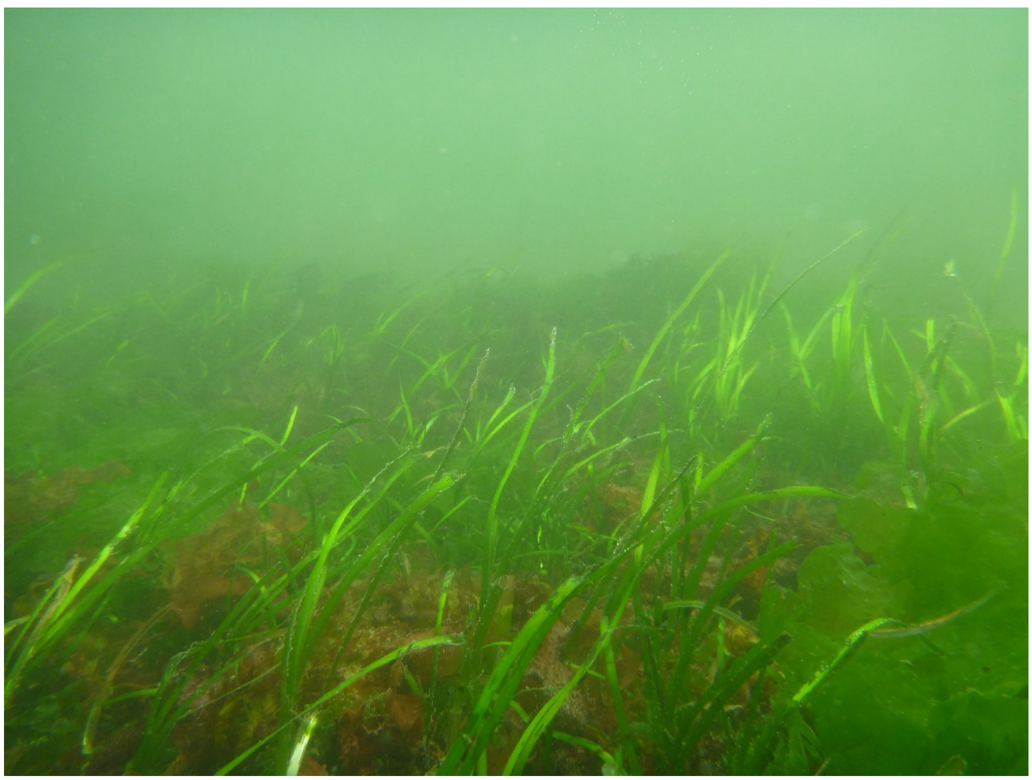
NATURAL RESOURCES

Extent and Magnitude of Nutrient and Contaminant Concentrations in Eelgrass (*Zostera marina* L.) in Puget Sound

March 30, 2016



WASHINGTON STATE DEPARTMENT OF
Natural Resources
Peter Goldmark - Commissioner of Public Lands

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Jeffrey Gaeckle

Nearshore Habitat Program
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EXECUTIVE SUMMARY

The Washington State Department of Natural Resources (DNR) is steward of 2.6 million acres of state-owned aquatic land. DNR manages these aquatic lands for the benefit of current and future citizens of Washington State. DNR's stewardship responsibilities include protection of eelgrass (*Zostera marina* L.), an ecologically important nearshore habitat in greater Puget Sound. In 2010, DNR became the monitoring lead for the Puget Sound eelgrass ecosystem indicator (Action Team 2010). DNR contributes to efforts that aim to achieve the goals set by the Puget Sound Partnership and supports the conservation and restoration of eelgrass.

The effects of certain stressors on eelgrass in Puget Sound are not well understood (Thom et al. 2011). Specifically, little is known about the concentration of nitrogen, carbon, metals (e.g., As, Cd, Cr, Cu, Fe, Hg, Ni, Pb, V, and Zn), and contaminants (e.g., PAHs, PCBs, PBDEs) in eelgrass and whether these concentrations are at levels considered toxic to eelgrass. In an effort to meet its stewardship responsibilities and to support the Partnership's goal to increase eelgrass area by 20% by 2020, DNR has identified a need for more information on the effects nutrients, metals, and contaminants have on eelgrass in greater Puget Sound. The current study assessed the concentration of carbon, nitrogen, 10 metals, and a suite of organic contaminants in the above- and belowground compartments of eelgrass at 15 sites throughout Puget Sound.

In general, the concentrations of carbon, nitrogen and metals in eelgrass were within the range observed in other seagrass, and eelgrass-specific, studies worldwide. The concentration of organic contaminants were low, and likely due to limited uptake and accumulation potential by seagrass because of the small concentration of lipids in seagrass biomass. With the exception of a very low C:N ratio that suggests high nutrient availability, it is not entirely clear how the measured concentrations of metals and organic contaminants in eelgrass will affect populations throughout Puget Sound. However, persistent, long-term effects of exposure to metal and organic contaminant concentrations may cause detrimental effects on eelgrass populations over time.

It was anticipated, based on the literature, that concentrations of nitrogen, metals, and organic contaminants would be higher in eelgrass above- and belowground compartments that were in close proximity to areas with greater shoreline development (e.g., industrial, commercial and metropolitan areas). For carbon and nitrogen isotopes, no clear pattern emerged. With the exception of one site, the C:N ratio in the aboveground biomass was consistently lower than 10, and in the belowground biomass the ratio was consistently below 15. The low C:N ratio indicates that eelgrass in Puget Sound is exposed to relatively high concentrations of nitrogen.

The concentration of some metals (Section 3.2) were highest near developed areas but the pattern was not consistent across all metals that were analyzed. Concentrations of copper in the above- and belowground biomass was highest at Four-Mile Rock; a site at the entrance of Elliot Bay which receives water that has circulated past the highly developed and industrialized waterfront of Seattle. Although levels of arsenic in eelgrass biomass were orders of magnitude less than a similar study conducted in 1994 by the US Fish and Wildlife Service, the aboveground eelgrass biomass at Padilla Bay had the highest concentrations of arsenic in contrast to low levels measured at March Point. It is not clear whether the arsenic at Padilla Bay could be from the residual fallout from the petroleum refineries on March Point, other activities or natural sources.

The concentration of PAHs in above- and belowground compartments of eelgrass were relatively low compared to results from the earlier USFWS study that measured PAHs in eelgrass in Fidalgo and Padilla Bays. However, at two sites adjacent to heavily developed uplands (e.g., Four-Mile Rock and Big Gulch), the PAH concentrations were significantly higher compared to other sample sites.

There was no clear pattern in the concentration of persistent organic pollutants (e.g., PCBs, PBDEs, DDTs, CHLDs, and HCHs) measured in eelgrass biomass relative to contamination sources. In some cases, eelgrass collected in close proximity to industrialized areas had higher levels of contamination (e.g., Four-Mile Rock) than other sites, but this pattern did not hold true for sites considered less developed and more pristine (e.g., Holly). In general, the concentration of persistent organic pollutants were low or undetectable, particularly in contrast to organism with higher lipid contents than eelgrass (e.g., mussels; Lanksbury et al. 2014).

This study provides the first soundwide assessment of the concentration of carbon, nitrogen, metal, and organic compounds in above- and belowground eelgrass compartments. Many factors affect the concentrations of carbon, nitrogen, metals and organic contaminants in seagrass such as their availability in the system (e.g., concentration in sediment and water), physical and chemical properties of the environment (e.g., temperature, salinity, pH), and plant health (e.g., physiology, growth rates, photosynthetic activity). Although the study was conducted at one point in time and at a limited number of sites, the results provide insight on the role of eelgrass in the cycling of these compounds throughout greater Puget Sound. The ability to use eelgrass as an indicator of ecosystem degradation due to the bioaccumulation of nutrients, metals and organic compounds has potential but requires sampling at greater frequency to understand seasonal variations and across smaller gradients to hone in on sources of pollution. In addition, sampling sediment and water would provide valuable information on the bioavailability of these substances to eelgrass in the environment where it grows.

1 INTRODUCTION

Eelgrass (*Zostera marina* L.), the dominant seagrass in the Pacific Northwest, is an important component of both public and private nearshore aquatic lands in greater Puget Sound. Eelgrass and other seagrasses are known to provide extensive ecosystem services worldwide (Costanza et al. 1997, Cullen-Unsworth and Unsworth 2013, Green and Short 2003, Larkum et al. 2006). It has been well documented that eelgrass stabilizes sediments and filters marine waters (Short and Short 1984). In Puget Sound specifically, eelgrass provides spawning grounds for Pacific herring (*Clupea harengus* pallasi) (Phillips 1984); shelter for egg-bearing Dungeness Crab (*Cancer magister*) (Armstrong et al. 1988, MacKay 1942); out-migrating corridors for juvenile salmon (*Oncorhynchus* spp.) (Simenstad 1994); and important feeding and foraging habitats for waterbirds such as the Black Brant (*Branta bernicla*) (Wilson and Atkinson 1995); and Great Blue Heron (*Ardea herodias*) (Butler 1995).

As steward of state-owned aquatic lands, the Washington State Department of Natural Resources (DNR) is committed to monitor eelgrass in Puget Sound through the Puget Sound Ecosystem Monitoring Program (PSEMP) and supports efforts to achieve the Puget Sound Partnership (PSP) goals. The Partnership has recognized the global ecological significance of eelgrass (Dennison et al. 1993, Krause-Jensen et al. 2005, Orth et al. 2006) and has identified it as one of 21 indicators for the health of Puget Sound.

Currently there are approximately 24,300 ha of eelgrass in Puget Sound but there has been evidence of declines at varying temporal and spatial scales (Nearshore Habitat Program 2015, 2016, Gaeckle et al. 2011, Thom and Hallum 1990, Wyllie-Echeverria et al. 2003). Declines in seagrass expand beyond Pacific Northwest, raising awareness of its loss throughout the world (Duarte 2002, Orth et al. 2006, Short and Burdick 1996, Waycott et al. 2009). Although research has been conducted that alludes to factors that cause eelgrass declines in Puget Sound (Schanz et al. 2010, Dooley et al. 2013), limited knowledge is available in regard to the effects of specific stressors on eelgrass (Thom et al. 2011). However, there is clear evidence elsewhere that losses to eelgrass (Short and Burdick 1996) and other seagrass populations are linked to pollution (Grady 1980, Marshall et al. 1993). Consequently, DNR has identified a need for more information on the impacts outfall effluent (e.g., nutrients, metals, and organic contaminants) have on eelgrass throughout the Sound. Baseline information, such as the concentration of nutrients, metals and organic compounds in eelgrass, will provide value in understanding the presence of these substances and their potential effect on eelgrass in Puget Sound. The additional eelgrass stressor data will improve management of this resource on state-owned aquatic lands and provide critical data towards the PSP's effort to increase eelgrass area by 20% by 2020.

The assessment of nutrients, metals, and organic contaminants in seagrass has been performed in many coastal areas throughout the world (Govers et al. 2014, Lewis and Devereux 2009, Ralph et al. 2006). The focus of many of these investigations has been to determine the concentration of nutrients, metals and contaminants in seagrass and to understand their effect on plant physiology and survival. There have also been efforts to understand the fate of nutrients, metals, and contaminants incorporated into seagrass tissue throughout the broader environment (Kaldy 2006), how much of a role seagrass plays in phytoremediation (Huesemann et al. 2009), and if bio-accumulation of metals or contaminants is a concern in trophic processes (Scarlett et al. 1999).

Limited research has demonstrated the physiological effects, particularly on photosynthetic processes, that nutrients, metals, and contaminants cause to eelgrass. However, many factors affect the availability, uptake, and toxicity of nutrients, metals, and contaminants. Little is known about the concentrations of these substances in eelgrass in the Pacific Northwest (Kaldy 2006) and, more specifically, greater Puget Sound. Basic nutrients such as nitrogen and phosphorus are abundant in Puget Sound, but whether these substances are at levels that cause toxicity in eelgrass is unknown. Similarly, it is likely that metals and contaminants abound in Puget Sound, particularly in the nearshore environment where most eelgrass grows, because of the abundance of effluent from outfalls and the proximity to riverine input and upland stormwater runoff. However, no research has been conducted locally to understand the concentration of these substances in eelgrass and the potential effect nutrients, metals, and contaminants have on the health of this dominant flora.

An initial baseline assessment of nutrients, metals, and contaminants in eelgrass is necessary to understand the concentrations of these substances in eelgrass, to determine whether these concentrations are approaching deleterious levels based on the literature and to understand their fate to effectively manage nearshore systems. Of particular interest are the concentrations of metals, organic contaminants and herbicides because of their potential as phytotoxins, their multi-chemical interactions on seagrass (Lewis and Devereux 2009), and their likely abundance in Puget Sound (Ecology and King County 2011, Mohamedali et al. 2011). Baseline data on nutrient, metal and contaminant concentrations in eelgrass will provide valuable information for decision makers to effectively protect this ecologically significant resource in Puget Sound.

Therefore, the objectives of this phase of the project were to 1) determine baseline concentrations of nutrients, metals, and organic contaminants in eelgrass throughout Puget Sound and to 2) relate these data to values in the literature to determine potential effects on eelgrass.

2 METHODS

Eelgrass was collected at 15 sites throughout the greater Puget Sound study area from January 7 through January 14, 2013 (Figure 1). Fourteen (14) of the sample sites were co-located with WDFW's Mussel Watch Pilot Expansion Project (Gaeckle 2013b, Lanksbury et al. 2012, 2014), and representated of a wide range of shoreline types and contaminant levels over a large geographical extent throughout Puget Sound.

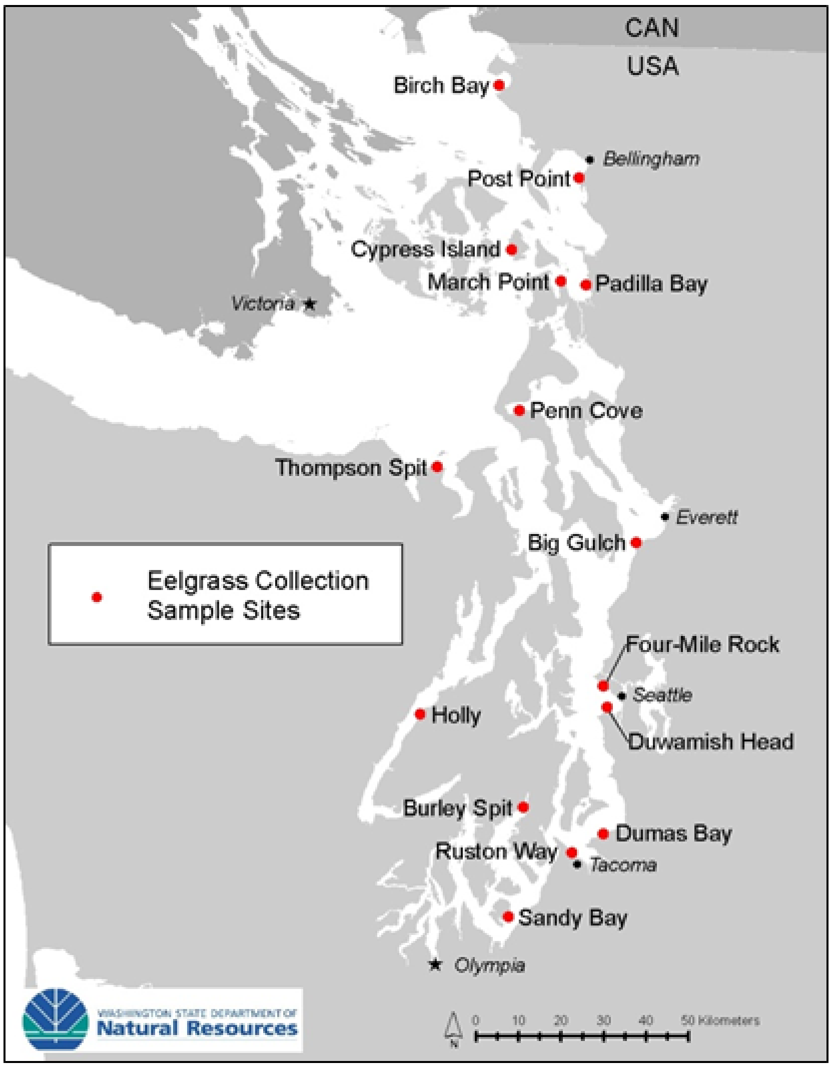

Figure 1. Location of the 15 eelgrass (*Zostera marina*) collection sites in the greater Puget Sound study area.

Burley Spit, a site at the northern end of Carr Inlet, did not have an associated mussel cage (Gaeckle 2013b, Lanksbury et al. 2014). A list of sites, their location, funding source and whether a mussel cage from WDFW's Mussel Watch Pilot Expansion Project was deployed at the site can be found in Table A-1 (Appendix A).

At each site, three replicate samples, separated by a miniumum distance of 100 m, of above- and belowground eelgrass were collected in a 0.25 m^2 quadrat at 0 m (MLLW) (Gaeckle 2013b). One quality control (QC) samples was collected at six randomly selected sites from the pool of 15 total sites. All samples (replicate and QC) were thoroughly rinsed with seawater at the site, stored in Ziplock bags in a cooler and returned to the Aquatic Botany Laboratory (Natural Resources Building, Olympia, WA) for initial processing (Gaeckle 2013a, 2013b). After processing, samples were shipped to different analytical labs to be assessed for carbon – nitrogen content (Stable Isotope Core Lab, Washington State University, Pullman, WA), ten metals (King County Environmental Laboratory, Seattle, WA), and a suite of organic contaminants (Northwest Fisheries Science Center, National Oceanic and Atmospheric Administration, Seattle, WA) (Gaeckle 2013a, 2013c, 2014). A complete description of sample collection and processing is available in Deliverable 2.1 – Field Sampling Summary (Gaeckle 2013b). A complete description of the laboratory analyses for carbon – nitrogen content, metals, and organic contaminants is available in the Quality Assurance Project Plan (Gaeckle 2013a).

Metal data reported as less than reporting detection limit (e.g., <RDL), but above the method detection limit (MDL) were maintained in the analyses. Organic contaminant data identified as analyte not detected above lower limit of quantitation (e.g., U analyte qualifier) were replaced with zeros (0s) prior to analysis. In cases where the lab calculated an estimate (e.g., J analyte qualifier), data were maintained in the analyses. Data were analyzed with a simple ANOVA using R statistical software (R Core Team 2015). In cases where the ANOVA generated a significant p-value, a Tukey's Honest Significant Difference (HSD) post-hoc analysis was conducted to determine significant differences between sites for each of the measured variables (e.g., C, N, metals and organic contaminants).

3 RESULTS

Three replicate eelgrass samples were successfully collected and processed from each of the 15 sites throughout greater Puget Sound in early January 2013 (Table 1). A quality control (QC) sample was also collected at 6 randomly selected sites of the 15 total sites (Table 1) (Gaeckle 2013b, 2013c, 2014). Samples were collected in early January to capture the signal of contaminants from the first winter runoff events that commence in early November and to coincide with WDFW's Mussel Watch Pilot Expansion Project sampling (Lanksbury et al. 2012).

Table 1. The sample site name, location, latitude and longitude, sample type [replicate sample, Rep, or quality control sample, (1)], and sample date for the 15 sites where eelgrass (*Zostera marina*) was collected in the Puget Sound study area. Sites are listed from north to south.

SITE	LOCATION	LATITUDE	LONGITUDE	TYPE	DATE
				Rep (QC)	
Birch Bay	Ferndale	48.8962	-122.7854	3	9 Jan 2013
Post Point	Fairhaven-Bellingham	48.7194	-122.5167	3	9 Jan 2013
Cypress Island	Strawberry Bay	48.5637	-122.7222	3 (1)	14 Jan 2013
March Point	Anacortes	48.4996	-122.5675	3	8 Jan 2013
Padilla Bay	Mt. Vernon	48.4924	-122.4866	3 (1)	8 Jan 2013
Penn Cove	Coupeville, Whidbey Island	48.2219	-122.6863	3	10 Jan 2013
Thompson Spit	Miller Peninsula, Gardiner	48.0967	-122.9394	3 (1)	9 Jan 2013
Big Gulch	Mukilteo	47.9107	-122.3222	3 (1)	8 Jan 2013
Four-Mile Rock	Magnolia, Seattle	47.6385	-122.4122	3	9 Jan 2013
Duwamish Head	Alki, West Seattle	47.5893	-122.3953	3	9 Jan 2013
Holly	Hood Canal	47.5706	-122.9715	3 (1)	11 Jan 2013
Burley Spit	Purdy	47.3791	-122.6399	3	9 Jan 2013
Dumas Bay	Federal Way	47.3290	-122.3905	3 (1)	10 Jan 2013
Ruston Way	Puget Creek, Tacoma	47.2811	-122.4771	3	11 Jan 2013
Sandy Bay	Anderson Island	47.1494	-122.6764	3	7 Jan 2013

3.1 Carbon and Nitrogen

The eelgrass aboveground $\delta^{13}C$ values ranged from -17.6 to -11.9‰ and the belowground $\delta^{13}C$ values ranged between -15.5 and -10.5‰ (Figure 2, Table 2). The lowest average aboveground $\delta^{13}C$ values were observed at Dumas Bay, Thompson Spit and Ruston Way and the highest was observed at Holly and Padilla Bay (Figure 2, Table 2). The lowest average belowground $\delta^{13}C$ value was observed at Penn Cove, Big Gulch and Ruston Way, and the highest $\delta^{13}C$ value was observed at Padilla Bay (Table 2 Figure 3).

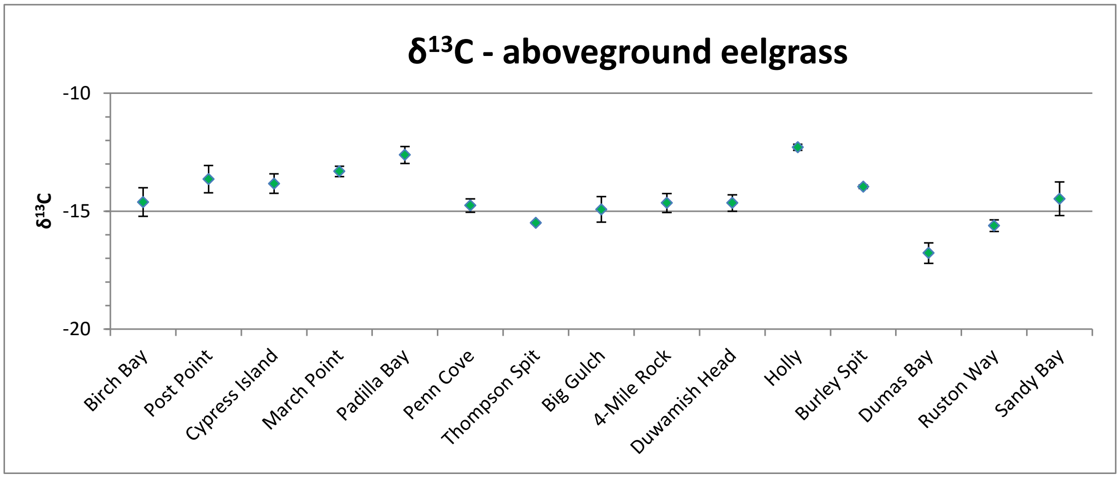

Figure 2**. The concentration (± SE) of** $\delta^{13}C$ in aboveground eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound.

Table 2. Average (± SE, n=3) carbon content (% dry weight) and $\delta^{13}C$ values for above- and belowground eelgrass (*Zostera marina*) compartments collected at 15 sample sites throughout Puget Sound.

SITES	ABOVEGROUND				BELOWGROUND			
	%C	(SE)	δ13C	(SE)	%C	(SE)	δ13C	(SE)
Birch Bay	38.5	0.7	-14.6	0.6	31.6	0.3	-12.8	0.5
Post Point	37.9	1.0	-13.6	0.6	34.8	0.7	-12.4	0.9
Cypress Island	36.2	1.8	-13.8	0.4	34.1	0.4	-13.2	0.4
March Point	37.7	0.6	-13.3	0.2	32.6	0.6	-13.1	0.4
Padilla Bay	38.3	0.2	-12.6	0.4	34.3	0.1	-11.1	0.3
Penn Cove	37.6	0.2	-14.8	0.3	34.5	0.0	-14.0	0.6
Thompson Spit	37.6	0.4	-15.5	0.0	29.7	0.4	-13.9	0.2
Big Gulch	37.6	0.2	-14.9	0.5	31.8	0.2	-14.0	0.9
Four-Mile Rock	37.0	0.3	-14.7	0.4	31.1	0.7	-12.8	0.5
Duwamish Head	33.9	1.9	-14.7	0.3	32.2	0.2	-13.3	0.1
Holly	37.2	0.4	-12.3	0.1	33.4	0.7	-11.9	0.6
Burley Spit	37.2	0.2	-14.0	0.1	31.2	0.3	-12.5	0.3
Dumas Bay	37.8	0.2	-16.8	0.4	31.3	0.2	-12.5	0.6
Ruston Way	36.5	0.2	-15.6	0.4	31.9	0.2	-14.1	0.4
Sandy Bay	38.1	0.2	-14.5	1.2	33.1	1.2	-13.6	0.7

The δ^{13}C signal in the aboveground eelgrass was significantly different among the 15 sample sites ($p < 0.001$; Figure 2). The results of the Tukey's HSD showed 24 significant paired site comparisons (Table 3).

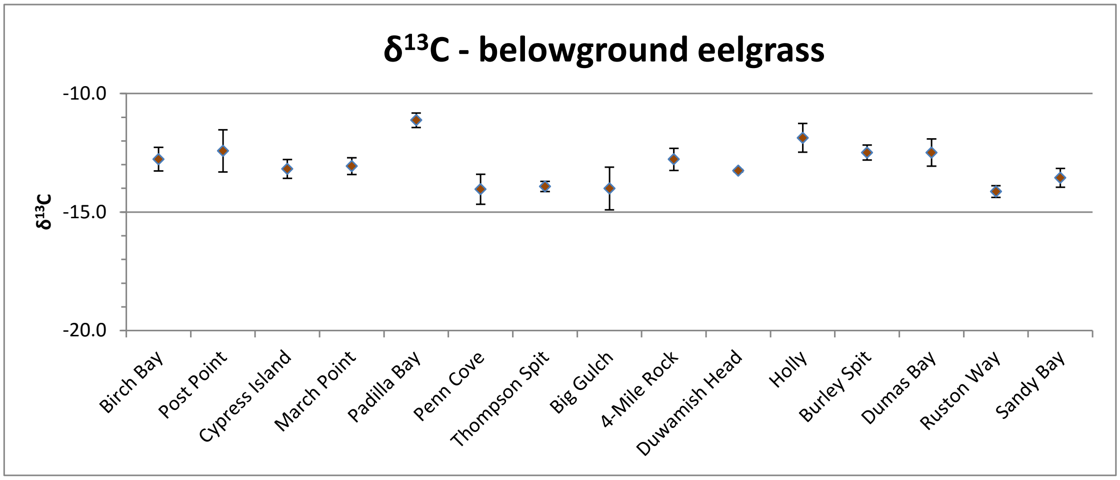

Figure 3**. The concentration (± SE) of δ^{13}**C in belowground eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound.

There was also a significant difference in the δ^{13}C signal in the belowground eelgrass compartment between sites ($p < 0.008$; Figure 3). However, the results of the Tukey's HSD test showed only four significant multiple site comparisons (Table 3).

Table 3. **Results of the Tukey HSD multiple comparison test for δ^{13}**C in the aboveground (green) and belowground (brown) eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound. For the aboveground **δ**^{13}C there were 24 paired site comparisons that were significant (green). There were four (4) paired site comparisons that were significant in the belowground **δ**^{13}C (brown).

δ^{13}C	BB	BG	BS	CI	DB	DH	FR	HY	MP	PB	PC	PP	RW	SB	TS
BB	1	1.00	1.00	0.99	0.04	1.00	1.00	0.02	0.62	0.08	1.00	0.92	0.90	1.00	0.96
BG	0.91	1	0.93	0.84	0.14	1.00	1.00	0.01	0.30	0.02	1.00	0.65	1.00	1.00	1.00
BS	1.00	0.72	1	1.00	<0.01	1.00	1.00	0.25	1.00	0.57	0.98	1.00	0.26	1.00	0.37
CI	1.00	1.00	1.00	1	<0.01	0.98	0.98	0.36	1.00	0.72	0.94	1.00	0.17	1.00	0.26
DB	1.00	0.72	1.00	1.00	1	0.05	0.05	<0.01	<0.01	<0.01	0.08	<0.01	0.77	0.02	0.64
DH	1.00	1.00	1.00	1.00	1.00	1	1.00	0.02	0.58	0.07	1.00	0.90	0.93	1.00	0.97
FR	1.00	0.91	1.00	1.00	1.00	1.00	1	0.02	0.58	0.07	1.00	0.90	0.93	1.00	0.97
HY	0.99	0.22	1.00	0.87	1.00	0.83	0.99	1	0.89	1.00	0.01	0.56	<0.01	0.04	<0.01
MP	1.00	0.99	1.00	1.00	1.00	1.00	1.00	0.93	1	1.00	0.46	1.00	0.02	0.77	0.04
PB	0.61	0.02	0.84	0.27	0.84	0.23	0.60	1.00	0.35	1	0.05	0.89	<0.01	0.14	<0.01
PC	0.89	1.00	0.69	1.00	0.69	1.00	0.90	0.20	0.99	0.02	1	0.81	0.97	1.00	0.99
PP	1.00	0.66	1.00	1.00	1.00	1.00	1.00	1.00	1.00	0.88	0.63	1	0.09	0.98	0.14
RW	0.84	1.00	0.61	0.99	0.61	0.99	0.84	0.16	0.97	0.02	1.00	0.54	1	0.79	1.00
SB	1.00	1.00	0.97	1.00	0.97	1.00	1.00	0.57	1.00	0.10	1.00	0.95	1.00	1	0.89
TS	0.95	1.00	0.79	1.00	0.79	1.00	0.95	0.27	1.00	0.03	1.00	0.73	1.00	1.00	1

The belowground $\delta^{13}C$ signal was significantly higher at Padilla Bay compared to the concentration in eelgrass at Penn Cove, Thompson Spit, Big Gulch, and Ruston Way (Table 3).

The aboveground $\delta^{15}N$ values ranged from 5.9 to 11.8‰, whereas the belowground $\delta^{15}N$ values ranged between 4.8 and 10.1‰ (Figures 4 & 5; Table 4). The lowest average above- and belowground eelgrass $\delta^{15}N$ values were observed at Cypress Island and the highest values were observed at Padilla Bay (Figures 4 & 5; Table 4).

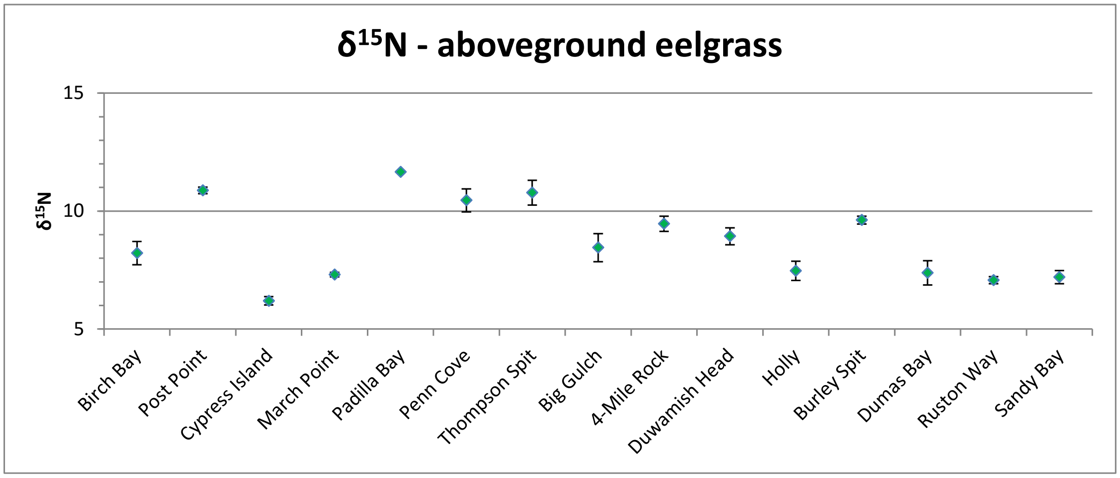

Figure 4. **The concentration (± SE) of δ^{15}N in aboveground eelgrass at 15 sites throughout Puget Sound.**

Table 4. **Average (± SE, n=3) nitrogen content (% dry weight) and δ^{15}N values for above- and belowground eelgrass (*Zostera marina*) compartments collected at 15 sample sites in Puget Sound.**

SITES	ABOVEGROUND				BELOWGROUND			
	%N	(SE)	δ^{15}N	(SE)	%N	(SE)	δ^{15}N	(SE)
Birch Bay	3.9	0.2	8.2	0.9	3.5	0.4	6.8	1.1
Post Point	3.9	0.1	10.9	0.2	3.3	0.3	8.8	0.5
Cypress Island	3.4	0.1	6.2	0.3	2.3	0.0	5.7	0.1
March Point	4.4	0.2	7.3	0.2	2.5	0.4	6.3	0.4
Padilla Bay	4.3	0.1	11.7	0.0	2.9	0.2	9.9	0.3
Penn Cove	4.3	0.3	10.5	0.8	3.3	0.3	8.5	0.2
Thompson Spit	3.3	0.0	10.8	0.9	3.9	0.1	8.8	0.8
Big Gulch	4.3	0.2	8.5	1.0	3.1	0.3	6.5	0.7
Four-Mile Rock	3.8	0.2	9.5	0.6	2.5	0.1	7.9	0.9
Duwamish Head	3.8	0.1	8.9	0.6	2.6	0.3	7.4	0.4
Holly	4.1	0.2	7.5	0.7	5.2	0.3	5.9	1.0
Burley Spit	3.6	0.1	9.6	0.3	2.6	0.3	9.1	0.8
Dumas Bay	4.2	0.2	7.4	0.9	2.8	0.1	5.7	0.8
Ruston Way	3.7	0.1	7.1	0.3	2.0	0.1	6.4	0.3
Sandy Bay	3.1	0.2	7.2	0.5	1.8	0.2	6.4	0.4

The $\delta^{15}N$ signal in aboveground eelgrass was significantly different among the 15 sample sites ($p < 0.001$, Figure 4). The results of the Tukey's HSD showed 51 significant multiple site comparisons (Table 5).

Similar results were observed in the $\delta^{13}C$ signal in the belowground eelgrass (Figure 5). The results of the Tukey's HSD showed 44 significant multiple site comparisons (Table 5).

Table 5. Results of the Tukey HSD multiple comparison test **for $\delta^{15}N$** in aboveground (green) and belowground (brown) eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound. For the aboveground **$\delta^{15}N$** concentration there were 51 significant paired site comparisons (green). There were 44 significant paired site com**parisons in the belowground $\delta^{15}N$** concentrations.

$\delta^{15}N$	BB	BG	BS	CI	DB	DH	FR	HY	MP	PB	PC	PP	RW	SB	TS
BB	1	1.00	0.33	0.03	0.94	0.98	0.51	0.97	0.89	<0.01	0.01	<0.01	0.64	0.79	<0.01
BG	1.00	1	0.61	0.01	0.73	1.00	0.79	0.82	0.64	<0.01	0.03	<0.01	0.35	0.50	0.01
BS	0.01	<0.01	1	<0.01	0.01	0.99	1.00	0.01	0.01	0.02	0.94	0.49	<0.01	<0.01	0.62
CI	0.77	0.98	<0.01	1	0.59	<0.01	<0.01	0.48	0.68	<0.01	<0.01	<0.01	0.91	0.80	<0.01
DB	0.74	0.97	<0.01	1.00	1	0.20	0.02	1.00	1.00	<0.01	<0.01	<0.01	1.00	1.00	<0.01
DH	1.00	0.91	0.16	0.16	0.15	1	1.00	0.27	0.15	<0.01	0.22	0.04	0.06	0.10	0.06
FR	0.79	0.41	0.61	0.02	0.02	1.00	1	0.03	0.01	0.01	0.82	0.31	<0.01	0.01	0.42
HY	0.89	1.00	<0.01	1.00	1.00	0.25	0.04	1	1.00	<0.01	<0.01	<0.01	1.00	1.00	<0.01
MP	1.00	1.00	<0.01	1.00	1.00	0.68	0.19	1.00	1	<0.01	<0.01	<0.01	1.00	1.00	<0.01
PB	<0.01	<0.01	0.97	<0.01	<0.01	0.01	0.04	<0.01	<0.01	1	0.56	0.96	<0.01	<0.01	0.91
PC	0.18	0.05	1.00	<0.01	<0.01	0.80	1.00	<0.01	0.02	0.38	1	1.00	<0.01	<0.01	1.00
PP	0.04	0.01	1.00	<0.01	<0.01	0.39	0.90	<0.01	<0.01	0.79	1.00	1	<0.01	<0.01	1.00
RW	1.00	1.00	<0.01	0.99	0.99	0.83	0.31	1.00	1.00	<0.01	0.03	0.01	1	1.00	<0.01
SB	1.00	1.00	<0.01	0.99	0.99	0.83	0.30	1.00	1.00	<0.01	0.03	0.01	1.00	1	<0.01
TS	0.04	0.01	1.00	<0.01	<0.01	0.38	0.89	<0.01	<0.01	0.80	1.00	1.00	0.01	0.01	1

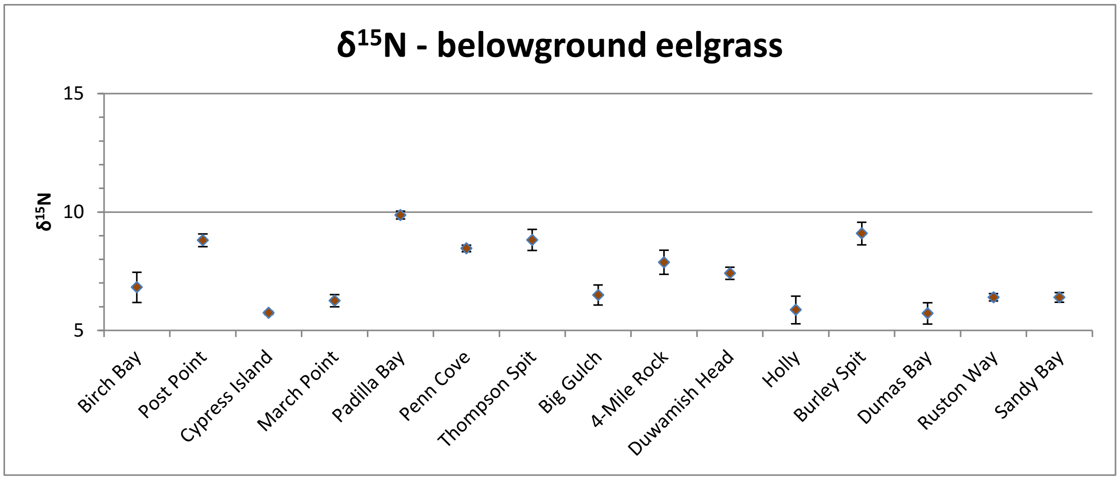

Figure 5. **The concentration (± SE) of δ**^{15}N in aboveground eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound.

The average (±SE) aboveground eelgrass compartment C:N ratio ranged from 7.3 (±0.2) to 10.7 (±0.7), and the average belowground C:N ratio ranged from 5.6 (±0.2) to 16.3 (±1.4) (Figure 6).

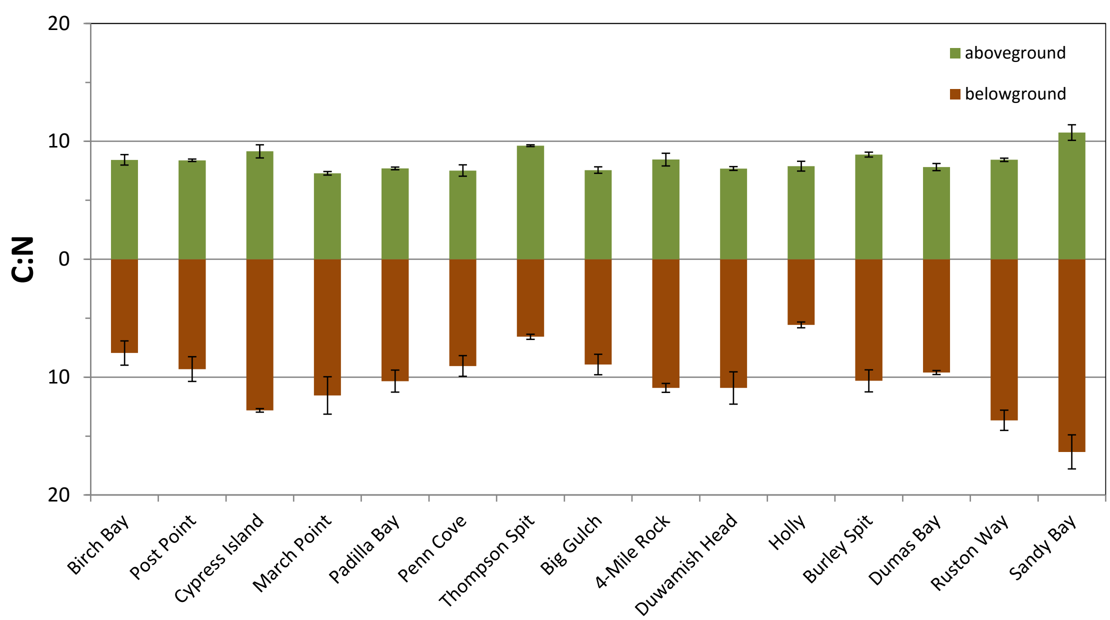

Figure 6. Average (± SE) above- and belowground eelgrass (*Zostera marina*) C:N ratio collected at 15 sites throughout Puget Sound.

3.2 Metals

The order of the average metal concentrations in the aboveground eelgrass biomass measured in Puget Sound was Fe > Zn > Cu > Cd > Ni > V > As > Pb > Cr > Hg, while the order of the belowground concentrations varied slightly, Fe > Zn > Cu > Cd > V > As >Ni> Cr > Pb > Hg. The average concentration (µg gdw^{-1}, ± SE) of metals in the aboveground compartment of eelgrass exhibited significant differences across the 15 sites (Hg, $p < 0.001$; As, $p < 0.001$; Cd, $p = 0.004$; Cr, $p < 0.001$; Cu, $p = 0.031$; Fe, $p = 0.002$; Pb, $p = 0.015$; Ni, $p < 0.001$; V, $p = 0.001$; Zn, $p = 0.001$). With the exception of lead (Pb), the concentration of metals in the belowground compartment of eelgrass exhibited significant differences across the 15 sites (Hg, $p < 0.001$; As, $p < 0.001$; Cd, $p = 0.002$; Cr, $p < 0.001$; Cu, $p < 0.001$; Fe, $p < 0.001$; Pb, $p = 0.131$; Ni, $p < 0.001$; V, $p = 0.001$; Zn, $p = 0.038$).

A list of analyte qualifiers for the metals is available in Deliverable 2.2 – Part I. Laboratory Analysis Data Metals and Nutrients (Gaeckle 2013c). There were only a few qualifiers from the metal analyses and these included:

1) Mercury – the minimum holding period for mercury samples exceeded the allowed 28 days. However, the samples were stored in a freezer at -20° C immediately after processing to remove any risk of volatile losses of the analyte.
2) Chromium – 62% of the samples (56 samples) were identified as being less than the reporting detection limit (<RDL) and 1% of the samples (1 sample, PP-3-BG) was identified as less than the method detection limit (<MDL).
3) Lead - 38% of the samples (34 samples) were identified as being less than the reporting detection limit (<RDL) and 1% of the samples (1 sample, BB-2-BG) was identified as less than the method detection limit (<MDL).

Mercury (Hg)

The average concentration of mercury in the aboveground eelgrass biomass ranged from 0.0045 ±0.0001 µg gdw^{-1} (±SE) at Cypress Island to 0.0158 ±0.0006 µg gdw^{-1} (±SE) at Dumas Bay (Figure 7). The average concentration of mercury in the aboveground compartment of eelgrass was significantly higher at Dumas Bay (Figure 7). The mercury concentration in the aboveground compartment was lowest, but not significantly, at Cypress Island, Holly, March Point, and Sandy Bay (Figure 7, Table 6).

The average concentration of mercury in the belowground eelgrass biomass ranged from 0.0024 ±0.0002 µg gdw^{-1} (±SE) at Holly to 0.0049 ±0.0002 µg gdw^{-1} (±SE) at

Post Point (Figure 7). The average concentration of mercury in the belowground compartment of eelgrass was lowest at Holly, but only significantly different than the concentrations measured at Post Point, Dumas Bay, and Big Gulch (Figure 7, Table 6). The average concentration of mercury measured at all other sites was not significantly different than observed at Holly.

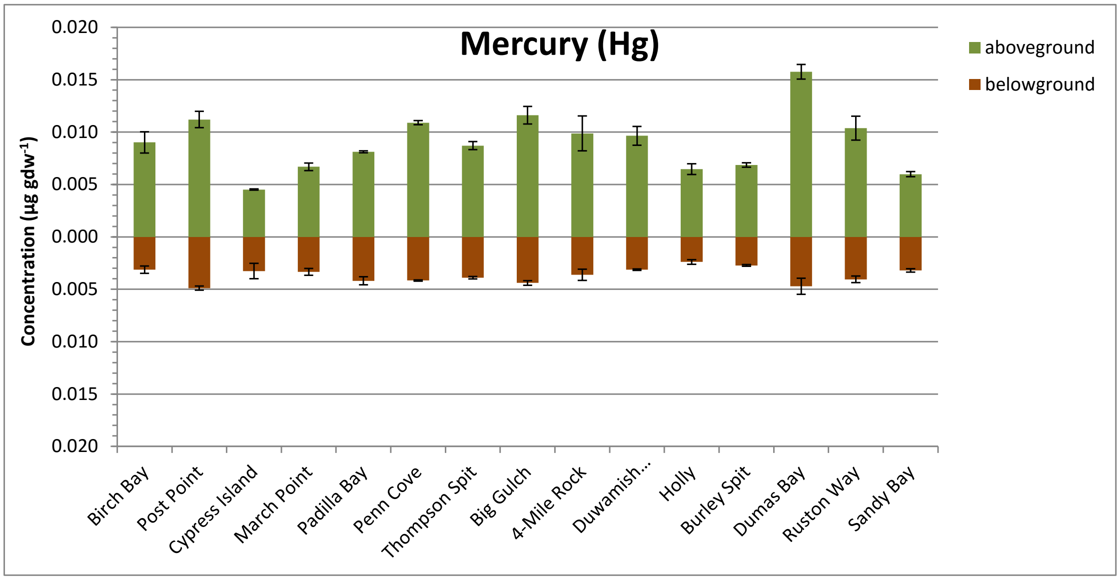

Figure 7. Average concentration (µg gdw^{-1}, ± SE) of mercury (Hg) in the above- and belowground compartments of eelgrass (*Zostera marina* L.) measured at 15 sites throughout Puget Sound.

The results of the Tukey's HSD showed 35 significant paired site comparisons in aboveground eelgrass Hg concentrations and only five significant paired site comparisons in the belowground eelgrass Hg concentractions (Table 6).

Table 6. Results of the Tukey HSD multiple comparison test for average mercury (Hg) concentration in the above- (green) and belowground (brown) eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound. Comparisons of the Hg concentration in the aboveground eelgrass was significant between 35 paired sites. Only five (5) paired sites showed a significant difference in the Hg concentration measured in the belowground eelgrass compartment.

Hg	BB	BG	BS	CI	DB	DH	FR	HY	MP	PB	PC	PP	RW	SB	TS
BB	1	0.51	0.76	0.01	<0.01	1.00	1.00	0.53	0.66	1.00	0.89	0.75	0.99	0.27	1.00
BG	0.49	1	0.01	<0.01	0.03	0.86	0.94	<0.01	0.01	0.12	1.00	1.00	1.00	<0.01	0.33
BS	1.00	0.14	1	0.66	<0.01	0.39	0.28	1.00	1.00	1.00	0.04	0.02	0.12	1.00	0.90
CI	1.00	0.65	1.00	1	<0.01	<0.01	<0.01	0.86	0.76	0.10	<0.01	<0.01	<0.01	0.98	0.03
DB	0.22	1.00	0.04	0.34	1	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	0.01	<0.01	<0.01	<0.01
DH	1.00	0.49	1.00	1.00	0.22	1	1.00	0.21	0.30	0.98	1.00	0.97	1.00	0.08	1.00
FR	1.00	0.97	0.89	1.00	0.80	1.00	1	0.14	0.21	0.93	1.00	0.99	1.00	0.05	1.00
HY	0.98	0.03	1.00	0.93	0.01	0.98	0.53	1	1.00	0.96	0.02	0.01	0.05	1.00	0.72
MP	1.00	0.73	1.00	1.00	0.41	1.00	1.00	0.89	1	0.99	0.03	0.01	0.08	1.00	0.83
PB	0.77	1.00	0.31	0.89	1.00	0.77	1.00	0.09	0.93	1	0.40	0.25	0.71	0.77	1.00
PC	0.84	1.00	0.37	0.93	1.00	0.84	1.00	0.12	0.96	1.00	1	1.00	1.00	0.01	0.74
PP	0.10	1.00	0.02	0.17	1.00	0.10	0.57	<0.01	0.22	0.99	0.97	1	1.00	<0.01	0.56
RW	0.89	1.00	0.45	0.96	1.00	0.89	1.00	0.15	0.98	1.00	1.00	0.95	1	0.02	0.95
SB	1.00	0.57	1.00	1.00	0.28	1.00	1.00	0.96	1.00	0.84	0.89	0.14	0.93	1	0.43
TS	0.96	1.00	0.61	0.99	0.97	0.96	1.00	0.25	1.00	1.00	1.00	0.87	1.00	0.98	1

Arsenic (As)

The average concentration of arsenic in the aboveground eelgrass biomass ranged from 0.56 ±0.12 μg gdw^{-1} (±SE) at Four-Mile Rock to 1.96 ±0.17 μg gdw^{-1} (±SE) at Padilla Bay (Figure 8). The average concentration of arsenic in the aboveground compartment of eelgrass was significantly higher at Padilla Bay compared to the levels measured at the 14 other sites (Figure 8, Table 7).

The average concentration of arsenic in the belowground eelgrass biomass ranged from 0.34 ±0.04 μg gdw^{-1} (±SE) at Holly to 0.98 ±0.04 μg gdw^{-1} (±SE) at Thompson Spit (Figure 8). The average concentration of arsenic in the belowground compartment of eelgrass was significantly higher at Thompson Spit compared to the concentration measured at all other sites except for Ruston Way (Figure 8, Table 7).

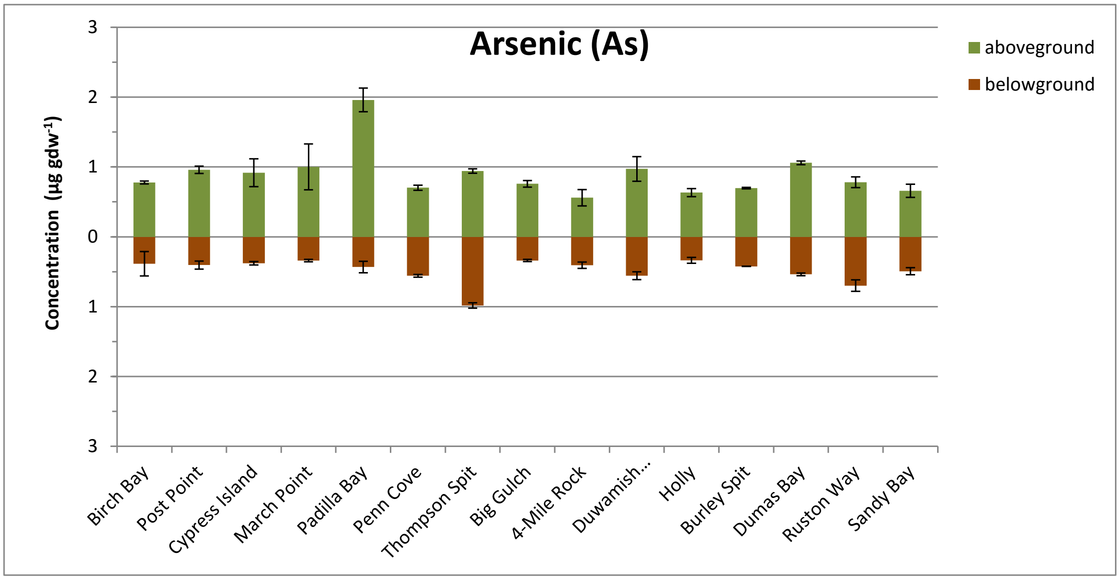

Figure 8. Average concentration (µg gdw^{-1}, ± SE) of arsenic (As) in the above- and belowground compartments of eelgrass (*Zostera marina* L.) measured at 15 sites throughout Puget Sound.

The results of the Tukey's HSD showed 14 significant paired site comparisons in the aboveground eelgrass and 16 significant paired site comparisons in the belowground eelgrass concentrations (Table 7).

Table 7. Results of the Tukey HSD multiple comparison test for average arsenic (As) concentration in the above- (green) and belowground (brown) eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound. Comparisons of the As concentration in the aboveground eelgrass was significant between 14 paired sites. Sixteen (16) paired sites showed a significant difference in the As concentration measured in the belowground eelgrass compartment.

As	BB	BG	BS	CI	DB	DH	FR	HY	MP	PB	PC	PP	RW	SB	TS
BB	1	1.00	1.00	1.00	0.96	1.00	1.00	1.00	0.99	<0.01	1.00	1.00	1.00	1.00	1.00
BG	1.00	1	1.00	1.00	0.93	1.00	1.00	1.00	0.99	<0.01	1.00	1.00	1.00	1.00	1.00
BS	1.00	1.00	1	1.00	0.79	0.97	1.00	1.00	0.93	<0.01	1.00	0.98	1.00	1.00	0.99
CI	1.00	1.00	1.00	1	1.00	1.00	0.80	0.95	1.00	<0.01	1.00	1.00	1.00	0.98	1.00
DB	0.92	0.65	0.99	0.89	1	1.00	0.32	0.56	1.00	<0.01	0.80	1.00	0.96	0.66	1.00
DH	0.84	0.51	0.97	0.79	1.00	1	0.62	0.85	1.00	<0.01	0.97	1.00	1.00	0.91	1.00
FR	1.00	1.00	1.00	1.00	0.97	0.93	1	1.00	0.51	<0.01	1.00	0.67	0.99	1.00	0.73
HY	1.00	1.00	1.00	1.00	0.64	0.50	1.00	1	0.77	<0.01	1.00	0.88	1.00	1.00	0.92
MP	1.00	1.00	1.00	1.00	0.66	0.52	1.00	1.00	1	<0.01	0.93	1.00	1.00	0.84	1.00
PB	1.00	1.00	1.00	1.00	1.00	0.98	1.00	1.00	1.00	1	<0.01	<0.01	<0.01	<0.01	<0.01
PC	0.83	0.50	0.97	0.78	1.00	1.00	0.92	0.49	0.51	0.98	1	0.98	1.00	1.00	0.99
PP	1.00	1.00	1.00	1.00	0.97	0.92	1.00	1.00	1.00	1.00	0.91	1	1.00	0.93	1.00
RW	0.08	0.02	0.18	0.06	0.88	0.95	0.13	0.02	0.02	0.21	0.95	0.12	1	1.00	1.00
SB	1.00	0.91	1.00	0.99	1.00	1.00	1.00	0.90	0.91	1.00	1.00	1.00	0.60	1	0.96
TS	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.16	<0.01	1

Cadmium (Cd)

The average concentration of cadmium in the aboveground eelgrass biomass ranged from 2.7 ±0.7 µg gdw^{-1} (±SE) at March Point to 6.7 ±0.7 µg gdw^{-1} (±SE) at Big Gulch (Figure 9). The average concentration of cadmium in the aboveground compartment of eelgrass was significantly higher at Big Gulch compared to Four-Mile Rock, March Point, Sandy Bay and Thompson Spit (Figure 9, Table 8). The average concentration of cadmium in the aboveground compartment of eelgrass was not significantly different among the other sites except between March Point and Burley Spit, where the concentration measured at Burley Spit was significantly higher (Figure 9, Table 8).

The average concentration of cadmium in the belowground eelgrass biomass ranged from 0.77 ±0.03 µg gdw^{-1} (±SE) at March Point to 2.32 ±0.15 µg gdw^{-1} (±SE) at Thompson Spit (Figure 9). The average concentration of cadmium in the belowground compartment of eelgrass was significantly higher at Thompson Spit compared to the concentration measured at Cypress Island, March Point, Padilla Bay and Sandy Bay (Figure 9, Table 8). The cadmium concentration in the belowground compartment measured at all other sites was not significantly different than the levels observed at Thompson Spit.

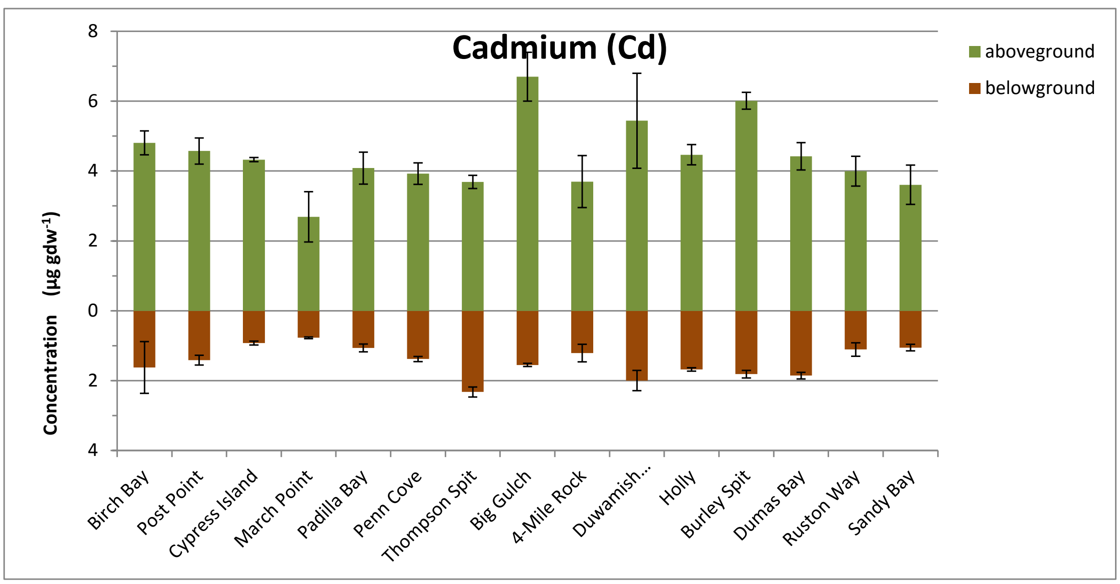

Figure 9. Average concentration (µg gdw^{-1}, ± SE) of cadmium (Cd) in the above- and belowground compartments of eelgrass (*Zostera marina* L.) measured at 15 sites throughout Puget Sound.

Table 8. Results of the Tukey HSD multiple comparison test for average cadmium (Cd) concentration in the above- (green) and belowground (brown) eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound. Comparisons of the Cd concentration in the aboveground eelgrass was significant between five (5) paired sites. The Cd concentration was significantly different between four (4) paired sites in the the belowground eelgrass compartment.

Cd	BB	BG	BS	CI	DB	DH	FR	HY	MP	PB	PC	PP	RW	SB	TS
BB	1	0.55	0.97	1.00	1.00	1.00	0.98	1.00	0.38	1.00	1.00	1.00	1.00	0.97	0.98
BG	1.00	1	1.00	0.22	0.28	0.95	0.04	0.30	<0.01	0.12	0.08	0.37	0.10	0.03	0.04
BS	1.00	1.00	1	0.72	0.79	1.00	0.26	0.82	0.02	0.53	0.40	0.88	0.46	0.21	0.25
CI	0.73	0.84	0.37	1	1.00	0.98	1.00	1.00	0.76	1.00	1.00	1.00	1.00	1.00	1.00
DB	1.00	1.00	1.00	0.30	1	0.99	1.00	1.00	0.68	1.00	1.00	1.00	1.00	1.00	1.00
DH	1.00	0.99	1.00	0.14	1.00	1	0.68	1.00	0.09	0.92	0.84	1.00	0.88	0.60	0.67
FR	0.99	1.00	0.88	1.00	0.82	0.56	1	1.00	0.99	1.00	1.00	1.00	1.00	1.00	1.00
HY	1.00	1.00	1.00	0.62	1.00	1.00	0.98	1	0.65	1.00	1.00	1.00	1.00	1.00	1.00
MP	0.44	0.58	0.17	1.00	0.13	0.05	0.99	0.34	1	0.90	0.96	0.56	0.94	1.00	0.99
PB	0.93	0.97	0.63	1.00	0.55	0.30	1.00	0.86	1.00	1	1.00	1.00	1.00	1.00	1.00
PC	1.00	1.00	0.99	0.98	0.98	0.86	1.00	1.00	0.87	1.00	1	1.00	1.00	1.00	1.00
PP	1.00	1.00	1.00	0.97	0.99	0.90	1.00	1.00	0.82	1.00	1.00	1	1.00	1.00	1.00
RW	0.96	0.99	0.72	1.00	0.64	0.37	1.00	0.91	1.00	1.00	1.00	1.00	1	1.00	1.00
SB	0.92	0.97	0.61	1.00	0.53	0.29	1.00	0.85	1.00	1.00	1.00	1.00	1.00	1	1.00
TS	0.72	0.58	0.96	0.01	0.98	1.00	0.11	0.82	<0.01	0.04	0.28	0.33	0.05	0.04	1

Chromium (Cr)

The average concentration of chromium in the aboveground eelgrass biomass ranged from 0.05 ±0.002 µg gdw^{-1} (±SE) at Ruston Way to 0.27 ±0.06 µg gdw^{-1} (±SE) at Holly. The average (µg gdw^{-1}, ± SE) concentration of chromium in the aboveground compartment of eelgrass was significantly higher at Cypress Island, Big Gulch, Holly, Burley Spit, and Dumas Bay relative to the other 10 sites (Figure 10, Table 9).

The average concentration of chromium in the belowground eelgrass biomass ranged from 0.04 ±0.01 µg gdw^{-1} (±SE) at Ruston Way to 0.35 ±0.02 µg gdw^{-1} (±SE) at Holly. The average (µg gdw^{-1}, ± SE) concentration of chromium in the belowground compartment of eelgrass was significantly higher at Holly, but not significantly different than the cadmium concentration measured at Cypress Island, Thompson Spit, Burley Spit, Dumas Bay, and Sandy Bay (Figure 10, Table 9).

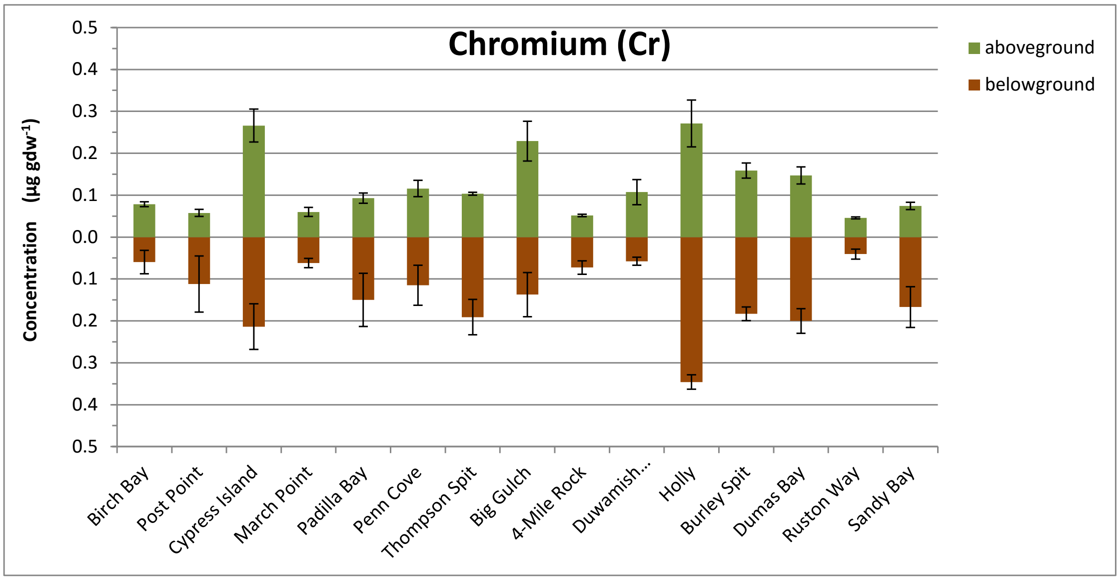

Figure 10. Average concentration (µg gdw^{-1}, ± SE) of chromium (Cr) in the above- and belowground compartments of eelgrass (*Zostera marina* L.) measured at 15 sites throughout Puget Sound.

Table 9. Results of the Tukey HSD multiple comparison test for average chromium (Cr) concentration measured in the above- (green) and belowground (brown) eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound. Comparisons of the Cr concentration in the aboveground eelgrass was significant between 27 paired sites. Only eight (8) paired sites showed a significant difference in the Cr concentration from the belowground eelgrass compartment.

Cr	BB	BG	BS	CI	DB	DH	FR	HY	MP	PB	PC	PP	RW	SB	TS
BB	1	0.01	0.62	<0.01	0.82	1.00	1.00	<0.01	1.00	1.00	1.00	1.00	1.00	1.00	1.00
BG	0.97	1	0.79	1.00	0.59	0.09	<0.01	1.00	<0.01	0.04	0.15	<0.01	<0.01	0.01	0.07
BS	0.59	1.00	1	0.20	1.00	0.98	0.21	0.15	0.31	0.86	1.00	0.28	0.15	0.55	0.96
CI	0.26	0.98	1.00	1	0.10	0.01	<0.01	1.00	<0.01	<0.01	0.01	<0.01	<0.01	<0.01	0.01
DB	0.39	1.00	1.00	1.00	1	1.00	0.36	0.08	0.50	0.96	1.00	0.46	0.27	0.76	0.99
DH	1.00	0.97	0.56	0.24	0.37	1	0.95	0.01	0.99	1.00	1.00	0.98	0.90	1.00	1.00
FR	1.00	1.00	0.74	0.38	0.54	1.00	1	<0.01	1.00	1.00	0.88	1.00	1.00	1.00	0.97
HY	<0.01	0.03	0.19	0.48	0.33	<0.01	<0.01	1	<0.01	<0.01	0.01	<0.01	<0.01	<0.01	<0.01
MP	1.00	0.98	0.61	0.28	0.41	1.00	1.00	<0.01	1	1.00	0.95	1.00	1.00	1.00	0.99
PB	0.92	1.00	1.00	1.00	1.00	0.91	0.97	0.05	0.93	1	1.00	1.00	0.99	1.00	1.00
PC	1.00	1.00	0.99	0.86	0.95	1.00	1.00	0.01	1.00	1.00	1	0.94	0.80	1.00	1.00
PP	1.00	1.00	1.00	0.91	0.97	1.00	1.00	0.03	1.00	1.00	1.00	1	1.00	1.00	0.99
RW	1.00	0.87	0.37	0.13	0.21	1.00	1.00	<0.01	1.00	0.75	0.98	1.00	1	1.00	0.94
SB	0.77	1.00	1.00	1.00	1.00	0.75	0.89	0.10	0.80	1.00	1.00	1.00	0.55	1	1.00
TS	0.49	1.00	1.00	1.00	1.00	0.47	0.65	0.25	0.52	1.00	0.98	0.99	0.29	1.00	1

Copper (Cu)

The average copper concentration in the aboveground eelgrass biomass ranged from 16.0 ±1.9 µg gdw^{-1} (±SE) at Penn Cove to a high of 74.1 ±4.4 µg gdw^{-1} (±SE) measured at Four-Mile Rock (Figure 11). The concentration of copper in the aboveground compartment of eelgrass at Four-Mile Rock was highest but not significantly different than the concentration measured at Birch Bay, March Point, Padilla Bay, Big Gulch, Burley Spit and Sandy Bay (Figure 11, Table 10).

The average copper concentration in the belowground eelgrass biomass ranged from a low of 2.4 ±1.1 µg gdw^{-1} (±SE) at Birch Bay to a high of 15.07 ±1.4 µg gdw^{-1} (±SE) measured at Four-Mile Rock (Figure 11). The average concentration of copper in the belowground compartment of eelgrass was significantly higher at Four-Mile Rock compared to the concentration measured at the other 14 sites (Figure 11, Table 10). The concentration of copper in the belowground compartment of eelgrass was significantly lower at Birch Bay but only compared to the concentration measured at Post Point, Ruston Way, and Sandy Bay (Figure 11, Table 10).

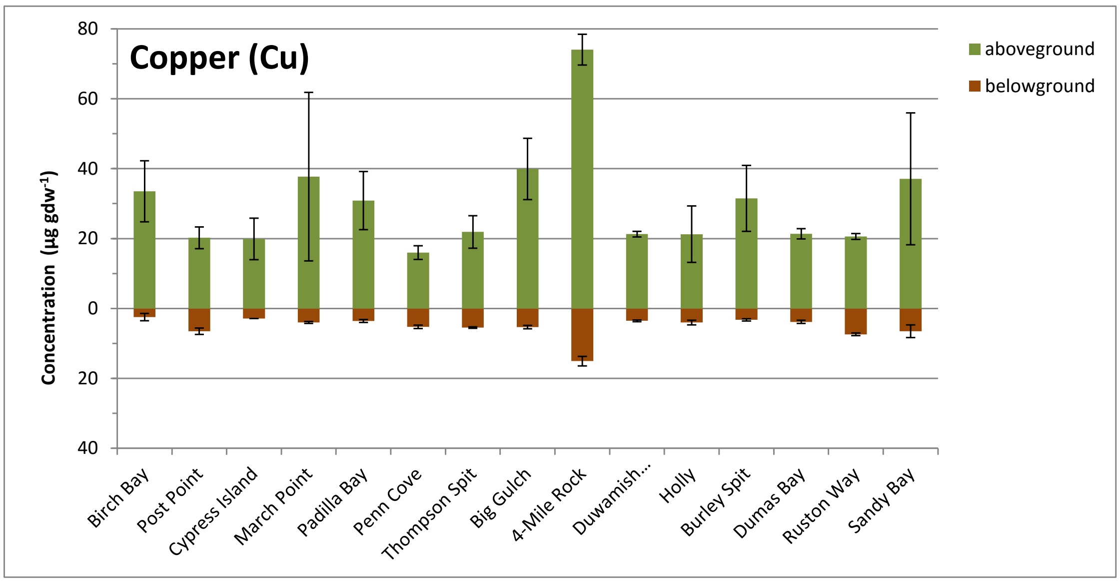

Figure 11. Average concentration (µg gdw^{-1}, ± SE) of copper (Cu) in the above- and belowground compartments of eelgrass (*Zostera marina* L.) measured at 15 sites throughout Puget Sound.

Table 10. Results of the Tukey HSD multiple comparison test for average copper (Cu) concentration measured in the above- (green) and belowground (brown) eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound. Comparisons of the Cu concentration in the aboveground eelgrass were significant between eight (8) paired sites. Nineteen (19) paired sites showed a significant difference in the Cu concentration from the belowground eelgrass compartment.

Cu	BB	BG	BS	CI	DB	DH	FR	HY	MP	PB	PC	PP	RW	SB	TS
BB	1	1.00	1.00	1.00	1.00	1.00	0.23	1.00	1.00	1.00	0.99	1.00	1.00	1.00	1.00
BG	0.37	1	1.00	0.97	0.99	0.99	0.47	0.99	1.00	1.00	0.90	0.98	0.98	1.00	0.99
BS	1.00	0.83	1	1.00	1.00	1.00	0.17	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
CI	1.00	0.64	1.00	1	1.00	1.00	0.03	1.00	0.99	1.00	1.00	1.00	1.00	0.99	1.00
DB	0.99	0.98	1.00	1.00	1	1.00	0.03	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
DH	1.00	0.93	1.00	1.00	1.00	1	0.03	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
FR	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	1	0.03	0.38	0.16	0.01	0.03	0.03	0.35	0.04
HY	0.97	1.00	1.00	1.00	1.00	1.00	<0.01	1	1.00	1.00	1.00	1.00	1.00	1.00	1.00
MP	0.98	0.99	1.00	1.00	1.00	1.00	<0.01	1.00	1	1.00	0.95	0.99	0.99	1.00	1.00
PB	1.00	0.95	1.00	1.00	1.00	1.00	<0.01	1.00	1.00	1	1.00	1.00	1.00	1.00	1.00
PC	0.40	1.00	0.85	0.67	0.99	0.94	<0.01	1.00	1.00	0.96	1	1.00	1.00	0.96	1.00
PP	0.04	1.00	0.20	0.11	0.48	0.31	<0.01	0.60	0.57	0.35	1.00	1	1.00	0.99	1.00
RW	0.01	0.82	0.04	0.02	0.12	0.06	<0.01	0.17	0.16	0.07	0.80	1.00	1	1.00	1.00
SB	0.04	1.00	0.21	0.11	0.49	0.32	<0.01	0.60	0.58	0.35	1.00	1.00	1.00	1	1.00
TS	0.30	1.00	0.76	0.55	0.97	0.88	<0.01	0.99	0.99	0.91	1.00	1.00	0.88	1.00	1

Iron (Fe)

The average aboveground iron concentration in eelgrass ranged from 91.8 ± 3.4 µg gdw^{-1} (± SE) at Ruston Way to 345.3 ±33.3 µg gdw^{-1} (± SE) at Dumas Bay. The iron concentration at Dumas Bay was significantly higher than the iron concentration in the aboveground compartment of eelgrass measured at Penn Cove, Four-Mile Rock, Duwamish Head, Ruston Way and Sandy Bay but it was not significantly higher than the iron concentration measured at the other nine sample sites (Figure 12, Table 11).

The highest average concentration of iron in the belowground compartment of eelgrass was observed at Thompson Spit 419.0 ±62.1 µg gdw^{-1} (± SE). The concentration measured at Thompson Spit was higher than observed iron concentrations at all other sites except for Padilla Bay and Holly (Figure 12, Table 11). The lowest iron concentrations measured in the belowground compartment was 66.5 ±29.9 µg gdw^{-1} (± SE) at Birch Bay (Figure 12); only significantly different than the concentration measured at Holly (Table 11).

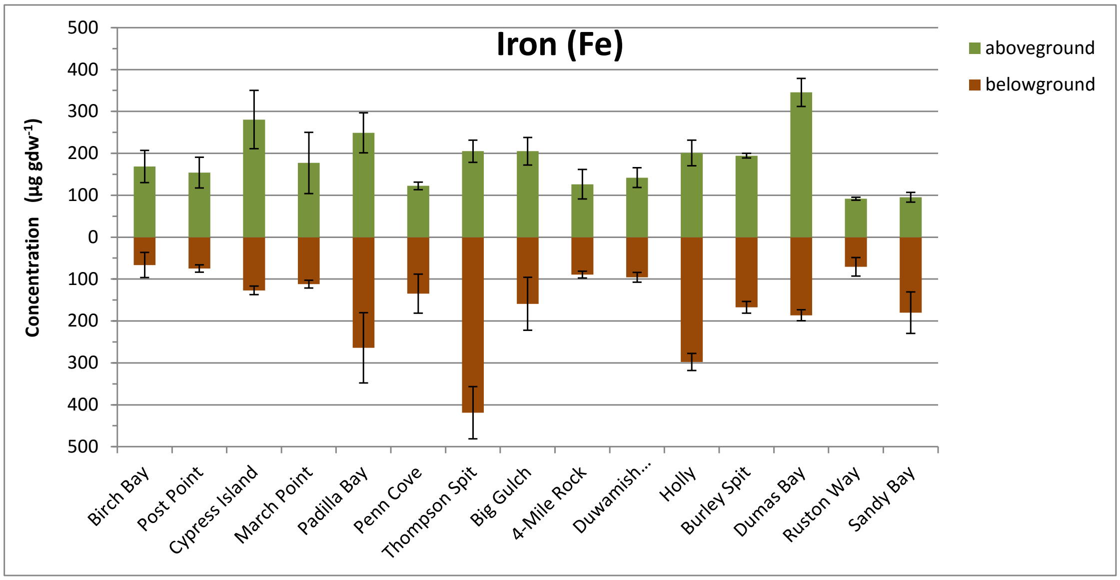

Figure 12. Average concentration (µg gdw^{-1}, ± SE) of iron (Fe) in the above- and belowground compartments of eelgrass (*Zostera marina* L.) measured at 15 sites throughout Puget Sound.

Table 11. Results of the Tukey HSD multiple comparison test for average iron (Fe) concentration measured in the above- (green) and belowground (brown) eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound. Comparisons of the Fe concentration in the aboveground eelgrass was significant between five (5) paired sites. Seventeen (17) paired sites showed a significant difference in the Fe concentration from the belowground eelgrass compartment.

Fe	BB	BG	BS	CI	DB	DH	FR	HY	MP	PB	PC	PP	RW	SB	TS
BB	1	1.00	1.00	0.72	0.11	1.00	1.00	1.00	1.00	0.96	1.00	1.00	0.98	0.98	1.00
BG	0.92	1	1.00	0.98	0.38	1.00	0.97	1.00	1.00	1.00	0.96	1.00	0.71	0.74	1.00
BS	0.86	1.00	1	0.94	0.28	1.00	0.99	1.00	1.00	1.00	0.99	1.00	0.82	0.85	1.00
CI	1.00	1.00	1.00	1	1.00	0.40	0.25	0.97	0.81	1.00	0.22	0.54	0.07	0.08	0.98
DB	0.66	1.00	1.00	1.00	1	0.04	0.02	0.34	0.15	0.88	0.01	0.06	<0.01	<0.01	0.38
DH	1.00	1.00	0.99	1.00	0.93	1	1.00	1.00	1.00	0.78	1.00	1.00	1.00	1.00	1.00
FR	1.00	0.99	0.98	1.00	0.89	1.00	1	0.98	1.00	0.59	1.00	1.00	1.00	1.00	0.97
HY	0.01	0.44	0.53	0.16	0.76	0.05	0.04	1	1.00	1.00	0.97	1.00	0.75	0.79	1.00
MP	1.00	1.00	1.00	1.00	0.98	1.00	1.00	0.09	1	0.99	1.00	1.00	0.94	0.96	1.00
PB	0.06	0.82	0.89	0.46	0.98	0.17	0.14	1.00	0.30	1	0.54	0.89	0.23	0.25	1.00
PC	0.99	1.00	1.00	1.00	1.00	1.00	1.00	0.21	1.00	0.55	1	1.00	1.00	1.00	0.96
PP	1.00	0.96	0.92	1.00	0.75	1.00	1.00	0.02	1.00	0.08	1.00	1	1.00	1.00	1.00
RW	1.00	0.94	0.89	1.00	0.71	1.00	1.00	0.02	1.00	0.07	1.00	1.00	1	1.00	0.71
SB	0.73	1.00	1.00	1.00	1.00	0.96	0.92	0.69	0.99	0.96	1.00	0.81	0.77	1	0.74
TS	<0.01	<0.01	0.01	<0.01	0.01	<0.01	<0.01	0.64	<0.01	0.27	<0.01	<0.01	<0.01	0.01	1

Lead (Pb)

The average aboveground lead concentrations (µg gdw^{-1}, ± SE) in eelgrass ranged from 0.07 ±0.2 µg gdw^{-1} (±SE) at Cypress Island to 0.50 ±0.3 µg gdw^{-1} (±SE) at Post Point (Figure 13). The comparison of Pb concentration in the aboveground eelgrass was significant ($p = 0.015$). However, the variability in lead concentrations measured in the aboveground eelgrass compartments was so great that there were no observed significant differences in concentrations between paired sites based on the Tukey's HSD post-hoc analysis (Table 12).

The highest average concentration for lead in the belowground compartment of eelgrass was observed at Ruston Way (0.35 ±0.18 µg gdw^{-1} (± SE)). The lowest lead concentration was 0.02 µg gdw^{-1} measured at Cypress Island. There is no error associated with the belowground lead concentration at Cypress Island because two samples had Pb concentrations lower than the method detection limit. The concentrations of lead in the belowground biomass were not statistically different between the 15 sample sites ($p = 0.13$; Figure 13) and therefore a Tukey's HSD post-hoc test was not performed.

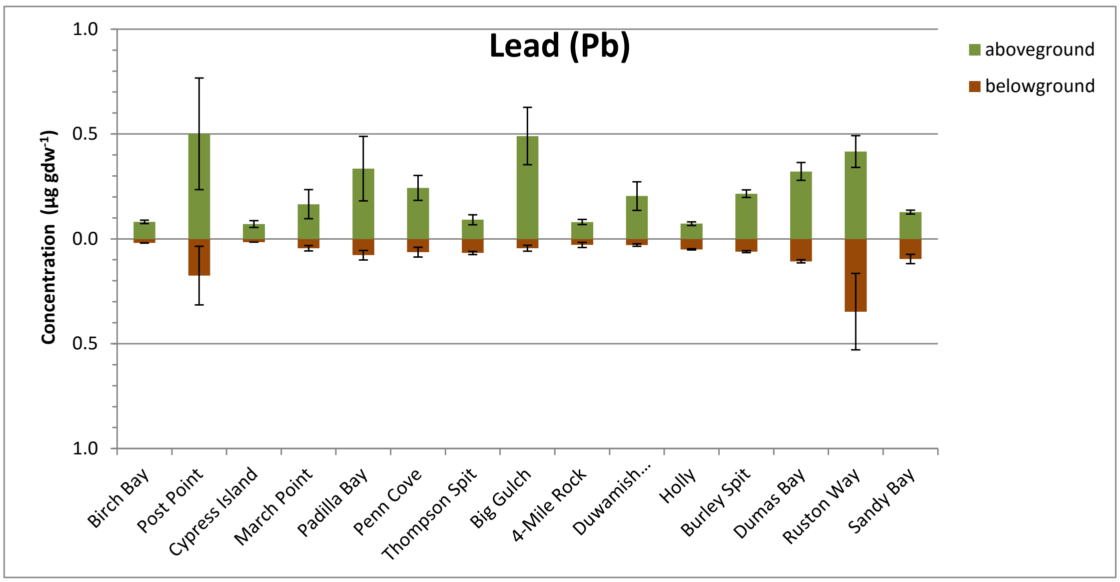

Figure 13. Average concentration (µg gdw^{-1}, ± SE) of lead (Pb) in the above- and belowground compartments of eelgrass (*Zostera marina* L.) measured at 15 sites throughout Puget Sound.

Table 12. Results of the Tukey HSD multiple comparison test for average lead (Pb) concentration in the aboveground eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound. There were no significant post-hoc comparisons between sample sites for Pb concentrations in the aboveground eelgrass. A post-hoc analysis was not performed on the belowground Pb concentration.

Pb	BB	BG	BS	CI	DB	DH	FR	HY	MP	PB	PC	PP	RW	SB	TS
BB	1	0.19	1.00	1.00	0.89	1.00	1.00	1.00	1.00	0.84	1.00	0.17	0.47	1.00	1.00
BG		1	0.76	0.17	0.99	0.71	0.19	0.17	0.52	1.00	0.86	1.00	1.00	0.35	0.22
BS			1	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	0.71	0.97	1.00	1.00
CI				1	0.85	1.00	1.00	1.00	1.00	0.80	0.99	0.14	0.43	1.00	1.00
DB					1	1.00	0.88	0.86	1.00	1.00	1.00	0.99	1.00	0.98	0.91
DH						1	1.00	1.00	1.00	1.00	1.00	0.66	0.95	1.00	1.00
FR							1	1.00	1.00	0.84	1.00	0.16	0.47	1.00	1.00
HY								1	1.00	0.81	0.99	0.15	0.43	1.00	1.00
MP									1	0.99	1.00	0.47	0.85	1.00	1.00
PB										1	1.00	0.99	1.00	0.96	0.87
PC											1	0.82	0.99	1.00	1.00
PP												1	1.00	0.31	0.19
RW													1	0.70	0.52
SB														1	1.00
TS															1

Nickel (Ni)

The average aboveground nickel concentrations (µg gdw^{-1}, ± SE) in eelgrass were significantly higher at Cypress Island (3.6 ±0.3 µg gdw^{-1} (±SE)) relative to the concentrations measured at the other 14 sites. The lowest aboveground nickel concentration measured in the aboveground eelgrass compartment was observed at March Point (0.7 ±0.2 µg gdw^{-1} (±SE); Figure 14). The low nickel concentration measured at March Point was similar to the concentration measured at the other sites except for Cypress Island, Big Gulch and Duwamish Head (Figure 14, Table 13).

The average nickel concentration in the belowground compartment of eelgrass at Cypress Island, 1.3 ±0.1 µg gdw^{-1} (±SE), was statistically higher than concentrations measured in eelgrass at the other sites (Figure 14, Table 13). The site with the second highest nickel concentration in the belowground compartment of eelgrass was Thompson Spit but nickel concentrations in belowground biomass were only significantly different from concentrations measured at Birch Bay, Cypress Island and March Point (Figure 14, Table 13).

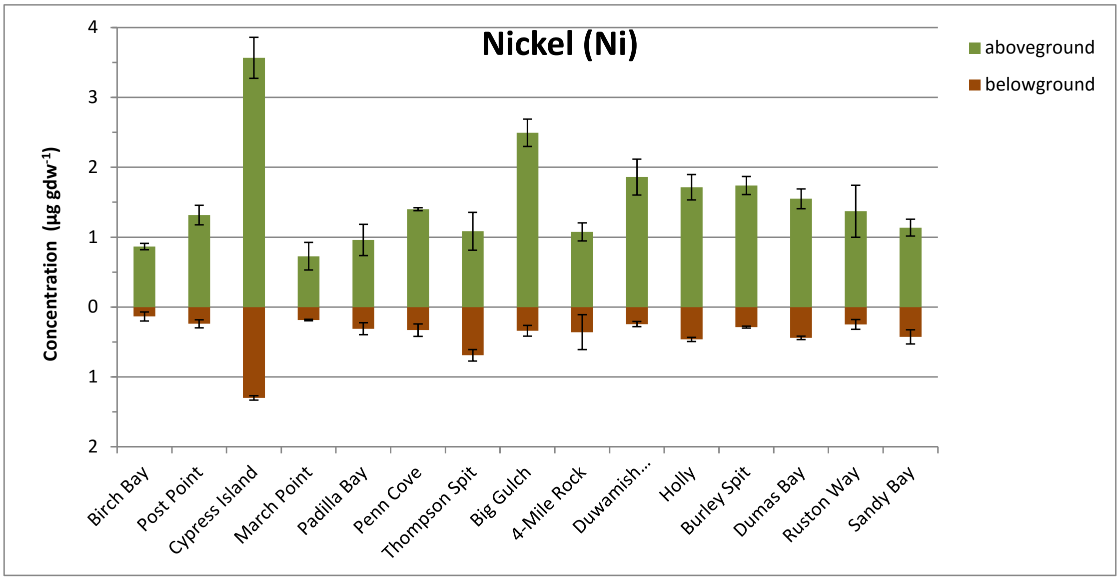

Figure 14. Average concentration (µg gdw^{-1}, ± SE) of nickel (Ni) in the above- and belowground compartments of eelgrass (*Zostera marina* L.) measured at 15 sites throughout Puget Sound.

Table 13. Results of the Tukey HSD multiple comparison test for average nickel (Ni) concentration measured in the above- (green) and belowground (brown) eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound. Comparisons of the Ni concentration in the aboveground eelgrass was significant between 24 paired sites. Sixteen (16) paired sites showed a significant difference in the Ni concentration in the belowground eelgrass compartment.

Ni	BB	BG	BS	CI	DB	DH	FR	HY	MP	PB	PC	PP	RW	SB	TS
BB	1	<0.01	0.19	<0.01	0.54	0.08	1.00	0.22	1.00	1.00	0.85	0.95	0.89	1.00	1.00
BG	0.93	1	0.39	0.04	0.12	0.65	<0.01	0.34	<0.01	<0.01	0.04	0.02	0.03	<0.01	<0.01
BS	0.99	1.00	1	<0.01	1.00	1.00	0.58	1.00	0.07	0.34	1.00	0.97	0.99	0.72	0.60
CI	<0.01	<0.01	<0.01	1	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
DB	0.49	1.00	0.99	<0.01	1	1.00	0.93	1.00	0.26	0.75	1.00	1.00	1.00	0.97	0.94
DH	1.00	1.00	1.00	<0.01	0.95	1	0.33	1.00	0.03	0.16	0.94	0.84	0.91	0.45	0.35
FR	0.88	1.00	1.00	<0.01	1.00	1.00	1	0.65	0.99	1.00	1.00	1.00	1.00	1.00	1.00
HY	0.39	1.00	0.98	<0.01	1.00	0.90	1.00	1	0.09	0.39	1.00	0.98	1.00	0.77	0.66
MP	1.00	0.99	1.00	<0.01	0.75	1.00	0.98	0.65	1	1.00	0.56	0.75	0.63	0.98	0.99
PB	0.98	1.00	1.00	<0.01	1.00	1.00	1.00	0.99	1.00	1	0.96	0.99	0.98	1.00	1.00
PC	0.96	1.00	1.00	<0.01	1.00	1.00	1.00	1.00	1.00	1.00	1	1.00	1.00	1.00	1.00
PP	1.00	1.00	1.00	<0.01	0.94	1.00	1.00	0.88	1.00	1.00	1.00	1	1.00	1.00	1.00
RW	1.00	1.00	1.00	<0.01	0.96	1.00	1.00	0.91	1.00	1.00	1.00	1.00	1	1.00	1.00
SB	0.58	1.00	1.00	<0.01	1.00	0.97	1.00	1.00	0.82	1.00	1.00	0.97	0.98	1	1.00
TS	0.01	0.30	0.14	<0.01	0.79	0.06	0.38	0.87	0.02	0.19	0.26	0.06	0.07	0.72	1

Vanadium (V)

The average concentration of vanadium in the aboveground compartment of eelgrass ranged from 0.5 ±0.1 µg gdw^{-1} (±SE) at Ruston Way to 2.2 ±0.6 µg gdw^{-1} (±SE) at Padilla Bay (Figure 15). Although the concentration of vanadium was significantly different between the 15 sites ($p = 0.001$), there were no significant paired sites results (Table 14).

The average concentration of vanadium in belowground eelgrass compartments ranged from a low of 0.09 ±0.01 µg gdw^{-1} (±SE) at Cypress Island to a high of 1.90 ±0.13 µg gdw^{-1} (±SE) at Thompson Spit (Figure 15). There were significant differences in the belowground eelgrass vanadium concentrations between sites with the highest belowground concentration measured at Thompson Spit. The concentrations of vanadium in the belowground eelgrass biomass at Padilla Bay, Duwamish Head, Holly, Burley Spit, Dumas Bay, and Sandy Bay were also high relative to other sites, but not as high as the concentration measured at Thompson Spit (Figure 15, Table 14). The Tukey's HSD post-hoc test showed vanadium was significantly higher at Thompson Spit compared to all the other sample sites (Table 14).

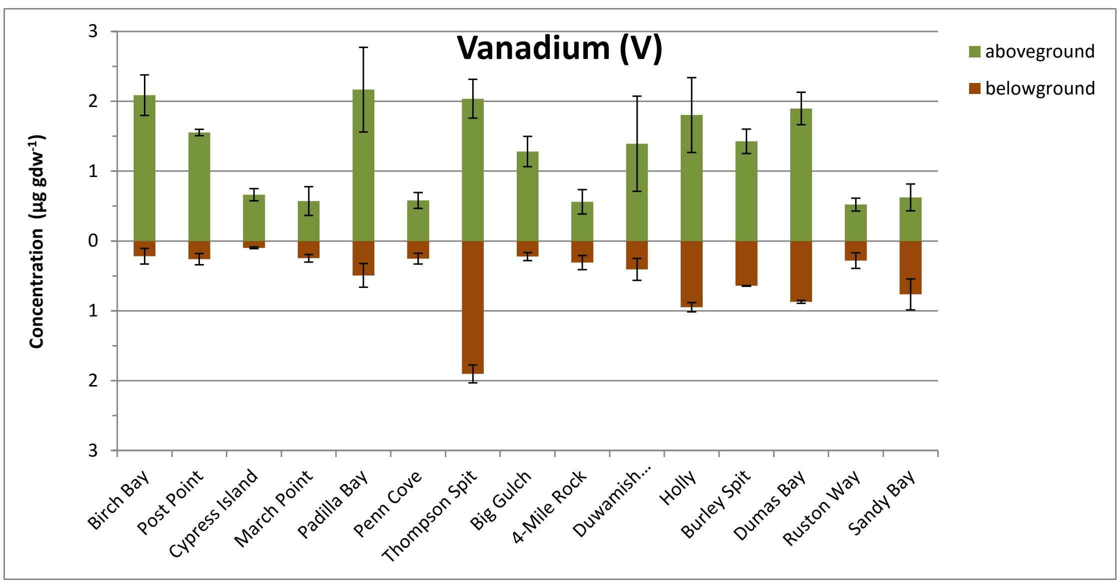

Figure 15. Average concentration (µg gdw^{-1}, ± SE) of vanadium (V) in the above- and belowground compartments of eelgrass (*Zostera marina* L.) measured at 15 sites throughout Puget Sound.

Table 14. Results of the Tukey HSD multiple comparison test for average vanadium (V) concentration in the above- (green) and belowground (brown) eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound. There were no significant difference between paired sites in the comparison of the V concentration in the aboveground eelgrass. Thirty (30) paired sites showed a significant difference in the V concentration measured in the belowground eelgrass compartment.

V	BB	BG	BS	CI	DB	DH	FR	HY	MP	PB	PC	PP	RW	SB	TS
BB	1	0.89	0.97	0.16	1.00	0.96	0.11	1.00	0.11	1.00	0.11	1.00	0.09	0.14	1.00
BG	1.00	1	1.00	0.99	0.99	1.00	0.95	1.00	0.96	0.81	0.96	1.00	0.93	0.98	0.93
BS	0.33	0.34	1	0.92	1.00	1.00	0.83	1.00	0.85	0.94	0.86	1.00	0.79	0.89	0.99
CI	1.00	1.00	0.07	1	0.34	0.94	1.00	0.46	1.00	0.12	1.00	0.81	1.00	1.00	0.20
DB	0.01	0.01	0.97	<0.01	1	1.00	0.24	1.00	0.25	1.00	0.26	1.00	0.20	0.30	1.00
DH	0.99	1.00	0.96	0.78	0.21	1	0.87	1.00	0.88	0.92	0.89	1.00	0.83	0.92	0.98
FR	1.00	1.00	0.69	0.98	0.05	1.00	1	0.34	1.00	0.07	1.00	0.68	1.00	1.00	0.13
HY	<0.01	<0.01	0.79	<0.01	1.00	0.08	0.02	1	0.35	1.00	0.36	1.00	0.29	0.41	1.00
MP	1.00	1.00	0.43	1.00	0.02	1.00	1.00	0.01	1	0.08	1.00	0.69	1.00	1.00	0.14
PB	0.89	0.90	1.00	0.43	0.49	1.00	1.00	0.23	0.95	1	0.08	0.99	0.06	0.10	1.00
PC	1.00	1.00	0.46	1.00	0.02	1.00	1.00	0.01	1.00	0.96	1	0.70	1.00	1.00	0.14
PP	1.00	1.00	0.48	1.00	0.03	1.00	1.00	0.01	1.00	0.97	1.00	1	0.62	0.76	1.00
RW	1.00	1.00	0.58	1.00	0.04	1.00	1.00	0.01	1.00	0.99	1.00	1.00	1	1.00	0.11
SB	0.07	0.08	1.00	0.01	1.00	0.59	0.23	1.00	0.10	0.90	0.12	0.13	0.17	1	0.17
TS	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	1

Zinc (Zn)

The average zinc concentration in the aboveground eelgrass biomass ranged from 56.6 ±1.0 µg gdw^{-1} (±SE) at Thompson Spit to a high of 106.6 ±15.8 µg gdw^{-1} (±SE) measured at Four-Mile Rock (Figure 16). There was a significant difference in zinc concentrations measured in the aboveground eelgrass biomass at the 15 sites (p = 0.001). The zinc concentrations measured in the aboveground eelgrass biomass at Four-Mile Rock were the highest, but only significantly different than the concentrations measured at Cypress Island, Thompson Spit, Holly, and Sandy Bay (Figure 16, Table 15). The only other significant paired site difference in measured zinc concentrations was observed between eelgrass from Thompson Spit and Duwamish Head (Table 15).

The zinc concentrations measured in the belowground eelgrass biomass ranged from 31.5 ±4.5 µg gdw^{-1} (±SE) at Sandy Bay to a high of 68.4 ±3.2 µg gdw^{-1} (±SE) measured at Sandy Bay (Figure 16). Although there was a significant difference between zinc concentrations measured in the belowground eelgrass biomass at the 15 sites (p= 0.038), the Tukey's HSD post-hoc comparison showed no significant differences between paired sites (Table 15).

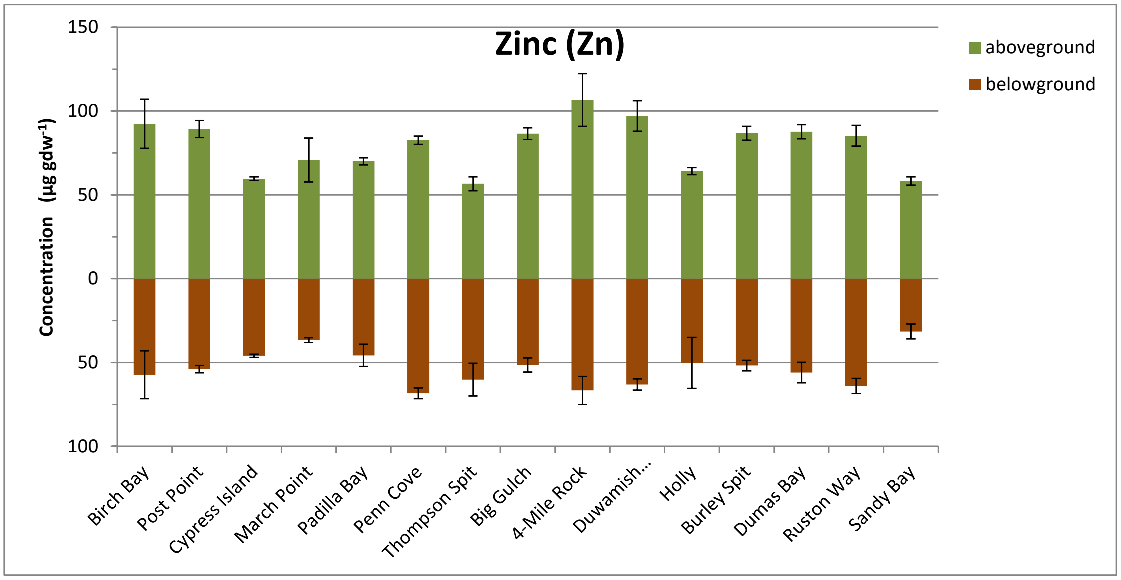

Figure 16. Average concentration (µg gdw^{-1}, ± SE) of zinc (Zn) in the above- and belowground compartments of eelgrass (*Zostera marina* L.) measured at 15 sites throughout Puget Sound.

Table 15. Results of the Tukey HSD multiple comparison test for average zinc (Zn) concentration measured in the above- (green) and belowground (brown) eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound. Comparisons of the Zn concentration in the aboveground eelgrass was significant between five (5) paired sites. There were no significant paired sites in the Zn concentration from the belowground eelgrass compartment.

Zn	BB	BG	BS	CI	DB	DH	FR	HY	MP	PB	PC	PP	RW	SB	TS
BB	1	1.00	1.00	0.19	1.00	1.00	0.99	0.39	0.78	0.74	1.00	1.00	1.00	0.15	0.11
BG	1.00	1	1.00	0.47	1.00	1.00	0.85	0.74	0.97	0.96	1.00	1.00	1.00	0.39	0.31
BS	1.00	1.00	1	0.45	1.00	1.00	0.86	0.72	0.97	0.96	1.00	1.00	1.00	0.38	0.30
CI	1.00	1.00	1.00	1	0.40	0.08	0.01	1.00	1.00	1.00	0.71	0.32	0.54	1.00	1.00
DB	1.00	1.00	1.00	1.00	1	1.00	0.90	0.67	0.95	0.93	1.00	1.00	1.00	0.33	0.26
DH	1.00	1.00	1.00	0.92	1.00	1	1.00	0.19	0.51	0.46	0.99	1.00	1.00	0.06	0.04
FR	1.00	0.97	0.97	0.77	1.00	1.00	1	0.03	0.11	0.09	0.64	0.94	0.79	0.01	0.01
HY	1.00	1.00	1.00	1.00	1.00	0.99	0.94	1	1.00	1.00	0.91	0.57	0.80	1.00	1.00
MP	0.77	0.97	0.97	1.00	0.83	0.41	0.23	0.99	1	1.00	1.00	0.91	0.99	1.00	0.99
PB	1.00	1.00	1.00	1.00	1.00	0.92	0.76	1.00	1.00	1	1.00	0.88	0.98	1.00	0.99
PC	1.00	0.93	0.94	0.66	0.99	1.00	1.00	0.89	0.17	0.65	1	1.00	1.00	0.62	0.53
PP	1.00	1.00	1.00	1.00	1.00	1.00	0.99	1.00	0.92	1.00	0.98	1	1.00	0.26	0.20
RW	1.00	0.99	1.00	0.89	1.00	1.00	1.00	0.99	0.36	0.89	1.00	1.00	1	0.46	0.37
SB	0.45	0.80	0.79	0.98	0.53	0.17	0.09	0.86	1.00	0.98	0.06	0.66	0.15	1	1.00
TS	1.00	1.00	1.00	0.98	1.00	1.00	1.00	1.00	0.59	0.98	1.00	1.00	1.00	0.29	1

Table 16. Average concentration (µg gdw^{-1}) (±SE, n=3) of mercury (Hg), arsenic (As), cadmium (Cd), chromium (Cr), and copper (Cu) measured in the aboveground compartments of eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound.

SITE	Hg		As		Cd		Cr		Cu	
	X	SE	X	SE	X	SE	X	SE	X	SE
Birch Bay	0.0090	0.0010	0.78	0.02	4.81	0.34	0.08	0.01	33.53	8.75
Post Point	0.0112	0.0008	0.96	0.05	4.57	0.37	0.06	0.01	20.23	3.10
Cypress Island	0.0045	0.0001	0.92	0.20	4.33	0.06	0.27	0.04	19.93	5.94
March Point	0.0067	0.0004	1.00	0.33	2.69	0.72	0.06	0.01	37.73	24.10
Padilla Bay	0.0081	0.0001	1.96	0.17	4.08	0.46	0.09	0.01	30.87	8.30
Penn Cove	0.0109	0.0002	0.70	0.04	3.92	0.31	0.12	0.02	16.00	1.98
Thompson Spit	0.0087	0.0004	0.94	0.03	3.69	0.19	0.10	0.00	21.90	4.66
Big Gulch	0.0116	0.0008	0.76	0.05	6.70	0.70	0.23	0.05	39.93	8.76
Four-Mile Rock	0.0099	0.0017	0.56	0.12	3.70	0.74	0.05	0.00	74.07	4.39
Duwamish Head	0.0097	0.0009	0.97	0.18	5.44	1.36	0.11	0.03	21.27	0.78
Holly	0.0065	0.0005	0.63	0.06	4.47	0.29	0.27	0.06	21.24	8.06
Burley Spit	0.0069	0.0002	0.70	0.01	6.01	0.24	0.16	0.02	31.50	9.42
Dumas Bay	0.0158	0.0007	1.06	0.03	4.42	0.39	0.15	0.02	21.37	1.50
Ruston Way	0.0104	0.0011	0.78	0.08	4.00	0.43	0.05	0.00	20.60	0.84
Sandy Bay	0.0060	0.0002	0.66	0.09	3.61	0.56	0.07	0.01	37.10	18.87

Table 17. Average concentration (µg gdw^{-1}) (±SE, n=3) of iron (Fe), lead (Pb), nickel (Ni), vanadium (V) and zinc (Zn) measured in the aboveground compartments of eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound.

SITE	Fe		Pb		Ni		V		Zn	
	X	SE	X	SE	X	SE	X	SE	X	SE
Birch Bay	168.67	38.48	0.08	0.01	0.86	0.04	2.09	0.29	92.40	14.60
Post Point	154.00	36.51	0.50	0.27	1.32	0.14	1.55	0.05	89.33	5.08
Cypress Island	280.67	69.84	0.07	0.02	3.57	0.29	0.66	0.09	59.63	1.04
March Point	177.07	73.24	0.17	0.07	0.73	0.20	0.57	0.21	70.83	13.12
Padilla Bay	249.00	47.51	0.34	0.15	0.96	0.22	2.17	0.61	70.00	2.06
Penn Cove	122.33	8.84	0.24	0.06	1.40	0.02	0.58	0.11	82.57	2.48
Thompson Spit	205.00	26.76	0.09	0.02	1.08	0.27	2.04	0.28	56.63	4.17
Big Gulch	205.00	32.72	0.49	0.14	2.49	0.20	1.28	0.22	86.50	3.54
Four-Mile Rock	126.30	35.07	0.08	0.01	1.08	0.13	0.56	0.18	106.63	15.75
Duwamish Head	142.00	23.58	0.20	0.07	1.86	0.26	1.39	0.68	97.03	9.11
Holly	201.00	30.44	0.07	0.01	1.71	0.18	1.80	0.54	64.13	2.11
Burley Spit	194.33	5.46	0.22	0.02	1.74	0.13	1.43	0.17	86.77	4.16
Dumas Bay	345.33	33.29	0.32	0.04	1.55	0.14	1.90	0.23	87.70	4.20
Ruston Way	91.80	3.39	0.42	0.08	1.37	0.37	0.52	0.09	85.27	6.16
Sandy Bay	95.07	11.69	0.13	0.01	1.14	0.12	0.62	0.19	58.23	2.47

Table 18. Average concentration (µg gdw^{-1}, ±SE, n=3) of mercury (Hg), arsenic (As), cadmium (Cd), chromium (Cr), and copper (Cu) measured in the belowground compartments of eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound.

SITE	Hg		As		Cd		Cr		Cu	
	X	SE	X	SE	X	SE	X	SE	X	SE
Birch Bay	0.0031	0.0004	0.39	0.17	1.62	0.74	0.06	0.03	2.44	1.05
Post Point	0.0049	0.0002	0.41	0.06	1.41	0.14	0.11	0.07	6.52	0.91
Cypress Island	0.0033	0.0007	0.38	0.02	0.92	0.05	0.21	0.05	2.89	0.02
March Point	0.0033	0.0003	0.34	0.02	0.77	0.03	0.06	0.01	3.99	0.29
Padilla Bay	0.0042	0.0004	0.43	0.08	1.07	0.11	0.15	0.06	3.59	0.43
Penn Cove	0.0042	0.0001	0.56	0.02	1.38	0.08	0.12	0.05	5.27	0.48
Thompson Spit	0.0039	0.0001	0.98	0.04	2.32	0.15	0.19	0.04	5.47	0.22
Big Gulch	0.0044	0.0002	0.34	0.02	1.55	0.05	0.14	0.05	5.32	0.48
Four-Mile Rock	0.0036	0.0005	0.41	0.05	1.21	0.25	0.07	0.02	15.07	1.39
Duwamish Head	0.0031	0.0001	0.56	0.06	2.00	0.29	0.06	0.01	3.52	0.25
Holly	0.0024	0.0002	0.34	0.04	1.68	0.05	0.35	0.02	4.03	0.66
Burley Spit	0.0027	0.0001	0.42	0.00	1.81	0.11	0.18	0.02	3.25	0.33
Dumas Bay	0.0047	0.0008	0.54	0.02	1.85	0.09	0.20	0.03	3.83	0.44
Ruston Way	0.0041	0.0003	0.70	0.08	1.11	0.19	0.04	0.01	7.41	0.38
Sandy Bay	0.0032	0.0002	0.49	0.05	1.06	0.10	0.17	0.05	6.52	1.80

Table 19. Average concentration (µg gdw^{-1}, ±SE, n=3) of iron (Fe), lead (Pb), nickel (Ni), vanadium (V) and zinc (Zn) measured in the belowground compartments of eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound.

SITE	Fe		Pb		Ni		V		Zn	
	X	SE	X	SE	X	SE	X	SE	X	SE
Birch Bay	66.47	29.87	0.02	0.00	0.14	0.06	0.22	0.11	57.27	14.24
Post Point	74.67	8.83	0.17	0.14	0.24	0.06	0.26	0.08	53.97	2.22
Cypress Island	127.00	10.02	0.02	-	1.30	0.03	0.10	0.01	46.00	0.93
March Point	111.87	9.40	0.05	0.01	0.19	0.01	0.25	0.05	36.60	1.46
Padilla Bay	264.00	83.76	0.08	0.02	0.31	0.09	0.49	0.17	45.77	6.66
Penn Cove	134.73	46.69	0.06	0.02	0.33	0.09	0.25	0.08	68.43	3.21
Thompson Spit	419.00	62.13	0.07	0.01	0.69	0.08	1.90	0.13	60.20	9.79
Big Gulch	159.03	63.04	0.04	0.01	0.34	0.08	0.22	0.06	51.53	4.24
Four-Mile Rock	89.30	7.93	0.03	0.01	0.36	0.25	0.31	0.10	66.67	8.37
Duwamish Head	95.67	11.69	0.03	0.01	0.24	0.04	0.41	0.16	63.17	3.35
Holly	297.67	20.42	0.05	0.00	0.46	0.03	0.95	0.07	50.27	15.25
Burley Spit	167.33	14.11	0.06	0.00	0.29	0.01	0.64	0.01	51.80	3.14
Dumas Bay	186.33	13.28	0.11	0.01	0.44	0.02	0.87	0.02	56.00	6.06
Ruston Way	70.67	22.13	0.35	0.18	0.25	0.07	0.28	0.11	64.00	4.55
Sandy Bay	180.43	49.50	0.10	0.02	0.43	0.10	0.76	0.22	31.50	4.48

3.3 Organics

3.3.1 Lipids

The ability for organic contaminants to bioaccumulate in organisms is partially dependent on the amount of lipids, or fatty tissue. Seagrasses typically have low lipid concentrations therefore, organic contamination is expected to be low relative to organisms with fatty tissue such as finfish, shellfish and marine mammals. The percent lipids in the above- and belowground eelgrass biomass was measured at only seven sites. The percent lipids were very low and ranged from 0.11 to 0.17% in the aboveground biomass and from 0.07 to 0.15% in the belowground biomass (Figure 17).

There was a significant difference in the average lipids (%) for the aboveground biomass between the seven sites ($p<0.001$). The Tukey's HSD multiple comparison test showed lipids were consistently lower at Post Point (PP) compared to lipids at four other sites (PB, PC, DH, and DB), but not significantly lower than lipids measured at Four-Mile Rock and Burley Spit (Figure 17 and Table 20). Lipid concentrations measured at Penn Cove and Duwamish Head were similar and not different than percent lipids measured in eelgrass at Padilla Bay, but in general percent lipids in eelgrass at these sites were higher than the other four sites. There was no significant difference in the average lipids (%) for the belowground biomass between the seven sites ($p<0.11$).

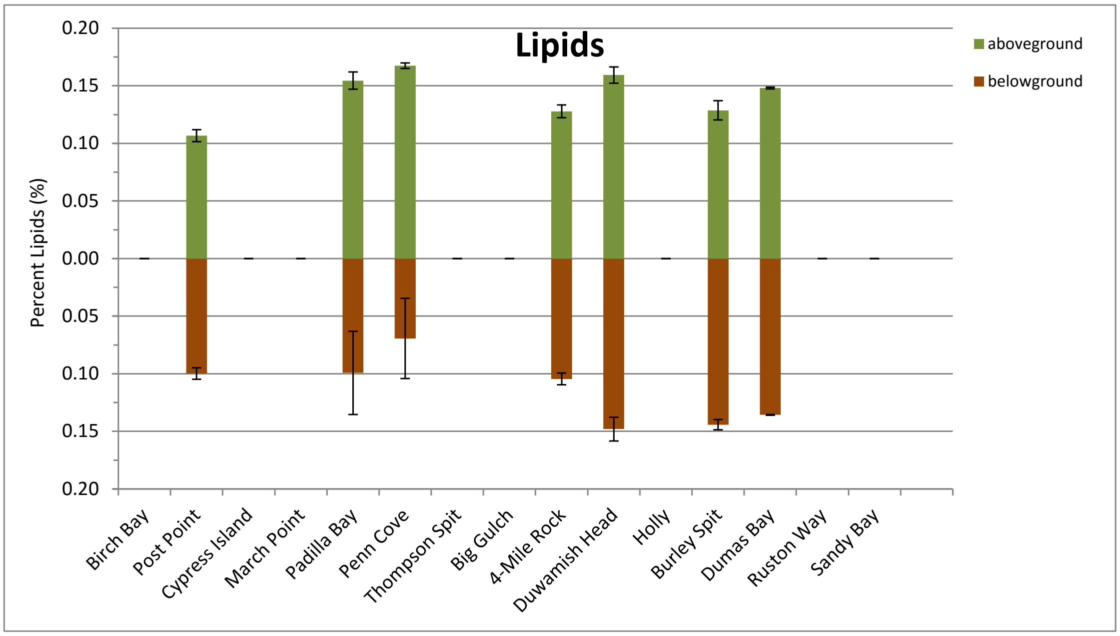

Figure 17. Average percent lipids (%,±SE) in the above- and belowground compartments of eelgrass (*Zostera marina* L.) measured at seven sites throughout Puget Sound.

Table 20. Results of the Tukey HSD multiple comparison test between the average lipids (%) measured in the aboveground eelgrass (*Zostera marina*) at seven sites throughout Puget Sound. A post-hoc test for lipids was not performed on the belowground biomass (p = 0.11).

LIPIDS	PP	PB	PC	FR	DH	BS	DB
PP	1	<0.01	< 0.01	0.22	<0.01	0.19	<0.01
PB		1	0.71	0.07	1.00	0.09	0.98
PC			1	<0.01	0.95	0.01	0.30
FR				1	0.02	1.00	0.25
DH					1	0.03	0.81
BS						1	0.30
DB							1

3.3.2 Polycyclic Aromatic Hydrocarbons

Low molecular weight PAHs

The above- and belowground low molecular weight (LMW) polycyclic aromatic hydrocarbons (PAHs) consisted of the sum of 22 analytes (Appendix A, Table A-2). The average aboveground concentration of LMW PAHs ranged from 8.3 ±1.41 ng gww^{-1} (±SE) at March Point to a high of 24.9 ±4.39 ng gww^{-1} (±SE) at Four-Mile Rock (Figure 18, Table 21). There was a significant difference in the average LMW PAH concentration for the aboveground biomass between sites ($p<0.001$). The LMW PAH concentration observed at Four-Mile Rock was significantly higher than the concentration measured at all other sites except for Penn Cove, Big Gulch, and Dumas Bay (Figure 18, Table 22).

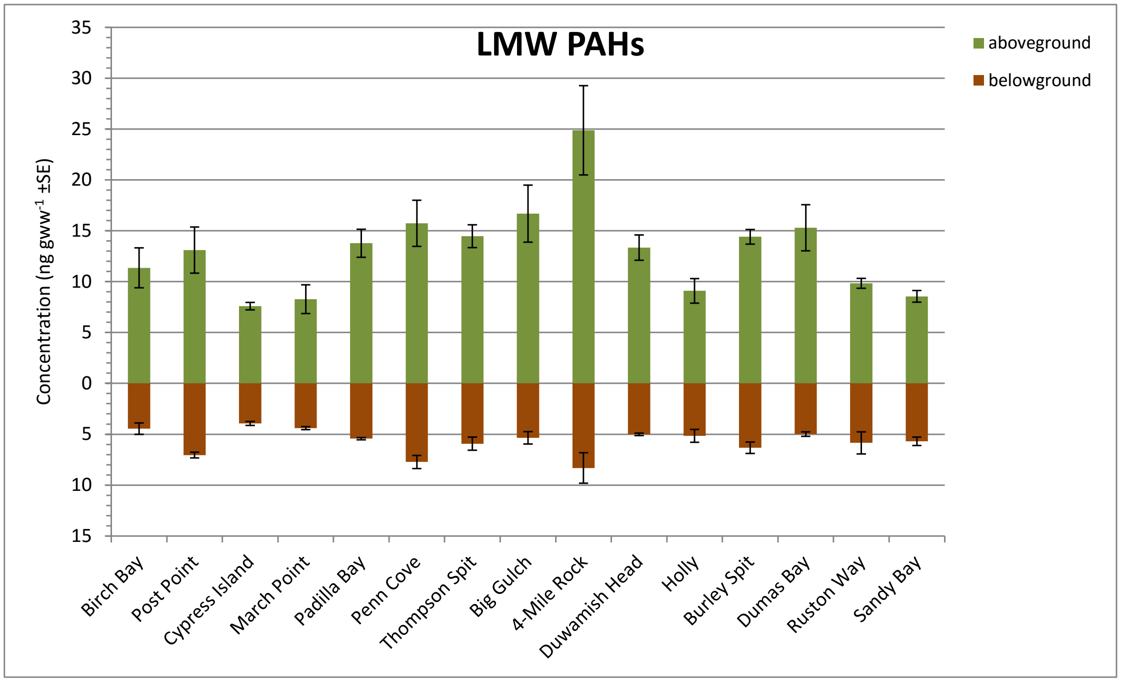

Figure 18. Average concentration (ng gww^{-1}, ±SE) of low molecular weight (LMW) polycyclic aromatic hydrocarbons (PAHs) in the above- and belowground compartments of eelgrass (*Zostera marina* L.) measured at 15 sites throughout Puget Sound.

The average belowground concentration of LMW PAHs ranged from 3.9 ±0.19 ng gww^{-1} (±SE) at Cypress Island to a high of 8.3 ±1.50 ng gww^{-1} (±SE) at Four-Mile Rock (Figure 17, Table 20). There significant difference in the average LMW PAH concentration for the aboveground biomass between sites ($p<0.001$). The LMW PAH concentration observed at Four-Mile Rock was significantly higher than Birch Bay, Cypress Island, and March Point, otherwise, there was no difference between the average LMW PAH concentration between Four-Mile Rock and the remaining 11 sites (Figure 17, Table 22).

Table 21. The average concentration (ng gww^{-1}, ±SE, n=3) of low molecular weight (LMW) polycyclic aromatic hydrocarbons (PAHs) in the aboveground and belowground compartment of eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound.

SITE	ABOVEGROUND		BELOWGROUND	
	X	SE	X	SE
Birch Bay	11.4	1.97	4.5	0.57
Post Point	13.1	2.27	7.1	0.28
Cypress Island	7.6	0.37	3.9	0.19
March Point	8.3	1.41	4.4	0.15
Padilla Bay	13.8	1.38	5.4	0.11
Penn Cove	15.7	2.27	7.7	0.65
Thompson Spit	14.5	1.12	5.9	0.65
Big Gulch	16.7	2.81	5.3	0.61
Four-Mile Rock	24.9	4.39	8.3	1.50
Duwamish Head	13.3	1.24	5.0	0.12
Holly	9.1	1.21	5.2	0.64
Burley Spit	14.4	0.72	6.3	0.56
Dumas Bay	15.3	2.27	5.0	0.22
Ruston Way	9.8	0.48	5.9	1.09
Sandy Bay	8.6	0.57	5.7	0.42

Table 22. Results of the Tukey HSD multiple comparison test between the average concentrations of low molecular weight (LMW) polycyclic aromatic hydrocarbon (PAH) measured in the above- (green) and belowground (brown) eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound. Comparisons of the LMW PAH concentrations in the aboveground eelgrass were significant between 11 paired sites. There were significant differences observed for the LMW PAH concentrations in the belowground eelgrass compartment at seven (7) paired sites.

LMW PAH	BB	BG	BS	CI	DB	DH	FR	HY	MP	PB	PC	PP	RW	SB	TS
BB	1	0.81	1.00	0.98	0.98	1.00	<0.01	1.00	1.00	1.00	0.94	1.00	1.00	1.00	1.00
BG	1.00	1	1.00	0.11	1.00	0.99	0.21	0.31	0.18	1.00	1.00	0.99	0.46	0.22	1.00
BS	0.73	1.00	1	0.47	1.00	1.00	0.04	0.81	0.63	1.00	1.00	1.00	0.92	0.70	1.00
CI	1.00	0.95	0.37	1	0.29	0.72	<0.01	1.00	1.00	0.62	0.22	0.77	1.00	1.00	0.46
DB	1.00	1.00	0.97	1.00	1	1.00	0.07	0.62	0.43	1.00	1.00	1.00	0.78	0.49	1.00
DH	1.00	1.00	0.97	1.00	1.00	1	0.01	0.96	0.86	1.00	1.00	1.00	0.99	0.90	1.00
FR	0.01	0.12	0.66	<0.01	0.05	0.05	1	<0.01	<0.01	0.02	0.11	0.01	<0.01	<0.01	0.04
HY	1.00	1.00	0.99	0.99	1.00	1.00	0.07	1	1.00	0.91	0.51	0.97	1.00	1.00	0.80
MP	1.00	1.00	0.70	1.00	1.00	1.00	0.01	1.00	1	0.77	0.33	0.89	1.00	1.00	0.61
PB	1.00	1.00	1.00	0.93	1.00	1.00	0.14	1.00	1.00	1	1.00	1.00	0.98	0.83	1.00
PC	0.06	0.38	0.96	0.01	0.20	0.20	1.00	0.27	0.05	0.44	1	1.00	0.68	0.39	1.00
PP	0.26	0.84	1.00	0.08	0.61	0.62	0.98	0.72	0.23	0.88	1.00	1	1.00	0.93	1.00
RW	0.96	1.00	1.00	0.71	1.00	1.00	0.33	1.00	0.94	1.00	0.73	0.99	1	1.00	0.92
SB	0.98	1.00	1.00	0.81	1.00	1.00	0.24	1.00	0.98	1.00	0.62	0.97	1.00	1	0.68
TS	0.94	1.00	1.00	0.66	1.00	1.00	0.38	1.00	0.92	1.00	0.78	0.99	1.00	1.00	1

Note: The p-values for the pairwise results in LMW PAH concentrations in belowground eelgrass biomass between Four-Mile Rock - Dumas Bay and Penn Cove - March Point were 0.0490, while the p-value between Four-Mile Rock – Duwamish Head was 0.0500.

High molecular weight PAHs

The above- and belowground high molecular weight (HMW) polycyclic aromatic hydrocarbons (PAHs) consisted of the sum of 20 analytes (Appendix A, Table A-2). The average aboveground concentration of HMW PAHs ranged from 1.5 ±0.41 ng gww^{-1} (±SE) at Sandy Bay to a high of 31.0 ±3.89 ng gww^{-1} (±SE) at Four-Mile Rock (Figure 19, Table 23). There was a significant difference in the average HMW PAH concentration for the aboveground biomass between sites ($p<<0.011$). The HMW PAH concentration observed at Four-Mile Rock was significantly higher than all other sites sampled (Figure 18, Table 24). Big Gulch had the second highest HMW PAH concentration measured in the aboveground compartment of eelgrass (Figure 18). The concentration of HMW PAH measured at Big Gulch was significantly higher than six other sites; Cypress Island, March Point, Padilla Bay, Thompson Spit, Holly, and Sandy Bay (Figure 19, Table 24).

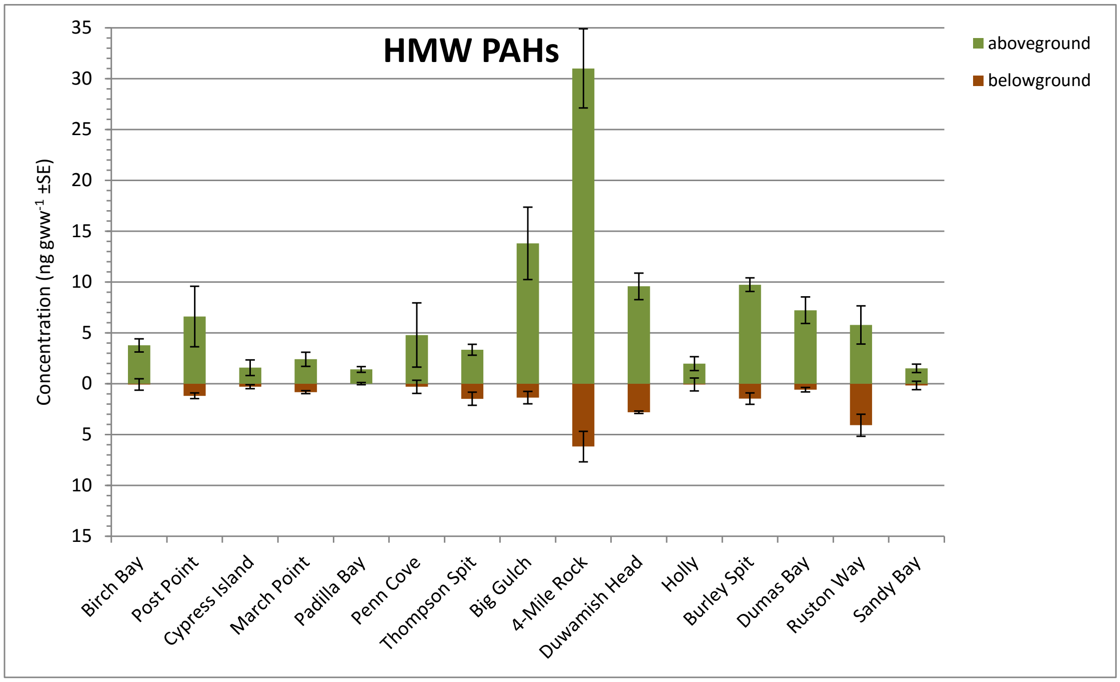

Figure 19. The average concentration (ng gww^{-1}, ±SE) of high molecular weight (HMW) polycyclic aromatic hydrocarbons (PAHs) in the above- and belowground compartments of eelgrass (*Zostera marina* L.) measured at 15 sites throughout Puget Sound.

The average belowground concentration of HMW PAHs ranged from 0.08 ±0.08 ng gww^{-1} (±SE) observed at Birch Bay and Holly to a high of 6.2 ±2.67 ng gww^{-1} (±SE) at Four-Mile Rock (Figure 19, Table 23). There was a significant difference in the average HMW PAH concentration for the belowground biomass between sites ($p=0.001$). The HMW PAH concentration observed at Four-Mile Rock was significantly higher than all sites except for Thompson Spit, Ruston Way, and Burley Spit (Figure 19, Table 24).

Table 23. The average concentration (ng gww^{-1}, ±SE, n=3) of high molecular weight (HMW) polycyclic aromatic hydrocarbons (PAHs) in the aboveground and belowground compartment of eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound. Blank cells indicate a value below the Limit of Quanitification (LOQ).

SITE	ABOVEGROUND		BELOWGROUND	
	X	SE	X	SE
Birch Bay	3.8	0.65	0.1	0.08
Post Point	6.6	2.98	1.2	0.08
Cypress Island	1.6	0.77	0.3	0.30
March Point	2.4	0.69	0.8	0.13
Padilla Bay	1.4	0.28		
Penn Cove	4.8	3.16	0.3	0.16
Thompson Spit	3.3	0.54	1.5	0.32
Big Gulch	13.8	3.56	1.4	0.42
Four-Mile Rock	31.0	3.89	6.2	2.67
Duwamish Head	9.6	1.31	2.8	0.45
Holly	2.0	0.67	0.1	0.08
Burley Spit	9.7	0.67	1.5	0.62
Dumas Bay	7.2	1.30	0.6	0.13
Ruston Way	5.8	1.88	4.1	2.07
Sandy Bay	1.5	0.41	0.2	0.18

Table 24. Results of the Tukey HSD multiple comparison test for high molecular weight (HMW) polycyclic aromatic hydrocarbon (PAH) concentration in the above- (green) and belowground (brown) eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound. Comparisons of the HMW PAH concentration in the aboveground eelgrass was significant between 20 paired sites. There were 10 significant paired sites in the HMW PAH concentration from the belowground eelgrass compartment.

HMW PAH	BB	BG	BS	CI	DB	DH	FR	HY	MP	PB	PC	PP	RW	SB	TS
BB	1	0.05	0.68	1.00	0.99	0.72	<0.01	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
BG	1.00	1	0.97	0.01	0.53	0.96	<0.01	0.01	0.02	0.01	0.12	0.40	0.24	0.01	0.04
BS	1.00	1.00	1	0.22	1.00	1.00	<0.01	0.29	0.37	0.20	0.88	1.00	0.98	0.21	0.58
CI	1.00	1.00	1.00	1	0.75	0.25	<0.01	1.00	1.00	1.00	1.00	0.86	0.96	1.00	1.00
DB	1.00	1.00	1.00	1.00	1	1.00	<0.01	0.83	0.90	0.71	1.00	1.00	1.00	0.74	0.98
DH	0.71	1.00	1.00	0.81	0.91	1	<0.01	0.31	0.40	0.22	0.90	1.00	0.98	0.24	0.62
FR	<0.01	0.05	0.05	0.01	0.01	0.40	1	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
HY	1.00	1.00	1.00	1.00	1.00	0.71	<0.01	1	1.00	1.00	1.00	0.92	0.98	1.00	1.00
MP	1.00	1.00	1.00	1.00	1.00	0.96	0.02	1.00	1	1.00	1.00	0.96	0.99	1.00	1.00
PB	1.00	1.00	1.00	1.00	1.00	0.68	<0.01	1.00	1.00	1	0.99	0.84	0.95	1.00	1.00
PC	1.00	1.00	1.00	1.00	1.00	0.81	0.01	1.00	1.00	1.00	1	1.00	1.00	1.00	1.00
PP	1.00	1.00	1.00	1.00	1.00	0.99	0.03	1.00	1.00	1.00	1.00	1	1.00	0.85	1.00
RW	0.17	0.73	0.76	0.24	0.35	1.00	0.94	0.17	0.46	0.15	0.24	0.64	1	0.96	1.00
SB	1.00	1.00	1.00	1.00	1.00	0.76	<0.01	1.00	1.00	1.00	1.00	1.00	0.20	1	1.00
TS	1.00	1.00	1.00	1.00	1.00	1.00	0.06	1.00	1.00	1.00	1.00	1.00	0.78	1.00	1

Note: The p-value for the pairwise result in HMW PAH concentrations in aboveground eelgrass between Birch Bay – Big Gulch was 0.053. In the results of the pairwise comparison for the belowground HMW PAH concentration, the p-value between Four-Mile Rock – Burley Spit was 0.052.

3.3.3 Persistent Organic Pollutants

There were five general types of persistent organic pollutants analyzed in the above- and belowground compartments of eelgrass at the 15 study sites throughout Puget Sound. These groups included:

- Polychlorinated biphenyls (PCBs) – used in a range of industrial uses such as electrical insulators, capacitors, and oils.
- Polybrominated diphenyl ethers (PBDEs) – fire retardants
- Dichlorodiphenyltrichloroethane (DDT) – insecticides
- Chlordanes (CHLDs) – pesticides
- Hexachlorocyclohexanes (HCHs) – insecticides

The analysis found inconsistencies in the results with interfering peaks in the chromatogram and values below the limit of quanitification (LOQ, Gaeckle 2012) for dichlorodiphenyltrichloroethane (DDTs), Chlordanes (CHLDs) and Hexachlorocyclohexanes (HCHs). Due to the inconsistencies in these data and values recorded below the LOQ, the results of these three groups (DDTs, CHLDs and HCHs) of persistent organic compounds are not presented.

Polychlorinated Biphenyls (PCBs)

The average concentration of PCBs in the above- and belowground compartments of eelgrass varied throughout the 15 sample sites in Puget Sound (Figure 19). The average aboveground concentration of 40 PCB congeners ranged from a low of 0.19 ±0.10 ng gww^{-1} (±SE) at Penn Cove to a high of 1.58 ±0.56 ng gww^{-1} (±SE) measured at Ruston Way (Figure 20, Table 25). The average concentration of the 40 PCBs in the aboveground eelgrass compartment was significantly different between sites ($p=0.002$). However, the average concentration of the 40 PCBs measured in the aboveground eelgrass at Ruston Way was only significantly higher than the concentration measured at Post Point, Padilla Bay, and Penn Cove (Figure 20, Table 26). There was no significant difference in the average concentration of 40 PCB congeners in the belowground eelgrass between the 15 sample sites ($p = 0.06$). At two of the sites, Post Point and Penn Cove, the analysis in the belowground eelgrass compartment was restricted due to a Limit of Quanititation (LOQ) in the level of the 40 PCB congeners measured (Figure 20, Table 25).

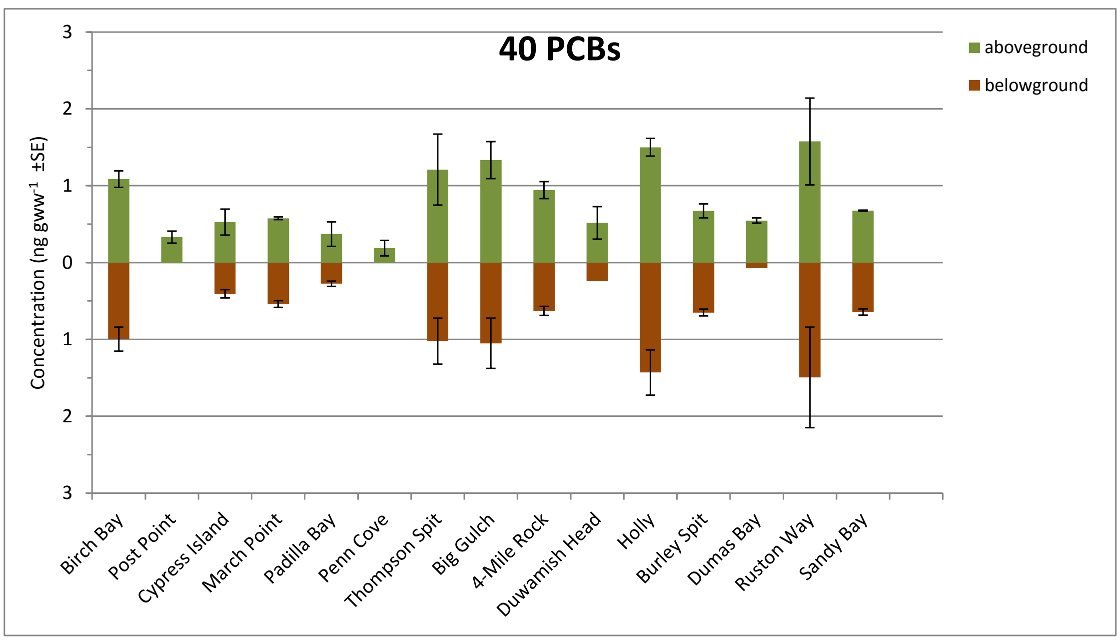

Figure 20. The average concentration (ng gww^{-1}, ±SE) of 40 polychlorinated biphenyls (PCBs) in the above- and belowground compartments of eelgrass (*Zostera marina* L.) measured at 15 sites throughout Puget Sound. Note: there are no values for the concentration of 40 PCBs in the belowground eelgrass at Post Point and Penn Cove as the sample analysis at these sites were below the Limit of Quantitation (LOQ). At Duwamish Head and Dumas Bay the belowground 40 PCB sample size (n) = 1, therefore a standard error could not be calculated for these two sites.

Table 25.The average concentration (ng gww^{-1}, ±SE, n=3) of 40 polychlorinated biphenyls (PCBs) in the aboveground and belowground compartment of eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound. Blank cells indicate a value below the Limit of Quanitification (LOQ) or the inability to calculate a standard error because sample size (n) = 1.

SITE	ABOVEGROUND		BELOWGROUND	
	X	SE	X	SE
Birch Bay	1.09	0.11	1.00	0.16
Post Point	0.33	0.08		
Cypress Island	0.53	0.17	0.41	0.05
March Point	0.58	0.02	0.54	0.04
Padilla Bay	0.37	0.16	0.28	0.03
Penn Cove	0.19	0.10		
Thompson Spit	1.21	0.46	1.02	0.30
Big Gulch	1.33	0.24	1.05	0.33
Four-Mile Rock	0.94	0.11	0.63	0.06
Duwamish Head	0.52	0.21	0.24	
Holly	1.50	0.12	1.43	0.29
Burley Spit	0.67	0.09	0.65	0.04
Dumas Bay	0.55	0.03	0.07	
Ruston Way	1.58	0.56	1.49	0.65
Sandy Bay	0.68	0.01	0.64	0.04

Table 26. Results of the Tukey HSD multiple comparison test for the average concentration of the sum of 40 PCB congeners (PCB 40) in the aboveground (green) eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound. Comparisons of the average PCB 40 concentration in the aboveground eelgrass was only significant between 3 paired sites. The average PCB 40 concentration in the belowground eelgrass was not significantly different between sites.

PCB 40	BB	BG	BS	CI	DB	DH	FR	HY	MP	PB	PC	PP	RW	SB	TS
BB	1	1.00	0.99	0.90	0.92	0.89	1.00	0.99	0.95	0.64	0.47	0.56	0.96	0.99	1.00
BG		1	0.75	0.46	0.50	0.44	1.00	1.00	0.56	0.21	0.15	0.17	1.00	0.76	1.00
BS			1	1.00	1.00	1.00	1.00	0.42	1.00	1.00	0.99	1.00	0.29	1.00	0.93
CI				1	1.00	1.00	0.99	0.20	1.00	1.00	1.00	1.00	0.12	1.00	0.71
DB					1	1.00	0.99	0.22	1.00	1.00	1.00	1.00	0.14	1.00	0.74
DH						1	0.99	0.19	1.00	1.00	1.00	1.00	0.12	1.00	0.68
FR							1	0.90	1.00	0.88	0.72	0.83	0.80	1.00	1.00
HY								1	0.26	0.07	0.06	0.06	1.00	0.43	1.00
MP									1	1.00	1.00	1.00	0.17	1.00	0.80
PB										1	1.00	1.00	0.04	1.00	0.40
PC											1	1.00	0.03	0.99	0.28
PP												1	0.03	1.00	0.33
RW													1	0.30	1.00
SB														1	0.93
TS															1

Total PCBs

A similar pattern emerged in the concentration of total PCBs (TOT PCBs) measured in the above- and belowground eelgrass compartments at the sample sites throughout Puget Sound.

The lowest average concentration of the total PCBs in the aboveground compartment of eelgrass was 0.13 ±0.04 ng gww^{-1} (±SE) at Penn Cove and the highest concentration of 1.83 ±0.54 ng gww^{-1} (±SE) was observed at Ruston Way (Figure 21, Table 27). There was a significant difference in the concentration of total PCBs in the aboveground eelgrass compartment between all the sample sites ($p<0.001$). Although the average concentration of total PCBs was highest at Ruston Way, the measured concentration was not significantly different than the concentration observed at Birch Bay, March Point, Thompson Spit, Big Gulch, Holly and Sandy Bay (Figure 21, Table 28).

The average concentration of the total PCBs in the belowground eelgrass biomass was significantly different between sites ($p=0.005$) and highest at Ruston Way, Birch Bay, Thompson Spit, Big Gulch and Holly compared to the other 10 sites (Figure 21, Table 27, Table 28). Data from four sites were not included in the statistical analyses due to insufficient data. At Post Point and Penn Cove the total PCBs measured in all three samples were below the LOQ. At Duwamish Head and Dumas Bay the total PCBs in two of the three samples were below the LOQ. The data presented for Duwamish Head and Dumas Bay is the total PCBs measured in only one sample ($n=1$) (Figure 21, Table 27).

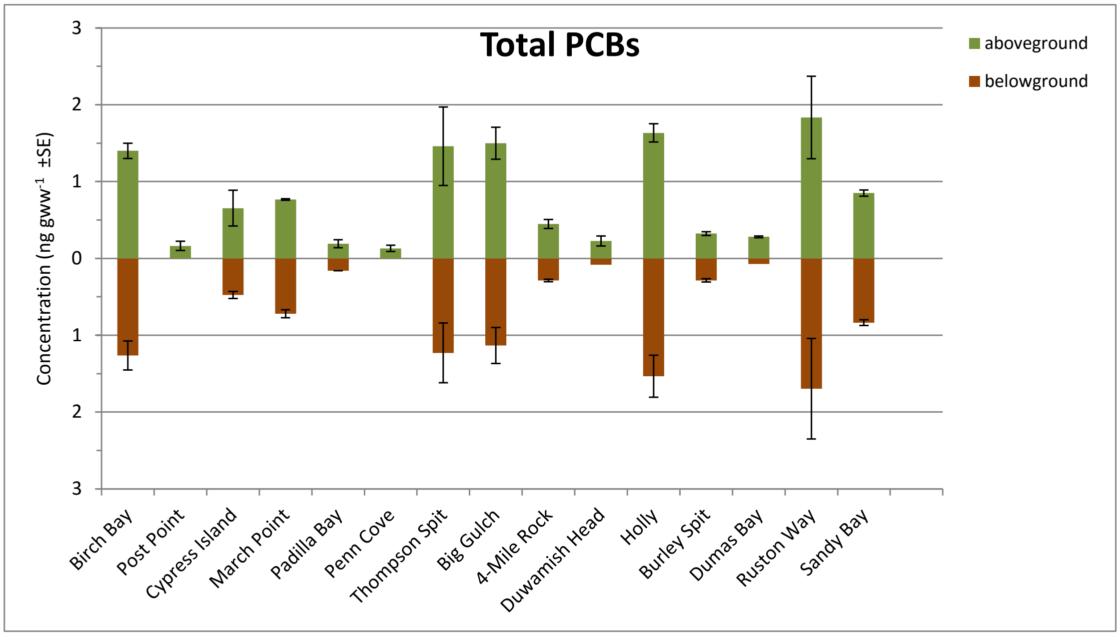

Figure 21. The average concentration (ng gww-1, ±SE) of all the polychlorinated biphenyls (PCBs) in the above- and belowground compartments of eelgrass (*Zostera marina* L.) measured at 15 sites throughout Puget Sound. The concentration of total PCBs in the belowground samples from Post Point and Penn Cove were less than the Limit of Quantitation (LOQ) and therefore were not included in the figure. At Duwamish Head and Dumas Bay the belowground 40 PCB sample size (n) = 1, therefore a standard error could not be calculated for these two sites.

Table 27. The average concentration (ng gww-1, ±SE, n=3) of all polychlorinated biphenyls (PCBs) in the aboveground and belowground compartment of eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound. Blank cells indicate a value below the Limit of Quanitification or the inability to calculate a standard error.

SITE	ABOVEGROUND		BELOWGROUND	
	X	SE	X	SE
Birch Bay	1.40	0.10	1.26	0.19
Post Point	0.16	0.06		
Cypress Island	0.65	0.23	0.48	0.05
March Point	0.77	0.01	0.72	0.05
Padilla Bay	0.19	0.05	0.16	0.00
Penn Cove	0.13	0.04		
Thompson Spit	1.46	0.51	1.23	0.39
Big Gulch	1.50	0.21	1.13	0.24
Four-Mile Rock	0.45	0.06	0.29	0.02
Duwamish Head	0.23	0.07	0.08	
Holly	1.63	0.12	1.53	0.27
Burley Spit	0.32	0.02	0.29	0.02
Dumas Bay	0.28	0.01	0.07	
Ruston Way	1.83	0.54	1.70	0.7
Sandy Bay	0.85	0.04	0.84	0.0

Table 28.Results of the Tukey HSD multiple comparison test for the average concentration of total polychlorinated biphenyls (TOT PCBs) in the above- (green) and belowground (brown) eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound. Comparisons of the TOT PCB concentration in the aboveground eelgrass was significant between 30 paired sites. There were two (2) significant paired sites in the HMW PAH concentration from the belowground eelgrass compartment.

TOT PCB	BB	BG	BS	CI	DB	DH	FR	HY	MP	PB	PC	PP	RW	SB	TS
BB	1	1.00	0.08	0.52	0.06	0.04	0.18	1.00	0.75	0.03	0.05	0.02	0.98	0.89	1.00
BG	1.00	1	0.04	0.33	0.03	0.02	0.09	1.00	0.55	0.01	0.03	0.01	1.00	0.72	1.00
BS	0.31	0.50	1	1.00	1.00	1.00	1.00	0.01	0.98	1.00	1.00	1.00	<0.01	0.91	0.05
CI	0.60	0.80	1.00	1	1.00	0.98	1.00	0.15	1.00	0.97	0.96	0.95	0.04	1.00	0.40
DB					1	1.00	1.00	0.01	0.95	1.00	1.00	1.00	<0.01	0.86	0.04
DH						1	1.00	0.01	0.90	1.00	1.00	1.00	<0.01	0.77	0.02
FR	0.31	0.50	1.00	1.00			1	0.04	1.00	1.00	1.00	1.00	0.01	0.99	0.12
HY	1.00	0.99	0.09	0.22			0.09	1	0.29	<0.01	0.01	<0.01	1.00	0.44	1.00
MP	0.92	0.99	0.98	1.00			0.98	0.56	1	0.85	0.86	0.80	0.08	1.00	0.63
PB	0.30	0.46	1.00	1.00			1.00	0.10	0.95	1	1.00	1.00	<0.01	0.70	0.02
PC											1	1.00	<0.01	0.73	0.03
PP												1	<0.01	0.64	0.02
RW	0.98	0.91	0.04	0.10			0.04	1.00	0.31	0.05			1	0.15	1.00
SB	0.98	1.00	0.92	1.00			0.92	0.74	1.00	0.86			0.48	1	0.79
TS	1.00	1.00	0.36	0.65			0.36	1.00	0.95	0.34			0.97	0.99	1

Note: The p-value for the pairwise result in TOT PCB concentrations in aboveground eelgrass between Birch Bay – Penn Cove was 0.050, whereas the p-value for the pairwise comparison between Burley Spit – Thompson Spit was 0.051. The p-value for the pairwise comparison in the belowground eelgrass biomass TOT PCB concentrations between Ruston Way – Padilla Bay was 0.045.

Polybrominated diphenyl ethers (PBDEs)

The average concentration of PBDEs in the aboveground eelgrass biomass ranged from a low of 0.06 ±0.01 ng gww^{-1} (±SE) at Holly to a high of 0.40 ±0.09 ng gww^{-1} (±SE) measured at Four-Mile Rock (Figure 22, Table 29). The concentration of PBDEs in the aboveground eelgrass biomass varied significantly across the 13 sample sites (p=0.032); Padilla Bay and Penn Cove were omitted from statistical analyses due to only one sample each. However, a post hoc assessment found the concentration of PBDEs in the aboveground biomass measured at Four-Mile Rock was only significantly different compared to the concentration measured at Holly and Burley Spit (Figure 22, Table 30).

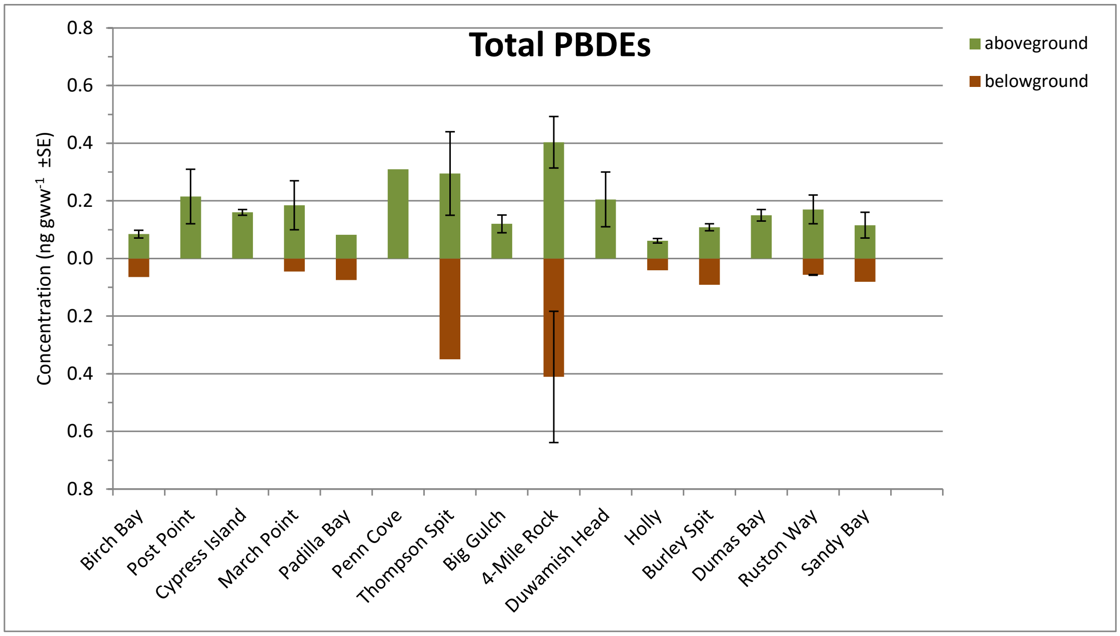

Figure 22. The average concentration (ng gww^{-1}, ±SE) of total polybrominated diphenyl ethers (PBDEs) in the above- and belowground compartments of eelgrass (*Zostera marina* L.) measured at 15 sites throughout Puget Sound. Note: In cases where standard errors (±SE) were not calculated for above- and belowground concentration of PBDEs, only one sample (n=1) of the three replicates analyzed measured a concentration above the Limit of Quantitation (LOQ). The concentration of total PBDEs in the three replicate belowground samples from Post Point, Cypress Island, Penn Cove, Big Gulch, Duwamish Head, and Dumas Bay was less than the LOQ so these data were not included in the figure.

There were limited PBDE results from the belowground eelgrass biomass sample analyses. PBDEs were present and above the LOQ in all three samples from Four-Mile Rock but only measurable in two samples from Ruston Way and one sample from seven other sites (Figure 21, Table 28). The average concentration of PBDEs in the belowground eelgrass biomass ranged from a low of 0.04 ng gww^{-1} (n=1) at Holly to a high of 0.41 ±0.23 ng gww^{-1} (±SE) measured at Four-Mile Rock (Figure 22, Table 29). Due to the limited results it was not possible to conduct statistical analyses on these data.

Table 29. The average concentration (ng gww^{-1}, ±SE, n=3) of polybrominated diphenyl ethers (PBDEs) in the aboveground and belowground compartment of eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound. Blank cells indicate a value below the limit of quanitification or the inability to calculate a standard error.

SITE	ABOVEGROUND		BELOWGROUND	
	X	SE	X	SE
Birch Bay	0.08	0.01	0.06	
Post Point	0.22	0.10		
Cypress Island	0.16	0.01		
March Point	0.19	0.09	0.05	
Padilla Bay	0.08		0.08	
Penn Cove	0.31			
Thompson Spit	0.30	0.15	0.35	
Big Gulch	0.12	0.03		
Four-Mile Rock	0.40	0.09	0.41	0.23
Duwamish Head	0.21	0.10		
Holly	0.06	0.01	0.04	
Burley Spit	0.11	0.01	0.09	
Dumas Bay	0.15	0.02		
Ruston Way	0.17	0.05	0.06	0.00
Sandy Bay	0.12	0.04	0.08	

Table 30. Results of the Tukey HSD multiple comparison test for the average concentration of total polybrominated diphenyl ethers (TOT PBDEs) in the above- (green) eelgrass (*Zostera marina*) at 13 sites throughout Puget Sound; Padilla Bay and Penn Cove were omitted from the statistical analyses. Comparisons of the TOT PBDEs concentration in the aboveground eelgrass was significant between 2 paired sites. Statistical tests were not performed on the belowground TOT PBDEs due to limited detection in samples.

TOT PBDE	BB	BG	BS	CI	DB	DH	FR	HY	MP	PP	RW	SB	TS
BB	1	1.00	1.00	1.00	1.00	0.98	0.06	1.00	1.00	0.96	1.00	1.00	0.58
BG		1	1.00	1.00	1.00	1.00	0.06	1.00	1.00	0.99	1.00	1.00	0.70
BS			1	1.00	1.00	0.99	0.04	1.00	1.00	0.98	1.00	1.00	0.62
CI				1	1.00	1.00	0.27	0.99	1.00	1.00	1.00	1.00	0.95
DB					1	1.00	0.12	0.99	1.00	1.00	1.00	1.00	0.88
DH						1	0.54	0.88	1.00	1.00	1.00	1.00	1.00
FR							1	0.01	0.41	0.61	0.32	0.11	0.98
HY								1	0.95	0.83	0.98	1.00	0.32
MP									1	1.00	1.00	1.00	0.99
PP										1	1.00	1.00	1.00
RW											1	1.00	0.97
SB												1	0.77
TS													1

<u>*Dichloro-diphenyl-trichloroethane* (DDT)</u>

The average concentration of DDT in the aboveground eelgrass biomass ranged from a low of 0.05 ng gww^{-1} (n=1) at March Point to a high of 0.12 ±0.01 ng gww^{-1} (±SE) measured at Padilla Bay and 0.12 ±0.02 ng gww^{-1} (±SE) measured at Dumas Bay (Figure 23, Table 31). Statistical tests were performed between the five sites that had concentrations of DDT measured in the aboveground eelgrass biomass and results showed no difference between sites (p = 0.186).

The average concentration of DDT in the belowground eelgrass biomass ranged from 0.04 ng gww^{-1} (n = 1) measured at March Point to a high of 0.09 ±0.01 ng gww^{-1} (±SE) measured at Burley Spit (Figure 23, Table 31).

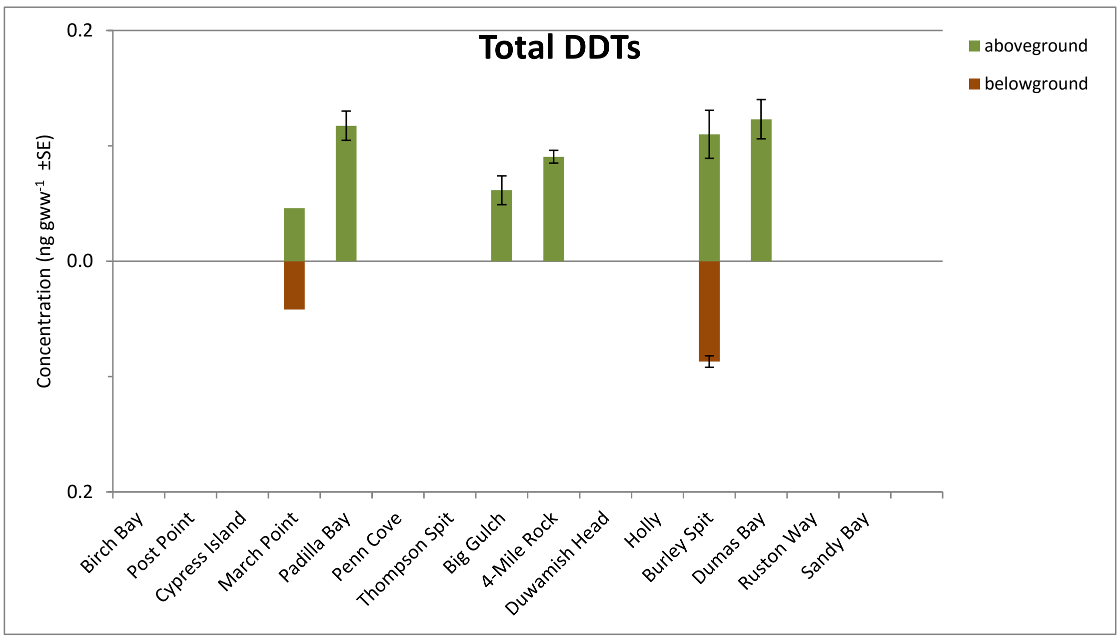

Figure 23. The average concentration (ng gww^{-1}, ±SE) of total dichloro-diphenyl-trichloroethane (DDT) in the above- and belowground compartments of eelgrass (*Zostera marina* L.) measured at 15 sites throughout Puget Sound. Note: Missing standard errors (±SE) indicate only one sample (n=1) of three replicates analyzed measured a concentration of DDT above the LOQ. No data indicates no replicates measured DDT concentration above the LOQ.

Table 31. The average concentration (ng gww^{-1}, ±SE, n=3) of dichloro-diphenyl-trichloroethane (DDT) in the aboveground and belowground compartment of eelgrass (*Zostera marina*) at 15 sites throughout Puget Sound. Blank cells indicate a value below the limit of quanitification or the inability to calculate a standard error.

SITE	ABOVEGROUND		BELOWGROUND	
	X	SE	X	SE
Birch Bay				
Post Point				
Cypress Island				
March Point	0.05		0.04	
Padilla Bay	0.12	0.01		
Penn Cove				
Thompson Spit				
Big Gulch	0.06	0.01		
Four-Mile Rock	0.09	0.01		
Duwamish Head				
Holly				
Burley Spit	0.11	0.02	0.09	0.01
Dumas Bay	0.12	0.02		
Ruston Way				
Sandy Bay				

4 DISCUSSION

4.1 Carbon and Nitrogen

The nutrient content, %C, %N, $\delta^{13}C$ and $\delta^{15}N$, in the above- and belowground eelgrass compartments varied between the 15 sites throughout Puget Sound. There was no obvious pattern indicating a gradient towards higher or lower areas of nutrient enrichment or loading. The values measured in Puget Sound were within the range observed in other studies (Tables 32 and 33). Carbon and nitrogen content of eelgrass have been measured throughout its range and the observed values are considerably more variable than values measured in other seagrass species (Duarte 1990). Previous studies found the range of percent carbon in eelgrass between 28 – 43% DW and nitrogen content between 1.2 – 5.6 % DW (Duarte 1990). For comparison, the average percent carbon measured in eelgrass across the 15 sites in Puget Sound was 37.3% DW and nitrogen was 3.9% DW.

Table 32. Summary of aboveground **percent carbon and nitrogen content and δ^{13}**C and **δ^{15}**N in eelgrass from different sources including this study.

Aboveground		Sample Size	%C	%N	C:N	$\delta^{15}N$	$\delta^{13}C$
		(n)	mean (±SE)	mean (±SE)	mean (±SE)	mean (±SE)	mean (±SE)
Tomales Bay, CA	Fourqurean et al. 1997 (Aug 1992)	23	38.4 (0.3)	2.37 (0.12)	20.0 (1.1)	9.5 (0.3)	-9.9 (0.3)
Tomales Bay, CA	Fourqurean et al. 1997 (Aug 1992 & Jul 1994)	72	36.3 (0.6)	2.33 (0.07)	19.7 (0.7)	9.7 (0.3)	-9.6 (0.2)
Yaquina Bay, OR	Kaldy 2014 (Feb-Apr 2008)	6-9		3.8 (0.3)	15-34	6 (0.1)	-9 (0.2)
Back Sound, NC	Kenworthy & Thayer 1984		42.8	2.02	21		
Global average	Duarte 1990	46	28-43	1.2-5.6	17		
Global average	Hemminga & Mateo 1996	17					-9.2 (1.7)
Samish Bay, WA	Conway-Cranos et al. 2015	10				7.6 (0.5)	9.3 (0.5)
Hamma Hamma, Hood Canal, WA	Conway-Cranos et al. 2015	6				6.0 (0.2)	-9.9 (0.7)
Dosewallips, Hood Canal, WA	Conway-Cranos et al. 2015	6				6.8 (0.2)	-8.9 (0.3)
Puget Sound, WA	Yang et al. 2013	85 (5 site^{-1})			9.4 (0.19) – 12.8 (0.52)		
Puget Sound, WA	this study (Jan 2013)	45 (3 site^{-1})	37.3 (0.2)	3.9 (0.07)	8.4 (0.16)	8.7 (0.3)	-14.4 (0.2)

The ratio of carbon to nitrogen is a potential indicator of nutrient enrichment in seagrass beds. Hemminga and Duarte (2000) suggested that C:N ratios less than 20 indicate nutrient enriched systems. The 15 site average C:N ratio measured in Puget Sound was 8.4 ± 0.16 (±SE) for the aboveground eelgrass biomass and 10.3 ±0.44 (±SE) for the belowground biomass suggesting high nutrient availability. These values may differ slightly if measured during the growing season as nutrients are quickly processed in nearshore waters. The relatively low C:N ratio indicates that eelgrass in Puget Sound is exposed to relatively high concentrations of nitrogen, compared to seagrass growing in other areas of the world. Puget Sound receives the majority of its nitrogen from natural sources (Roberts et al. 2014). The low C:N ratios indicate eelgrass may be vulnerable to increases in nitrogen load relative to the already high background concentrations in Puget Sound (Mackas and Harrison 1997, Short et al. 2014). While there is no clear evidence of eelgrass decline due to high nitrogen concentrations in Puget Sound, it is well documented that nutrient enrichment is one of the primary factors for seagrass declines worldwide (Short and Wyllie-Echeverria 1996, Orth et al. 2006, Waycott et al. 2009, Short et al. 2014, Unsworth et al. 2014).

Stable isotope ratios of nitrogen are often used for source tracking of nutrients from anthropogenic sources. Sewage and manure often have relatively high $\delta^{15}N$ ratios compared to natural background levels (Lapoint et al. 2004). Nutrients derived from fertilizer have low $\delta^{15}N$ since they are derived from atmospheric nitrogen when manufactured (McClelland et al. 1997). While $\delta^{15}N$ ratios in eelgrass leaves vary from 5.9 to 11.8‰, there is no clear gradient in $\delta^{15}N$ over the 15 sites measured in our study that would suggest a relationship to sources of human or agriculture waste.

Table 33. Summary of belowground percent carbon and nitrogen content **and δ^{13}**C and **δ^{15}**N in eelgrass from different sources including this study.

Belowground		Sample Size	%C	%N	C:N	**δ^{15}N**	**δ^{13}C**
		(n)	mean (±SE)	mean (±SE)	mean (±SE)	mean (±SE)	mean (±SE)
Tomales Bay, CA	Fourqurean et al. 1997 (Aug 1992)	23					
Tomales Bay, CA	Fourqurean et al. 1997 (Aug 1992 & Jul 1994)	72					
Yaquina Bay, OR	Kaldy 2014 (Feb-Apr 2008)	6-9		1.4 (0.2)	26 (1.5)	6 (0.4)	-10 (0.3)
Back Sound, NC	Kenworthy & Thayer 1984 (rhizome)		43.4	0.66	65		
Back Sound, NC	Kenworthy & Thayer 1984 (roots)		40	0.92	43		
Global average	Duarte 1990	46					
Puget Sound, WA	this study (Jan 2013)	45 (3 $site^{-1}$)	32.5 (0.2)	2.9 (0.13)	10.3 (0.44)	7.3 (0.2)	-13 (0.2)

4.2 Metals and Contaminants

Metals and contaminants are incorporated into seagrass leaves and vascular tissue through assimilation from the water column and the sediment (Bester 2000, Brinkhuis et al. 1980, Schwartzschild et al. 1994). Studies have demonstrated that metal and contaminant concentrations in seagrass tissue often reflect the availability of these substances in the environment based on the similarities between concentrations in plant compartments (e.g., leaves, rhizomes, and roots) and the environment (sediment and water) (Bester 2000, Pergent-Martini and Pergent 2000, Marín-Guirao et al. 2005, Lyngby and Brix 1982, Sanchiz et al. 2001). Depending on the availability of metals and contaminants and physiological conditions of eelgrass at a site, uptake of excess metals can cause toxicity and metabolic or morphological effects.

Although the order of metal concentrations in the aboveground and belowground compartments of eelgrass differed slightly, there was general agreement in the order of the most abundant metals (Fe, Zn, and Cu) and these results were consistent with findings in other global studies (Brix and Lyngby 1984, DeCasabianca et al. 2004, Kaldy 2006, McRoy 1970). In some studies, the rank order of metal concentrations did not align with the results observed in Puget Sound because samples were collected adjacent to contaminated sites. Brix and Lyngby (1982) showed that Cd, Pb, and Zn bio-accumulated in eelgrass while Cu concentrations declined with age due to translocation or leaf senescence. In another study in Denmark, the concentrations of four metals were similar between the leaf and rhizome-root compartments measured across a gradient of pollution (Zn > Cu > Pb > Cd), while the levels of three metals (Zn, Cu, and Pb) were significantly elevated near an industrial city center and a wastewater discharge (Brix et al. 1983). Elevated levels of Cd, Cu, Pb, and Zn were also observed in eelgrass at a site within close proximity of mining activities in southern Spain compared to sites in more pristine areas (Stenner and Nickless 1975).

In general, the concentration of the 10 metals analyzed from eelgrass aboveground and belowground biomass in Puget Sound fell within the range observed in other global studies that assessed metals in eelgrass (Appendix A Table A-3). Some of the differences can be attributed to the relative location of a sample site to a contaminated or industrial site, metropolitan areas, and the season during which sampling took place.

A discussion of the concentration of metals in the above- and belowground eelgrass follows with comparison to concentrations measured in eelgrass previously measured at a few sites in Puget Sound (USFWS 1994) and elsewhere throughout its range (Govers et al. 2014, Lewis and Devereux 2009). Information of the physiological effects of excess metal concentrations in eelgrass biomass is limited. Most research has focused on plant response to the exposure of a specific metal concentration over a controlled study period (duration), than assess the physiological response to certain metal concentrations in seagrass biomass (Lewis and Devereux 2009). Therefore, the ability to identify sites where metal concentrations pose a threat to eelgrass physiology in Puget Sound will be limited to the availabity of observations from other studies.

In addition, a comparison of the concentration of six metals (mercury, arsenic cadmium, copper, lead, and zinc) measured in mussels as part of the Mussel Watch Pilot Expansion Project (Lanksbury et al. 2014) relative to the concentrations measured in eelgrass will be discussed.

Mercury (Hg)

Mercury concentrations measured in Puget Sound were slightly lower than values found in the literature. The mercury concentrations measured at March Point in this study were considerably lower than concentrations measured near the same location in 1994 (Table 34; USFWS). However, the mercury concentrations were similar at the Padilla Bay site between the two studies.

Nearly all (99%) of the mussels deployed throughout Puget Sound for the Mussel Watch Pilot Expansion Project had higher mercury concentrations than baseline conditions after ~60 days of exposure (Lanksbury et al. 2014). The highest concentrations of mercury in mussels measured as part of the Mussel Watch Pilot Expansion Project (Lanksbury et al. 2014) were 3 to 5 times greater than concentrations measured in eelgrass. One similarity between the two studies was the relatively low mercury concentrations observed at the Cypress Island site in both mussels and eelgrass.

Arsenic (As)

Arsenic concentrations in this study were more similar to thoses measured in the Piscataqua River and Great Bay Estuary (Johnston et al. 1994a, 1994b; Short 1994) than those measured from the 1994 research at Padilla Bay and March Point (Appendix A, Table A-3; Table 34; USFWS 1994). Arsenic concentrations from the 1994 sampling by USFWS were orders of magnitude higher than measured in this study.

Although regulations control discharge limits on the March Point petroleum refineries, the significantly higher concentrations of arsenic measured in aboveground eelgrass biomass at Padilla could be from residual levels in the system. The refineries at March Point went online in 1958, and considering predominant winds are from the southwest during months with the greatest precipitation (October – March), the fallout from refinery activities likely ended up on the Padilla Bay eelgrass meadow. The low arsenic levels observed in the eelgrass from the March Point site may be artifact of weather patterns and how metals precipitate out from the refinery discharge. A similar pattern was not observed in the Mussel Watch Pilot Expansion Project (Lanksbury et al. 2014). Arsenic concentrations in mussels from Padilla Bay (4.8-5.4 µg gdw^{-1}) were lower than concentrations in mussels at March Point (5.5-5.7 µg gdw^{-1}).

Cadmium (Cd)

Cadmium is another chemical that is commonly discharged into Puget Sound through stormwater conduits. Its primary source within the watershed is from roofing materials (Ecology and King County 2011). Even with the high population density along the eastern shores of Puget Sound, measured cadmium concentrations fell with in the range observed in other studies that measured cadmium in eelgrass (Appendix A, Table A-3). It is not clear why cadmium concentration at Big Gulch was statistically higher than four other sites (Four-Mile Rock, March Point, Sandy Bay, and Thompson Spit). Cadmium concentrations were higher in the current study then values measured at the comparative sites in 1994 (Table 34; USFWS).

Cadmium concentrations measured in eelgrass can reach levels related to concentrations found in the environment where the plants grow. Cadmium is readily absorbed by eelgrass through the leaves and rhizome-roots and over certain exposure times and concentrations, cadmium is translocated from the aboveground biomass to the belowground biomass (Faraday and Churchill 1979). After 72 hours of exposure in a controlled experiment, cadmium concentrations in eelgrass reached a peak of 48 µg gdw^{-1} in rhizomes-roots and 94 µg gdw^{-1} in leaves (Faraday and Churchill 1979). In Puget Sound, the concentrations of cadmium in eelgrass were higher compared to concentrations found in the seagrass literature (Brix et al. 1983, Govers et al. 2014, Lewis and Devereux 2009). The average cadmium concentration in aboveground eelgrass in Puget Sound was 1.9 – 8.1 µg gdw^{-1}, while in Denmark the concentrations in eelgrass ranged from 0.1 – 2.9 µg gdw^{-1} (Brix et al. 1983). Other studies found cadmium aboveground concentrations between 0.5 – 5 µg gdw^{-1} (Govers et al. 2014). In the belowground eelgrass biomass, average cadmium concentrations in Puget Sound were 0.1 – 2.6 µg gdw^{-1} and in Denmark the cadmium concentrations in belowground biomass were 0.1 – 0.9 µg gdw^{-1} (Brix et al. 1983).

Whole eelgrass shoot concentrations (combined above- and belowground concentrations) measured in Puget Sound ranged between 2.6 to 10.6 µg gdw^{-1}. Althought the lowest cadmium concentrations measured in a whole shoot overlapped the range of cadmium found in mussels (1.6 to 4.0 µg gdw^{-1}), the highest whole shoot concentrations were 2.5 times greater than measured in mussels (Lanksbury et al. 2014).

Chromium (Cr)

Chromium concentrations were low compared to the range of values observed in other studies (Appendix A, Table A-3). In Puget Sound, the chromium concentration in the aboveground biomass ranged between 0.04 – 0.36 µg gdw^{-1}, while a review of literature found values ranging between 0.5 – 25 µg gdw^{-1} (Govers et al. 2014). Three sites, Big Gulch, Cypress Island, and Holly, had statistically higher chromium concentrations than other sites with no evident causal factor. Furthermore, the upland use is quite different between the three sites with the highest residential footprint adjacent to Big Gulch and the lowest on Cypress Island and Holly. The chromium concentrations previously measured in 1994 at March Point and Padilla Bay were approximately 120 to 170 times higher than values measured in this study (Table 34; USFWS 1994). It is not clear exactly why the large difference in values except that the 1994 study focused samples near creosote pilings which may have elevated levels in seagrass biomass. It is also possible that the 1994 samples were contaminated.

Copper (Cu)

Copper concentrations measured in the aboveground biomass in Puget Sound were the highest among all other studies that measured copper concentrations in eelgrass (Govers et al. 2014, Lewis and Devereux 2009); even studies that focused sampling adjacent to contaminated or industrial areas (Brix et al. 1983, Stenner and Nickless 1975). The highest copper concentration in Puget Sound was at Four-Mile Rock, a site down drift from the Seattle waterfront; the most urbanized and populated area of Puget Sound. Copper is found in roofing materials, brake pads, residential and industrial herbicides and fertilizers, and marine anti-fouling paints (Ecology and King County 2011). The primary pathway that copper reaches Puget Sound is through stormwater runoff (Ecology and King County 2011). Most of the water in Elliot Bay that orginated from upland sources (e.g., stormwater and the Duwamish River) along with tidally exchanged water, circulates counter-clockwise due to tidal patterns and predominant winds (Dexter et al. 1981, Hinchey et al. 1980). These waters pass the shipping terminal in the southern portion of Elliot Bay and transport pollutants northward past the Seattle waterfront towards Smith Cove, another industrial area, and exit Elliot Bay just beyond the Four-Mile Rock site. It is possible that the high copper concentrations measured in eelgrass at Four-Mile Rock are released from a wide range of residential and commercial sources within the Seattle metropolitan area. Copper concentrations in eelgrass were four times higher in this study at March Point, Padilla Bay and on average at all the sites between the current study and the 1994 USFWS assessment (Table 34).

The copper concentrations measured in eelgrass (aboveground biomass was 5.1 – 85.9 µg gdw^{-1} and belowground biomass was 0.3 – 17.8 µg gdw^{-1}) exceeded concentrations found in mussels (4.0 – 10.5 µg gdw^{-1}; Lanksbury et al. 2014). There was a significant relationship between copper concentrations in mussels and urban growth areas and a similar pattern was observed with significantly higher concentrations of copper measured in eelgrass at Four-Mile Rock.

Copper is a trace metal, readily taken up by seagrass and essential for seagrass photosynthetic activity (Prange and Dennison 2000). However, excess quantities of copper in seagrass caused toxicity and affected photosynthetic function in five seagrass species native to subtropical Australia (Prange and Dennison 2000). Although the research has not investigated the effects of copper on photosynthetic activity in eelgrass, concentrations measured in Puget Sound exceeded concentrations measured in Australian seagrasses where effects were observed.

Iron (Fe)

The iron concentration in eelgrass varied widely throughout Puget Sound (whole shoot = 128 – 770 µg gdw^{-1}), with concentrations more similar to the low range measured in eelgrass globally (34 µg gdw^{-1}), and well below the upper end of the range measured in eelgrass (10,300 µg gdw^{-1}; Appendix A, Table A-3). The iron concentrations within this study were 30 to 75 times lower than concentrations measured in the 1994 UWFWS study (Table 34) and it is not clear what caused these differences.

Iron can reach concentrations considered toxic to some seagrass species. The photosynthetic responses of two subtropical seagrass, *Halophila spinulosa* and *H. ovalis*, were noticeably affected with whole shoot concentrations in excess of 2,000 µg gdw^{-1} (Prange and Dennison 2000). The effects of high iron concentrations observed in eelgrass have not been assessed to date.

Lead (Pb)

Lead concentrations measured in the above- and belowground eelgrass biomass in Puget Sound were low relative to other studies but within the range observed globally. Lead concentrations measured in Puget Sound were up to 50 times lower than those measured adjacent to contaminated areas in New Hampshire, USA (Johnston et al. 1994a, 1994b, Short 1994), Turkey (Güven et al. 1993), and Denmark (Brix and Lyngby 1984). The lead concentrations in the aboveground eelgrass biomass measured in this study were 3 to 13 times lower than concentrations measured in Padilla Bay and March Point in 1994 (Table 34; USFWS).

The combined lead concentration in the above- and belowground eelgrass biomass (0.04 – 1.4 µg gdw^{-1}) was similar to the concentrations observed in mussels (0.1 – 1.4 µg gdw^{-1}, Lanksbury et al. 2014). However, high concentrations of lead were observed in mussels from Edmonds, Bremerton and Quartermaster Harbor and the highest levels in eelgrass were observed at Post Point and Ruston Way.

Nickel (Ni)

Nickel concentrations measured in this study were not nearly as high as values measured in eelgrass in Oregon (Kaldy 2006), Turkey (Güven et al. 1993), and in Padilla Bay (USFWS 1994). The relatively high nickel concentration values measured at the Cypress Island site in this study could be a result of a serpentine rich sediment source on the southern end of Cypress Island. Serpentine rich sediments are derived from mafic and ultramafic rock that typically produce high concentrations of nickel when eroded (DNR 2007, Cornwall 1967). There was also a large range of differences (6 to 17 times) between the nickel concentrations in eelgrass measured in the 1994 USFWS study and in this study (Table 34).

Vanadium (V)

The current study is the only research to date to document vanadium concentrations in eelgrass. Although there were no differences in vanadium concentrations in the aboveground eelgrass biomass at the 15 sample sites, the concentrations of vanadium in the belowground eelgrass biomass at Thompson Spit were clearly much higher than measured at any other site. It turns out that vanadium is the second most abundant transition metal in oceans and a primary enzyme in nitrogen fixation (Winter and Moore 2009). Vanadium is an important component of marine phytoplankton, algae and other organisms (as cited in Wang and Wilhelmy 2009) and a major element of heavy fuels (Mamane and Pirrone 1998) and marine distillates (Nigam et al. 2007). Vanadium particles are released into the atmosphere and marine environment during the combustion of heavy fuels and diesel fuels (Mamane and Pirrone 1998, Nigam et al. 2007).

It is not entirely clear why there was a spike in vanadium concentrations in the belowground eelgrass at Thompson Spit relative to the other sample sites. It is possible there was some previous fuel oil contamination or pooling of marine phytoplankton that caused an increase in the substrate where the eelgrass was sampled.

Zinc (Zn)

Zinc concentrations measured in Puget Sound eelgrass were within the range measured in other studies, but not as high as concentrations measured from Poland (300-820 µg gdw^{-1}, Bojanowski 1973) or documented in the literature (Govers et al.

2014, Lewis and Devereux 2009). Zinc concentrations measured in eelgrass were slightly more than twice as high in this study compared to the 1994 USFWS study (Table 34).

Similar to copper concentrations, the highest zinc concentrations in Puget Sound were observed at Four-Mile Rock; a site down current from the Seattle metropolitan area (Hinchey et al. 1980, Dexter et al. 1981). Also like copper, one of the major sources of zinc within the Puget Sound watershed is roofing materials and its primarily pathway to Puget Sound is via stormwater runoff (Ecology and King County 2011). Zinc is also used on marine vessels as a sacrificial anode to minimize electrochemical corrosion to other critical metals (e.g., propeller, drive shaft, electronics sensors). Water enters Elliot Bay by means of stormwater discharge pipes, the Duwamish River, precipitation and tidal exchange and then circulates in a counter-clockwise pattern past the marine shipping terminal, the Seattle waterfront, Smith Cove and the Elliot Bay Marina before passing Four-Mile Rock site. Therefore, the eelgrass at Four-Mile Rock are likely exposed to waters with potentially higher concentrations of zinc.

The combined zinc concentration in the above- and belowground eelgrass biomass (81 - 205 µg gdw^{-1}) was greater than the concentrations observed in mussels (68 – 137 µg gdw^{-1}, Lanksbury et al. 2014). The relationship between zinc accumulation in mussels relative to urban growth areas coincides with the significantly higher zinc concentrations observed in eelgrass at Four-Mile Rock relative to sites with lower urban growth footprints on the adjacent uplands (e.g., Holly and Sandy Bay).

Table 34. A comparison of metal concentrations (µg gdw^{-1} dry weight) measured in eelgrass at Padilla Bay (PB) and March Point (MP) from the 1994 USFWS study and this study. Sites with 1994 suffix are from the 1994 USFWS study. Sites with the 2013 suffix are from this study.

SITE	COMPART - MENT	Hg	As	Cd	Cr	Cu	Fe	Ni	Pb	Zn
		µg gdw^{-1}	µg gdw^{-1}	µg gdw^{-1}	µg gdw^{-1}	µg gdw^{-1}	µg gdw^{-1}	µg gdw^{-1}	µg gdw^{-1}	µg gdw^{-1}
		mean (±SE)	mean (±SE)	mean (±SE)	mean (±SE)	mean (±SE)	mean (±SE)	mean (±SE)	mean (±SE)	mean (±SE)
PB2-1994	Aboveground	0.01	7.3	1.9	16.0	7.2	7620	9.1	1.0	34.0
PB-2013	Aboveground	0.01 (0.0)	2.0 (0.2)	4.1 (0.5)	0.1 (0.0)	30.9 (8.3)	249.0 (47.5)	1.0 (0.2)	0.3 (0.2)	70.0 (2.1)
PB-2013	Belowground	0.0 (0.0)	0.4 (0.1)	1.1 (0.1)	0.2 (0.1)	3.6 (0.4)	264.0 (83.8)	0.3 (0.1)	0.1 (0.0)	45.8 (6.7)
MP3-1994	Aboveground	0.022	3.4	2.0	17.0	9.4	8420	12.0	2.7	29.9
MP-2013	Aboveground	0.01 (0.0)	1.0 (0.3)	2.7 (0.7)	0.1 (0.0)	37.7 (24.1)	177.1 (73.2)	0.7 (0.20	0.2 (0.1)	70.8 (13.1)
MP-2013	Belowground	0.0 (0.0)	0.3 (0.0)	0.8 (0.0)	0.1 (0.0)	4.0 (0.3)	111.9 (9.4)	0.2 (0.0)	0.1 (0.0)	36.6 (1.5)
USFWS-1994	Aboveground	0.15 (0.002)	4.1 (0.9)	2.1 (0.3)	14.5 (3.4)	8.0 (0.9)	6157 (965)	9.8 (1.8)	1.4 (0.2)	30.6 (1.6)
PS-2013	Aboveground	0.01 (0.0)	0.89 (0.06)	4.43 (0.19)	0.12 (0.01)	29.82 (2.95)	183.84 (12.89)	1.52 (0.11)	0.23 (0.03)	79.58 (2.76)
PS-2013	Belowground	0.00 (0.0)	0.49 (0.03)	1.45 (0.08)	0.14 (0.01)	5.27 (0.48)	162.94 (16.48)	0.40 (0.05)	0.09 (0.02)	53.54 (2.18)

4.3 Organics

Polycyclic aromatic hydrocarbons (PAHs), polychlorinated biphenyls (PCBs) and persistent organic pollutants (POPs) enter the nearshore system by means of stormwater runoff and, in some cases, nonpoint sources such as atmospheric deposition. After entering Puget Sound, these organic pollutants accumulate in sediments and biota or cycle through the food web. The presence of most contaminants depends on land use patterns and point source pollution resulting in an abundance of contaminants that originate from petroleum based activities, industrial sources, and agriculture. A recent assessment of toxic loading to Puget Sound found the highest loading rates of PAHs from residential sources such as surface and stormwater runoff (EnvironVision et al. 2008, Hart Crowser et al. 2007). As for PCBs, the greatest loading is from residential and industrial/commercial sources (Pelletier and Mohamedali 2009). Although seagrass is shielded from certain contaminants because of the structural integrity of the plant and antibacterial biofilm on the leaves (Gunnarsson et al. 1999), impacts from contaminants can range from smothering by oils spills to various physiological implications caused by herbicides. Although some of these contaminants (e.g., oil) typically enter the marine environment through sources other than outfalls (a focus of this study), it is important to consider their effects on eelgrass.

A discussion of the concentration of organic contaminants follows with comparison to concentrations measured in eelgrass previously in Puget Sound (USFWS 1994) and relative to values in the literature (Govers et al. 2014, Lewis and Devereux 2009). In addition, the discussion will provide a brief comparison of the organic contaminant concentrations measured in eelgrass compared to the concentration measured in mussels from the Mussel Watch Pilot Expansion Project (Lanksbury et al. 2014).

Low and High molecular weight PAHs

The only other study that measured PAHs in eelgrass in Puget Sound was conducted by the USFWS in Padilla and Fidalgo Bays (1994). The 1994 study reported results as a combination of low and high molecular weight PAHs. Levels of PAHs measured from the 1994 study were low (~ 0.05 – 0.18 µg gww^{-1}), however, the study found evidence of elevated PAHs at the two sites near March Point relative to sites in Padilla and Samish Bays (USFWS 1994). The 1994 study concluded that the elevated PAHs were consistent with oil industry activites and associated infrastructure (e.g., piers) within the area. The concentrations of PAHs measured in eelgrass at the Padilla Bay and March Point sites in 2013 were low compared to the concentrations measured in the 1994 study. The only concentration of PAHs measured in eelgrass from Puget

Sound that were similar to the levels measured in the 1994 study were observed from the eelgrass collected at Four-Mile Rock (Figures 18 & 19). The PAH concentrations measured in eelgrass at Four-Mile Rock were statistically higher relative to most other sites (Tables 22 & 24). High concentrations of PAHs were also observed in mussel at five sites along the Elliot Bay shoreline (Lanksbury et al. 2014). At Four-Mile Rock, the concentration of both high and low molecular weight PAHs (Σ_{42}PAHs) in mussels was 4,526 ng gdw^{-1} (Lanksbury et al. 2014), compared to 70 ng gdw^{-1} in eelgrass (combined values from Figures 18 and 19). Similar to the elevated concentrations of copper and zinc measured in eelgrass at the Four-Mile Rock, high PAHs concentrations in eelgrass may be that the site is located down current from the largest metropolitan area in Puget Sound. PAHs enter Elliot Bay through the Duwamish River, marine industry, shipping traffic and stormwater from the surround residential and commercial development and are circulated past Four-Mile Rock.

In Florida, research has shown that PAHs are often below the detectable limit in most seagrasses (Lewis et al. 2007), while others have observed higher values of PAHs in seagrass closer to anthropogenic sources (e.g., harbor, city; Pergent et al. 2011). One mesocosm study with eelgrass found the uptake of PAHs in the above- and belowground biomass matched sediment PAH concentrations within a 60 day period (Huesemann et al. 2009).

It is unknown whether the concentrations of PAHs measured in eelgrass in Puget Sound pose a toxic threat to plant physiology and health. There has been some research on the physiological effects PAHs have on seagrasses (Cambridge et al. 1986, Hatcher and Larkum 1982, Thorhaug et al. 1986, Thorhaug and Marcus 1987, Ralph and Burchett 1998b), but little has been conducted specific to eelgrass. Oil spills in the Persian Gulf show no effect on seagrass plant photosynthesis (Durako et al. 1993), species composition, abundance or distribution (Kenworthy et al. 1993), while other studies found seagrass exposed to minute concentrations of oil residues experienced significantly reduced leaf growth rates (Cambridge et al. 1986). In the case of the *Exxon Valdez* oil spill, there were a range of impacts from a reduction in eelgrass biomass and density of reproductive shoots (Dean et al. 1998) to a total loss of eelgrass in some areas (Juday and Foster 1990). The resilience of seagrass in the wake of an oil spill is probably dependent on factors that differ relative to environmental and climatic conditions.

Persistent Organic Pollutants

Persistent Organic Pollutants (POPs) are lipophilic compounds that accumulate in fatty tissue of living organisms. Low concentrations of measured POPs in eelgrass were anticipated as eelgrass naturally has very low fat content (Section 3.3.1, Figure

17; ~1% described in Felger and Moser 1973). Therefore, sites with high levels of POPs concentration in eelgrass biomass might be an indicator of disproportionately high contamination.

Higher levels of PCBs were measured in the above- and belowground eelgrass biomass at five sites within Puget Sound (Figure 21), however the connection to a source of contamination was not entirely clear. Sources of PCBs from industrial applications are likely greater near the Ruston Way, Big Gulch and Birch Bay sites, but it is not clear why PCB concentrations in eelgrass were high at Holly and Thompson Spit. The concentration of PCBs in mussels at Four-Mile Rock (45 ng gdw^{-1}) were low relative to mussels sampled from Elliot Bay (59-74 ng gdw^{-1}), Sinclair Inlet (113 ng gdw^{-1}), and Hylobos Waterway (178-533 ng gdw^{-1}) (Lanksbury et al. 2014).

A possible connection to a source was observed in the concentration of PBDEs (flame retardants) in eelgrass. Although the concentrations of PBDEs in the above- and belowground eelgrass biomass at Four-Mile Rock was only significantly higher than two sites (Figure 22, Table 30), the high values measured at this site could be a result of many residential and commercial sources of PBDEs discharged into Elliot Bay. Again, the concentration of PBDEs in mussels (1.7 – 3.5 ng gdw^{-1}) were considerably higher than concentrations measured in eelgrass (0.1 – 0.8 ng gdw^{-1}). There was also a significant relationship between the concentration of PBDEs in mussels and urban growth areas (Lanksbury et al. 2014).

The DDT concentrations measured in eelgrass produced limited data for analyses (Figure 23). The concentration of DDTs in mussels (1 – 46 ng gdw^{-1}) was considerably higher than concentrations measured in eelgrass (0.1 – 0.2 ng gdw^{-1}). There was also a significant relationship between DDT concentrations in mussels and urban growth areas versus less developed sites (Lanksbury et al. 2014).

Seagrasses will absorb and accumulate some fraction of organic compounds in the system but the ability to incorporate these compounds is severly limited because of the low percent of lipids within the above- and belowground plant compartments. Low levels of organic compounds are not limited to eelgrass, but indicative of seagrasses on a whole. A study in Australia found concentrations of PCBs below detectable levels in three different seagrasses; *Cymodocea serrulata*, *Halodule uninervis*, and *Zostera muelleri* (Haynes et al. 2000). Similarly, undetectable results for PCBs were found in a study that assessed PCBs in *Thalassia testudinum* and *Halodule wrightii* at 13 sites along the Florida pan handle (Lewis et al. 2007) and PCBs in eelgrass at the mouth of the Piscataqua River, Maine (Johnston et al. 1994).

5 CONCLUSION

Eelgrass is an important habitat in Puget Sound and supports numerous ecosystem functions. It is considered a significant indicator of ecosystem health (Dennison et al. 1993, Krause-Jensen et al. 2005, Orth et al. 2006), and has been identified as an indicator to track the recovery of Puget Sound. To further assess stressors that cause eelgrass decline in the Sound (Thom et al. 2011), it is critical to understand the effects of outfall construction and effluent on eelgrass. Outfalls that discharge residential, commercial, and industrial wastewater along with upland stormwater are abundant throughout developed coastal areas, particularly in Puget Sound (Gaeckle et al. 2015). Furthermore, outfall construction and discharge can affect marine organisms and processes, specifically eelgrass (Gaeckle 2012). The impacts outfalls have to eelgrass range from physical effects on the environment where it grows to effects on the plants from increased turbidity, changes in flow patterns and an excess of chemicals that could affect seagrass physiology.

The current study assessed the concentration of carbon, nitrogen, 10 metals, and a suite of organic contaminants in the above- and belowground compartments of eelgrass at 15 sites throughout Puget Sound. In general, the concentrations of carbon, nitrogen and metal in eelgrass were within the range observed in other seagrass, and eelgrass specific, studies worldwide. The concentration of organic contaminants were low, but the low levels observed may be due to limited uptake and accumulation potential by seagrass because of generally low lipids in seagrass biomass. With the exception of a very low C:N ratio that suggests a naturally high nutrient environment, it is not entirely clear how the measured concentrations of metals and organic contaminants in eelgrass will affect populations throughout Puget Sound. There is a paucity of global studies that show how specific concentrations of metals and organic contaminants in eelgrass biomass affect plant physiology. In Puget Sound, there have been no studies that demonstrate the effect of these substances on eelgrass, yet the Sound continues to receive inputs loaded with metals and organic contaminants (Ecology and King County 2011).

In a few cases, results from the study suggested a correlation between high concentration of metals and organics measured in eelgrass biomass relative to site location. Relatively high concentrations of copper, zinc, and PAHs at Four-Mile Rock suggest a correlation between contamination source and higher concentrations measured in eelgrass as observed in other studies that assessed metals and contaminants in eelgrass (Govers et al. 2014, Lewis and Devereux 2009). In contrast, it is not entirely clear what caused high vanadium concentrations in eelgrass at Thompson Spit or POP concentrations measured at Holly. A study with higher replication stratified over an assumed pollution gradient at these sites may identify sources of contamination. In addition, more stable isotope work may aid in identifying sources of pollution. Overall, the data collected provide a baseline for future assessments that could track changes in concentrations and sources of pollution over different spatial and temporal gradients. The study also establishes the need to understand what concentration of metals and organic contaminants impede seagrass physiology and resilience in a changing Puget Sound.

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7 APPENDIX

Table A-1. List of fifteen (15) sample sites, their locations, funding source and weather a mussel cage as part of WDFW Mussel Watch Pilot Expansion Project was deployed at the site.

SITE	LOCATION	FUNDING SOURCE	MUSSEL CAGE
Birch Bay	Ferndale	DNR Aquatic Reserves Program	yes
Post Point	Fairhaven-Bellingham	US Environmental Protection Agency	yes
Cypress Island	Strawberry Bay	DNR Aquatic Reserves Program	yes
March Point	Anacortes	US Environmental Protection Agency	yes
Padilla Bay	Mt. Vernon	US Environmental Protection Agency	yes
Penn Cove	Coupeville, Whidbey Island	US Environmental Protection Agency	yes
Thompson Spit	Miller Peninsula, Gardiner	DNR Aquatic Reserves Program	yes
Big Gulch	Mukilteo	US Environmental Protection Agency	yes
Four-Mile Rock	Magnolia, Seattle	US Environmental Protection Agency	yes
Duwamish Head	Alki, West Seattle	US Environmental Protection Agency	yes
Holly	Hood Canal	US Environmental Protection Agency	yes
Burley Spit	Purdy	DNR Eelgrass Stressor Response Program	no
Dumas Bay	Federal Way	US Environmental Protection Agency	yes
Ruston Way	Puget Creek, Tacoma	US Environmental Protection Agency	yes
Sandy Bay	Anderson Island	DNR Aquatic Reserves Program	yes

Table A-2. Name and abbreviation of low (LMW) and high molecular weight (HMW) polycyclic aromatic hydrocarbon (PAH) analytes measured in eelgrass (*Z. marina*) compartments (leaves, rhizomes, roots) at 15 sites throughout Puget Sound, WA, USA.

LMW PAH NAME	ABBREVIATION	HMW PAH NAME	ABBREVIATION
naphthalene	NPH	fluoranthene	FLA
C1-naphthalenes	C1NPH	pyrene	PYR
C2-naphthalenes	C2NPH	C1-fluoranthenes/pyrenes	C1FLA
C3-naphthalenes	C3NPH	C2-fluoranthenes/pyrenes	C2FLA
C4-naphthalenes	C4NPH	C3-fluoranthenes/pyrenes	C3FLA
acenaphthylene	ACY	C4-fluoranthenes/pyrenes	C4FLA
acenaphthene	ACE	benz[a]anthracene	BAA
fluorene	FLU	chrysene	CHR
C1-fluorenes	C1FLU	C1-benzanthracenes/chrysenes	C1CHR
C2-fluorenes	C2FLU	C2-benzanthracenes/chrysenes	C2CHR
C3-fluorenes	C3FLU	C3-benzanthracenes/chrysenes	C3CHR
dibenzothiophene	DBT	C4-benzanthracenes/chrysenes	C4CHR
C1-dibenzothiophenes	C1DBT	benzo[b]fluoranthene	BFF
C2-dibenzothiophenes	C2DBT	benzo[k]fluoranthene	BKF
C3-dibenzothiophenes	C3DBT	benzo[e]pyrene	BEP
C4-dibenzothiophenes	C4DBT	benzo[a]pyrene	BAP
phenanthrene	PHN	perylene	PER
anthracene	ANT	indeno[1,2,3-cd]pyrene	IDP
C1-phenanthrenes/anthracenes	C1PHN	dibenz[a,h]anthracene	DBA
C2-phenanthrenes/anthracenes	C2PHN	benzo[ghi]perylene	BZP
C3-phenanthrenes/anthracenes	C3PHN		
C4-phenanthrenes/anthracenes	C4PHN		

Table A-3. Metallic composition (ppm dry weight, µg gdw^{-1}) of eelgrass (*Z. marina*) compartments (leaves, rhizomes, roots) from the literature and the current study. Table modified from Lewis and Devereux (2009) and Brix and Lyngby (1984) with the addition of other data as listed.

REFERENCE	LOCATION	COMPARTMENT	VALUE	Al	As	Au	Ca	Cd	Ce	Co	Cr
		(leaves, rhizome/roots)	(single, mean, range)	Aluminum	Arsenic	Gold	Calcium	Cadmium	Cesium	Cobalt	Chromium
Augier et al. (1983)	Mediterranean Sea	leaves									
Bellester et al. (1980)[††]	Catalonia Coast (Spain)	whole plant	single					0.26			
Bojanowski (1973)	Baltic (Poland)	leaves	mean				11,800			1.91	
			range				9,300-21,800			0.27-6.80	
Brix et al. (1983)	Bay of Aarhus (Denmark)*	leaves	mean					1.03			1.4
		rhizome/roots	range					0.1-0.9			
Brix and Lingby (1984)	Limfjord (Denmark)	leaves	mean				13,000	0.62			2.2
			range				4,600-36,300	0.09-2.92		2.5-15.7	0.07-9.8
Damyanova et al. (1981)	Black Sea (Bulgary)	leaves	mean		0.21	0.0044		7.15	0.72	6.34	1.53
			range		0.07-0.43	0.0024-0.0072		6.00-9.20	0.53-0.90	5.66-7.10	1.25-1.75
DeCasabianca et al. (2004)	Thau Lagoon (France)	leaves	mean								0.3
DeCasabianca et al. (2004)	Thau Lagoon (France)	rhizome/roots	mean								2
Dieckmann (1982	Kiel Fjord (Germany)	leaves	mean					0.49-2.22			
Drifmeyer et al. (1980)	Beaufort (North Carolina)	leaves	mean								
Gorham et al. (1980)	English Channel	leaves	mean				5130				
Güven et al. (1993)	Bosphorus Strait (Turkey)*	leaves						2.3			13.6
Güven et al. (1993)	Bosphorus Strait (Turkey)*	whole plant	mean					1.9-2.3			8.3-13.6
Johnston et al. 1994a, 1994b, and Short 1994	Piscataqua River and Great Bay Estuary (Maine/New Hampshire)*	leaves	mean	51.3	0.9			0.9			0.6
		leaves	range	9.0-120.0	0.6-1.4			0.3-1.9			0.3-0.9
		rhizome/roots	mean	577.7	4.1			0.5			4.5
		rhizome/roots	range	203.0-938.0	1.5-10.9			0.3-0.8			1.7-9.7
Kaldy (2006)	Yaquina Bay (Oregon)	leaves	range								3-15
McRoy (1966)[†]								0.23		0.03	
Stenner & Nickless 1975	Spain							2.0			
Stenner & Nickless 1975	Spain*							5.3			
US FWS (1994)	Padilla and Fidalgo Bays		Range	1,905-7,320	1.6-7.8			1.1-3.6			5.5-35.2

Water Quality Institute (1978)	Bay of Køge (Denmark)	leaves	mean					0.5			0.8
			range					0.1-1.7			<0.4-1.8
Wolfe et al. (1975)[††]	Newport River estuary (Oregon)	whole plant	mean								
Wolfe et al. (1976)	Beaufort (North Carolina)	leaves	mean								
This Study (2013)	Puget Sound (Washington)	leaves	range		0.44-2.29			1.91-8.10			0.04-0.36
This Study (2013)	Puget Sound (Washington)	leaves	mean		0.89			4.43			0.12
This Study (2013)	Puget Sound (Washington)	rhizome / roots	range		0.04-1.06			0.14-2.59			0.00-0.37
This Study (2013)	Puget Sound (Washington)	rhizome / roots	mean		0.49			1.45			0.14

* = samples taken from or near contaminated sites, [†] = cited in Burrell and Schubel (1977), [††] = cited in Lewis and Devereux (2009)

REFERENCE	LOCATION	COMPARTMENT	VALUE	Cu	Fe	Hg	K	Mg	Mn	Na	Ni
		(leaves, rhizome/roots)	(single, mean, range)	Copper	Iron	Mercury	Potassium	magnesium	manganese	sodium	nickel
Augier et al. (1983)	Mediterranean Sea	leaves									
Bellester et al. (1980)[††]	Catalonia Coast (Spain)	whole plant	single	5.6							2.1
Bojanowski (1973)	Baltic (Poland)	leaves	mean	15.2	480		34,700	9,900	990	24,300	4.6
			range	8.0-33.5	120-1,540		12,800-53,500	8,200-11,200	130-2,270	10,000-34,800	1.3-11.8
Brix et al. (1983)	Bay of Aarhus (Denmark)*	leaves		5.86	296						
		rhizome/roots	range	1.8-19.3							
Brix and Lingby (1984)	Limfjord (Denmark)	leaves	mean	4.91	390	0.012	35,000	7,960	1,820	33,300	
			range	1.86-16.6	80-2,990	0.005-1.14	13,600-49,800	6,900-11,000	480-5,770	24,400-49,500	
Damyanova et al. 1981	Black Sea (Bulgary)	leaves	mean	9.89	670	0.48			50		
			range	7.76-11.65	559-789	0.38-0.59			41-60		
DeCasabianca et al. (2004)	Thau Lagoon (France)	leaves	mean	10	186						0.6
DeCasabianca et al. (2004)	Thau Lagoon (France)	rhizome/roots	mean	9	921						1
Dieckmann (1982	Kiel Fjord (Germany)	leaves	mean	7.9	1,240				154		
Drifmeyer et al. (1980)	Beaufort (North Carolina)	leaves	mean	6.4	810						
Gorham et al. (1980)	English Channel		mean				29,100	12,000		55,900	
Güven et al. (1993)	Bosphorus Strait (Turkey)*	leaves		39.8							17.5
Güven et al. (1993)	Bosphorus Strait (Turkey)*	whole plant	mean	23.4-39.8							12.9-17.5
Johnston et al. 1994a, 1994b, and Short 1994	Piscataqua River and Great Bay Estuary (Maine/New Hampshire)*	leaves	mean	20.0	294.3	0.01			96.2		1.4
		leaves	range	8.8-62.6	58.0-590.0	0.01-0.02			14.0-265.0		0.4-2.3
		rhizome/roots	mean	17.6	3,624.4	0.03			57.2		2.1
		rhizome/roots	range	8.3-36.7	1,280.0-6,200.0	0.01-0.05			15.0-240.0		1.1-3.0
Kaldy (2006)	Yaquina Bay (Oregon)	leaves	range	10-20							2-120

McRoy (1966)†				7.50	34-345	1.33			34-1,845		0.4
Stenner & Nickless (1975)	Spain			9-36							
Stenner & Nickless (1975)	Spain*			1350							
US FWS (1994)	Padilla and Fidalgo Bays		range	5.05-11.8	2,835-10,300	0.009-0.022			118-324		4.6-21.0
Water Quality Institute (1978)	Bay of Køge (Denmark)	leaves	mean	4.5		0.19					
			range	2.0-9.3		0.07-0.50					
Wolfe et al. (1975)††	Newport River estuary (Oregon)	whole plant	mean	7.9							
Wolfe et al. (1976)	Beaufort (North Carolina)	leaves	mean								
This Study (2013)	Puget Sound (Washington)	leaves	range	5.1-85.9	62.9-411.0	0.004-0.017					0.51-4.08
This Study (2013)	Puget Sound (Washington)	leaves	mean	29.82	183.84	0.01					1.52
This Study (2013)	Puget Sound (Washington)	rhizome / roots	range	0.34-17.8	7.92-540.0	0.002-0.006					0.01-1.35
This Study (2013)	Puget Sound (Washington)	rhizome / roots	mean	5.27	162.94	0.00					0.40

* = samples taken from or near contaminated sites, † = cited in Burrell and Schubel (1977), †† = cited in Lewis and Devereux (2009)

REFERENCE	LOCATION	COMPARTMENT	VALUE	Pb	Sb	Sc	Se	Sm	Sr	V	Zn
		(leaves, rhizome/roots)	(single, mean, range)	lead	antimony	scandium	selenium	samarium	strontium	vanadium	zinc
Augier et al. (1983)	Mediterranean Sea	leaves									
Bellester et al. (1980)††	Catalonia Coast (Spain)	whole plant	single	0.79							
Bojanowski (1973)	Baltic (Poland)	Leaves	mean						240		300
			range						155-480		80-820
Brix et al. (1983)	Bay of Aarhus (Denmark)*	leaves	mean								
		rhizome/roots	range	0.4-30							25-125
Brix and Lingby (1984)	Limfjord (Denmark)	leaves	mean	1.07							78
			range	0.47-37.5							41-175
Damyanova et al. (1981)	Black Sea (Bulgary)	leaves	mean		0.91	0.018	0.22	0.38			37
			range		0.75-1.01	0.014-0.021	0.10-0.33	0.27-0.53			31-45
DeCasabianca et al. (2004)	Thau Lagoon (France)	leaves	mean	1							83
DeCasabianca et al. (2004)	Thau Lagoon (France)	rhizome/roots	mean	2							44
Dieckmann (1982	Kiel Fjord (Germany)	leaves	mean								70
Drifmeyer et al. (1980)	Beaufort (North Carolina)	leaves	mean								
Gorham et al. (1980)	English Channel		mean								
Güven et al. (1993)	Bosphorus Strait (Turkey)*	leaves		32							91
Güven et al. (1993)	Bosphorus Strait (Turkey)*	whole plant	mean	26.1-32.1							48.7-91.3
Johnston et al. 1994a, 1994b, and Short 1994	Piscataqua River and Great Bay Estuary (Maine/New Hampshire)*	leaves	mean	1.3							63.7
		leaves	range	0.8-2.1							51.4-79.2
		rhizome/roots	mean	7.4							48.4
		rhizome/roots	range	1.7-14.0							24.2-75.9
Kaldy (2006)	Yaquina Bay (Oregon)	leaves	range								20-40
McRoy (1966)†											27-56
Stenner & Nickless (1975)	Spain			6-16							100-215
Stenner & Nickless (1975)	Spain*			1,800							1,480

US FWS (1994)	Padilla and Fidalgo Bays		range	0.9-2.7			<0.2-0.4				21.5-34.8
Water Quality Institute (1978)	Bay of Køge (Denmark)	leaves	mean	3.4							66
			range	0.4-13.0							33-120
Wolfe et al. (1975)[††]	Newport River estuary (Oregon)	whole plant	mean								70
Wolfe et al. (1976)	Beaufort (North Carolina)	leaves	mean								
This Study (2013)	Puget Sound (Washington)	leaves	range	0.05-1.0						0.4-3.4	51.1-128
This Study (2013)	Puget Sound (Washington)	leaves	mean	0.2						1.3	79.6
This Study (2013)	Puget Sound (Washington)	rhizome / roots	range	0.02-0.7						0.02-2.2	21.2-77.0
This Study (2013)	Puget Sound (Washington)	rhizome / roots	mean	0.1						0.5	53.5

* = samples taken from or near contaminated sites, [†] = cited in Burrell and Schubel (1977), [††] = cited in Lewis and Devereux (2009)