxides; residue from fraction 2 was extracted with 0.04 M $NH_2OH.HCl$ in 25% HOAc at 96 °C.

Fraction 4: metals bound to organic matter and sulfides; residue from fraction 3 was extracted for 5 hr with 30 % H_2O_2 (85 °C) adjusted at pH 2 with 20 % HNO_3.

Fraction 5: "residual metals"; the residue from fraction 4 was extracted with aqua regia at 96 °C.

Detailed experimental procedures for the first 4 fractions and that of fraction 5 are given in Tessier et al,. (1979, 1980) and Nriagu et al., (1979), respectively. The Pb and Fe concentrations in the extracts were determined by flame atomic absorption spectrometry. Samples yielding low amounts of Pb were determined by flameless AAS. Background correction was used for all Pb analyses but not for Fe.

2.2.2 *X-ray Fluorescence Spectrometry.* Dried sediment subsamples were pressed into pellets and analyzed by X-ray fluorescence spectrometry (Phillips PW 1450) to determine the total Fe, Pb and S content in the sediments. The accuracy of the analyses was checked by running two Canadian reference standards, BCSS-1 and MESS-1, and a South African standard, NIM-G.

The sediment C content was determined with a Leco Carbon Analyzer (model WR-12). Inorganic C was assumed to be equal to the difference between total and organic C concentrations, the latter being measured after acid treatment with 6N HCl for 30 min.

2.3 MUSSEL ANALYSES

Similar size mussels (65 ± 3 mm, shell length) were selected and thawed at room temperature. The bivalves were shucked using stainless steel instruments, the tissues dried (70 °C), weighed and ground in an agate mortar. A total of 30 mussels were pooled for each sampling station. Approximately 300 mg subsamples of the pooled samples were digested with concentrated HNO_3 (BDH, Aristar) in Teflon bombs (Borg et al., 1981). The tissue digests were analyzed on a Perkin-Elmer 603 flame atomic absorption spectrometer equipped with deuterium background correction. Tissues with low Pb concentrations were analyzed on a HGA graphite furnace assembly.

2.4 QUALITY CONTROL

To minimize contamination, all glassware in contact with the samples and standard were soaked in 7N HNO_3 for a minimum of 48 hr, rinsed 3 times with doubled distilled water (DDW), stored in 2N HNO_3 and rinsed again with DDW before use.

3. Results

3.1 TRACE METAL DISTRIBUTION IN THE SEDIMENTS

The results from the sequential extraction of Fe did not show any major differences among the sediment samples (Table 1). Except for fractions 3 and 4 which yielded comparable amounts of Fe, progressively more Fe was leached from the sediments as the extractant became more vigorous. Approximately twice as much Fe was extracted in fraction 5, than in the first four fractions (F1(Fe) < F2(Fe) < F3(Fe) ≈ F4(Fe) << F5(Fe)). Generally a third of the total Fe was extracted from the sediment samples. The distribution of a given metal among the various fractions obtained by the sequential extraction procedure does not necessarily reflect the scavenging action of discrete sediment phases, but rather should be considered as operationally defined by the methods of extraction (Tessier *et al.*, 1979).

Table 1. Tissue Pb concentrations in *Mytilus edulis* and physicochemical characteristics of the < 100 μm granulometric fraction of the sediments sampled in the Dalhousie-Belledune area. All results are based on tissue or sediment dry weight; [M(F1)] - [M(F5)] represent metal concentrations in fractions 1-5 following the sequence given in the text; [M(S5)] represents the sum of metal concentration for the five fractions; Percent Extracted M represents the percentage of metal extracted by the sequential extraction procedure as determined by ([M(S5)]/Total M) 100; Total M & Total S represents the total metal concentrations as determined by X-ray fluorescence spectrometry; $C_{organic}$ and $C_{inorganic}$ represent organic and inorganic carbon concentrations, respectively.

Site	Tissue Pb	[M(F1)]		[M(F2)]		[M(F3)]		[M(F4)]		[M(F5)]	
	(μg.g^{-1})										
	Mean ± std	Pb	Fe	Pb	Fe	Pb	Fe	Pb	Fe	Pb	Fe
DH1	16.4 ± 2.8	<0.5	11.5	83.2	243	85.3	7781	137.7	6719	188.8	10004
DH2	10.8 ± 1.7	<0.5	9.4	45.1	174	101.8	5226	157.6	6297	203.2	9831
DH3	8.8 ± 1.4	<0.5	13.6	21.4	185	117.6	6248	105.4	6923	55.9	11924
DH4	9.3 ± 1.2	<0.5	8.4	27.2	223	122.4	6528	98.6	7184	61.8	8451
BH1	428.5 ± 68.1	<0.5	15.1	107.3	201	130.0	4336	270.3	5529	218.2	13959
BH2	387.4 ± 44.4	<0.5	11.1	141.1	245	153.7	5249	264.9	5779	326.5	14015
BH3	167.8 ± 33.7	<0.5	9.7	73.3	251	187.7	5306	175.8	6103	123.5	11398
B1	83.6 ± 15.9	<0.5	8.2	78.3	217	97.8	4812	86.1	5334	121.9	17396
B2	55.1 ± 15.1	<0.5	10.3	85.6	219	101.5	5193	115.4	5981	98.7	10944
C1	525.3 ± 79.2	<0.5	18.6	175.5	238	173.8	5784	216.3	6229	154.7	7024
C2	227.6 ± 36.0	<0.5	9.5	168.7	271	89.9	6872	100.8	6551	115.5	9813
C3	102.4 ± 27.5	<0.5	16.3	35.8	209	21.7	4195	51.0	5206	7.7	10093
C4	13.7 ± 2.7	<0.5	17.0	24.1	222	23.3	4378	41.8	5003	8.1	8996

Site	[M(S5)] (μg.g^{-1})		Total M (μg.g^{-1})		Percent Extracted M		$C_{organic}$	$C_{inorganic}$	Total S
	Pb	Fe	Pb	Fe	Pb	Fe	(%)	(%)	(%)
DH1	495	24759	531	67300	93%	37%	2.86	0.76	2.43
DH2	508	21537	551	63900	92%	34%	3.15	0.61	2.68
DH3	300	25294	352	66100	85%	38%	3.04	0.82	1.96
DH4	310	22394	345	70100	90%	32%	2.98	0.71	1.89
BH1	726	24040	725	63700	100%	38%	2.25	0.95	1.03
BH2	886	25299	882	71300	100%	35%	2.91	1.19	1.11
BH3	560	23068	558	64500	100%	36%	3.20	1.06	1.05
B1	384	27767	392	65900	98%	42%	2.83	0.99	0.87
B2	401	22347	422	65300	95%	34%	3.61	0.72	0.79
C1	720	19294	722	62100	100%	31%	3.19	0.66	0.92
C2	475	23517	486	71200	98%	33%	2.81	0.45	0.63
C3	81	20719	93	67700	87%	31%	3.00	0.58	0.75
C4	97	18616	104	63800	94%	29%	2.16	0.74	0.51

The superficial sediments collected within Belledune Harbour yielded approximately twice as much Pb than those collected outside of the harbour (B1 and B2) or at Dalhousie Harbour (Table I). No detectable amount of Pb was extracted from Fraction 1 in any of the sediment samples. Aside from Fractions 4 and 5 which yielded similar amounts of Pb, progressively more Pb was extracted from the sediments as the extractant became more vigorous ($F1(Pb) << F2(Pb) < F3(Pb) < F4(Pb) \approx F5(Pb)$). Over 90 % of the Pb was extracted from most of the sediment samples, with the highest percentage of Pb obtained in the sediments collected within Belledune Harbour and nearest to the active outfall (BH1-BH3 and C1).

The sediments collected at Dalhousie Harbour contained the highest S concentrations (Table I). The S levels in the samples collected nearest to the ore loading facility (DH1 and DH2; Figure 1) were slightly higher than the other two samples collected in the eastern portion of the harbour. In Belledune, the highest S concentrations were recorded in the sediments collected within the harbour. The organic C averaged about 5 times higher than the inorganic C content in the sediments. However, there was no discernable pattern for either of these two parameters (Table 1).

3.2 LEAD LEVELS IN THE TISSUES

The Pb concentrations in the mussels collected from Dalhousie Harbour decreased from west to east (Table 1). The mussels nearest to the ore loading facility in the western portion of the harbour (Figure 1) sequestered significantly ($p < 0.05$) higher Pb levels than the mussels collected from the eastern portion of Dalhousie Harbour. The tissue Pb levels in the mussels collected from the Belledune area were approximately 1 to 2 orders of magnitude higher than those collected within Dalhousie Harbour. The Pb levels in mussels collected from Belledune Harbour ranged from 2 to 9 times higher than the Pb levels in mussels sampled just outside of the harbour (B1 and B2). The highest tissue Pb concentration was recorded in mussels collected nearest to the current outfall, at station C1 (Figure 1). The Pb concentration in the mussels collected south of Belledune Harbour (C1 - C4) decreased rapidly in relation to their distance from the smelter outfall.

3.3 BIVALVE-SEDIMENT RELATIONSHIPS

Tissue Pb levels were first compared with the Pb concentrations in each of the four extractions ([Pb(F2)] to [Pb(F5)]) and with the partial sums of these concentrations ([Pb(S3)] to [Pb(S5)]). Little information was gained by using multiple regressions because of the high multicollinearity existing among the various sediment fractions. Therefore the concentrations in each sediment fraction or combination were normalized with respect to the iron oxide, organic and inorganic C, and total S content in the sediments. Bivariate analyses between the normalized parameters and the tissue concentrations were then performed. The correlation coefficient values (r) were calculated in each case and the data were examined graphically to verify the spread and linearity. The mean tissue concentrations were used for all of the calculations.

Significant correlations occurred between tissue Pb concentrations and sediment fractions 2 and 3 (Table 2). Correlation coefficients were not calculated with Pb(F1) as the values in this fraction were always below the detection limit (Table I). Lead extracted from the second fraction provided the highest correlation of the un-normalized sediments components (r: 0.821). This relationship was only slightly improved (r: 0.896) when Pb(F2) was normalized with respect to the Fe extracted from Fraction 3 (i.e., Pb(F2)/Fe(F3). In most cases, normalizing the Pb obtained from the various sediment fractions with respect to either the Fe or organic C content in the sediments did not greatly improve the correlations of the bivariate analyses. Conversely, the total S content within the sediments improved the predictive ability of Pb uptake by Mytilus edulis much more effectively (Table 2). Only 26 % of the variability was explained when tissue Pb was regressed with the Pb extracted from Fraction 4. However the highest correlation observed in this study (r: 0.915) resulted when Pb(F4) was normalized with the total S (Pb(F4)/tot S; Figure 2) increasing the explained variability to 86 %.

4. DISCUSSION

Based on the overall Pb levels in the sediments, it is not readily apparent why the mussels collected from Dalhousie Harbour sequestered 1 to 2 orders of magnitude less Pb than those collected from the Belledune region. When tissue Pb levels are plotted against the Pb extracted from either Pb(F2) or Pb(F4) however, the Dalhousie data points plot below the general relationship

Table 2. Correlation coefficients between tissue Pb in *Mytilus edulis* and Pb in sediments. Values significantly different from zero at the 5% (regular type) and 1% (italicized) levels are distinguished from those not significantly different from zero at the 5% level (in parentheses). Sum 4 = F2 + F3 + F4.

Tissue Pb *vs*	Pb(F*X*)	Pb(F*X*)/ Fe(F3)	Pb(F*X*)/ C_{org}	Pb(F*X*)/ Total S
Fraction 2	*0.821*	*0.896*	*0.833*	*0.874*
Fraction 3	(0.311)	(0.382)	(0.336)	*0.726*
Fraction 4	0.511	0.599	0.527	*0.916*
Fraction 5	(0.423)	(0.412)	(0.321)	0.646
Sum 4	0.579	*0.756*	*0.611*	*0.901*

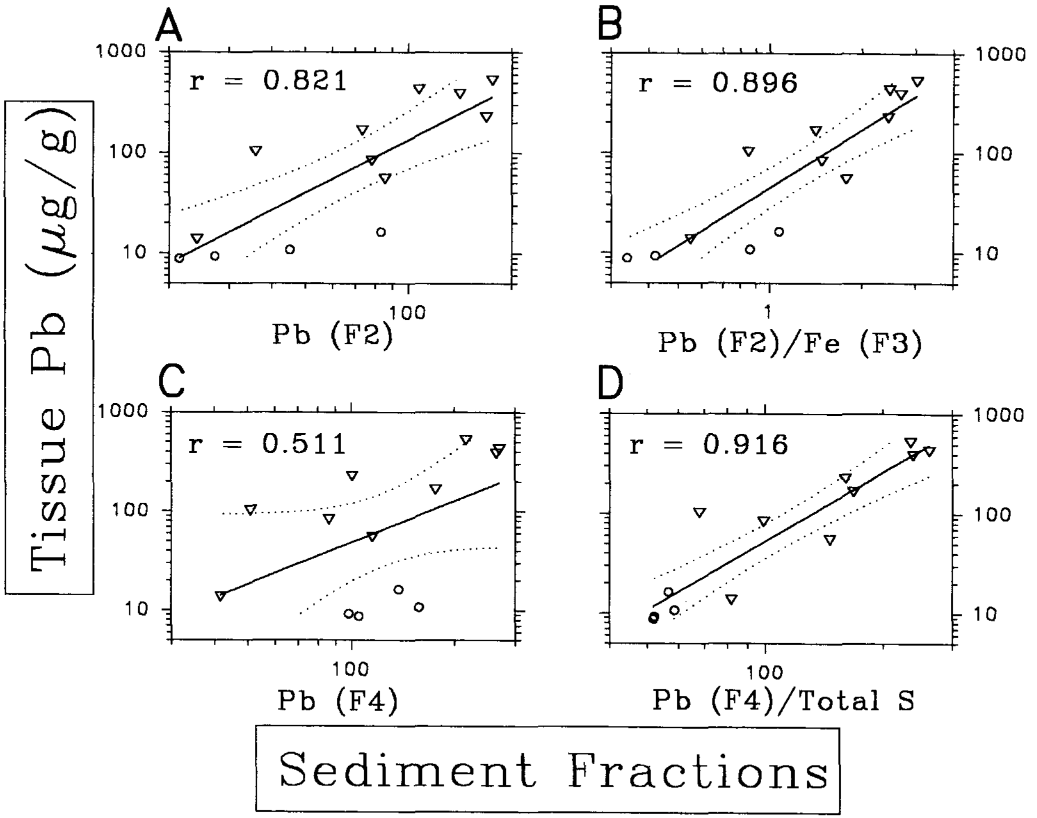

Figure 2. Relationship between the Pb concentrations in Mytilus edulis and various fractions of sediments collected in the Dalhousie-Belledune area (dotted lines represent 95% confidence limits). Circles represent data from Dalhousie Harbour; triangles represent data from the Belledune region. **A)** Lead concentrations in the tissues and in Fraction 2 (Y = 1.8X - 1.4). **B)** Lead concentrations in the tissues and the ratio [Pb(F2)]/[Fe(F3)] (Y = 1.9X + 1.7). **C)** Lead concentrations in the tissues and in Fraction 4 (Y = 1.4X - 1.1). **D)** Lead concentrations in the tissues and the ratio [Pb(F4)]/[Total S] (Y = 2.3X - 2.9).

suggesting that the Pb in Dalhousie sediments are comparatively less available to the indigenous mussel population (Figure 2 A&C). Ferguson (1983) observed that Pb compounds from smelter effluent were labile or weakly complexed and displayed a higher bioavailability. Others (Ray et al., 1981) have shown the Pb and other sedimentary trace metals near Dalhousie Harbour had a lower bioavailability because much of it occurred as sulphide. The isolated cluster created by the Dalhousie data points when tissue Pb is plotted against the Pb extracted from the sulphide-organic phase (Pb(F4)) supports this observation (Figure 2C).

Extracting the sediments and normalizing with respect to sediment components (e.g. Fe(F3), tot S)) increased the explained variance in tissue Pb over that explained by simply using the extracted metal value. This was particularly true for the total S content which when used as a normalizing factor, yielded significant correlations for all 4 extracted fractions (Table 2). The effectiveness of this sediment component in increasing the predictive ability of Pb uptake in Mytilus edulis is illustrated in Figure 2D in which the isolated cluster of the Dalhousie data points in Figure 2C has been drawn into the general relationship.

Others (Luoma and Bryan, 1978; Tessier et al., 1984) have suggested that the increase in the correlation coefficient values when the trace metal concentrations extracted from the sediments were normalized with respect to particular sediment fraction implied a "protective" or "competitive" role for that particular sediment fraction. While this explanation may also apply in this study, we believe that normalizing the Pb levels in fraction 4 with the total S content corrected for the large amount of Pb extracted from the sulphide fraction. Its been noted that even a weak acid leach such as 1N HCl may overestimate the Pb bioavailability in sulphide-rich sediments (Bourgoin, in press). It must be stressed however, that the partitioning of Pb among the various fractions is operationally defined and that others (Rapin et al., 1986) have noted that the selectivity of the extraction procedure used in this study deteriorates markedly in sulphide-rich (anoxic) sediments. While only oxic sediments were analyzed in this study, the high sulphide content due to the ore concentrate does warrant some caution to the selectivity of the procedure.

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