INFLUENCE OF SPECIES AND SEX ON METAL RESIDUES IN FRESHWATER MUSSELS (F. UNIONIDAE) FROM THE ST. LAWRENCE RIVER, WITH IMPLICATIONS FOR BIOMONITORING PROGRAMS

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MANAGEMENT PERSPECTIVE

Marine bivalves have long been used as sentinel organisms in "mussel watch" programs around the world. The goals of these programs are to assess the levels of contamination in coastal areas and warn of potentially hazardous conditions, and to support environmental regulation and enforcement processes. The success of these programs can largely be attributed to the amount of effort expended on developing sound sampling and analytical protocols. Freshwater mussels have similar potential for monitoring in freshwater systems, but to date they have been under-utilized and there are no standard protocols available for their use.

Studies on marine mussels have shown that biological factors can significantly influence the bioaccumulation of contaminants by these organisms, and must be standardized or accounted for in the design of mussel monitoring programs. Otherwise, spatial or temporal trends in pollution may be masked by biological variability. Similar studies on freshwater mussels are virtually absent. The present study investigated the effects of species and sex on metal residues in *E. complanata* and *L. r. radiata* (Bivalvia: Unionidae) from the St. Lawrence River. The impetus behind the study was the requirement for biomonitoring techniques identified in the St. Lawrence Action Plan and the Cornwall, Ontario and Massena, New York Remedial Action Plans for the St. Lawrence River Area of Concern. Under these Plans, industries throughout the river have been targeted for reductions in their discharge of toxic effluents, and hazardous waste sites are scheduled for cleanup. Mussel biomonitoring may be an effective means of demonstrating the success of remedial activities and tracking improvements in the river over time.

Mussels were collected in 1989 and 1990 from sites representing a wide range of types and degrees of metal pollution. In 1989, composite samples of five specimens (normally three males and two females combined)/species/site were analyzed for residues of 12 metals in their soft tissues to determine the effects of species on metal accumulation. In 1990, males and females were analyzed separately to determine the importance of sex as a source of variability in the data. Interspecific differences in bioaccumulation were observed for most metals, however, concentrations were frequently correlated between species and the differences could therefore be quantified. *E. complanata* demonstrated a broader response range to the same exposures than *L. r. radiata* for most metals, suggesting that it may be more sensitive to changes in pollution status. Differences in metal uptake between the sexes were less pronounced than differences between species, and male specimens displayed less variability than females. Consideration of these factors in mussel biomonitoring programs should greatly improve precision and sensitivity.

Abstract - The implementation of freshwater mussel watch programs has been hindered by a lack of information on biological factors affecting the levels of contaminants accumulated by these organisms. This study investigated the influence of species and sex on metal residues in *Elliptio complanata* and *Lampsilis radiata radiata* (F. Unionidae) from the St. Lawrence River. Mussels were collected from sites representing a wide range of types and degrees of metal pollution. Composite samples of five specimens (males and females combined)/species/site and five specimens/sex/species/site were analyzed for residues of 12 metals in the soft tissues to determine the effects of species and sex, respectively, on variability in the data. Interspecific differences in bioaccumulation were observed for most metals, however, concentrations were frequently correlated between species and the differences could therefore be quantified. *E. complanata* demonstrated a broader response range to the same exposures than *L. r. radiata* for most metals, suggesting that it may be more sensitive to changes in pollution status. Differences in metal uptake between the sexes were less pronounced than differences between species, and male specimens displayed less variability than females. Consideration of these factors in mussel biomonitoring programs should greatly improve sensitivity and precision.

Keywords - bioaccumulation metals freshwater mussels unionids biomonitoring

INTRODUCTION

Marine bivalves have long been used as sentinel organisms for monitoring contaminant levels in coastal areas around the world. In the United States, for example, "mussel watch" programs at the national level [1,2] and in the state of California [3] have been in place since the mid-1970s using mussels and oysters. Similar programs are under development in Australia [4] and South Africa [5], and have also been proposed for Canada [6]. All mussel watch programs share two common objectives, namely, to assess the level of contamination in coastal areas and to support environmental regulation and enforcement processes [3]. The success of these programs can largely be attributed to the development of sound sampling and analytical protocols.

The main advantages of using marine bivalves as monitoring organisms include the following: wide distributions of closely related species maximize data comparability; sedentary habits ensure site-specific information; large, stable populations permit repeated sampling; specimens are readily sampled, handled and identified; tolerances to many contaminants are high compared with other aquatic organisms, bioconcentration factors are high and bivalves provide a measure of contaminant bioavailability near the entry level of the food chain [1,2]. As these characteristics also apply to freshwater bivalves [7], similar mussel watch programs might be developed for freshwater systems. In fact, zebra mussels have recently been used to monitor inland waterways in Europe [8] and the exotic Asiatic clam, *Corbicula* sp., has been recommended for large-scale monitoring in the thirty-three American states in which it now occurs [9].

Most freshwater bivalves belong to the family Unionidae. Unionids are widely distributed in all types of freshwater systems, particularly large rivers, and have been used with increasing frequency as contaminant biomonitors [7]. Unfortunately, there have been few systematic attempts to develop standardized protocols for their use.

The accumulation of metals by mussels is highly dependent on the physico-chemical factors which affect exposure, and also on biological factors which determine the organisms' response to exposure. Variability due to biological factors can mask spatial or temporal patterns in metal pollution, and must therefore be eliminated or accounted for in the design of biomonitoring programs. The purpose of this study was to compare metal accumulation by the two dominant species of unionids in the St. Lawrence River, and to investigate the importance of sex as a source of intraspecific variability in metal residues. The impetus behind the study was the requirement for biomonitoring techniques identified in the St. Lawrence Action Plan [10], and the Cornwall, Ontario and Massena, New York Remedial Action Plans for the St. Lawrence River Area of Concern [11,12]. Under these Plans, industries throughout the river have been targeted for reductions in their discharge of toxic effluents, and hazardous waste sites are scheduled for cleanup. Mussel biomonitoring may be an effective means of demonstrating the success of remedial activities and tracking improvements in the river over time.

METHODS

Study sites

In 1989, 11 sites in four stretches of the St. Lawrence River were selected for study (sites 1-5 and 7-12, Fig.1). In 1990, five of these sites were revisited (sites 3, 4, 7, 8 and 9) and a new site (13) was added. Study sites were chosen to represent a variety of types and degrees of metal pollution, such that inter- and intraspecific differences in metal accumulation by mussels could be determined over a wide range of exposures. With the exception of sites 1 and 11, which represented the background influence of Lake Ontario and the Ottawa River, respectively, all sites were located immediately above or below point sources of metal pollution. These point sources are identified by industrial sector in Fig. 1. All of the industries would be classified as major polluters. Industry P is a sulphate (kraft) paper mill with the second highest daily flow of process effluent (126,000 m^3/d) among Ontario's 27 pulp and paper mills [13]; industry O was recently identified as the most significant discharger in Ontario's organic chemical manufacturing sector, with loadings of Zn, Hg and Pb greatly exceeding provincial water quality objectives [14]; industries A1, A2 and A3 are scheduled for cleanup under USEPA Superfund administrative orders [12]; industries E1, E2, F1, F2 and I are among 50 industries in the Québec portion of the river that have been targeted for effluent reductions [10]. The latter three industries alone account for nearly half of the total metal loadings to the St. Lawrence River between Cornwall and Sorel, especially Pb (97%), Ni (80%), Fe (67%), Cr (61%), Zn (55%) and Cu (34%) [15].

Study organisms

Elliptio complanata (Subf. Ambleminae) and *Lampsilis radiata radiata* (Subf. Lampsilinae) co-occurred in roughly equal proportions at most of the sites sampled in 1989, accounting for 97% of all mussels encountered. Therefore, the study focused on these two dominant species. All unionids are filter-feeders. Tessier et al. [16] determined that *E. complanata* feeds on particles $< 70\ \mu m$ in size, but no information is available for *L. r. radiata*. Both species are long-lived, with maximum life-spans of 14 to 19 years reported for *E. complanata* [17-20] and 11 to 16 years for *L. radiata* [17,21]. According to Clarke [22], *L. r. radiata* prefers gravel and sand substrates whereas *E. complanata* may be found on gravel, sand, clay or mud. In the present study, divers observed greater densities of *L. r. radiata* in zones of higher velocity and larger proportions of *E. complanata* further inshore (L.C. Grapentine, personal observation).

L. r. radiata is dioecious and sexually dimorphic [22]. Sexual allocation in *E. complanata* is more complicated, with dioecious, hermaphroditic and sequentially hermaphroditic organisms co-existing at ratios that may vary among populations [23]. Downing et al. [23] determined that 94% of specimens in Lac de l'Achigan, 60 km north of Montréal, were either true males or females (20%) or functional males or females (defined as having $< 10\%$ or $> 90\%$ female tissue in their gonads, respectively). As females of both species are gravid in late spring or early summer [22,24], and as sampling was conducted during the third week of June in both years of this study, it was relatively easy to separate the sexes. Fully gravid specimens of *E.*

complanata were considered to be females, whereas non-gravid specimens were designated as males. All specimens of *L. r. radiata* with a female shell shape were found to be gravid.

Collection of mussels and preparation of samples for metals analysis

Mussels were collected by SCUBA divers from all sites except 7 and 8 in 1989, where oyster tongs were used due to poor visibility. In 1989, attempts were made to obtain 10 specimens of each species from each of the 11 study sites. The only basis for selection in the field was that very small (< 6.0 cm shell length) and very large (> 10.0 cm) individuals were not taken. Mussels were rinsed clean of sediment using river water, wiped dry with Kimwipes®, wrapped in pre-fired (550°C) and hexane-rinsed aluminum foil, placed in plastic food storage bags and immediately frozen on dry ice. This procedure was used because samples were destined for analysis of both organic and inorganic contaminants. As mussels remained tightly closed, there was no contact between soft tissues and foil. In the laboratory, mussels were thawed for 30 min, then opened, sexed and shucked individually into acid-washed glass jars and weighed. Five specimens were pooled for analysis, which is the minimum number recommended for determining trace metal trends in the U.S. Mussel Watch Program [25]. Two females and three males/species/site were composited, with the following exceptions: only four specimens of *E. complanata* were available from site 2 and two from site 12, and all were males; no female *L. r. radiata* were available from sites 10 and 11 and only one was available from site 8, whereas only two males of this species were available from sites 7 and 12. Composite samples were homogenized for 3 to 5 min using a stainless steel blender, and subsamples of 20 to 25 g were freeze-dried.

In 1990, five male and five female *L. r. radiata* and 15 *E. complanata* (the latter could not be sexed in the field) were collected from each of the six study sites, and samples consisting of five specimens/sex/species/site were composited for analysis. For *E. complanata*, the first five males and females shucked were taken. Sample handling and preparation followed the methods described above except that specimens were combined after freeze-drying rather than before, and composites were ground for 1 min in a Bel-Art Micro-Mill® with stainless steel blades and grinding chamber.

In both years, mussels were frozen immediately after collection without permitting them to clear their digestive tracts. In most studies which consider whole-body burdens of metals, organisms are depurated before analysis in order to eliminate any bias due to the presence of metals in the gut contents. This may be particularly important for small animals such as insects, where the gut contents represent a significant proportion of the body weight [26,27]. Tessier et al. [16] estimated that the intestinal tract and its contents accounted for about 10% of the total body weight in *E. complanata*. If this were a major contributor to whole-body burdens, one would expect concentrations in the viscera to be high relative to other organs. In fact, concentrations of metals in the viscera of undepurated freshwater mussels from both clean and contaminated sites are consistently lower than those in all other organs [28-31]. Two studies that directly compared body burdens in depurated vs. non-depurated mussels found that concentrations of Cd, Cu, Ni and Zn in *Mytilus edulis* and Cd in *E. complanata* did not change after 2d of gut-clearing [32,33]. This suggests that undigested material in the gut contains a negligible portion of the whole body content of metals, and that gut purging may not be necessary for these organisms. Several studies have shown that depuration may itself be a source of error due to the partial elimination of some biologically-incorporated metals during the depuration period. Metals with short biological half-lives, such as Cu, may be quickly excreted from tissues [34]. Losses of Mn from the mantle/gonad complex of *M. edulis* from a relatively clean site [32] and Cd from the gills and kidneys of *E. complanata* from an experimentally-dosed lake [33] were demonstrated after only 2d of depuration.

Analytical methods

Mussel samples were analyzed for metal residues by Environment Canada's National Laboratory for Environmental Testing (NLET), Burlington, Ontario, using standard procedures described in their Analytical Methods Manual [35]. Briefly, the analytical methods and associated detections limits (DLs) on a μg/g dry weight basis for the tested elements were: Hg - cold vapour atomic absorption (AA) spectroscopy, DL = 0.03; As and Se - atomic emission spectroscopy using an inductively coupled argon plasma system, DL = 0.50 for both elements; remaining metals - direct aspiration AA spectroscopy, DLs = 0.20 (Cd), 0.50 (Ni), 1.0 (Pb), 2.0 (Cr, Cu, Zn), 10.0 (Fe, Mn) and 50.0 (Al).

Lead was not detected in the 1989 samples using the above analytical method, due to interference from high concentrations of Fe and Mn in mussel tissues (Jacques Carrier, NLET, personal communication). Therefore, samples were reanalyzed using graphite furnace AA spectroscopy, which is appropriate for both Cd and Pb (DLs = 0.01 and 0.20, respectively). Three of the 22 samples could not be reanalyzed due to insufficient material. Lead was detected in all other samples. Using a paired-difference test to compare concentrations of Cd determined by both methods, no significant difference was observed ($t = 2.08$, $df = 18$, $p < 0.05$). Thus, Cd values obtained using the original method were reported as they were available for all 22 samples. The graphite furnace technique was subsequently applied to the 1990 samples for analysis of both Cd and Pb.

Samples were analyzed in accordance with the routine quality assurance (QA) procedures of the NLET, which include duplicate analyses to determine sample homogeneity, analysis of three reference materials to determine accuracy, spike-recovery tests to assess interference, and analysis of blanks to determine contamination due to laboratory procedures. Samples which do not meet the QA objectives are reanalyzed, and those which still do not meet the standards are rejected. No samples from this study had to be rejected. However, results for As were less than optimum in terms of accuracy and interference, and should therefore be interpreted with less confidence. Complete QA reports are available from the author.

Statistical analyses

Concentrations of 12 metals in composite samples of *E. complanata* and *L. r. radiata* (sexes combined) collected from 11 sites on the St. Lawrence River in 1989 were examined to determine: 1) whether there were interspecific differences in the tissue concentrations of any metals, 2) whether concentrations accumulated by the two species were correlated, regardless of any differences in actual levels, thus indicating the same site-to-site trends in metal bioavailability and 3) whether both species demonstrated the same range of responses to the tested exposures, thus offering similar capacities for site discrimination. Interspecific differences were determined using a paired-difference test, where concentrations of a given metal in both species from the same site constituted a pair; the test statistic was $t = \bar{d} - 0/(SD \div \sqrt{n})$, where $\bar{d}$ = mean difference in concentrations among n study sites. Significance of correlation in metal concentrations between species was determined using a t-test; the test statistic was $t = r \sqrt{(n-2)} / \sqrt{(1-r^2)}$, where r = correlation coefficient. Where correlations were significant, the slopes (β) of the regression lines relating concentrations of metals in *L. r. radiata* (y) to those in *E. complanata* (x) were used to indicate relative capacities for site discrimination as follows: where $\beta = 1.0$, the magnitude of the difference between species remained constant over all exposures, therefore both species demonstrated the same range of responses; where $\beta < 1.0$, the response of *L. r. radiata* was proportionately lower than that of *E. complanata*; where $\beta > 1.0$, the response of *E. complanata* was proportionately lower than that of *L. r. radiata*.

Concentrations of 12 metals in composite samples of male and female *E. complanata* and *L. r. radiata* (sexes separate) collected from 6 sites on the St. Lawrence River in 1990 were examined to determine the importance of sex as a source of variability in tissue metal concentrations. A paired-difference test was again used to determine differences in metal concentrations between species for each sex and between sexes for each species, while a t-test was used to determine the significance of correlation in metal concentrations between species and sexes.

RESULTS

Interspecific comparisons of metal concentrations in mussels - 1989

E. complanata accumulated significantly higher concentrations of Al, Cr, Fe, Hg and Ni than *L. r. radiata*, while *L. r. radiata* accumulated significantly higher concentrations of As, Cu, Mn and Zn (Table 1). Differences were highly significant ($p < 0.01$) for all metals except As. Concentrations were higher in the more contaminated species by average factors ranging from 1.2 to 2.5 X, depending on the metal. There were no significant differences between species for Cd, Se and Pb.

Correlations between concentrations in *E. complanata* and *L. r. radiata* were highly significant for Al, Cd, Cu, Fe and Pb, significant for Cr, Hg and Ni ($p < 0.05$) and not significant for As, Mn, Se and Zn (Table 2). If the information from Tables 1 and 2 is combined, four possible relationships with implications for biomonitoring programs emerge. These relationships are described below and illustrated with examples in Fig. 2, where study sites are arranged in descending order of contamination based on concentrations of metals in *E. complanata*:

1). Concentrations do not differ between species and are significantly correlated (Cd, Pb). In this situation, mussels of either species could be used interchangeably to monitor trends in metal bioavailability. The slopes of the regression lines relating concentrations in the two species should be approximately 1.0, and this was true for Cd (Fig. 2a). However, concentrations of Pb were substantially lower in *L. r. radiata* at one of the eight sites tested, and this resulted in a β of 0.53 for this metal.

2). Concentrations differ between species and are significantly correlated (Al, Cr, Cu, Fe, Hg, Ni). In this case, a regression equation could be used to convert concentrations in one species to those in the other. For Cu, β was approximately 1.0, indicating that the difference between species remained fairly constant over all exposures (Fig. 2b). For the remaining metals, *E. complanata* demonstrated a broader range of responses than *L. r. radiata*. The difference in response ranges was least pronounced for Fe (β = 0.74) and most pronounced for Hg (β = 0.24, Fig. 2c).

3). Concentrations do not differ between species and are not correlated (Se, Fig. 2d). This situation could occur if interspecific differences in bioaccumulation capacity for Se were minor and exposure levels were similar at all sites, or if Se were regulated to similar levels by both species over a range of exposures.

4). Concentrations differ between species and are not correlated (As, Mn, Zn). In this case, the two species appear to provide conflicting information on the bioavailability of these elements among the study sites. Concentrations of As (Fig. 2e) and Mn were higher in *L. r. radiata* at most sites, but by varying amounts. For Zn (Fig. 2f), *L. r. radiata* clearly displayed a broader range of responses than *E. complanata*; the lack of correlation between species was likely due to the nearly constant response of the latter species at all sites.

Sex as a source of variability in metal concentrations in mussels - 1990

Interspecific differences in tissue metal concentrations observed in 1989 without standardizing for sex were generally confirmed in 1990 when the sexes were tested separately (Table 3). Due to the smaller number of sites sampled in 1990 (6 vs. 11), a greater mean difference between species was required to achieve statistical significance in the second year. Nevertheless, differences between species were significant for Hg, Ni and Zn and not significant for Pb in both years, including both sexes in 1990. Significant differences observed for Al, As, Cr, Cu, Fe and Mn in 1989 were again observed for at least one sex in 1990, with the exception of Cu. In 1990, concentrations of Cd and Se were significantly higher in *L. r. radiata* vs. *E. complanata* males and in *E. complanata* vs. *L. r. radiata* females, respectively, whereas differences between species were not significant for either metal in 1989. For *E. complanata*, differences in metal concentrations between the sexes were not significant, with the exception of higher concentrations of Cu in females (Table 3). For *L. r. radiata*, however, concentrations of Cd, Fe, Se and Zn were significantly higher in male specimens.

Correlations between species were significant for Cd, Fe and Pb and not significant for As and Mn in both sexes (Table 4), confirming the findings from the previous year. Concentrations of Al, Cr and Ni were significantly correlated between species in 1989 and also in 1990 for males, but not females. Significant correlations observed for Cu and Hg in 1989 were not observed in either sex in 1990. No correlations between species were observed for Se or Zn in 1989; in contrast, correlations were significant for Se in males and for Zn in both sexes in

1990. Correlations between sexes were significant for Cd, Fe, Pb and Zn in both species and also for Al, Cr, Cu and Ni in *E. complanata* and for Hg and Mn in *L. r. radiata* (Table 4).

If Tables 3 and 4 are considered together, the implications of these findings for biomonitoring programs become more evident. The various relationships are illustrated with examples in Fig. 3. To facilitate comparisons among the four species/sex combinations, sites are arranged throughout in descending order of contamination based on concentrations of metals in male *E. complanata*. Concentrations of Cd, Fe, Pb and Zn were significantly correlated between species for each sex and between sexes for each species, therefore all mussels indicated the same site-to-site trends in bioavailability for these metals. As there were no significant differences in actual concentrations for Pb, mussels of either species or sex could be used interchangeably as biomonitors for this metal. Concentrations of Cd, Zn and Fe were significantly higher in male than female *L. r. radiata*, but did not differ between the sexes for *E. complanata*. Therefore, regression equations would be needed to convert concentrations in one sex to those in the other for *L. r. radiata*, whereas male and female *E. complanata* could be interchanged. Concentrations of Cd (Fig. 3a) and Zn were significantly higher in males of *L. r. radiata* than *E. complanata* and Zn was also higher in females. Concentrations of Fe (Fig. 3b) were higher in females of *E. complanata* than *L. r. radiata*. In these cases, regression equations would be needed to convert concentrations in one species to those in the other.

Concentrations of Al (Fig. 3c), Cr and Ni were significantly correlated between species for males, but not females. Actual levels did not differ between the sexes for either species, but were significantly correlated for *E. complanata* only. Thus, males of both species and female *E. complanata* indicated the same trends among sites, while female *L. r. radiata* deviated. As concentrations of these metals were significantly higher in male *E. complanata* than male *L. r. radiata*, regression equations would be required for conversion. Concentrations of Se (Fig. 3d) did not differ between species and were highly correlated when males were compared. No other correlations were observed, therefore male mussels of either species could be used interchangeably to monitor Se bioavailability among sites, but females deviated from each other and from males of the same species.

Mercury concentrations were higher in *E. complanata* than *L. r. radiata* of both sexes, whereas Mn levels (Fig. 3e) were higher in *L. r. radiata*. Otherwise, the two metals showed similar relationships: concentrations were not correlated between species for either sex, did not differ between the sexes for either species and were correlated between the sexes for *L. r. radiata* only. Thus, only male and female *L. r. radiata* indicated the same site to site trends. Conversely, the only correlation observed for Cu was between male and female *E. complanata*, with the latter accumulating higher concentrations. Arsenic concentrations were higher in male *L. r. radiata* than male *E. complanata* (Fig. 3f). No correlations and no other differences occurred, therefore each species and sex described a different trend for As.

DISCUSSION

The two dominant species of unionids in the St. Lawrence River, *E. complanata* and *L. r. radiata*, differed in their bioaccumulation capacities for most metals. Concentrations of Al, Cr, Fe, Hg and Ni were significantly higher in *E. complanata* collected from 11 sites along the river in 1989, whereas concentrations of As, Cu, Mn and Zn were significantly higher in *L. r. radiata* and no differences were observed for Cd, Pb or Se. After standardizing for sex in a subsequent interspecific comparison at six sites in 1990, differences were again significant for Hg, Ni and Zn and not significant for Pb in both sexes. Trends observed for Al, As, Cr, Fe and Mn in 1989 were also observed in 1990, although differences were not significant in both sexes. Lack of statistical significance could be partly due to the smaller number of sites sampled in 1990. For Cd and Se, however, differences between species were apparent only when the sexes were tested separately. In an earlier study on the Ottawa River, which is a major tributary to the St. Lawrence River, Metcalfe-Smith et al. [36] also reported significantly higher concentrations of Cr and Ni in *E. complanata*, As, Cu and Zn in *L. radiata*, and no differences between the species for Cd. Mercury and Se residues appeared to be higher in *E. complanata*, but only at one site, and Al, Fe, Mn and Pb were not analyzed. Only two other studies have directly compared metal accumulation by these two species [37,38], and both were conducted in less contaminated systems. Their findings were in agreement for Cd, Cr and Zn. However, Friant [37] observed opposite trends for both Cu and Hg, and Heit et al. [38] found that concentrations of most metals did not differ between species.

In the Ottawa River study [36], concentrations of As, Cr, Cu, Ni and Zn varied between species by factors ranging from 1.5 to 2.5 X. Similar factors of 1.2 to 2.5 X were determined for these metals in the present study. Maximum factors of 2 to 3 X are most often observed by investigators comparing metal uptake among various other freshwater mussel species, although differences of one to two orders of magnitude have occasionally been reported [36]. Factors of 1.2 to 2.5 X may not seem large, however, they are quite significant when compared with the magnitude of site-to-site differences in tissue residues. For most metals, concentrations in mussels of a given species varied over the study sites by factors ranging from only 2 to 10 X (Table 5). The ability to distinguish differences in bioavailability among sites would therefore be diminished if interspecific differences in accumulation were not accounted for in a biomonitoring program. To indicate the environmental conditions over which these conclusions are valid, the range of total metal concentrations in sediment are also presented in Table 5. Relationships between concentrations of metals in sediment and mussels are extremely complex [7,16], and will be the topic of a later paper.

Lobel et al. [39] collected two closely-related species of marine mussels, *M. edulis* and *Mytilus trossulus*, from a lagoon in Newfoundland and analyzed approximately 20 similar-sized individuals of each for residues of 25 elements. Concentrations of all elements except Mn were higher in *M. trossulus* by a factor of about 1.5 X. Because virtually all differences were in the same direction and of a similar magnitude, they were believed to be due to age and growth rate differences (*M. trossulus* are slower-growing and the specimens analyzed were older) rather than to any species-specific differences in metabolic capabilities. Using the same argument, differences in metal uptake between *E. complanata* and *L. r. radiata* in the present study could be attributed to metabolic factors because the species having the higher concentration varied from metal to metal and the magnitude of the difference also varied among metals. Furthermore, *E. complanata* displayed a broader range of responses over the 11 study sites for Al, Cr, Fe, Ni, Pb and especially Hg, where concentrations ranged from 0.07 to 0.33 μg/g vs. only 0.05 to 0.13 μg/g in *L. r. radiata* (Table 5). The latter species displayed a broader response range for only two metals, Mn and particularly Zn, where concentrations ranged from 190 to 511 μg/g vs. only 130 to 280 μg/g in *E. complanata*. These results suggest that in general *L. r. radiata* may be more capable of regulating metals than *E. complanata*. If so, it would be less suitable as an indicator of the availability of metals in the environment [40]. Life history factors could still contribute to the interspecific differences observed, however, little comparative information is available for these two species. Magnin et Stanczykowska [17] reported that *E. complanata* from Lac des Deux Montagnes, a riverine lake at the confluence of the Ottawa and St. Lawrence Rivers, grew more slowly and were somewhat longer-lived than co-occurring *L. radiata*. Perhaps residues of some metals are lower in *L. r. radiata* due to the effects of growth dilution. Alternatively, *E. complanata*'s preference for quieter waters suggests that it may feed on smaller, possibly more contaminated, food particles. More information is needed to identify the mechanisms responsible for interspecific differences in metal uptake.

Mussels collected from the St. Lawrence River during this study were in immediate pre-spawn condition. At this time of year, concentrations of Cu were higher in female than male *E. complanata* and concentrations of Cd, Fe, Se and Zn were higher in male than female *L. r. radiata*. No other differences were observed. In the only other study on a freshwater mussel, Jones and Walker [30] reported no differences in the accumulation of Cd, Fe, Mn or Zn by male vs. female *Velesunio ambiguus* from the River Murray in South Australia. Studies on a variety of filter-feeding marine bivalves have shown that concentrations of metals in both male and female specimens are highest immediately prior to spawning, but that differences between the sexes are at a minimum during this period. For example, Simpson [41] found that levels of Pb and Zn in *M. edulis* were highest before spawning and decreased over the summer. Lobel and Wright [42] also observed peak concentrations of Zn in this species just prior to spawning and attributed this to their poor nutritional status after overwintering. Orren et al. [43] reported that concentrations of Cu, Fe, Pb and Zn in both sexes, Mn in males and Cd in females of the black mussel, *Choromytilus meridionalis*, were higher prior to spawning than six months later. Levels of Cu, Fe, Mn and Zn in females were twice those in males at the latter date, but no differences were observed for any metal before spawning. Similarly, of eight metals analyzed in pre-spawn specimens of the hairy mussel, *Trichomya hirsuta*, only Cu was higher in one sex (female) than the other [4]. Lobel et al. [44] analyzed residues of 25 elements in post-spawn specimens of *M. edulis* from Bellevue, Newfoundland, and found that the majority had some association with sex. Of interest to the present study, concentrations of As, Cu, Mn, Se and Zn were higher in females, Pb was higher in males, and Al and Cd did not differ between species. In a subsequent study [45], Lobel et al. determined the relative effects of several biological factors on concentrations of 24 elements in *M. edulis*, and found that sex was a significant predictor for over half. They concluded that the sexes should be analyzed separately in mussel watch programs, and that it would be best to sample prior to spawning when element concentrations will be uniformly high and before dilution effects of the growing season lead to erratic values. Although these findings cannot be directly applied to freshwater mussel watch programs, they suggest that the sampling time chosen for the St. Lawrence River study may have been optimal in terms of maximizing response while minimizing individual variability.

Interspecific differences in the bioaccumulation capacities of mussels for metals do not necessarily preclude the use of more than one species in a monitoring program. Having a choice of several organisms would be advantageous in terms of broadening the geographic range of a program or ensuring better coverage of the available habitats. Actual concentrations accumulated by different species are less important than relative concentrations, i.e. provided that differences between species can be quantified over the range of conditions to be monitored, intercomparability can be achieved using conversion factors or regression equations. For example, in the U.S. Mussel Watch program [1] transplant experiments were used to calibrate the response of the east coast blue mussel, *M. edulis*, against that of the west coast California mussel, *Mytilus californianus*, thus permitting pollution trends to be evaluated at the national level.

In the present study, there were significant differences in the actual concentrations accumulated by *E. complanata* vs. *L. r. radiata* (sexes combined) for the majority of metals. However, concentrations were significantly correlated between species over the 11 study sites in most cases. It was determined that the two species could be used interchangeably to monitor site-to-site trends in the bioavailability of Cd and Pb, and after conversion by means of regression equations to monitor Al, Cr, Cu, Fe, Hg and Ni. Where a lack of correlation was observed (As, Mn, Se, Zn), further investigation would be required before the more appropriate species for biomonitoring could be chosen.

In a subsequent study at six St. Lawrence River sites, standardizing for sex was shown to have significant consequences for biomonitoring applications even during the pre-spawn period. Differences between species were more pronounced (i.e. higher level of significance, or significant vs. non-significant) when male specimens were compared for six of the tested metals and when female specimens were compared for only three. Paradoxically, correlations between species were significant for eight metals in males vs. only four in females. These findings imply that male mussels exhibit less individual variability in their body burdens of metals than female mussels, thus allowing the true differences between species and among sites to be more readily observed. There was evidence that most of the variability in the data for female mussels could be attributed to female *L. r. radiata*. If male specimens were considered, *E. complanata* and *L. r. radiata* could be used interchangeably to monitor site-to-site trends in bioavailability for Fe, Pb and Se, and with the application of appropriate conversion factors for monitoring Al, Cd, Cr, Ni and Zn. As female *E. complanata* could be directly exchanged with their male counterparts for all of these metals except Se, all three species/sex combinations would indicate the same trends. However, female *L. r. radiata* were significantly correlated with their male counterparts for only four of the metals, Cd, Fe, Pb and Zn, and could be directly substituted solely for Pb.

In general, results in terms of both interspecific differences in metal residues and correlations between species were similar in both years of this study. If sex were an important source of variability in the data, standardization for this factor might have been expected to improve statistical significance. Unfortunately, this effect would have been partly offset by the smaller number of sites sampled. Findings were more consistent between 1989 composites and 1990 males than females, and it should be noted that the 1989 composites were in fact dominated by male specimens. Of the 22 samples analyzed, 15 consisted of three males and two females, one included four males and one female, four contained males only, and just two consisted of more females (3) than males (2).

Agencies responsible for monitoring and assessment in this system would undoubtedly find it useful to be able to rank sites according to their relative contamination with metals. Relative ranks for the 1989 study sites based on residues of metals in *E. complanata*, *L. r. radiata* and sediment are presented in Table 6. As some sites were more contaminated with certain metals than others, it is acknowledged that considerable data reduction was used to generate overall rankings for each site. Nevertheless, there was good agreement between the two species. In contrast, ranks based on sediment contamination were very different from those for mussels. The bioavailability of metals to aquatic organisms depends on their partitioning and chemical forms in water and sediment and on the primary route of exposure (direct contact, respiration, ingestion), such that strong correlations between concentrations in organisms and sediment seldom occur. This illustrates the importance of analyzing living organisms rather than, or in conjunction with, abiotic components of the environment in order to determine biologically significant trends in metal pollution.

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REFERENCES

1. **Goldberg, E.D.** 1986. The mussel watch concept. *Environ. Monit. Assess.* **7**:91-103.
2. **Lauenstein, G.G.** and **T.P. O'Connor.** 1988. Measuring the health of U.S. coastal waters. *Sea Technology* **29**:29-32.
3. **Martin, M.** 1985. State mussel watch: toxic surveillance in California. *Mar. Pollut. Bull.* **16**:140-146.
4. **Klumpp, D.W.** and **C. Burdon-Jones.** 1982. Investigations of the potential of bivalve molluscs as indicators of heavy metal levels in tropical marine waters. *Aust. J. Mar. Freshwater Res.* **33**:285-300.
5. **Watling, H.R.** and **R.J. Watling.** 1976. Trace metals in *Choromytilus meridionalis*. *Mar. Pollut. Bull.* **7**:91-94.

6. **Sojo, L.E., M.C. Hamilton** and **D. Files.** 1990. National mussel watch program, Phase 1: Overview of existing mussel watch programs. Environment Canada, Environmental Protection, Dartmouth, Nova Scotia.

7. **Elder, J.F.** and **J.J. Collins.** 1991. Freshwater molluscs as indicators of bioavailability and toxicity of metals in surface-water systems. *Rev. Environ. Contam. Toxicol.* **122**:37-79.

8. **Mersch, J., A. Jeanjean, H. Spor** and **J.-C. Pihan.** 1992. The freshwater mussel *Dreissena polymorpha* as a bioindicator for trace metals, organochlorines and radionuclides. *Limnologie aktuell* **4**:227-244.

9. **Doherty, F.G.** 1990. The Asiatic clam, *Corbicula* spp., as a biological monitor in freshwater environments. *Environ. Monit. Assess.* **15**:143-181.

10. **Gonthier, C.** 1991. Bilan provisoire de la réduction des rejets des 50 industries du Plan d'action Saint-Laurent. Équipe d'intervention Saint-Laurent, Direction des services techniques, Montréal, Québec.

11. **St. Lawrence River Remedial Action Plan Team.** 1992. Remedial Action Plan for the Cornwall-Lake St. Francis Area, Stage I report: Environmental conditions and problem definition. Kingston, Ontario.

12. **New York State Department of Environmental Conservation.** 1990. St. Lawrence River at Massena Remedial Action, Stage I. Albany, NY.

13. **Ontario Ministry of the Environment.** 1991. Municipal-Industrial Strategy for Abatement (MISA), Preliminary report on the first six months of process effluent monitoring in the MISA pulp and paper sector (January 1, 1990 to June 30, 1990). Report No. PIBS 1436. Water Resources Branch, Toronto, Ontario.

14. **Ontario Ministry of the Environment.** 1992. Municipal-Industrial Strategy for Abatement (MISA), Six-month monitoring data report, organic chemical manufacturing sector (Period covered October 01, 1989 to March 31, 1990). Report No. PIBS 1815. Water Resources Branch, Toronto, Ontario.

15. **Environnement Canada.** 1985. Inventaire des connaissances sur les sources de pollution dans le fleuve Saint-Laurent, Tronçon Cornwall-Sorel. Service de la protection de l'environnement, Montréal, Québec.

16. **Tessier, A., P.G.C. Campbell, J.C. Auclair** and **M. Bisson.** 1984. Relationships between the partitioning of trace metals in sediments and their accumulation in the tissues of the freshwater mollusc *Elliptio complanata* in a mining area. *Can. J. Fish. Aquat. Sci.* **41**:1463-1472.

17. **Magnin, E.** et **A. Stanczykowska.** 1971. Quelques données sur la croissance, la biomasse et la production annuelle de trois mollusques Unionidae de la région de Montréal. *Can. J. Zool.* **49**:491-497.

18. **Ghent, A.W., R. Singer** and **L. Johnson-Singer.** 1978. Depth distributions determined with SCUBA, and associated studies of the freshwater unionid clams *Elliptio complanata* and *Anodonta grandis* in Lake Bernard, Ontario. *Can. J. Zool.* **56**:1654-1663.

19. **Strayer, D.L., J.J. Cole, G.E. Likens** and **D.C. Buso.** 1981. Biomass and annual production of the freshwater mussel *Elliptio complanata* in an oligotrophic softwater lake. *Freshwater Biol.* **11**:435-440.

20. **Wren, C.D.** and **H.R. MacCrimmon.** 1986. Comparative bioaccumulation of mercury in two adjacent freshwater ecosystems. *Wat. Res.* **20**:763-769.

21. **Nalepa, T.F.** and **J.M. Gauvin.** 1988. Distribution, abundance, and biomass of freshwater mussels (Bivalvia: Unionidae) in Lake St. Clair. *J. Great Lakes Res.* **14**:411-419.

22. **Clarke, A.H.** 1981. *The Freshwater Molluscs of Canada.* National Museums of Canada, Ottawa, Canada.

23. **Downing, J.A., J.-P. Amyot, M. Pérusse** and **Y. Rochon.** 1989. Visceral sex, hermaphroditism, and protandry in a population of the freshwater bivalve *Elliptio complanata. J. N. Am. Benthol. Soc.* **8**:92-99.

24. **Trdan, R.J.** 1981. Reproductive biology of *Lampsilis radiata siliquoidea* (Pelecypoda: Unionidae). *The American Midland Naturalist* **106**:243-248.

25. **Goldberg, E.D., M. Koide, V. Hodge, A.R. Flegal** and **J. Martin.** 1983. U.S. Mussel Watch: 1977-1978 results on trace metals and radionuclides. *Estuarine, Coastal and Shelf Science* **16**:69-93.

26. **Luoma, S.N.** 1983. Bioavailability of trace metals to aquatic organisms - a review. *The Science of the Total Environment* **28**:1-22.

27. **Smock, L.A.** 1983. The influence of feeding habits on whole-body metal concentrations in aquatic insects. *Freshwater Biol.* **13**:301-311.

28. **Anderson, R.V.** 1977. Concentration of cadmium, copper, lead, and zinc in six species of freshwater clams. *Bull. Environ. Contam. Toxicol.* **18**:492-496.

29. **Adams, T.G., G.J. Atchison** and **R.J. Vetter.** 1981. The use of the three-ridge clam (*Amblema perplicata*) to monitor trace metal contamination. *Hydrobiologia* **83**:67-72.

30. **Jones, W.G.** and **K.F. Walker.** 1979. Accumulation of iron, manganese, zinc and cadmium by the Australian freshwater mussel *Velesunio ambiguus* (Phillipi) and its potential as a biological monitor. *Aust. J. Mar. Freshwater Res.* **30**:741-751.

31. **V.-Balogh, K.** 1988. Heavy metal pollution from a point source demonstrated by mussel (*Unio pictorum* L.) at Lake Balaton, Hungary. *Bull. Environ. Contam. Toxicol.* **41**:910-914.

32. **Latouche, Y.D.** and **M.C. Mix.** 1982. The effects of depuration, size and sex on trace metal levels in bay mussels. *Mar. Pollut. Bull.* **13**:27-29.

33. **Malley, D.F., P.S.S. Chang** and **R.H. Hesslein.** 1989. Whole lake addition of cadmium-109:radiotracer accumulation in the mussel population in the first season. *The Science of the Total Environment* **87/88**:397-417.

34. **Lobel, P.B., S.P. Belkhode, S.E. Jackson** and **H.P. Longerich.** 1991. Sediment in the intestinal tract: a potentially serious source of error in aquatic biological monitoring programs. *Mar. Environ. Res.* **31**:163-174.

35. **National Laboratory for Environmental Testing.** 1992. Analytical methods manual, Volume 2, Group 2C, Trace metal analyses in fish. Environment Canada, Burlington, Ontario.

36. **Metcalfe-Smith, J.L., J.C. Merriman** and **S.P. Batchelor.** 1992. Relationships between concentrations of metals in sediment and two species of freshwater mussels in the Ottawa River. *Water Poll. Res. J. Canada* **27**:845-869.

37. **Friant, S.L.** 1979. Trace metal concentrations in selected biological, sediment, and water column samples in a northern New England river. *Water, Air, and Soil Pollution* **11**:455-465.

38. **Heit, M., C.S. Klusek** and **K.M. Miller.** 1980. Trace element, radionuclide, and polynuclear aromatic hydrocarbon concentrations in Unionidae mussels from northern Lake George. *Environ. Sci. Technol.* **14**:465-468.

39. **Lobel, P.B., S.P. Belkhode, S.E. Jackson** and **H.P. Longerich.** 1990. Recent taxonomic discoveries concerning the mussel *Mytilus*: implications for biomonitoring. *Arch. Environ. Contam. Toxicol.* **19**:508-512.

40. **Bryan, G.W.** 1979. Bioaccumulation of marine pollutants. *Phil. Trans. R. Soc. Lond. B.* **286**:483-505.

41. **Simpson, R.D.** 1979. Uptake and loss of zinc and lead by mussels (*Mytilus edulis*) and relationships with body weight and reproductive cycle. *Mar. Pollut. Bull.* **10**:74-78.

42. **Lobel, P.B.** and **D.A. Wright.** 1982. Gonadal and nongonadal zinc concentrations in mussels. *Mar. Pollut. Bull.* **13**:320-323.

43. **Orren, M.J., G.A. Eagle, H.F.-K.O. Hennig** and **A. Green.** 1980. Variations in trace metal content of the mussel *Choromytilus meridionalis* (Kr.) with season and sex. *Mar. Pollut. Bull.* **11**:253-257.

44. **Lobel, P.B., S.P. Belkhode, S.E. Jackson** and **H.P. Longerich.** 1989. A universal method for quantifying and comparing the residual variability of element concentrations in biological tissues using 25 elements in the mussel *Mytilus edulis* as a model. *Mar. Biol.* **102**:513-518.

45. **Lobel, P.B., C.D. Bajdik, S.P. Belkhode, S.E. Jackson** and **H.P. Longerich.** 1991. Improved protocol for collecting mussel watch specimens taking into account sex, size, condition, shell shape and chronological age. *Arch. Environ. Contam. Toxicol.* **21**:409-414.

Table 1. Differences in metal concentrations between *E. complanata* and *L. r. radiata* collected from the St. Lawrence River in 1989.

Metal	Species with higher concentration	Frequency (# sites)	t value	Average conversion factor[1]
Cr	*E. complanata*	11	5.73**	2.5X
Ni	*E. complanata*	10	5.55**	2.1X
Fe	*E. complanata*	11	4.36**	1.6X
Hg	*E. complanata*	11	4.00**	2.0X
Al	*E. complanata*	9	3.24**	1.3X
Se	*E. complanata*	8	2.01	-
Pb	*E. complanata*	5	1.21	-
Zn	*L. r. radiata*	11	4.57**	2.0X
Cu	*L. r. radiata*	9	4.39**	1.2X
Mn	*L. r. radiata*	10	3.55**	1.7X
As	*L. r. radiata*	8	2.47*	1.3X
Cd	*L. r. radiata*	7	1.10	-

study sites = 11 for all metals except Pb, where # study sites = 8.
*concentrations significantly higher at $p < 0.05$; $t_{crit} = 2.228$ (10 df), 2.365 (7 df).
**concentrations significantly higher at $p < 0.01$; $t_{crit} = 3.169$ (10 df), 3.499 (7 df).
[1]factor converts concentration in less contaminated species to concentration in more contaminated species.

Table 2. Significance of correlation between metal concentrations in *E. complanata* and *L. r. radiata* collected from the St. Lawrence River in 1989, and regression equations predicting concentrations in *L. r. radiata* (y) from those in *E. complanata* (x).

Metal	Correlation coefficient (r)	t value	Regression equation
Fe	0.98	14.77**	y = -205.16 + 0.74x
Pb	0.97	9.77**	y = 0.61 + 0.53x
Cd	0.95	9.13**	y = -0.10 + 1.13x
Al	0.84	4.64**	y = 91.00 + 0.53x
Cu	0.80	4.00**	y = 0.35 + 1.14x
Hg	0.71	2.98*	y = 0.04 + 0.24x
Cr	0.68	2.79*	y = 0.21 + 0.44x
Ni	0.64	2.50*	y = 0.45 + 0.39x
Zn	0.49	1.69	-
Mn	0.47	1.60	-
Se	0.40	1.31	-
As	0.15	0.46	-

study sites = 11 for all metals except Pb, where # study sites = 8.
*correlation significant at $p < 0.05$; $t_{crit} = 2.262$ (9 df), 2.447 (6 df).
**correlation significant at $p < 0.01$; $t_{crit} = 3.250$ (9 df), 3.707 (6 df).

Table 3. Differences in metal concentrations between male and female *E. complanata* and *L. r. radiata* collected from the St. Lawrence River in 1990.

	Species (sp.) or sex with higher concentration							
	E. complanata ♂ vs. *L. r. radiata* ♂		*E. complanata* ♀ vs. *L. r. radiata* ♀		*E. complanata* ♂ vs. ♀		*L. r. radiata* ♂ vs. ♀	
Metal	Sp.	t value	Sp.	t value	Sex	t value	Sex	t value
Cr	E	3.92*	E	2.53^+	♀	0.86^+	-	0.13
Ni	E	7.15**	E	3.05*	♀	1.87^+	♀	0.82^+
Fe	E	2.54^+	E	3.19*	♂	2.09^+	♂	2.83*
Hg	E	6.24**	E	4.13**	♂	2.27^+	-	0.00
Al	E	2.75*	E	1.31^+	♂	1.64^+	♀	1.91^+
Se	E	2.26^+	E	3.14*	♂	0.62^+	♂	5.83**
Pb	E	1.08^+	E	1.70^+	♀	0.60^+	-	0.96
Zn	L	8.52**	L	3.71*	-	0.64	♂	3.92*
Cu	L	1.92^+	E	1.63^+	♀	2.89*	♂	1.37^+
Mn	L	2.26^+	L	4.35**	♂	1.34^+	-	0.80
As	L	2.92*	L	1.90^+	-	0.80	♀	0.42^+
Cd	L	5.93**	L	1.16^+	-	0.25	♂	3.07*

E = *E. complanata*; L = *L. r. radiata*.
# study sites = 6.
$^+$concentrations higher at a majority of sites, but not statistically significant.
*concentrations significantly higher at $p < 0.05$; $t_{crit} = 2.571$ (5 df).
**concentrations significantly higher at $p < 0.01$; $t_{crit} = 4.032$ (5 df).

Table 4. Significance of correlation between metal concentrations in male and female *E. complanata* and *L. r. radiata* collected from the St. Lawrence River in 1990.

	E. complanata ♂ vs. *L. r. radiata* ♂		*E. complanata* ♀ vs. *L. r. radiata* ♀		*E. complanata* ♂ vs. ♀		*L. r. radiata* ♂ vs. ♀	
Metal	Correlation coefficient (r)	t value	Correlation coefficient (r)	t value	Correlation coefficient (r)	t value	Correlation coefficient (r)	t value
Fe	1.00	25.76**	0.99	14.04**	1.00	19.92**	1.00	19.92**
Pb	1.00	25.76**	1.00	14.04**	1.00	22.29**	1.00	14.04**
Cd	0.97	7.98**	0.90	4.13*	0.93	5.06**	0.98	9.85**
Al	0.91	4.64**	0.44	0.98	0.92	4.69**	0.62	1.58
Cu	0.10	0.19	0.16	0.32	0.86	3.37*	0.06	0.11
Hg	0.65	1.71	0.75	2.27	0.72	2.08	0.99	14.04**
Cr	0.89	3.90*	0.01	0.01	0.92	4.69**	0.47	1.06
Ni	0.98	9.85**	0.21	0.43	0.94	5.51**	0.32	0.68
Zn	0.89	3.90*	0.98	9.85**	0.97	7.98**	0.90	4.13*
Mn	0.20	0.41	0.40	0.87	0.47	1.06	0.85	3.23*
Se	0.96	6.86**	0.18	0.37	0.58	1.42	0.79	2.58
As	0.80	2.67	0.79	2.58	0.68	1.85	0.57	1.39

study sites = 6.
*correlation significant at $p < 0.05$; $t_{crit} = 2.776$ (4 df).
**correlation significant at $p < 0.01$, $t_{crit} = 4.604$ (4 df).

Table 5. Ranges of total metal concentrations ($\mu g/g$ dry weight) in *E. complanata*, *L. r. radiata* and sediment from the 1989 study sites.

Metal	Concentration range and magnitude of difference between lowest and highest value		
	E. complanata	*L. r. radiata*	Sediment[1]
Al	153 - 696 (5X)	146 - 534 (4X)	41800 - 70,000 (2X)
As	2.8 - 7.5 (3X)	4.2 - 8.6 (2X)	1.2 - 4.7 (4X)
Cd	0.4 - 7.1 (18X)	0.6 - 8.3 (14X)	0.20 - 0.85[2] (4X)
Cr	3.29 - 18.8 (6X)	2 - 12.6 (6X)	25.9 - 324 (12X)
Cu	7.8 - 13.6 (2X)	8.7 - 16.2 (2X)	3.91 - 148 (38X)
Fe	1140 - 9980 (9X)	771 - 7120 (9X)	19500 - 101000 (5X)
Hg	0.07 - 0.33 (5X)	0.05 - 0.13 (3X)	0.01 - 0.33 (33X)
Mn	1430 - 4690 (3X)	1740 - 7380 (4X)	365 - 733 (2X)
Ni	2.1 - 7 (3X)	1.2 - 3.9 (3X)	7.96 - 144 (18X)
Pb	0.88 - 13.2 (15X)	0.71 - 7.5 (11X)	6.56 - 81.3 (12X)
Se	2 - 5.2 (3X)	2 - 3 (2X)	0.2 - 0.8 (4X)
Zn	130 - 280 (2X)	190 - 511 (3X)	33.3 - 216 (6X)

[1] J.L. Metcalfe-Smith, unpublished data.
[2] concentrations of "extractable" metal (i.e. extracted with 0.5 N HCl) are reported for Cd in sediment.

Table 6. Relative ranks of the 1989 study sites in terms of overall metal contamination based on residues in *E. complanata* vs. *L. r. radiata* vs. sediment.

Sites ranked from most to least contaminated[1]		
E. complanata	*L. r. radiata*	Sediment
1	11	8
11	1]	7]
8	10]	10]
7	8	11
10	3	2
3	7]	5
9	9]	1
4	2	9
5	4	4
2	5	3

[1]overall ranks obtained from cumulative ranks for all metals except Pb (no data available for *E. complanata* from 3 sites) and all sites except 12 (no data available for metals in sediment); sites having the same ranks are connected by a bracket.

FIGURE LEGENDS

Fig. 1. Locations of the study sites (●) and point sources of metal pollution (▲). Point sources by industrial sector are: A = aluminum smelter, E = electroplating, F = foundry, I = inorganic chemical, O = organic chemical, P = pulp and paper, S = sewage treatment plant.

Fig. 2. Concentrations of selected metals in composite samples of *E. complanata* vs. *L. r. radiata* (sexes combined) collected from 11 sites on the St. Lawrence River in 1989. Sites arranged in descending order of contamination based on residues in *E. complanata*.

Fig. 3. Concentrations of selected metals in composite samples of male (m) and female (f) *E. complanata* (E.c.) vs. male and female *L. r. radiata* (L.r.r.) collected from six sites on the St. Lawrence River in 1990. Sites arranged in descending order of contamination based on residues in male *E. complanata*.

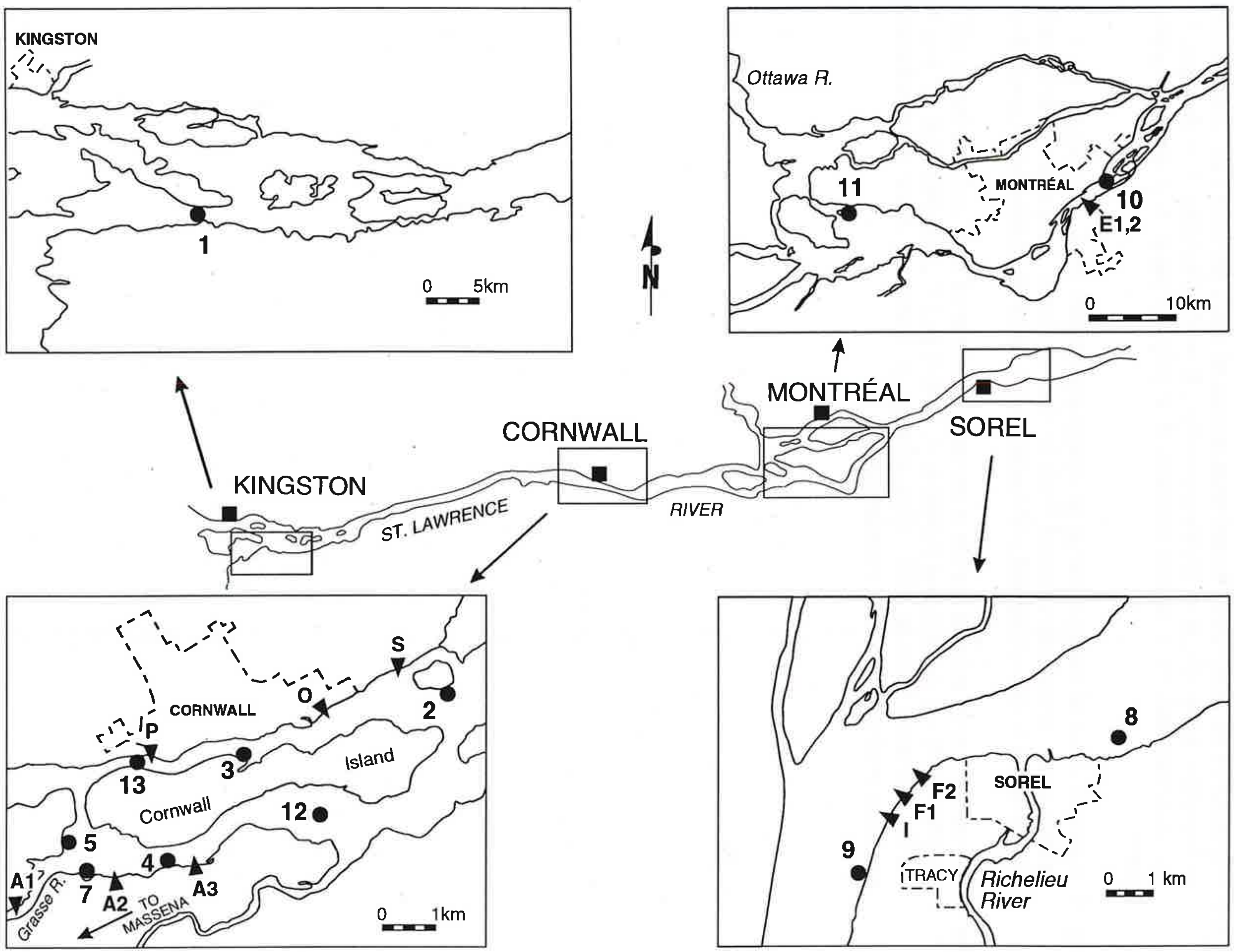

KINGSTON
1
0 5km
N
Ottawa R.
11
MONTRÉAL
10
E1,2
0 10km
KINGSTON
CORNWALL
MONTRÉAL
SOREL
ST. LAWRENCE
RIVER
CORNWALL
S
O
P
2
13
3
Island
Cornwall
12
5
4
A1
7
A2
A3
Grasse R.
TO MASSENA
0 1km
8
SOREL
F2
F1
I
9
TRACY
Richelieu River
0 1 km

Fig 1.

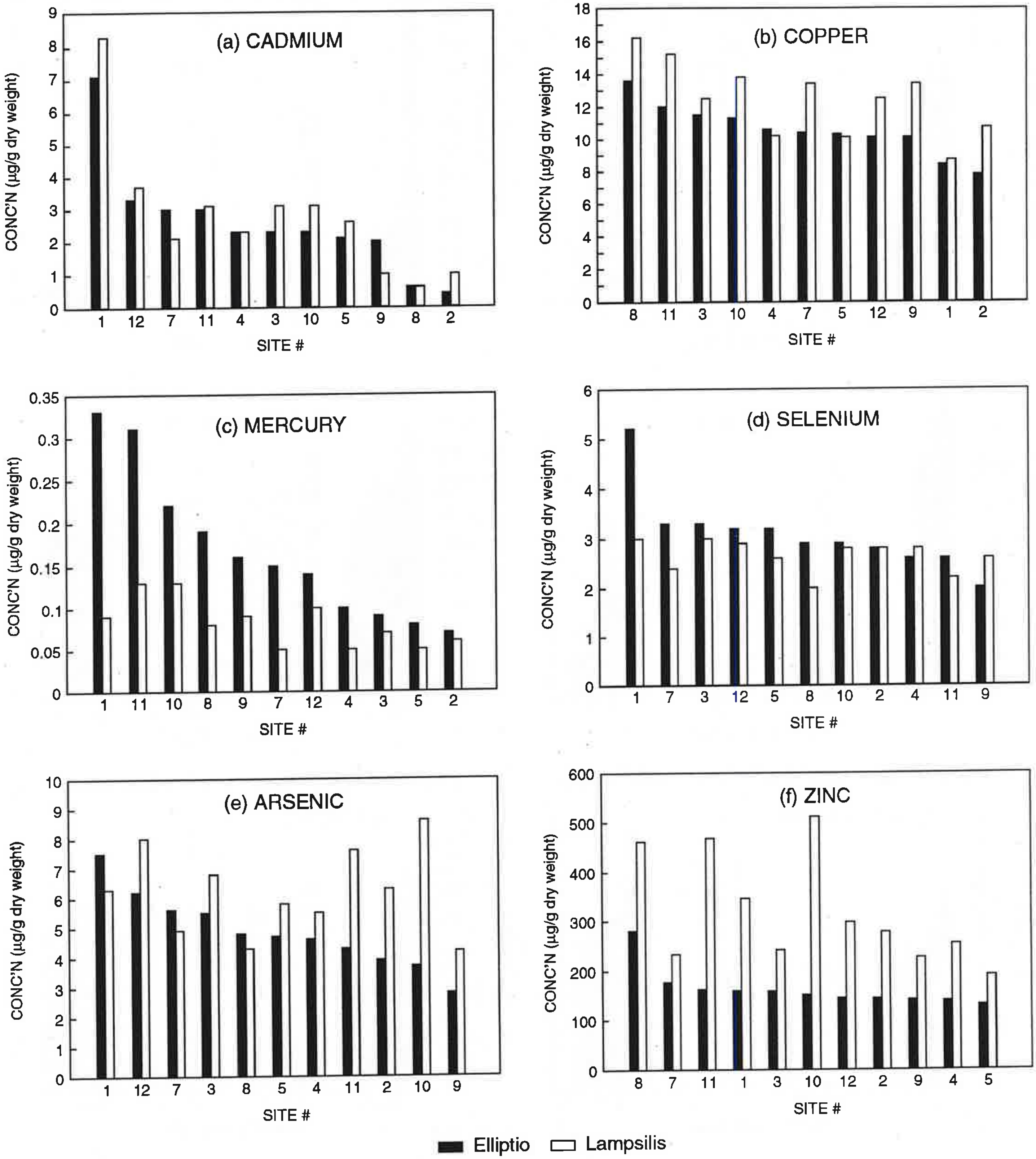
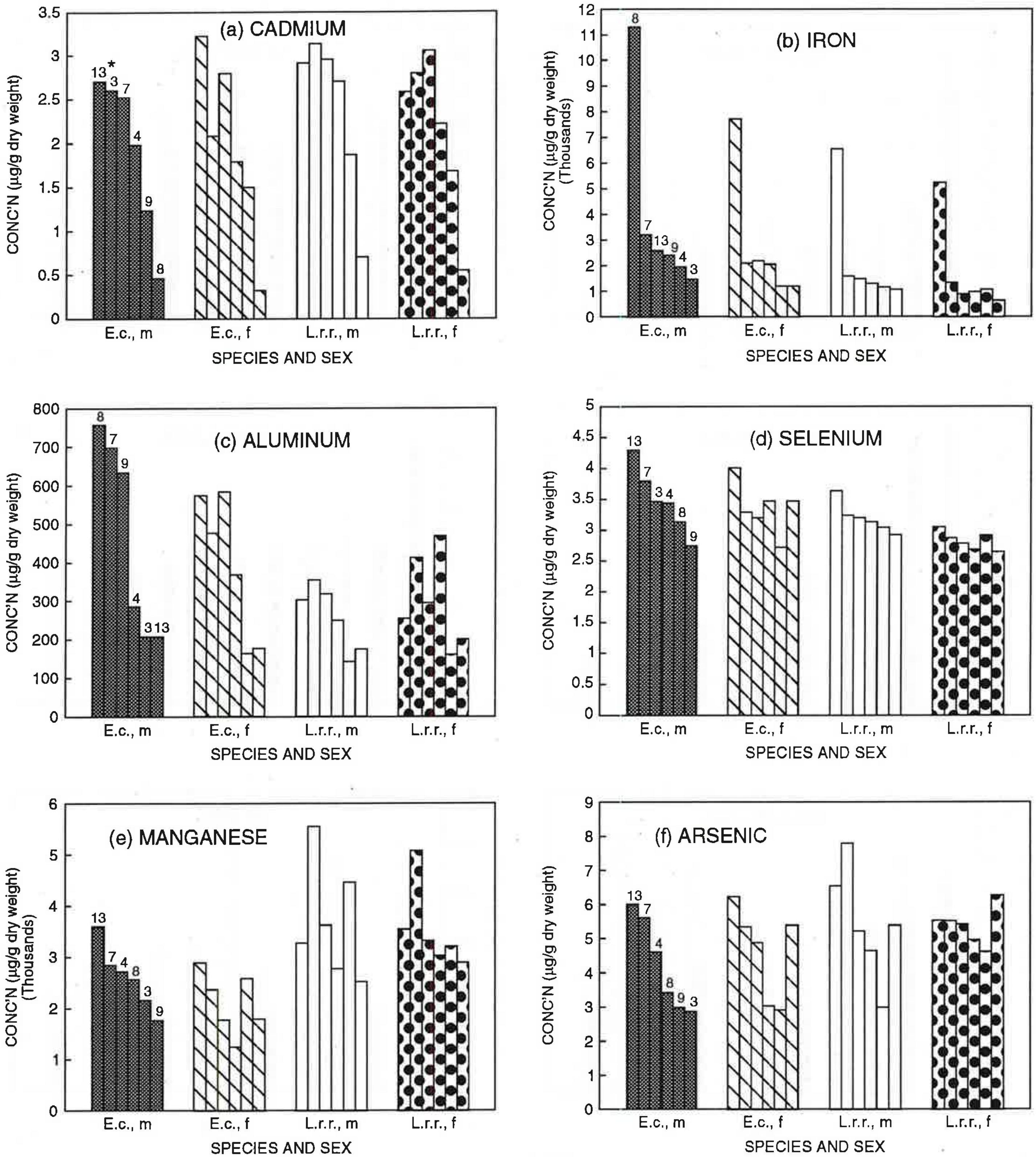

(a) CADMIUM
CONC'N (µg/g dry weight)
0 1 2 3 4 5 6 7 8 9
1 12 7 11 4 3 10 5 9 8 2
SITE #
(b) COPPER
CONC'N (µg/g dry weight)
0 2 4 6 8 10 12 14 16 18
8 11 3 10 4 7 5 12 9 1 2
SITE #
(c) MERCURY
CONC'N (µg/g dry weight)
0 0.05 0.1 0.15 0.2 0.25 0.3 0.35
1 11 10 8 9 7 12 4 3 5 2
SITE #
(d) SELENIUM
CONC'N (µg/g dry weight)
0 1 2 3 4 5 6
1 7 3 12 5 8 10 2 4 11 9
SITE #
(e) ARSENIC
CONC'N (µg/g dry weight)
0 1 2 3 4 5 6 7 8 9 10
1 12 7 3 8 5 4 11 2 10 9
SITE #
(f) ZINC
CONC'N (µg/g dry weight)
0 100 200 300 400 500 600
8 7 11 1 3 10 12 2 9 4 5
SITE #
Elliptio
Lampsilis

Fig. 2.


(a) CADMIUM
CONC'N (µg/g dry weight)
13* 3 7 4 9 8
(b) IRON
CONC'N (µg/g dry weight) (Thousands)
8 7 13 9 4 3
(c) ALUMINUM
CONC'N (µg/g dry weight)
8 7 9 4 3 13
(d) SELENIUM
CONC'N (µg/g dry weight)
13 7 3 4 8 9
(e) MANGANESE
CONC'N (µg/g dry weight) (Thousands)
13 7 4 8 3 9
(f) ARSENIC
CONC'N (µg/g dry weight)
13 7 4 8 9 3
E.c., m
E.c., f
L.r.r., m
L.r.r., f
SPECIES AND SEX

*SITE #

Fig. 3

APPENDICES

Appendix I. Concentrations of metals in composite samples of mussels collected in 1989 (ug/g dry weight).

SITE	SPECIES	Al	Zn	Fe	Mn	Cd (AA)	Cd (GF)	Cr	Cu	Ni	Pb (GF)	As	Se	Hg
1	1	153	159	2600	4310	7.1	6.94	18.8	8.4	3.5	2.64	7.5	5.2	0.33
1	2	252	346	2420	5490	8.3	8.08	7.2	8.7	2.3	3.24	6.3	3	0.09
2	1	469	134	1140	1430	0.4		3.8	7.8	2.2		3.9	2.8	0.07
2	2	334	277	869	3180	1	0.98	2	10.7	1.3	1.29	6.3	2.8	0.06
3	1	405	158	2020	3550	2.3	2.32	7.4	11.5	3.4	1.57	5.5	3.3	0.09
3	2	230	242	989	3920	3.1	3.27	3.6	12.5	1.6	1.58	6.8	3	0.07
4	1	291	139	1820	2070	2.3	2.35	8.2	10.6	3.2	1.51	4.6	2.6	0.1
4	2	166	254	916	3350	2.3	2.24	3.5	10.2	1.3	1.37	5.5	2.8	0.05
5	1	230	130	1300	1710	2.1	2.04	7.6	10.3	3.1	0.88	4.7	3.2	0.08
5	2	294	190	771	2560	2.6	2.55	2.7	10.1	1.8	1.14	5.8	2.6	0.05
6	1	576	169	1230	1120	0.44		3.29	11	3.95		3.8	2.9	0.35
7	1	497	177	2430	4690	3	3.34	14.9	10.4	2.8	1.88	5.6	3.3	0.15
7	2	387	233	1400	3940	2.1	2.13	2.3	13.4	1.7	0.94	4.9	2.4	0.05
8	1	488	280	9980	1920	0.6	0.92	17.1	13.6	7	13.2	4.8	2.9	0.19
8	2	380	461	7120	3900	0.6	0.81	12.6	16.2	3.9	7.5	4.3	2	0.08
9	1	684	140	2610	1540	2	2.03	13.3	10.1	3.2	1.3	2.8	2	0.16
9	2	534	226	1210	1740	1	1.07	6.1	13.4	1.3	0.71	4.2	2.6	0.09
10	1	515	151	2080	2080	2.3	2.44	8.5	11.3	4.2	2.06	3.7	2.9	0.22
10	2	291	511	1180	5560	3.1	3.44	4.1	13.8	1.3	1.67	8.6	2.8	0.13
11	1	696	162	3170	3240	3		8.8	12	4.1		4.3	2.6	0.31
11	2	449	468	2420	7380	3.1	3.77	5.9	15.2	1.2	1.89	7.6	2.2	0.13
12	1	193	144	2020	2740	3.3		12.5	10.1	2.1		6.2	3.2	0.14
12	2	146	297	1400	5920	3.7	3.77	6.1	12.5	2.4	1.73	8	2.9	0.1

NOTES: species 1 = E. complanata; species 2 = L. radiata
AA = atomic absorption spectroscopy, standard method
GF = atomic absorption spectroscopy, graphite furnace method

Appendix II. Concentrations of metals in composite samples of mussels collected in 1990 (ug/g dry weight).

SITE	SPECIES	SEX	Al	Zn	Fe	Mn	Cd	Cr	Cu	Ni	Pb	As	Se	Hg
3	2	1	160	220	631	3210	2.79	4.6	10.7	2.1	1.24	6.27	2.78	0.07
3	2	2	142	312	1050	4460	3.13	4.1	15	1.7	1.15	5.39	3.2	0.07
3	1	1	164	155	1190	2580	2.08	8.7	12.6	4.6	1.23	5.39	3.2	0.1
3	1	2	207	131	1470	2160	2.6	5.5	9.1	3.9	1.09	2.87	3.46	0.11
4	2	1	468	234	1060	3320	2.22	10.2	11.4	5	1.15	5.42	2.68	0.08
4	2	2	249	341	1150	3620	2.7	4	11	2.2	1.39	5.22	3.13	0.08
4	1	1	368	152	1190	1770	1.79	6.2	14.2	4.5	1.5	4.88	3.47	0.1
4	1	2	286	161	1960	2720	1.98	10.2	9.2	4	1.48	4.6	3.44	0.13
7	2	1	411	268	1330	5080	3.05	5	14.5	2.4	1.08	5.52	2.87	0.08
7	2	2	355	301	1580	5550	2.95	4.7	13.3	1.8	1.01	7.79	3.24	0.08
7	1	1	476	155	2090	2370	2.79	12.1	13.4	4.7	1.35	5.34	3.29	0.14
7	1	2	698	160	3200	2840	2.52	9.9	12.2	4	1.57	5.62	3.79	0.18
8	2	1	254	441	5240	3030	0.55	9	13.4	3.3	5.13	4.96	2.91	0.11
8	2	2	303	492	6550	2770	0.7	14.3	13.9	4.8	8.01	4.64	3.04	0.11
8	1	1	574	185	7710	1250	0.32	23.1	14.6	8.2	9.97	3.03	2.72	0.13
8	1	2	757	275	11300	2560	0.46	27.5	15.6	8.3	12.2	3.42	3.13	0.21
9	2	1	294	234	975	2890	1.68	6	11.2	2.3	0.57	4.62	2.64	0.13
9	2	2	318	323	1300	2520	1.87	7	12.6	2.4	0.66	2.99	2.92	0.14
9	1	1	584	153	2050	1790	1.5	12.6	14.5	3.7	1.22	2.91	3.47	0.21
9	1	2	633	144	2400	1760	1.23	11.8	11.5	4	1.08	2.99	2.74	0.2
13	2	1	200	220	875	3540	2.58	2.9	11.9	2	1.31	5.54	3.05	0.08
13	2	2	175	226	1480	3270	2.91	3.8	13.8	1.4	0.87	6.54	3.64	0.07
13	1	1	177	152	2180	2890	3.22	15.9	11.1	4.5	2.15	6.23	4.01	0.16
13	1	2	207	145	2580	3590	2.7	14.9	8	2.9	1.38	6	4.29	0.18

NOTES: species 1 - E. complanata; species 2 = L. radiata
sex 1 = female/gravid; sex 2 = male/non-gravid