Arsenic and mercury bioaccumulation in the aquatic plant, *Vallisneria neotropicalis*

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**ABSTRACT**

Arsenic (As) and mercury (Hg) are among the most toxic metals/metalloids. The overall goal of this study was to investigate the bioaccumulation of these trace elements in *Vallisneria neotropicalis*, a key trophic species in aquatic environments. For this purpose, As and Hg concentrations were determined in sediments and natural populations of *V. neotropicalis* in sub-estuaries of Mobile Bay (Alabama, USA), differing with respect to past and present anthropogenic impact. Analyses indicate that the Fish River is the most contaminated among the sub-estuaries investigated; levels of As found in Fish River sediments fall within a range that could potentially cause adverse effects in biota. Sediment As concentrations were only moderately correlated with those in *V. neotropicalis*; no correlation was found between sediment and plant Hg levels. However, several parameters could have masked such potential relationships (e.g., differences in sediment characteristics and “biological dilution” phenomena). Results presented herein highlight the numerous parameters that can influence metal/metalloids accumulation in aquatic plants as well as species-specific responses to trace element contamination. Finally, this study underscores the need for further investigation into contaminant bioaccumulation in ecologically and economically important coastal environments.

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1. Introduction

Estuarine environments are considered ecologically and economically important by virtue of their exceptional biological diversity and productivity. They support both fresh and saltwater organisms, and serve as nursery habitat for many commercially important species. However, these areas are threatened by human activities and associated chemical waste. Estuaries are widespread along the southern coast of the United States; they are formed by the mixing of freshwater discharge from some of North America’s largest rivers (e.g., the Mississippi, Brazos, and Rio Grande Rivers) with saline ocean water of the Gulf of Mexico. Estuaries of the southern coast of the United States are particularly threatened by trace element contamination, and more specifically by arsenic (As, a metalloid) and mercury (Hg, a metal). In 2007, the highest concentrations of total Hg wet deposition in the United States were found off the Gulf Coast (NADP, 2007). The entire coastline of the Gulf is under a Hg advisory (Lewis and Chancy, 2008), and several of its waterbodies are included on the US Clean Water Act 303(d) list as being impaired with respect to Hg and/or As (US EPA, 2009a). As and Hg are harmful, non-essential elements. Due to their persistent nature and tendency toward bioaccumulation, they are rated among the most toxic metals/metalloids (Soros et al., 2003; Walcek et al., 2003; Mishra et al., 2008; Gutierrez-Mejia et al., 2009). They rank first (As) and third (Hg) on the 2007 CERCLA (Comprehensive Environmental Response, Compensation, and Liability Act) priority list of hazardous substances (ATSDR, 2009). Pollution of aquatic ecosystems by As and Hg causes serious environmental and human health concerns on a worldwide scale (Graeme and Pollack, 1998; Wolfe et al., 1998; Zahir et al., 2005; Zahir et al., 2006).

Sediments act as a major reservoir for trace elements and, thus, provide an integrated indication of the condition of estuaries (Buggy and Tobin, 2008). However, their use is limited regarding the information they can provide on the bioavailability of contaminants. Despite the development and improvement of new approaches, no consensus yet exists concerning the most suitable methods for such an estimation (Almeida et al., 2005). By monitoring trace element concentrations in tissues of living organisms, we gain direct information on the fraction of contaminants that is of direct ecotoxicological relevance (i.e., the bioavailable forms – Rainbow, 1995; Blackmore et al., 1998). Aquatic plants are unique bioindicator species for the evaluation of water quality and subsequent risk assessment. Not only can we determine the bioavailability of contaminants, but plant biomonitor allow for the detection of early signs of environmental disturbance, before upper trophic
levels are affected. *Vallisneria americana* Michaux (Hydrocharitaceae), an aquatic angiosperm that forms extensive meadows in freshwater and mesohaline environments from Central America to Canada (Korschgen and Green, 1988; Hauxwell et al., 2007), has been proposed as a potential biomonitor of metal contamination (St-Cyr and Campbell, 1994; Hudon, 1998; St-Cyr and Campbell, 2000). This species has been shown to concentrate trace metals more efficiently than other species of submerged aquatic vegetation (SAV), and to reflect environmental contamination levels (St-Cyr and Campbell, 1994; Hudon, 1998; St-Cyr and Campbell, 2000). However, very few studies have investigated the potential of *V.* *americana* for such a role, and, to our knowledge, no previous study specifically considered the trace elements, As and Hg. Furthermore, the aforementioned studies were conducted in northern areas of the *V.* *americana* distribution range (i.e., Canada and north of the United States); morphological and molecular characters differ among northern and southern populations (i.e., well-described ecotypes exist; Smart and Dorman, 1993; Smart et al., 2005; McFarland, 2006), and a recent study of the genus recognizes *V. americana* as being distinct from the Gulf Coast species *V. neotropicalis* Marie-Vict. (Les et al., 2008).

The overall aim of the present study was to investigate the As and Hg bioaccumulation in the key trophic species, *V. neotropicalis*.

For this purpose, a field study was conducted to determine As and Hg concentrations in sediments and *V. neotropicalis* populations in sub-estuaries of Mobile Bay (Alabama, USA) that are exposed to different levels of anthropogenic pressure.

2. Materials and methods

2.1. Study area and sampling locations

The study area is part of the Mobile Bay Estuary system, a coastal transition zone between the Mobile Bay watershed and the Gulf of Mexico. It includes the lower Mobile–Tensaw Delta and Weeks Bay (Fig. 1). The Mobile Bay watershed drains two thirds of the state of Alabama and portions of Mississippi, Georgia, and Tennessee; it is the fourth largest watershed in the United States in terms of freshwater discharge volume (1756 m$^3$ s$^{-1}$ on average), and the sixth largest river system in area in the United States (draining 113 084 km$^2$; Mullins et al., 2002). The study area is a dynamic and complex system affected by tidal influx, salinity fluctuations, and freshwater discharge.

Six sampling locations were chosen in the lower Mobile–Tensaw Delta (Delvan Bay DB: 30° 41' N, 88° 00' W – Chacalochee Bay West CBW: 30° 41' N, 87° 59' W – Chacalochee Bay East CBE:...
30°40’ N, 87°58’ W – Meaher Park MP: 30°39’ N, 87°56’ W – Shellbank River SR: 30°41’ N, 87°55’ W – Bay Minette BM: 30°41’ N, 87°55’ W) and one in Weeks Bay (Fish River FR: 30°25’ N, 87°49’ W, Fig. 1). The Mobile–Tensaw Delta is formed by the merging of the Alabama and Tombigbee rivers at the terminus of the Mobile River Basin (O’Neil and Mettee, 2008). The Weeks River sub-estuary receives direct freshwater from the Fish (a 397 km² source of freshwater, accounting for approximately 73% of the flow) and Magnolia Rivers (Schooder, 1996). These locations are characterized by a complex of braided river channels, off-channel bays and bayous, interconnecting streams, and forested and emergent wetlands serving as the interface between fresh upland waters and the brackish waters of Mobile Bay. Sampling locations differ with respect to anthropogenic impact. The north-western side of Mobile Bay is highly urbanized and industrialized relative to the eastern side that is nearly undeveloped. Indeed, the city of Mobile, the second largest metropolitan area in the state of Alabama (US Census Bureau, 2009), is located on the northwest edge of Mobile Bay (Fig. 1). Moreover, the Mobile Field, that accounts for most of the coal production in Alabama (the coal from Warrior Bank River SR: 30°41’ N, 87°58’ W – Meaher Park MP: 30°39’ N, 87°56’ W – Shellbank River SR: 30°41’ N, 87°55’ W – Bay Minette BM: 30°41’ N, 87°55’ W) and one in Weeks Bay (Fish River FR: 30°25’ N, 87°49’ W, Fig. 1). The Mobile–Tensaw Delta is formed by the merging of the Alabama and Tombigbee rivers at the terminus of the Mobile River Basin (O’Neil and Mettee, 2008). The Weeks River sub-estuary receives direct freshwater from the Fish (a 397 km² source of freshwater, accounting for approximately 73% of the flow) and Magnolia Rivers (Schooder, 1996). These locations are characterized by a complex of braided river channels, off-channel bays and bayous, interconnecting streams, and forested and emergent wetlands serving as the interface between fresh upland waters and the brackish waters of Mobile Bay. Sampling locations differ with respect to anthropogenic impact. The north-western side of Mobile Bay is highly urbanized and industrialized relative to the eastern side that is nearly undeveloped. Indeed, the city of Mobile, the second largest metropolitan area in the state of Alabama (US Census Bureau, 2009), is located on the northwest edge of Mobile Bay (Fig. 1). Moreover, the Mobile Field, that accounts for most of the coal production in Alabama (the coal from Warrior River Basin (O’Neil and Mettee, 2008). The Weeks River sub-estuary receives direct freshwater from the Fish (a 397 km² source of freshwater, accounting for approximately 73% of the flow) and Magnolia Rivers (Schooder, 1996). These locations are characterized by a complex of braided river channels, off-channel bays and bayous, interconnecting streams, and forested and emergent wetlands serving as the interface between fresh upland waters and the brackish waters of Mobile Bay. Sampling locations differ with respect to anthropogenic impact. The north-western side of Mobile Bay is highly urbanized and industrialized relative to the eastern side that is nearly undeveloped. Indeed, the city of Mobile, the second largest metropolitan area in the state of Alabama (US Census Bureau, 2009), is located on the northwest edge of Mobile Bay (Fig. 1). Moreover, the Mobile Field, that accounts for most of the coal production in Alabama (the coal from Warrior River Basin (O’Neil and Mettee, 2008). The Weeks River sub-estuary receives direct freshwater from the Fish (a 397 km² source of freshwater, accounting for approximately 73% of the flow) and Magnolia Rivers (Schooder, 1996). These locations are characterized by a complex of braided river channels, off-channel bays and bayous, interconnecting streams, and forested and emergent wetlands serving as the interface between fresh upland waters and the brackish waters of Mobile Bay. Sampling locations differ with respect to anthropogenic impact. The north-western side of Mobile Bay is highly urbanized and industrialized relative to the eastern side that is nearly undeveloped. Indeed, the city of Mobile, the second largest metropolitan area in the state of Alabama (US Census Bureau, 2009), is located on the northwest edge of Mobile Bay (Fig. 1). Moreover, the Mobile Field, that accounts for most of the coal production in Alabama (the coal from Warrior

2.2. Study design and statistical analyses

The study was designed for a two-factor nested analysis of variance (location and site; ANOVA; STATISTICA™). Locations consisted of general sampling areas (Fig. 1), while sites were specific, randomly chosen areas, at least 100 m apart, within each location. Three sites were nested in each of seven locations (n = 21 sites total); Tukey’s HSD post hoc comparison tests were performed on data to determine where significant differences occurred. Pearson’s correlation coefficients were determined to reveal potential correlations. All statistical tests were assessed at the P = 0.05 level.

2.3. Environmental data collection

Measurements of depth (meter-stick), temperature, salinity, dissolved oxygen, conductivity (YSI – 85 handheld DO/Conductivity Instrument), pH (YSI – Environmental EcoSense handheld pH100), and alkalinity and hardness (titrimetric kits from CHEMetrics Inc.) were made for each site in July/August 2008, and coincident with plant and sediment collection.

2.4. Sampling and sample preparation

2.4.1. Sediment

For each sampling location, three sediment cores per site (n = 9 per location) were extracted within V. neotropicalis meadows (between V. neotropicalis plants). These extractions were made using a suction coring device made of acrylic plastic to avoid any potential contamination (Onuf et al., 1996). Each core was carefully extruded. Only the upper layer of sediment (about the first 2 cm) was considered for measurements as it corresponds to the sediment part which is situated inside the root-system. The center part of the upper layer was placed into a centrifuge tube for subsequent trace element analyses (i.e., the center part of the core was preferentially chosen to avoid any potential contamination from the corer surface), and the remaining part of the upper layer was placed into a sample bag for subsequent sediment characterization.

In the laboratory, large debris such as obvious plant/wood fragments, snails or shells were removed. Samples for trace element analyses were frozen, freeze-dried (6L Labconco Console Freeze-Dry system from Labconco Corp.), and disaggregated and homogenized. Samples for sediment characterization were dried using fans, disaggregated, and homogenized.

2.4.2. Vallisneria neotropicalis

This submerged aquatic plant is the dominant native species in the study area. V. neotropicalis is rooted in the sediment, with rosette-type ribbon-like leaves, and a short vertical stem. It is a perennial, producing new leaves (i.e., above-ground tissue) each year. In this study, only mature leaf material was used for the trace element analyses.

In each sampling location, nine shoots of V. neotropicalis were collected per site (n = 27 per location). Epiphytes and sediment were carefully removed from leaves using a glass slide. Samples were subsequently rinsed with ultra-pure water, placed in sample bags, frozen and freeze-dried (6L Labconco Console Freeze-Dry system from Labconco Corp.). Finally, samples were manually reduced to a coarse powder by hand-pressing and grinding on sealed bags.

2.5. Sediment characterization analysis

Grain-size analysis was carried out using the standard pipette and sieve method (Coventry and Fett, 1979; details as described by Haywick, 1996). Organic matter content (OM) was determined by the loss-on-ignition method (Schumacher, 2002).

2.6. Trace element analysis

2.6.1. Arsenic (As)

As concentrations in sediment and V. neotropicalis were measured by inductively coupled plasma mass spectrometry (ICP-MS), using the US EPA Method 6020 (US EPA, 2007).

2.6.2. Mercury (Hg)

Hg concentrations were measured with a Direct Mercury Analyzer 80 (DMA 80; Milestone Inc.) and a Hydra-C DMA (Teledyne Leeman Labs) in sediment and V. neotropicalis samples, respectively. These two instruments employ the “thermal decomposition – amalgamation – atomic absorption spectrophotometry” process as described in the US EPA Method 7473 (US EPA, 1998).

<table>
<thead>
<tr>
<th>Table 1</th>
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<tr>
<td><strong>Recoveries of As and Hg from certified standard reference materials.</strong></td>
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</tbody>
</table>

<table>
<thead>
<tr>
<th>CRM</th>
<th>As Certified</th>
<th>Measured</th>
<th>Recovery (%)</th>
<th>Hg Certified</th>
<th>Measured</th>
<th>Recovery (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lagarosiphon major BCR-060</td>
<td>8.0</td>
<td>N.D.</td>
<td>N.D.</td>
<td>0.34 ± 0.04</td>
<td>0.32 ± 0.01</td>
<td>94</td>
</tr>
<tr>
<td>Lobster hepatopancreas TORT-2</td>
<td>21.6 ± 1.8</td>
<td>19.4 ± 0.5</td>
<td>90</td>
<td>0.27 ± 0.06</td>
<td>0.31 ± 0.00</td>
<td>115</td>
</tr>
<tr>
<td>Dogfish muscle DORM-2</td>
<td>18.0 ± 1.1</td>
<td>N.D.</td>
<td>N.D.</td>
<td>4.04 ± 0.26</td>
<td>4.69 ± 0.06</td>
<td>101</td>
</tr>
</tbody>
</table>

Results of certified and measured trace element concentrations are expressed in µg g⁻¹ dry wt. (mean value ± 95% confidence interval).

* Uncertified value – N.D.: not determined.
2.6.3. Quality control

To avoid any possible contamination, all glassware and equipment was acid-soaked with 5% HNO₃ (Omnitrace Ultra™ Nitric Acid 67–70% from EMD Chemicals, Inc.) for at least 24 h, and thoroughly rinsed with ultra-pure water prior to use. Certified reference materials (Lagorosiphon major BCR-060 from the Community Bureau of Reference – Commission of the European Communities; Lobster Hepatopancreas TORT-2 and Dogfish Muscle DORM-2 from the National Research Council Canada) were routinely digested and analyzed alongside field-collected samples to determine the recovery rate of trace elements and verify the analytical procedure (Table 1).

3. Results

3.1. Environmental variables

Values for the environmental variables measured at each of the seven sampling locations are presented in Table 2. Depth ranged from 0.7 (Chacalochee Bay East) to 0.9 m (Bay Minette). Temperature values ranged from 28.7 (Meaher Park) to 32.3 °C (Shellbank River). Salinity ranged from 2.4 to 6.6‰, while conductivity ranged from 5.2 to 13.0 mS cm⁻¹: the lowest and highest values were recorded in Shellbank River and Delvan Bay, respectively. Water column pH was on the basic side, slightly increasing from east to west (pH₃[H₂O] < pH₅[H₂O] < pH₇[H₂O] < pH₉[H₂O] < pH₁[H₂O]). Alkalinity was lowest in Fish River (33 mg L⁻¹ as CaCO₃) and highest in Chacalochee Bay West (77 mg L⁻¹ as CaCO₃), whereas hardness was lowest in Shellbank River (437 mg L⁻¹ as CaCO₃) and highest in Delvan Bay (1000 mg L⁻¹ as CaCO₃). Dissolved oxygen concentrations varied between 5.1 (Bay Minette) and 8.8 mg L⁻¹ (Chacalochee Bay West).

3.2. Sediment characteristics

With respect to grain size, the majority of sediment samples fell into the sand and silty-sand fields of the Shepard diagram (Shepard, 1954; Fig. 2). Sediments from sites within Chacalochee Bay West and Delvan Bay locations showed the coarsest granulometry, with sand fraction ranges of 79–89% and 76–87%, respectively. The finest sediments were found in Bay Minette (silt + clay: 70–84%) and Fish River (silt + clay: 69–84%); according to the Shepard diagram, these sediments are classified as sand–silt–clay/clayey-silt sediments (Fig. 2). The composition of sediments in Shellbank River was very heterogeneous; samples from site C contained 98% of fines (silt + clay), while those from sites A and B only contained 25% and 40% of this fraction, respectively (Fig. 2).

The organic matter content was low for most locations (Delvan Bay, Chacalochee Bay West, Chacalochee Bay East, Meaher Park, and Shellbank River, except for site C; Table 3). The highest values were found in Fish River (21–28%) and Bay Minette (11–14%) relative to all other locations (Table 3). Organic matter content was highly correlated with the grain size components (i.e., high significant positive correlations with silt and clay fractions, a high significant negative correlation with sand fraction; Table 4).

3.3. Trace element concentrations

Spatial variability of As and Hg concentrations found in sediments and V. neotropicalis are shown in Fig. 3. In general, the concentrations measured in sediments were higher than those measured in V. neotropicalis.
Regarding the sediments, Fish River displayed the highest As and Hg concentrations ([As]FR = 10.51 ± 1.03 µg g⁻¹; [Hg]FR = 72.7 ± 11.6 ng g⁻¹); the differences were significant relative to others locations (Fig. 3 and Table 5A). In the Delta, As concentrations were significantly higher in eastern ([As]BM = 5.83 ± 0.50 µg g⁻¹; [As]DB = 8.1 ± 0.86 µg g⁻¹) than western locations ([As]MP = 1.82 ± 0.40 µg g⁻¹; [As]SR = 2.00 ± 0.38 µg g⁻¹; [As]CBE = 0.71 ± 0.13 µg g⁻¹; [As]CBW = 1.06 ± 0.19 µg g⁻¹; Fig. 3 and Table 5A). The same trend was observed for Hg concentrations; higher values were noted in the east ([Hg]BM = 47.2 ± 3.6 ng g⁻¹ and [Hg]SR = 45.9 ± 8.1 ng g⁻¹) than in the west ([Hg]MP = 21.4 ± 4.8 ng g⁻¹; [Hg]DB = 19.3 ± 1.9 ng g⁻¹; [Hg]SBW = 9.7 ± 1.8 ng g⁻¹; [Hg]CBE = 12.1 ± 1.9 ng g⁻¹; Fig. 3 and Table 5A).

No clear east–west gradient was mirrored by trace element concentrations in *V. neotropicalis*. As concentrations ranged from 1.05 ± 0.21 µg g⁻¹ (Chacalochee Bay West) to 2.47 ± 0.25 µg g⁻¹ (Bay Minette); levels in Fish River (2.16 ± 0.20 µg g⁻¹) were not significantly different from those in Bay Minette (Fig. 3 and Table 5C). The highest Hg concentration was measured in Fish River (15.78 ± 0.95 ng g⁻¹), followed by that in Chacalochee Bay West (12.86 ± 1.28 ng g⁻¹). The lowest Hg concentration was recorded in Shellbank River (1.76 ± 0.27 ng g⁻¹; Fig. 3 and Table 5C).

Strong positive correlations were found between sediment As and Hg concentrations and the silt fraction, clay fraction, and organic matter content of sediments (Table 4). Strong negative correlations were found between sediment As and Hg concentrations and the silt fraction, clay fraction, and organic matter content of sediments (Table 4). A significant positive correlation, albeit not strong, was found between As concentrations in *V. neotropicalis* and As concentrations in sediments (r = 0.51, P < 0.05). Conversely, no significant correlation was found between Hg concentrations in *V. neotropicalis* and Hg concentrations in sediments (Table 4).
As a matter of fact, several facilities in the area reported releases through the United States and its territories (US EPA, 2009b). The prevalence of organic matter coatings around sediment particles (silt and clay) due to the high volume-specific surface of fine-grained particles and organic matter in our study. Therefore, the nature of the sediments in Fish River, Bay Minette, and site C in Shellbank River (all characterized by fine fractions that are rich in organic matter) would promote a higher retention of As and Hg, and might explain our results. We can set potential limits to the importance of sediment granulometry in explaining differences in trace element concentrations by normalizing concentrations to percent content of fine material (silt + clay fraction). After doing so, Fish River still appears to be the most contaminated area with respect to As; regardless of location, no other significant differences exist relative to As levels. Although no significant differences exist among locations with respect to Hg contamination, the Fish River still displays the highest contamination levels (Table S8). Thus, the east–west patterns in As and Hg contamination are most likely explained by differences in sediment composition. Fish River still has significantly higher “normalized” sediment As concentrations than any other location, perhaps, pointing to either high anthropogenic input or a geochemical anomaly in this location.

The study locations differed in their sediment characteristics: Fish River and Bay Minette had finer sediments that were richer in organic matter. It is well known that there is a direct correlation between grain size and the abundance of elements in sediments (Cauwet, 1987; Cho et al., 1999; Fang and Hong, 1999; Rubio et al., 2000). Most elements are preferentially linked to fine-grained particles (silt and clay) due to the high volume-specific surface of the particles, favoring adsorption processes and ionic attraction. The prevalence of organic matter coatings around sediment particles further contributes to element binding (Wangersky, 1986; Buggy and Tobin, 2008). This could also be the case for As and Hg as their concentrations in sediments increased with the abundance of fine-grained particles and organic matter in our study. Therefore, the nature of the sediments in Fish River, Bay Minette, and site C in Shellbank River (all characterized by fine fractions that are rich in organic matter) would promote a higher retention of As and Hg, and might explain our results. We can set potential limits to the importance of sediment granulometry in explaining differences in trace element concentrations by normalizing concentrations to percent content of fine material (silt + clay fraction). After doing so, Fish River still appears to be the most contaminated area with respect to As; regardless of location, no other significant differences exist relative to As levels. Although no significant differences exist among locations with respect to Hg contamination, the Fish River still displays the highest contamination levels (Table S8). Thus, the east–west patterns in As and Hg contamination are most likely explained by differences in sediment composition. Fish River still has significantly higher “normalized” sediment As concentrations than any other location, perhaps, pointing to either high anthropogenic input or a geochemical anomaly in this location.

Contrary to the positive correlations between metal concentrations (Cd, Cr, Cu, Ni, Pb, and Zn) in sediments and in V. americana found by St-Cyr and Campbell (2000), our results showed a moderate positive correlation for As and no correlation for Hg. Thus,
V. neotropicalis tissues do not appear to reflect sediment levels of As and Hg contamination. Several factors could explain this result. First, we measured total concentrations of As and Hg in the sediment, but only one fraction of the total concentration might be available to biota. Numerous parameters are involved in the retention and release of sediment-associated contaminants, thereby, influencing their bioavailability. Along with sediment composition, hydrodynamic, biogeochemical, and physico-chemical processes (e.g., diffusion, redox conditions, pH, interstitial water hardness – Samiullah, 1990; Bryan and Langston, 1992; US EPA, 2000) play an important role in the adsorption onto or release of contaminants from sediment particles to subsequently affect availability to biota. Thus, even though few differences in habitat variables (i.e., depth, temperature, salinity, conductivity, pH, alkalinity, hardness, and dissolved oxygen) were detected among locations, the influence of the environment on As and Hg availability cannot be discounted.

Second, the absence of a relationship between element concentrations found in plants and those found in sediments could reflect an important foliar contribution to contaminant bioaccumulation by V. neotropicalis. Indeed, V. neotropicalis is a rooted aquatic plant and can thus absorb elements both through its leaves, via the surrounding water, and through its roots, via the sediment. However, an important trace element uptake from the water column seems unlikely to occur as it has been previously shown that sediment has a greater potential pool of contaminants than the overlying water column, and that rooted submerged plants with well-developed root systems, would primarily extract elements from the sediment with subsequent translocation to above-ground tissues (Jackson, 1998).

In addition to these factors, differences in biomass and growth rates among V. neotropicalis plants from different locations may have also played a role in determining results. Despite our sampling strategy, which was designed to minimize the impact of natural variability among locations (i.e., plants were collected at similar depths, and during similar time periods, the physico-chemical characteristics of the water column were quite similar – Table 2), plants often exhibited large differences in biomass, size, and presumably growth rates. High growth rates could lead to a "biological dilution" of contaminant contents in leaf material (Tinker, 1981; Hudson, 1998; Ferrat et al., 2003; Duman et al., 2006). To set limits to the potential importance of such a dilution we can calculate the total quantity of trace elements in the sample (i.e., multiply the trace element concentration by the total dry weight for each sample). After this calculation, there is still no correlation between levels of trace elements in V. neotropicalis leaves and in sediments ($r_{As} = 0.12, P > 0.05; r_{Hg} = 0.08, P > 0.05$; Pearson’s correlation). However, given the other factors that could mask potential significant relationships between concentrations in leaves of V. neotropicalis and sediments, this result is quite unsurprising.

While some studies suggest aquatic plants are effective biomonitors of trace element contamination (Jackson et al., 1991), the present study and others (e.g., Outridge and Noller, 1991) show that correlations between tissue trace element concentrations and those in water and/or sediments are often hard to establish. Although it might be difficult to draw any general conclusions concerning the sensitivity and reliability of V. neotropicalis as a bioindicator of As and Hg contamination from these data, such results will be informative and of value to future field studies in ecotoxicology. Results presented herein indicate that bioaccumulation processes can be both trace element- and species-specific.

It is noteworthy that the examination of trace element contamination in Vallisneria spp. is of importance because these macrophytes represent a key trophic resource for many primary consumers (e.g., macroherbivores such as turtles, manatees, muskrats, waterfowl; herbivorous fish; herbivorous and detritivorous invertebrates – Roberts et al., 1995; Feldman, 2001; Sponberg and Lodge, 2005; McFarland, 2006; Cremona et al., 2008). In turn, herbivorous fish and invertebrates that feed upon Vallisneria spp. are prey for many higher trophic level consumers (Rozas and Odum, 1988; Duffy and Baltz, 1998; Vanderkooy et al., 2000; Chaplin and Valentine, 2009). Consequently, even if low concentrations of trace elements are found in these macrophytes, the transfer of contaminants to higher trophic levels could lead to cautionary levels for human consumption. Thus, Vallisneria spp. might serve as a warning sign of potentially serious trace element contamination in commercially and recreationally important species of finfish and shellfish, and hence, could be valuable models for effective decision-making and environmental resource management in freshwater and estuarine systems. Clearly, further investigation into metal/metalloid bioaccumulation in high-impacted coastal systems is warranted.

To better understand metal/metalloid uptake, accumulation, and tolerance in V. neotropicalis, and in addition to sediment-associated measurements, future studies should include estimates of pore water and water column metal/metalloid concentrations. Moreover, bioavailability and toxicity of As and Hg depend upon chemical form. For example, arsenite, arsenate, and monomethylarsenate are considered to be the most toxic forms of As (Shaw et al., 2007), while methyl-mercury is the most toxic form of Hg and readily assimilated and biomagnified in aquatic food webs. Thus, it is recommended that future studies provide data for the most toxic forms of these elements, as opposed to measurements of total As and Hg, to address the issue of safety.

Acknowledgments

This work was supported by a grant from the Alabama Center for Estuarine Studies (“R83-0651”), a Center supported by the US Environmental Protection Agency, and a grant from the “Collectivité Territoriale de Corse”. The authors thank Drs. D. Haywick and E. Cioffi, from the University of South Alabama, for their assistance with sediment analyses and access to analytical equipment. We also thank Dr. David W. Evans, from NOAA, for his help with some metal analyses. Additionally, we wish to thank Dr. S. Phipps and E. Brunen from Weeks Bay NERR, S. Glenos, M. Mintz, K. VanDevlen, D. Marino, L. Moore, A. McDonald, B. Christiaen and J. Coff for their assistance in the field and/or lab. We also thank the editor of Chemosphere and the anonymous reviewers for their helpful comments and suggestions which improved this manuscript.

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