Tacoma-Pierce County Health Department

Final Report for the Mussel Watch Gradient Project
Ecology Grant Number G1200564

A sub-project of the Mussel Watch Pilot Expansion Study 2012-2013
Hylebos Waterway and Ruston Way
Tacoma, WA

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Executive Summary

The Tacoma-Pierce County Health Department (Health Department) participated in a complementary study to the **NOAA Mussel Watch Pilot Expansion Study 2012-2013** called the Mussel Watch Gradient Project. The goal of this project was to make progress toward defining the length of shoreline that represents a "site" for mussel contamination sampling as well as serving as an indicator for measuring impacts to nearshore biota. The study includes the same parameters as the Mussel Watch Pilot Expansion Study. This study was conducted by the Health Department with the assistance of numerous partners, including University of Washington Tacoma, Washington Department of Fish and Wildlife, Port of Tacoma, Pierce County Surface Water Management and the City of Tacoma and it was funded by Washington State Department of Ecology.

This was a spatial gradient study conducted over two sites in Tacoma, Ruston Way and the Hylebos Waterway. Nine mussel cages were placed at each site and one sample of naturally-occurring mussels was collected on the Hylebos Waterway. Composite tissue samples of both caged and native mussels were analyzed for metals and organic parameters.

The results from this study indicated a difference between the Hylebos Waterway site, which is in an industrial area, and the Ruston Way site, which is in a commercial/residential area. Therefore, the land use may be an important influence on contaminant loading to mussels in the intertidal zone. The concentrations of contaminants in the mussel tissue at both sites may be attributed in part to many of the current and historical activities adjacent to the water, and possibly to the discharge of contaminants from upland activities through stormwater outfalls.

The results showed some variability in contaminant concentrations between cages within each site but this variability was more subtle than the variability between the two sites.

In general the contaminant results for the Hylebos native mussel sample was higher than the results for the caged mussels for both the Hylebos Waterway and Ruston Way. Various reasons could account for the relatively high native mussel results and may have important implications if caged mussels are anticipated to be used for the assessment of land use impacts on near shore biota. Although these data are very limited, caged mussels may significantly underestimate contaminant loading to the near shore biota .Further monitoring with replicate cages and native mussels is recommended to better assess both intra-site variability and variability between caged mussels and native mussels.

Introduction

Toxic substances enter Puget Sound from a variety of pathways including: (1) non-point sources such as surface water runoff, groundwater releases, and air deposition; (2) point sources such as discharges from wastewater treatment plants and combined sewer overflows (CSOs); and, (3) meso-point sources such as marinas and ferry terminals. Alternatively, toxic substances may be recirculated via existing legacy pollutants residing in sediments. These toxic substances can cause harm to people, fish, other animals and plants. There is a large body of knowledge about toxic substances in marine deep water sediments and biota; however, the condition of contaminants in nearshore biota has long been recognized as a monitoring gap in Washington State. Understanding how contaminants enter and move through the marine food web (the fate and transport of chemicals), and what damage they cause once they are there, would improve our ability to make cost-effective decisions to mitigate the harm pollution causes Puget Sound's animals and plants. Additionally, understanding the level of pollutants in the nearshore environment would 1) provide a baseline of existing contamination and 2) indicate potential pollutant sources that may need source tracking and correction.

Bivalves are sessile organisms and filter feeders and they are well known bio-indicators of water quality and environmental conditions. Blue mussels (*Mytilus* spp.) and other sessile, filter-feeding bivalves have been used to monitor contaminant conditions in nearshore biota worldwide.

Over the past three years, staff from the Washington Department of Fish and Wildlife's Puget Sound Ecosystem Monitoring Program (WDFW-PSEMP) - Toxics in Fish team have worked with the National Mussel Watch Program to monitor contaminants in Washington State. Although data from the national program have been useful for broadly characterizing contaminant conditions in Washington, the 17 mussel collection sites in Puget Sound (one located in Pierce County) are not sufficient to answer regional questions regarding the locations and level of chemical contaminants in nearshore urbanized waters.

During the winter of 2012-13 the WDFW-PSEMP conducted a one-time pilot expansion of the National Mussel Watch Program model. This Mussel Watch Pilot Expansion Study increased the number of stations monitored from 17 to over 110 sites in Puget Sound.

This increased coverage will allow WDFW-PSEMP to evaluate the geographic extent and magnitude of nearshore contamination on a regional scale, covering a wide range of nearshore land-use conditions including rural, undeveloped, agricultural, urban, and industrial areas. Data from this single synoptic assessment of contaminants on a regional/watershed scale will be of interest to organizations responsible for managing municipal stormwater, industrial stormwater, as well as prioritizing upland source control efforts. (WDFW 2012)

The primary goal of the Pilot Expansion Study was to evaluate the geographic extent and magnitude of chemical contamination in near shore biota using caged Pacific blue mussels (*Mytilus trossulus*) as an indicator organism across a wide range of upland land use types including rural, undeveloped, agricultural, urban and industrial areas. Washington State Department of Fish and Wildlife (WDFW) was the lead for this project and partnered with 38 other groups and many citizen science volunteers.

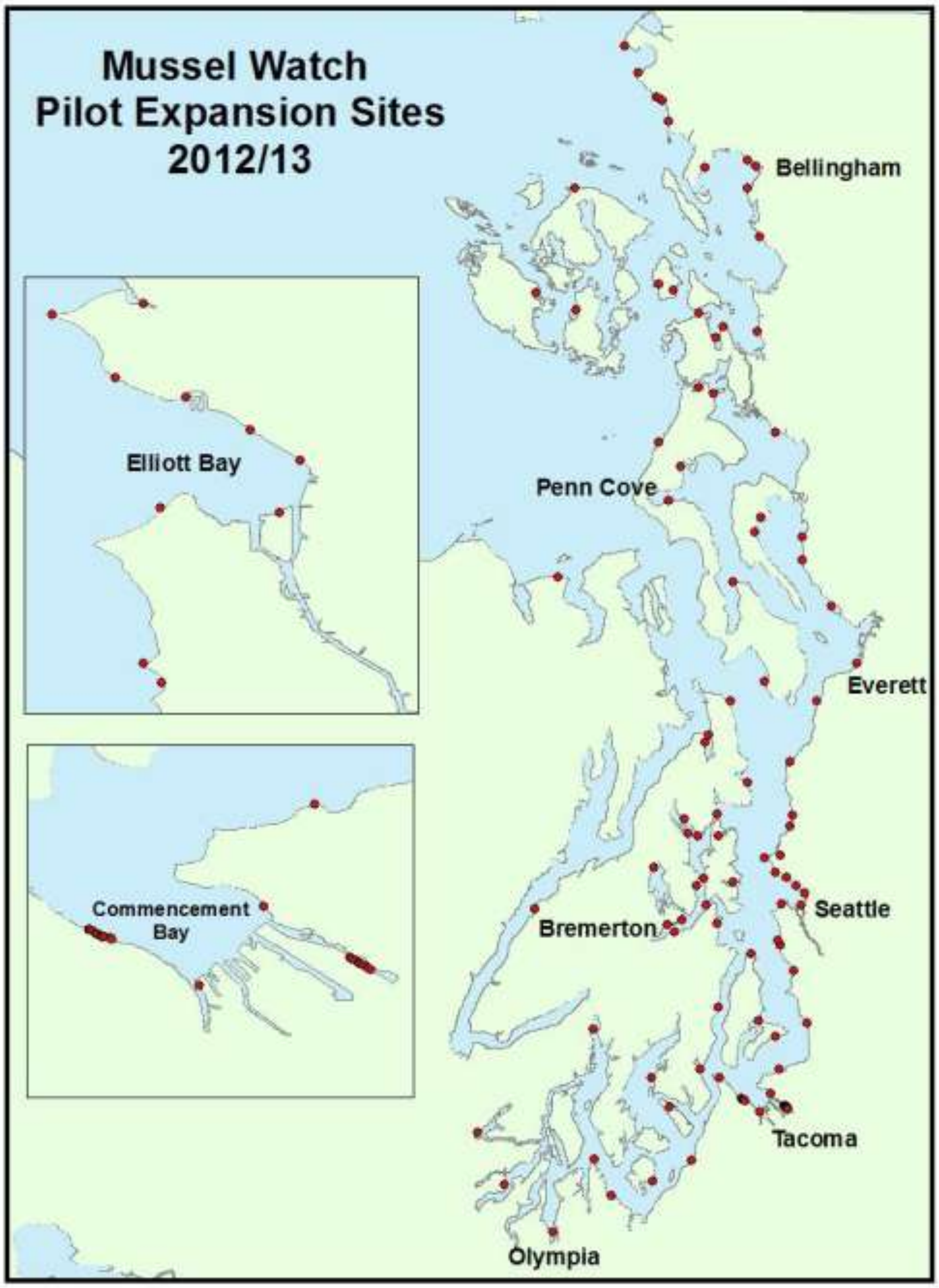
Due to limitations in funding and an interest in building a network of mussel monitoring entities within Washington State, the Mussel Watch Pilot Expansion Study solicited the participation of local agencies and groups willing to sponsor additional sites in their areas of interest.

Mussel Watch Gradient Study

The Tacoma-Pierce County Health Department (Health Department) participated in a complementary study, the Mussel Watch Gradient Project, which was funded by Ecology. The goal of this study was to make progress toward defining the length of shoreline that represents a "site" for mussel contamination sampling. The Mussel Watch Gradient Study was conducted by the Health Department with the assistance of numerous partners, including University of Washington Tacoma, WDFW, Port of Tacoma, Pierce County Surface Water Management, and the City of Tacoma.

The Mussel Watch Gradient Project was a spatial gradient study using caged mussels as an indicator for measuring impacts to nearshore biota in Puget Sound while measuring all the same parameters as the Mussel Watch Pilot Expansion Study. The project monitored two sites, one along Ruston Way and the other in the Hylebos Waterway. Nine mussel cages were placed at each site. In addition, one sample of naturally-occurring mussels was collected near two of the caged mussel locations at the Hylebos Waterway, in close proximity to Cages 1 and 2 (see Figure 3). The project utilized the Quality Assurance Project Plan developed by WDFW for the Mussel Watch Expansion Project.

Figure 1. Mussel Watch Pilot Expansion Sites 2012-2013



Methods

Site selection

Site selections were based on study criteria specifying shoreline areas with a variety of commercial, residential, and industrial land uses. Initial sites considered included: Middle Waterway in Commencement Bay, Hylebos Waterway, Dash Point, Ruston Way, Titlow Beach, and Chambers Bay. Following a cursory evaluation, the sites were narrowed down to two possible locations on Ruston Way and five possible locations on the Hylebos Waterway. After much communication with project partners (WDFW, University of Washington Tacoma (UWT), Ecology, Port of Tacoma, City of Tacoma, and Pierce County Surface Water Management) and property owners, the final site selection was made.

Ruston Way – The site chosen on Ruston Way in the City of Tacoma is located on Commencement Bay and is a residential/commercial site (Figure 2). It is located east of North Alder Way and the Silver Cloud Hotel. The property owners consist of Pierce County Parks, Ram Restaurant/Harbor Lights, City of Tacoma and Tacoma Metro Parks. Gaining access to each parcel on the Ruston Way site proved to be a challenge. The Ram/Harbor Lights parcel owner denied access for the study therefore accommodations were made for the mussel cage placement to be outside of that parcel boundary.

Right of Entry and Hold Harmless Agreements were obtained for those parcel owners which required the agreements for access. Communication about the study details and timelines were relayed to the parcel owners on a regular basis prior to cage deployment. Although the participating property owners were accommodating and tried to work with project staff, securing agreements was a very cumbersome and time-consuming process.

Ruston Way cages were placed an average of 326 feet apart, but actual distances between cages ranged from 250 to 485 feet. The different distances were due to working around properties where the owner didn’t want to participate and avoiding creosote pilings.

Figure 2. Ruston Way – Cage Placement Locations

Hylebos Waterway – This site (Figure 3), located on the western shore slightly over halfway down the waterway, is the former Arkema site and includes the property next to it. The site was formerly home to a chemical manufacturing plant which made products such as chlorine, sodium hydroxide (caustic soda), hydrochloric acid, and Penite (an herbicide containing arsenic). Production stopped in 1997 and most of the buildings have been removed. The Port of Tacoma purchased the property in 2007, assuming liability for the cleanup. (https://fortress.wa.gov/ecy/gsp/Sitepage.aspx?csid=3405)

Properties adjacent to this study site include US Gypsum to the west and a log yard to the east. Both properties within the study site have a sub-tidal and intertidal cap where cage placement took place. Since there was a possibility of disturbing the integrity of the caps due to the use of rebar to secure the cages to the substrate during deployment, the EPA was notified of the study. The caps do not have a protective membrane and approval was granted from EPA to move forward.

Gaining access to the Hylebos Waterway site required obtaining a Hold Harmless Agreement with the Port of Tacoma. Communication about the study details and timelines were relayed to the Port of Tacoma prior to cage deployment. This site is a secured, fenced site and required the Health department to contact Port Security prior to any work. The Port was helpful in working through the agreement and details but, as with the Ruston properties, this took much more time and effort than originally anticipated.

The Hylebos Waterway cages were placed an average of 328 feet apart. As with the Ruston Way cages, the distances between cages was quite variable, ranging from 250 to 485 feet, to avoid creosote pilings and the large dock.

Figure 3. Hylebos Waterway – Cage Placement Locations

Scope of Work Finalized

WDFW finalized the overall QAPP for the caged and resident mussels and Tacoma-Pierce County Health Department (Health Department) staff developed a work plan that was provided to the Port of Tacoma for the Hylebos Waterway as well as Ruston Way property owners.

Mussel Preparation

The Puget Sound native Pacific blue mussel (*Mytilus trossulus*) was used for this study. All *M. trossulus* were donated by Penn Cove Shellfish and came from their commercial shellfish aquaculture facility on Whidbey Island near Coupeville, Washington. From October 22–29, 2012, the WDFW-PSEMP team and citizen science volunteers prepared live mussels, provided from the daily harvest routine of Penn Cove Shellfish, for field deployment. Only living mussels (i.e., able to close their shells upon stimulation) with intact shells that measured between 50–60 mm in length were used in this study. Two groups of eight mussels (16 total) were placed into a polyethylene mesh grow-out bag. Each bag was subsequently labeled with a unique bag ID number. The shell lengths of all mussels placed into each bag and the corresponding bag ID number were recorded on a datasheet. The bags of mussels were attached approximately 20 cm apart to grow-out lines that were hung from an aquaculture raft in Penn Cove for 10 days. The 10 day period was intended to allow the bagged mussels time to reattach their byssal threads and recover from the stress of handling prior to deployment.

Reference Mussels

Thirty-one bags of mussels were left hanging on lines at the Penn Cove Shellfish aquaculture facility after deployment of all other mussel bags to their designated sites. These mussels were retained as potential replacements if deployed mussels were lost, and as a control for growth effects from the caging and translocation process.

Baseline Mussels

Twenty bags (containing 320 total mussels) were removed from the Penn Cove Shellfish aquaculture raft on November 15, 2012 and saved to allow for determination of the Condition Index (CI) and tissue contaminant residue of mussels prior to deployment (i.e. initial contaminant condition). The day after removal from Penn Cove, 100 of these mussels were taken from their bags, inspected, rinsed with tap and deionized water, and processed immediately at WDFW's Marine Resources Laboratory in Olympia. To determine CI, individual mussels were assigned a unique Fish Identification (FishID) number and their total shell length (TSL) was measured using digital calipers. Mussels were then opened using a scalpel blade inserted between the two valves to reveal the soft tissue. Any remaining byssal fibers were cut from the byssal gland and discarded. All soft tissue was scraped from the shells into a pre-weighed aluminum drying pan and weighed to the nearest tenth of a gram (0.1 g).

Mussel Cage Deployment

Due to the steep terrain and accessibility issues, the Hylebos Waterway site was first accessed during the day to determine cage placement. The strategy for cage placement with this site was somewhat different than Ruston Way due in part to there being only one property owner, the historical contamination at the site, the steeper intertidal zone slopes, and the secured site access. The site was first visited to consider the terrain, upland areas to avoid, and possible placement locations. Latitude and longitude coordinates were taken with a GPS unit at that time and subsequently placed on a satellite map to determine the approximate distance between cages. The Hylebos site is a fenced and secured site so any entrance onto the site was coordinated with Port Security.

The pre-bagged mussels for the Ruston Way site, sediment sample vials, data sheets, gloves, cages, zip-ties, helical anchors and rebar were retrieved from Penn Cove Shellfish on the afternoon of November 13, 2012, the same day the Ruston Way deployment was scheduled to take place. Mussels were kept in coolers, on ice, while being transported. The mussels for the Hylebos Waterway site, along with the pertinent supplies and tools, were collected the following afternoon, on November 14, 2012 for deployment that night. Deployment of the mussels and cages for both sites occurred at night during the low tide interval, to enable the placement of the cages between 0.0 and -1.5 feet mean sea level.

Staff and volunteers supplied additional materials needed to complete cage deployment such as safety equipment, gloves, boots and waders, life vests, flashlights and headlamps, cameras, GPS units, mallets, and wire cutters.

Mussel deployment for each site was organized separately. Health Department staff coordinated with Dr. James Gawel from the University of Washington, Tacoma for the Ruston Way site. Dr. Gawel managed the deployment of these cages on the night of November 13, 2012, using volunteers from UW Tacoma. Health Department staff managed the mussel cage deployment on the Hylebos Waterway at low tide on the night of November 14, 2012.

Staff and volunteers split up into teams to accomplish the cage installments within approximately one hour around the low tide. Four mussel bags were zip-tied into the top third of each cage and cage lids were secured with zip-ties. This work was completed at the site before the cages were taken to the beach locations for installation.

Installation of the cages took place at the 0’ to -1.5’ tide range for each site close to the GPS coordinates obtained in the preliminary scouting visit and, in the case of the Hylebos locations, orange marker flags. Care was taken to place the mussel cages well away from creosote pilings. Helical anchors were installed first, and the cages were attached to the anchors by zip-ties. Rebar stakes were then installed into the substrate through the cage on the opposite side or corner, and were also secured to the cage with zip-ties. At one site on the Hylebos it was necessary to use a cinder block for additional anchorage due to the conditions of the area (steep hills, rocky substrate).

Data and observations for each deployed cage were recorded on data sheets provided by WDFW. GPS coordinates were updated following cage placement and pictures of the installed cages were taken when possible. Copies of the cage placement data sheets and associated pictures were given to WDFW.

Figure 4. Picture of cage placement at the Hylebos Waterway

Cage Checks

Approximately one month after placement the mussel cages were checked for integrity as well as the possible invasion by sea stars. Both sites fared well with all of the mussel cages intact and no, or minimal, sea star invasion. The sites were checked on the following dates:

- *November 27, 2012- Ruston Way*
- *December 14, 2012- Hylebos Waterway*

The only signs of predation were found in three of the Ruston Way cages at the time of cage retrieval:

Cage #1- predation by 2 sea stars; 73% survival rate
Cage #5- P. producta *predation; 89% survival rate*
Cage #8- P. producta *and* P. helianthoides *predation; 84% survival rate*

Cage and Mussel Retrieval

During deployment of the Hylebos Waterway cages, Health Department staff noted a sizeable population of resident, or native, mussels in close proximity to several of the cages. A proposal was developed to also collect a native mussel sample at the Hylebos Waterway site. This proposal was supported by Ecology and WDFW.

Retrieval of mussel cages was organized separately in the same manner as cage deployment. Cages at both locations were retrieved at low tide on the evening of January 9, 2013.

Teams returned to each cage site to remove the rebar stakes and helical anchors. The four bags from each cage were kept together in labeled plastic bags and placed in a cooler with ice for transport to WDFW in Olympia the next morning.

Surface sediment samples were collected from select sites during the mussel cage retrieval to accommodate an ancillary study conducted and funded by one of the study partners, Dr. Gawel from UW Tacoma. Using the protocol developed by Dr. Gawel, volunteers collected a sediment sample from the top 2 cm of substrate at or near the mussel cage location. The sediment samples were held in the -20° C freezer at WDFW as they arrived, but were later transferred to the 5° C cold room. These samples were stored in the cold room until they were delivered to Dr. Gawel at UW Tacoma on January 31, 2013 for analysis.

Native mussels approximately 1.5" in length were collected for a comparison to the caged mussels near Hylebos Cage locations 1 and 2. Forty mussels were collected near Cage 1, and sixty mussels were collected near Cage 2. These mussels were placed in separate bags and stored overnight in the coolers with the caged mussels.

Data and observations for each cage were recorded prior to their complete removal from the deployment site. Pictures of cages were taken before removal when possible.

Figure 5. Picture of resident mussel sorting on the Hylebos Waterway

WDFW Laboratory Processing

Mussels from both the Ruston Way and Hylebos Waterway sites were transported in coolers the next morning (January 10, 2013) to WDFW Marine Resources Laboratory at the Natural Resources Building (1111 Washington St SE, 6th floor) in Olympia, Washington. The routine chains of custody forms were used.

After being checked in, all mussels were stored in the walk-in refrigerator to wait processing. Initial processing procedures involved selecting eight mussels from each of the four bags per cage, rinsing them in tap water, then removing any gaping, empty, dead, or cracked mussels and replacing them with live ones from the same cage. The excess byssal threads protruding from the shells were then removed and the mussels were rinsed again thoroughly with deionized water. These cleaned mussels were measured with calipers and their lengths were recorded. Each mussel was then opened, the tissue gently extracted into a cup on a tared scale, and the mass recorded. Once all tissue had been removed from mussels into the cup, the composition was blended into slurry and weighed out into separate jars and bags for distribution to laboratories for further testing.

The samples were delivered to contracted analytical laboratories (NOAA Montlake Laboratory and King County Environmental Laboratory) for analyses. Chemical contaminants analyzed included polychlorinated biphenyls (PCBs), polybrominated diphenylethers (PBDEs), polycyclic aromatic hydrocarbons (PAHs), organochlorine pesticides such as dichlorodiphenyltrichloroethane compounds (DDTs), and a suite of metals including mercury, lead, arsenic, zinc, copper, cadmium. Important biological covariates included CI, tissue lipid content, percent solids, and select stable isotopes.

Laboratory Processing of Native Mussels

The naturally occurring native mussels were rinsed with tap and deionized water following the standard protocol described above. These mussels were then measured and sorted according to length (size range was approximately 20–60 mm) and 44 of the longest mussels were then selected for processing. Of these 44 naturally occurring mussels, 12 were processed for Condition Index (CI) and 32 were stored in the -20° C freezer for future resection and contaminant analysis.

Laboratory Processing of Reference Mussels

Mussels that were not deployed in November but left hanging at the Penn Cove aquaculture rafts were also retrieved during the week of cage retrieval and processed as a control sample. Twenty-one bags of these reference mussels underwent sorting and rinsing following the protocols outlined above. Five mussels from each bag (100 total) were set aside for determination of CI. The remaining mussels were frozen for future resection and contaminant analysis.

Results

For the larger WDFW Project, and including the Ruston Way and Hylebos Waterway cages, mussel survival ranged from 63 – 97%. The survival rates for the Ruston Way and Hylebos Waterway cages are shown in Tables 1 through 3.

Dead mussels were sorted into four categories; 1) empty, 2) rotten, 3) gaping and 4) cracked. Descriptions of the four categories are as follows: 1) empty - mussels or shell fragments contained no living tissue, 2) rotten – mussels with putrid or rotting tissue, 3) gaping – open mussels were considered dead if they did not respond to stimulation, and 4) cracked - mussels had cracks or holes in their shell. Of the total 7,023 mussels that were returned and counted, 798 (11%) were empty, 22 (0.3%) were rotten, 27 (0.4%) were gaping, 34 (0.5%) had cracked shells, and 6,142 (87%) were alive and intact. Predation was noted in mussels from 12 sites during the sorting process. Predation was identified by the presence of drill holes in the shells, possibly from Japanese oyster drills (*Ceratostoma inornatum*).

If no empty shell valves or fragments were found in a bag and the total number of mussels in that bag was less than 16, then WDFW speculated that either the original number of mussels in the bag was miscounted or mussels were removed from the bag by a predator. Thus, in bags with fewer than 16 mussels WDFW used the total number of mussels remaining in each bag, minus any dead mussels, to calculate survival for that bag.

Table 1. Hylebos Waterway Survival Rates

Cage	Percent (%) Survival
1	84%
2	88%
3	84%
4	88%
5	80%
6	81%
7	86%
8	91%
9	77%

Table 2. Ruston Way Survival Rates

Cage	Percent (%) Survival
1	73%
2	81%
3	75%
4	92%
5	89%
6	81%
7	90%
8	84%
9	88%

Table 3. Percent Mortality & Percent Survival Data

	Hylebos	Ruston
Mean % Mortality	14.95	16.17
Mean % Survival	85.05	83.83
Median % Mortality	12.50	15.63
Median % Survival	87.50	84.38
Range % Mortality	0% - 31.25%	0% - 31.25%
Range % Survival	68.75% -100%	68.75% - 100%

Baseline and Reference Mussels

The bags of mussels taken from Penn Cove Shellfish during the deployment period (baseline) had a total of 18 dead and 318 living mussels (95% survival rate). The reference mussels taken from Penn Cove Shellfish during the retrieval period had a total of 22 dead (i.e. empty) and 310 living mussels (93% survival rate).

Condition Index (CI) Data

The equation used by WDFW to calculate the CI was:

*(Dry Tissue Weight/Shell Length)*100*

The dry weight CI was chosen for this study so as to remove variability related to differences in water retention between mussels. The CI is used to indicate the relative condition of the mussels over time. There doesn't seem to be an accepted scale or range of reference for CI data in any related studies. A comparison between high and low CI values is the accepted method of determining overall mussel health. The native mussel sample had a smaller sample size and age was unknown therefore the comparison to the caged mussel samples was not applicable.

Table 4. Average Dry Weight CI per site

Site	Average CI
Ruston Way	27.73
Hylebos Waterway*	25.81
Native Hylebos Population	17.00

*7 of 12 mussels from Hylebos Cage 7 did not have their lengths recorded and were not included in the calculation of that cage's CI value.

Metals

The raw data tables are provided in the Appendix. The native mussel sample from the Hylebos Waterway is referred to as sample number 13CPS_HYW2-MXW01B and in the table below. The native mussel results were not included in the All Sites samples results since they were not from the same original population and were not caged.

Wet and dry weight tables were included in the data received from WDFW for metals concentration comparisons. However, in the literature there doesn't seem to be consensus when comparing data whether to use wet weight or dry weight in the analyses. We used only wet weight data for our calculations of means and ranges for metals concentrations (mg/Kg or ppm). The All Sites data included the data from the Hylebos Waterway as well as Ruston Way. The Penn Cove data are the baseline reference samples. See the tables below:

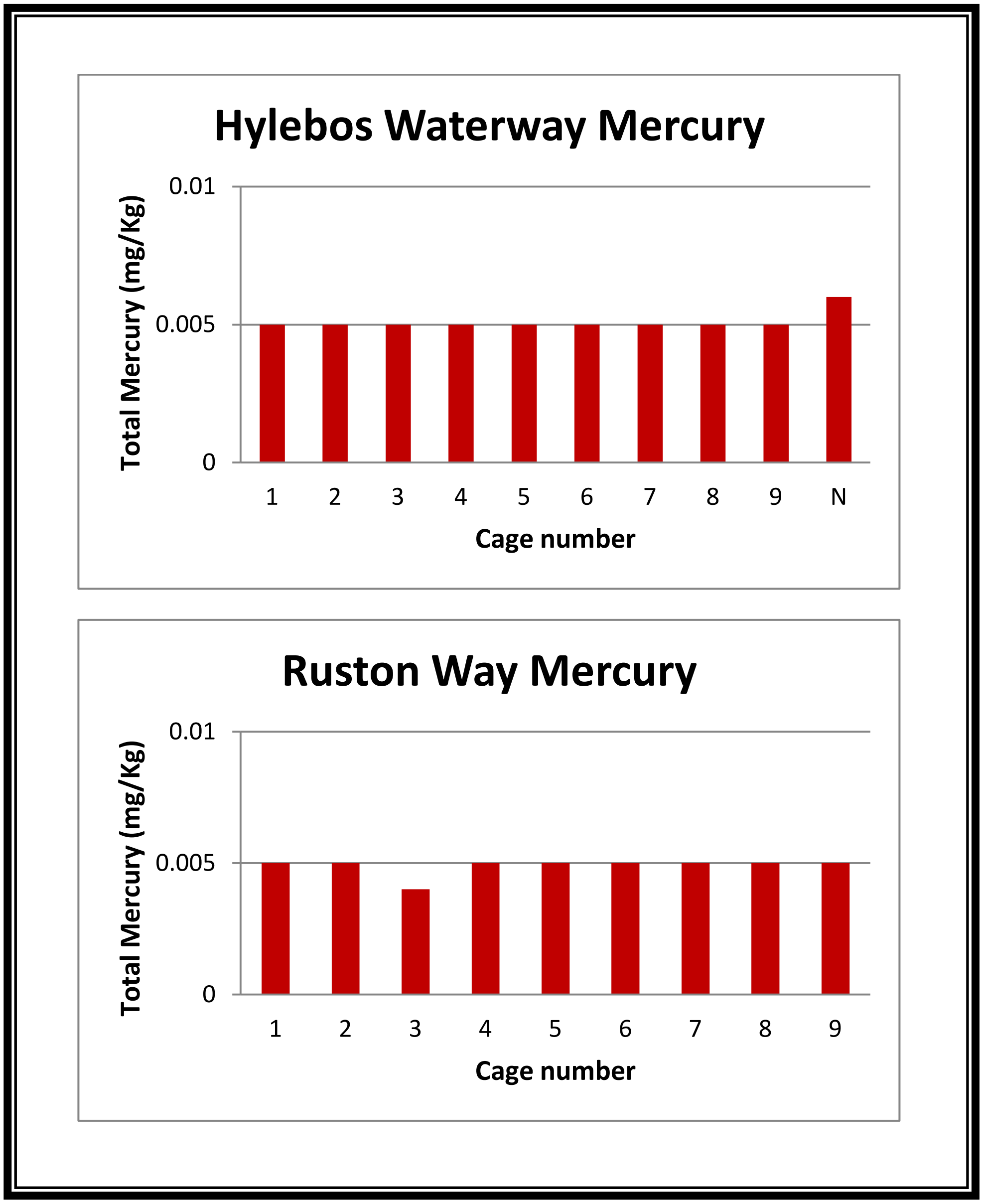
Table 5. Range and Mean Metal Concentrations (mg/Kg Wet Wt.)

Metals	**Ruston**		**Hylebos**		**All Sites**		**Penn Cove (Baseline)**	
	Mean	*Range*	*Mean*	*Range*	*Mean*	*Range*	*Mean*	*Range*
Mercury	0.005	0.004-0.005	0.005	0.005-0.005	0.006	0.004-0.011	0.005	0.005-0.005
Arsenic	0.83	0.78-0.89	0.95	0.84-1.12	0.87	0.65-1.2	0.85	0.83-0.88
Cadmium	0.36	0.32 - 0.40	0.30	0.27-0.34	0.31	0.24-0.42	0.33	0.31-0.36
Copper	0.96	0.83-1.09	1.12	0.93-1.75	0.90	0.60-1.8	0.80	0.74-0.84
Lead	0.07	0.061-0.095	0.04	0.035-0.06	0.05	0.02-0.14	0.02	0.019-0.023
Zinc	12.2	10.6-13.5	16.27	13.0-18.4	13.0	9.5-18.0	12.1	12.0-13.0

Table 5a. Hylebos Waterway Native Mussel Sample Metal Concentrations (mg/Kg Wet Wt.)

Metals	**Concentration**
Mercury	0.01
Arsenic	0.82
Cadmium	0.23
Copper	1.64
Lead	0.09
Zinc	31.8

Figure 6. Mercury Concentrations (mg/Kg wet wt.) in Ruston and Hylebos Mussels



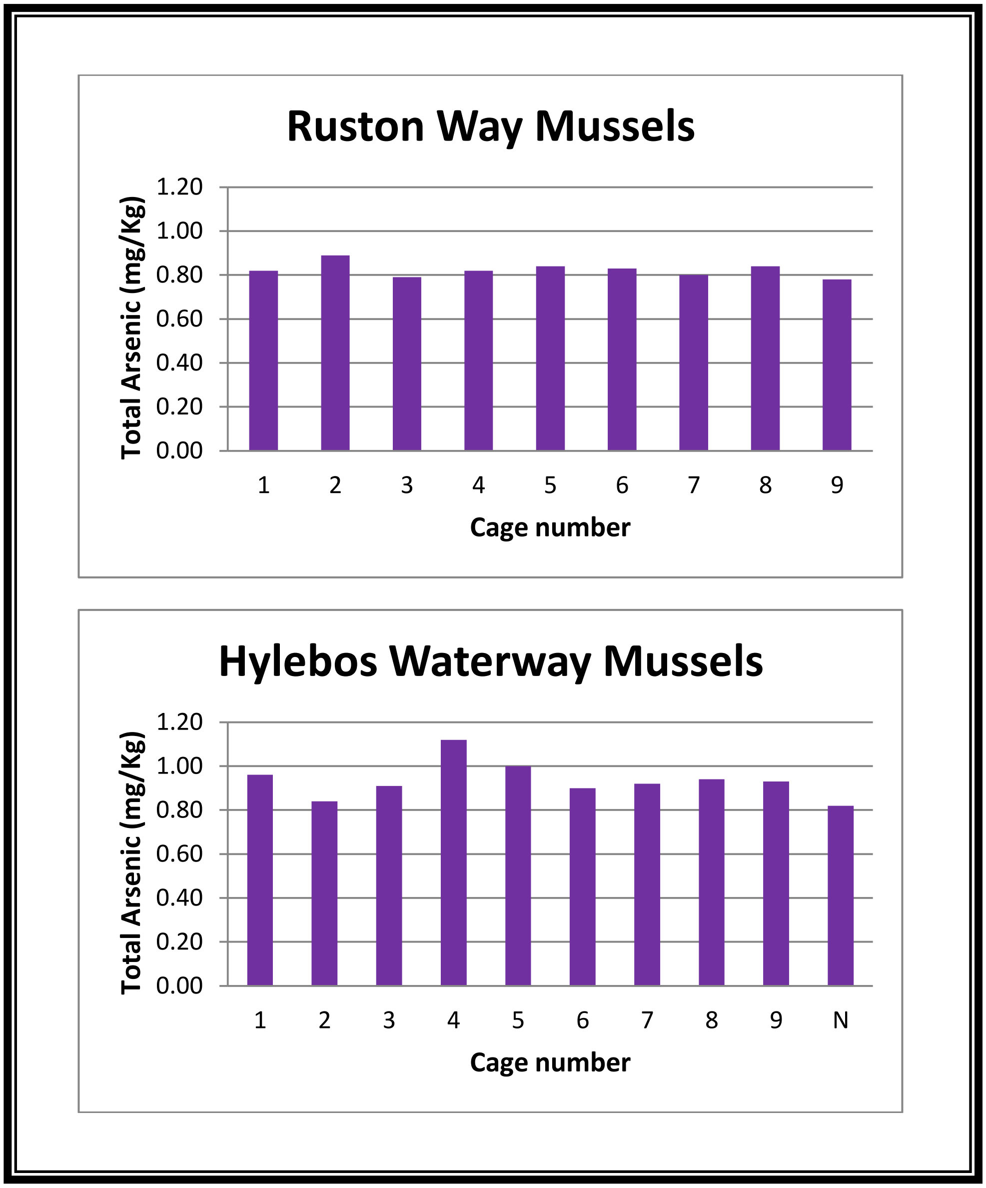
Mercury is mostly found in its organic form as methyl mercury in fish and seafood. Exposure to mercury, even in small amounts, is a great danger to humans and wildlife. When mercury enters the body it acts as a neurotoxin. Most mercury pollution is produced by coal-fired power plants and other industrial processes. The most common way humans are exposed to mercury is by eating contaminated fish (NRDC 2010).

We rounded the Mercury values to the hundredths therefore, the mean mercury concentrations for the Hylebos Waterway site and the Ruston Way site were the same at 0.005mg/Kg. Both the Hylebos

Waterway site and the Ruston Way site had mean mercury concentrations lower than the All Sites mean of 0.0062 mg/Kg.

The Hylebos native mussel sample had a mercury concentration of 0.006 mg/Kg. This was higher than any of the caged mussel samples from the Hylebos Waterway site or Ruston Way site but was within the range observed for All Sites. The Hylebos Waterway and Ruston Way caged mussel mercury concentrations were also similar to the concentrations of the Penn Cove baseline samples. Mercury was found in 100% of the All Sites samples as well as the Penn Cove baselines samples.

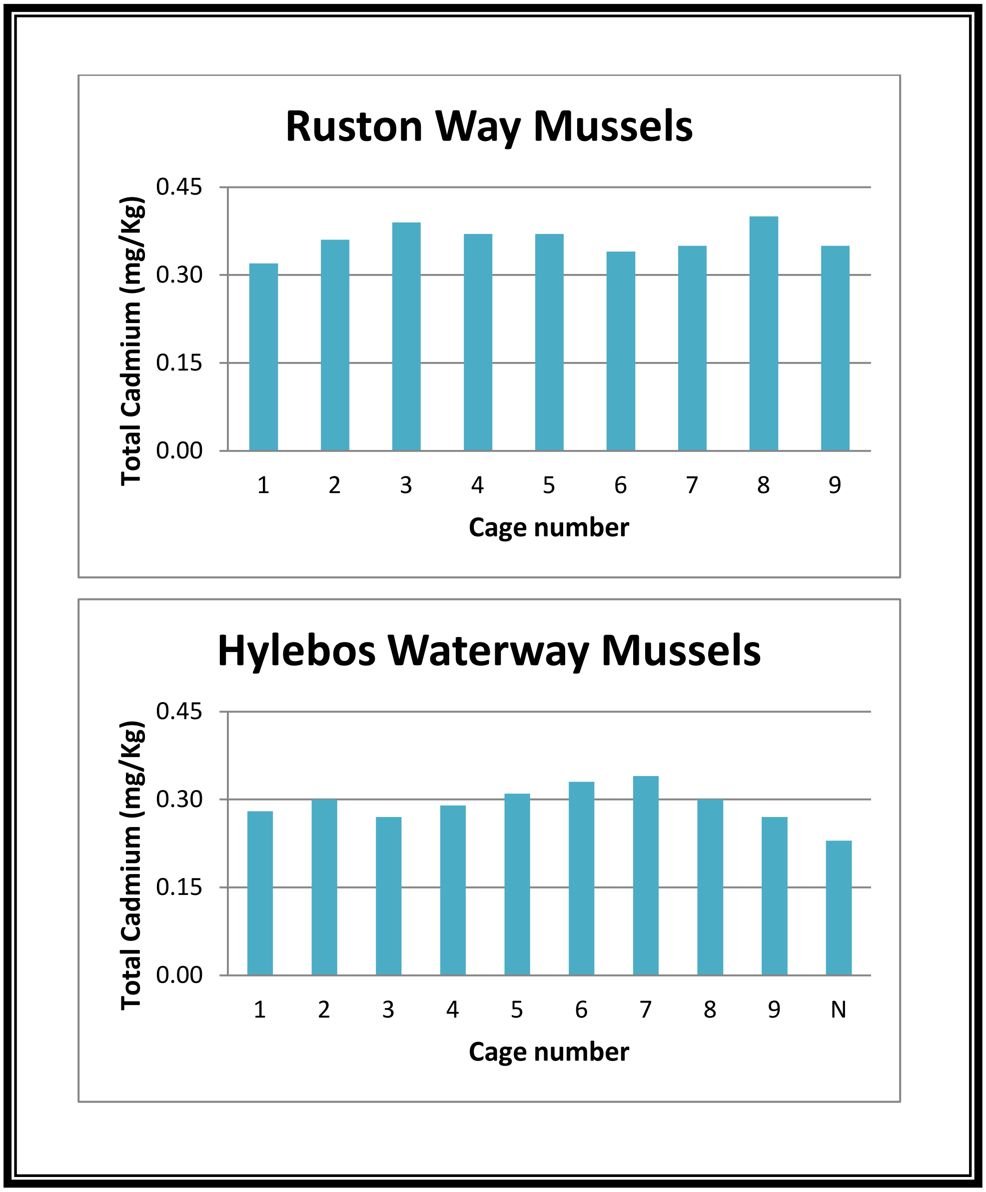
Figure 7. Arsenic Concentrations (mg/Kg wet wt.) in Ruston and Hylebos Mussels



Arsenic is a naturally occurring element and is found throughout the environment which includes marine waters. Most people are exposed to arsenic through food and possibly in drinking water (EPA 2013). In the past, arsenic has been used in wood preservation products as well as agricultural products.

The mean concentration of arsenic differed between sites, with the Hylebos Waterway site mean being higher (0.95 mg/Kg) than the Ruston Way site (0.83 mg/Kg). The mean for All Sites was 0.87mg/Kg and the Penn Cove baseline mean was 0.85 mg/Kg. The Hylebos native mussel result for arsenic was 0.82 mg/Kg and was within the Ruston Way site range but below the range for the Hylebos Waterway site. Arsenic was found in 100% of the All Sites samples and Penn Cove Baseline Samples.

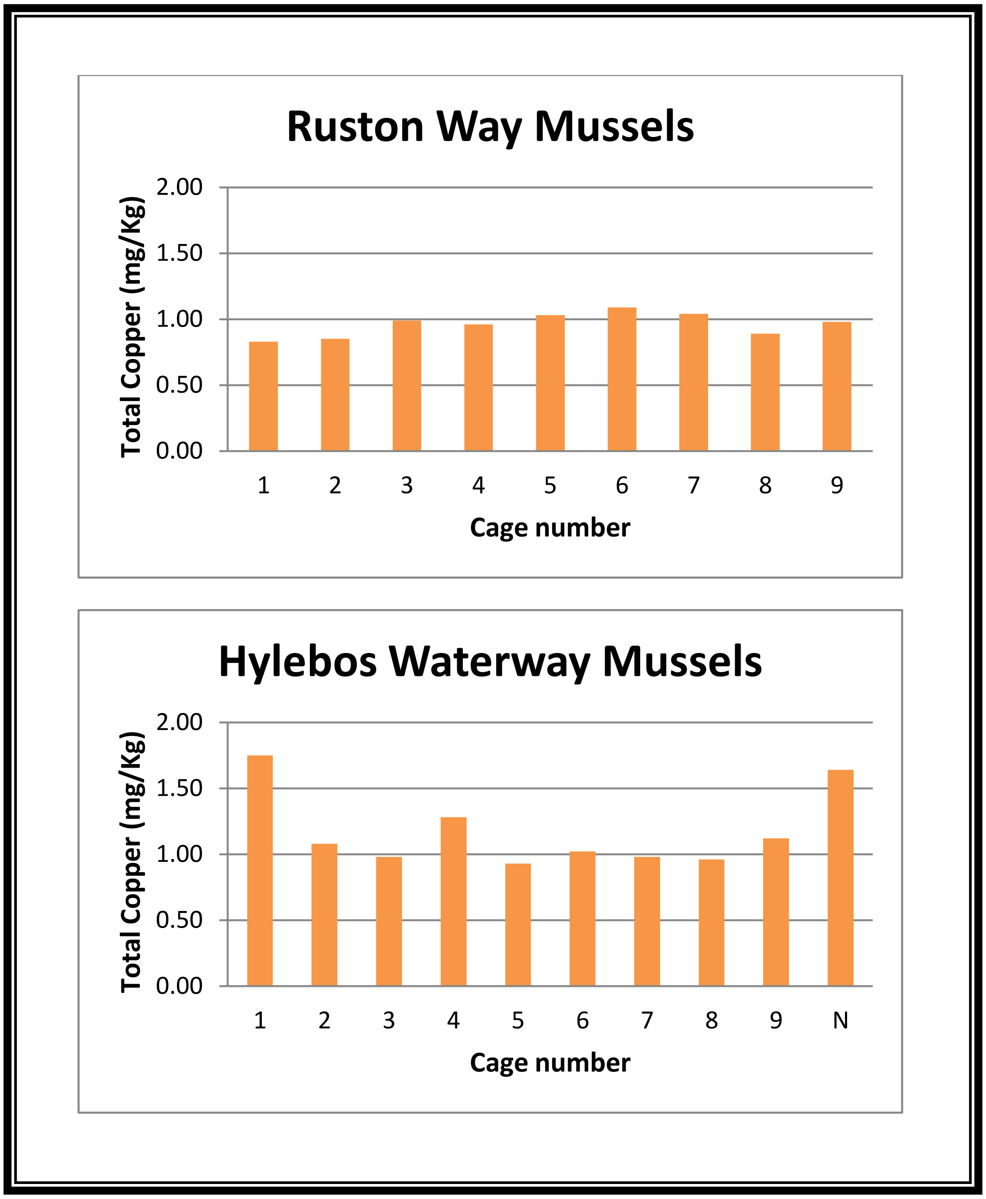
Figure 8. Cadmium Concentrations (mg/Kg wet wt.) in Ruston and Hylebos Mussels



Cadmium is widely used in industrial processes as an anticorrosive agent, a stabilizer in PVC products, a color pigment, a neutron-absorber in nuclear power plants, and in the fabrication of nickel-cadmium batteries and naturally occurring in marine waters. High levels of cadmium are often associated with industrial activities such as emissions and the use of fertilizer. Studies of human exposure to cadmium have shown kidney damage and bone demineralization (J. Godt 2006).

The mean concentration of cadmium for the Ruston Way site was 0.36 mg/Kg and was higher than the Hylebos Waterway site mean of 0.30 mg/Kg. The All Sites mean of 0.31 mg/Kg and the Penn Cove baseline mean of 0.33 mg/Kg fell in between Ruston Way and the Hylebos. Cadmium was found in 100% of the All Sites samples and Penn Cove baseline samples.

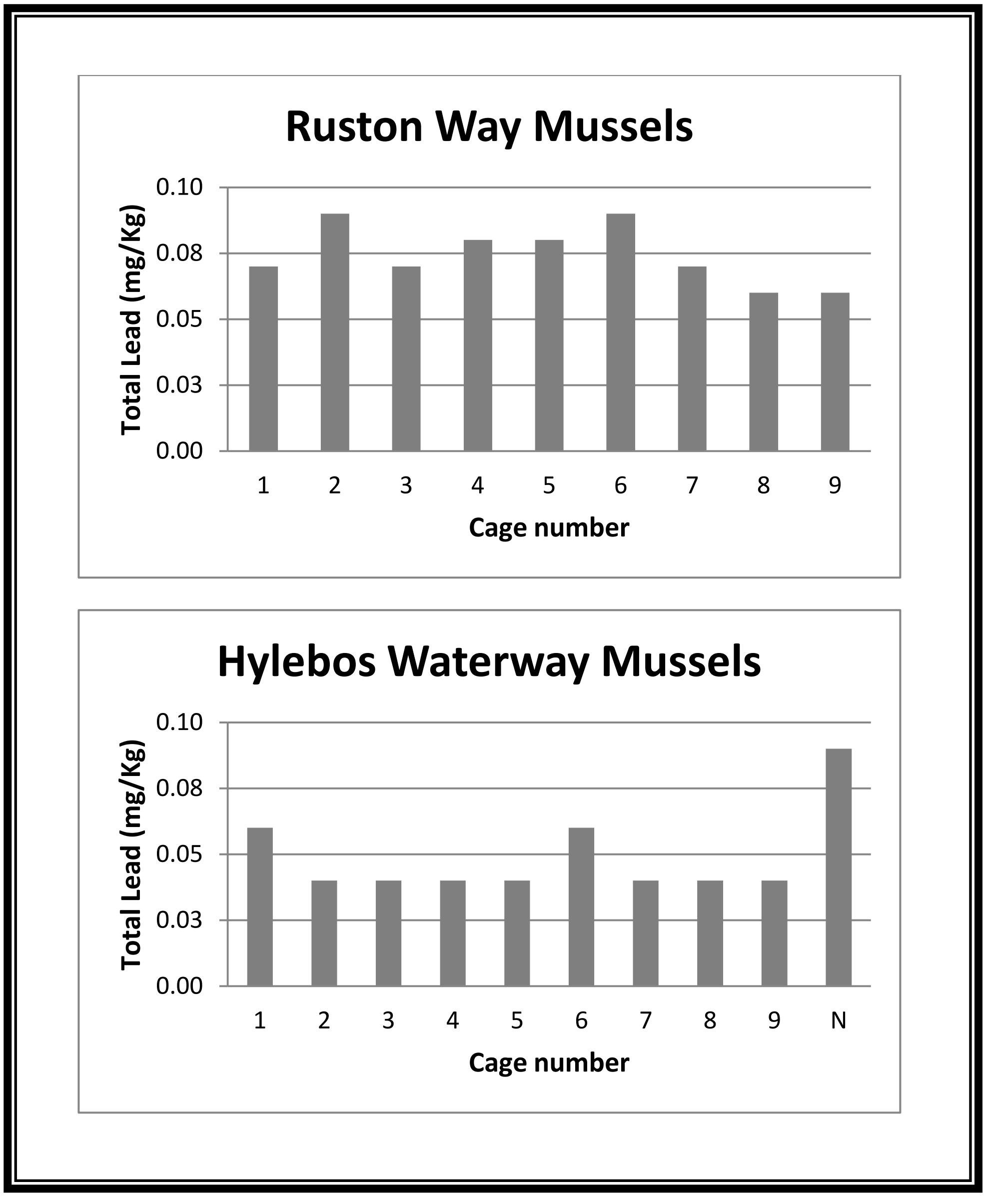
Figure 9. Copper Concentrations (mg/Kg wet wt.) in Ruston and Hylebos Mussels



Copper affects the olfactory ability of salmon during homing migration, and their ability to detect predators. Copper found in the environment can come from metal recycling and brake linings as well as various industrial processes. Although copper is an essential element to human health, it can be harmful in high doses.

Hylebos Cage 1 had the highest copper concentration at 1.75 mg/Kg. This may have been related to the location immediately adjacent to a large trucking facility parking area. The mean copper concentrations for both the Hylebos Waterway, 1.12 mg/Kg, and Ruston Way, 0.96 mg/Kg, caged mussels were above the All Sites mean of 0.90 mg/Kg as well as the Penn Cove Baseline mean of 0.80 mg/ kg. Copper was found in 100% of the All Sites samples and Penn Cove baseline samples.

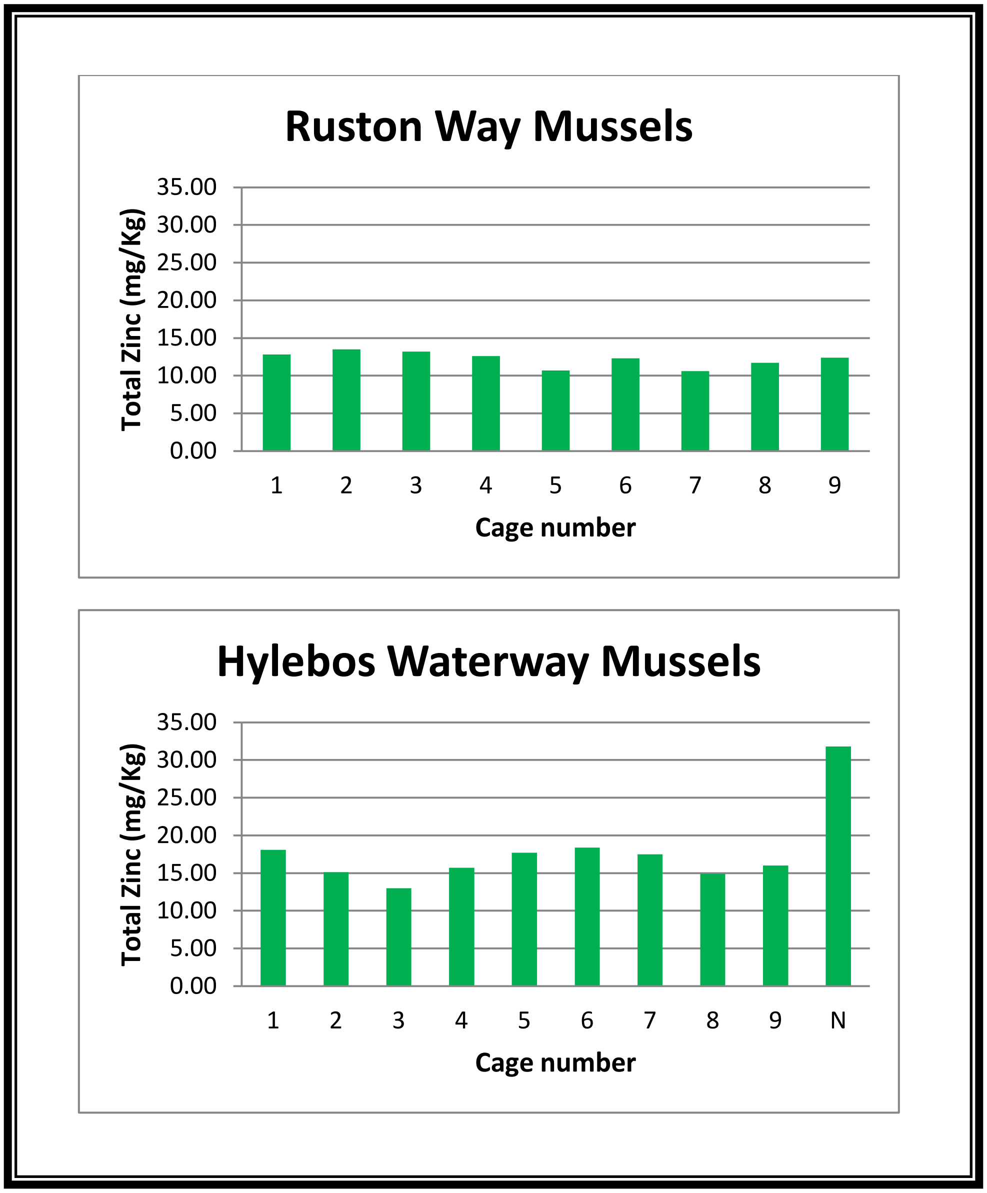
Figure 10. Lead Concentrations (mg/Kg wet wt.) in Ruston and Hylebos Mussels



Lead is a bio-accumulative heavy metal which has no known biological function and is a serious public health risk due to its toxicity. Lead exposure reduces cognitive development in children and increases blood pressure and cardiovascular diseases in adults. Lead is present in the environment due to paints, coatings, and industrial practices. (Moses 2009)

Ruston Way had a higher mean lead concentration, 0.07 mg/Kg, than the Hylebos Waterway site, 0.04 mg/Kg. The All Sites mean lead concentration was 0.047 mg/Kg, and the mean lead concentration for the Penn Cove baseline sample was 0.02 mg/Kg. Lead was found in 100% of the All Sites samples as well as the Penn Cove baseline samples.

Figure 11. Zinc Concentrations (mg/Kg wet wt.) in Ruston and Hylebos Mussels



Zinc is similar to copper in its anthropogenic origin and its effects on human health. Zinc is a contaminant in stormwater which originates from metal fencing, metal roofing, and industrial facilities such as scrap metal recycling.

The mean zinc concentration for Ruston Way, 12.20 mg/Kg, was much lower than the Hylebos Waterway site, 16.27 mg/Kg, and was comparable to the All Sites mean of 13.0 mg/Kg as well as the Penn Cove baseline mean of 12.1 mg/Kg. The Hylebos native mussel sample had a zinc concentration of 31.9 mg/Kg, which was much higher than any of the caged mussel samples. Zinc was found in 100% of All Sites and Penn Cove baseline

PCBs, PBDEs, PAHs, Contaminants of Emerging Concern, DDTs, Chlordane, HCHs and others

The organic results were reported in wet weight only. This resulted in a variation of the Method Detection Limit (MDL) from sample to sample, even for the same constituent. Standard reference materials (SRM) as well as laboratory control samples (LCS) and method blanks were run with each sample for quality control.

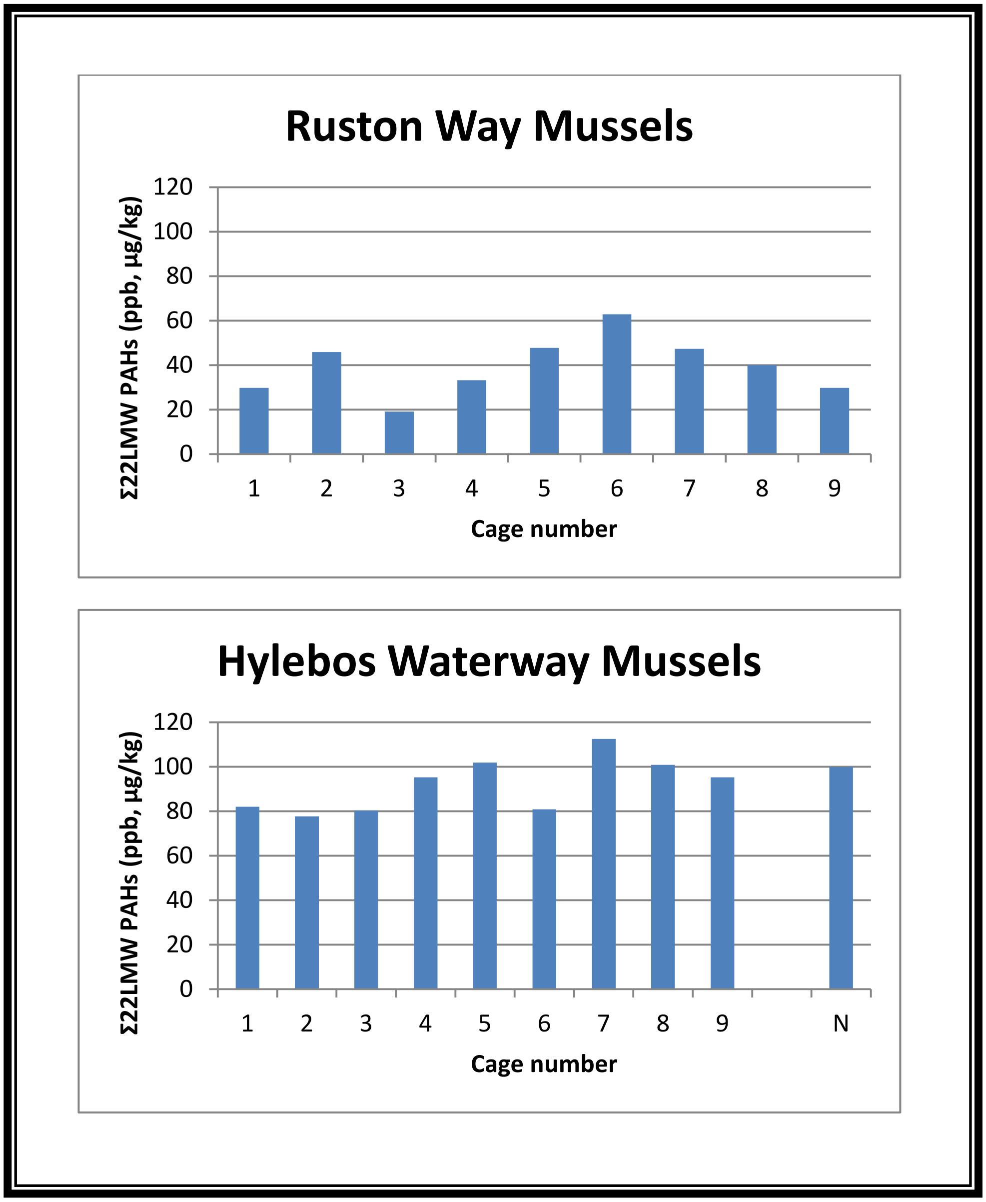
Table 6. Mean, Min, and Max Chemical Contaminant Concentrations (ppb or µg /Kg Wet Wt.)

Chemical Contaminants	Ruston		Hylebos		All Sites		Penn Cove baseline	
	Mean	*Range*	*Mean*	*Range*	*Mean*	*Range*	*Mean*	*Range*
LMWPAHs	40	19 - 63	92	90 - 113	39.2	3.3 - 290	9.26	6.9-14
HMWPAHs	48	27 - 71	142	118 - 175	57.2	1.1 - 480	1.71	0.94-2.8
Hexochlorobenzene	ND	ND	0.23	0.21 - 0.27	0.23	0.21 – 0.27	ND	ND
HCHs	ND	ND	ND	ND	ND	ND	ND	ND
Chlordanes	ND	ND	0.96	0.75 – 1.20	ND	0.13 – 1.6	ND	ND
DDTs	0.33	0.29 – 0.41	6.8	5.10 – 8.70	0.69	0.21 – 8.7	0.18	0.16-0.19
PCBs	4.4	3.2 – 9.7	32	27 - 35	5.44	0.57 - 35	1.54	0.74-2.6
Dieldrin	ND	ND	0.38	0.30 – 0.46	0.287	0.13 – 0.46	ND	ND
Mirex	ND	ND	ND	ND	0.24	0.24 – 0.24	ND	ND
PBDEs	1.28	0.99 – 1.8	3.67	3.10 – 4.00	1.38	0.24 – 5.4	0.44	0.24-0.79
Values with ND were below the detection limit								

Table 6a. Hylebos Waterway Native Mussel Sample Organic Concentrations (ppb, µg/kg)

Organics	Concentration
LMWPAHs	100
HMWPAHs	190
Hexachlorobenzene	0.19
HCHs	ND
Chlordanes	2
DDTs	11
Est. Total PCBs	75
Dieldrin	0.31
Mirex	0.61
PBDEs	14
Values with ND were below the detection limit	

Figure 12. Total LMW PAH Concentrations (ppb, µg/kg) in Ruston and Hylebos Mussels

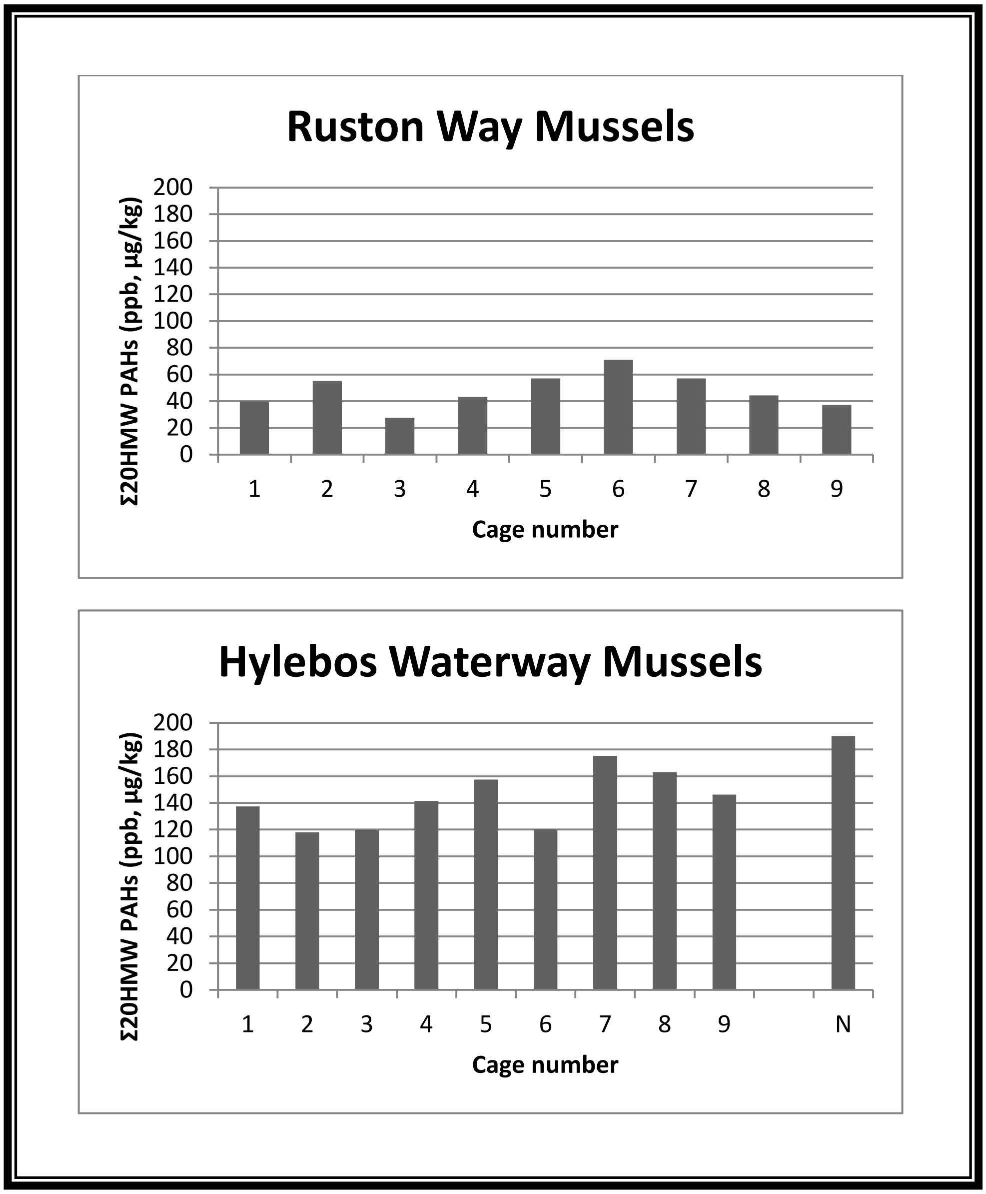


Polycyclic aromatic hydrocarbons (PAHs), also known as poly-*aromatic* hydrocarbons, are one of the most widespread group of organic pollutants. Some compounds have been identified as carcinogenic, mutagenic and teratogenic. In the environment PAHs will be found primarily in soil and sediment, but also occur in oil, coal, and tar deposits. When these are burned as fuel, the byproduct is atmospheric particulate air pollutants, which can also contain PAHs.

High Molecular Weight (HMW) PAHs are lipophilic, hydrophobic compounds with the ability to accumulate in sediments as well as organisms. Low Molecular Weight (LMW) PAHs have acutely toxic properties (Mozes, A. 2009). A potential source of PAH contamination in the environment is creosote, commonly used to preserve marine structures from decay. It is a complex mixture of chemicals, many of which are toxic to fish and other marine organisms.

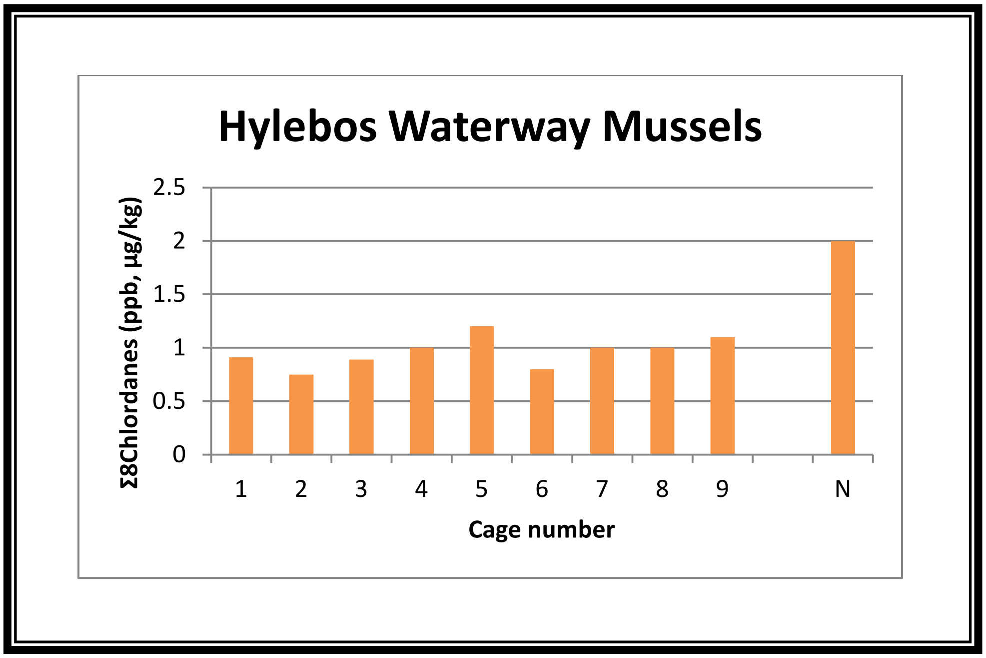
The SumLMW PAHs mean concentration for the Hylebos Waterway caged mussel samples of 92 ppb greatly exceeded the Ruston Way mean of 40 ppb, the All Sites mean of 39.2 ppb and the Penn Cove baseline mean of 9.26 ppb. The SumLMW PAHs concentration in the Hylebos native mussel sample of 100 ppb far exceeded that of the Hylebos Waterway caged mussel samples. LMWPAHs were present at detectable concentrations in all of the caged mussels for All Sites and the Penn Cove baseline samples.

Figure 13. Total HMW PAH Concentrations (ppb, µg/kg) in Ruston and Hylebos Mussels



The SumHMW PAHs mean concentration for the Hylebos Waterway caged mussel samples, at 142 ppb, greatly exceeded the mean for the Ruston Way samples (48 ppb), the All Sites mean (57.2 ppb) and the Penn Cove baseline mean (1.71 ppb). The Hylebos native mussel sample had a SumHMW PAHs concentration of 190 ppb which was much higher than the Hylebos Waterway caged mussel samples. The SumHMW PAHs were found in 100% of the All Sites samples and the Penn Cove baseline samples.

Figure 14. Total *Chlordane Concentrations (ppb, µg/kg) in Hylebos Waterway Mussels
All Ruston Way mussel samples had chlordane concentrations below the detection limit



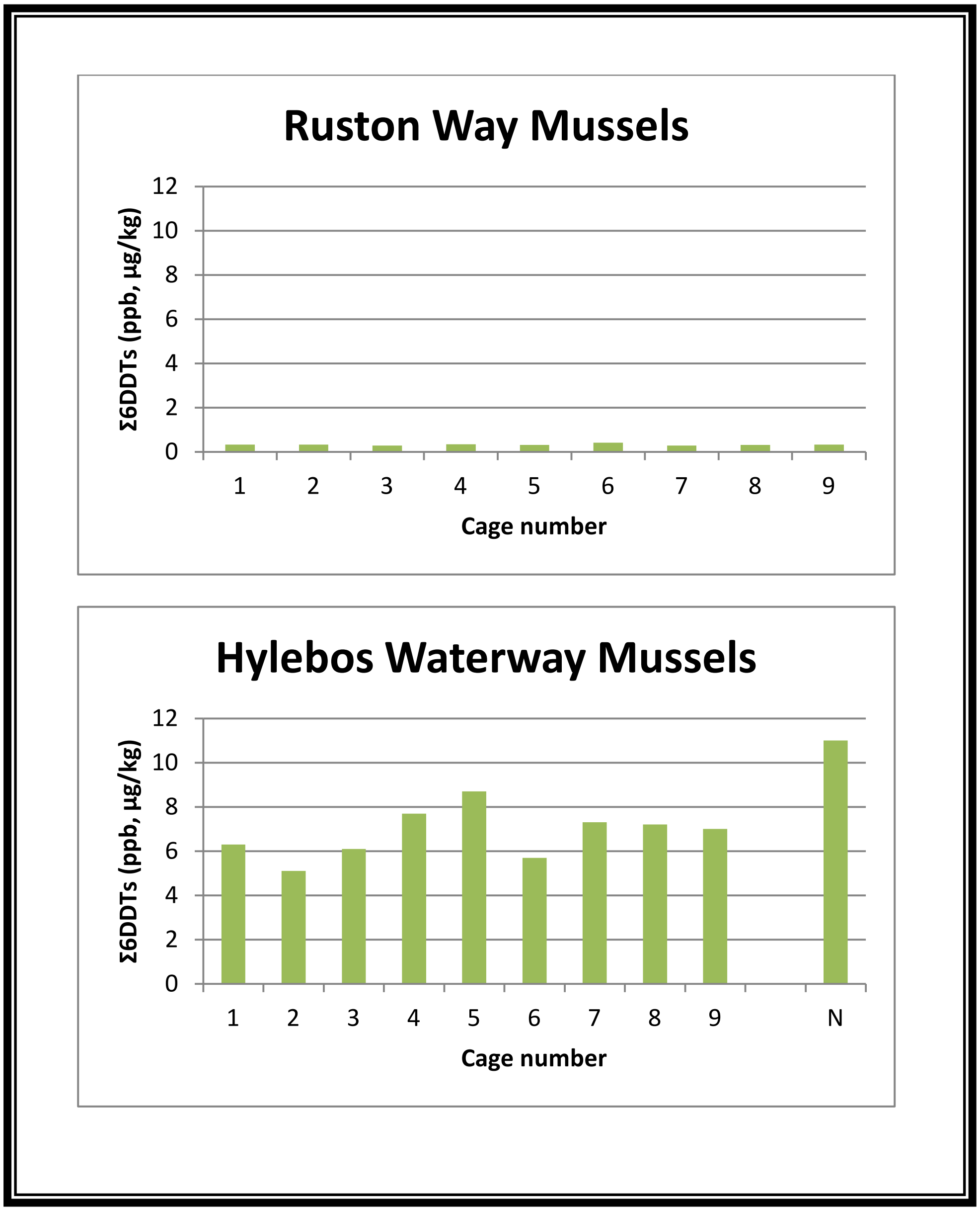
Chlordane was widely used as a pesticide, insecticide, and fumigating agent from 1948 until its ban by the EPA in 1988. It was included in products made for both crops and domestic gardens. Chlordane persists in the environment for many years after use, including in crops grown on farmland where chlordane was applied as a pesticide and in the air of homes treated with chlordane compounds during fumigation. It has a low solubility in water, allowing it to adhere to soil particles and slowly permeate into groundwater (ATSDR 1994).

Exposure to chlordane may result from ingesting foods grown in contaminated soil or foods containing a high amount of fat, and inhaling the air in/near a contaminated home. It may also be passed from a pregnant mother to a fetus through absorption in the placenta, or from a nursing mother to a child through breast milk. Health effects of exposure to chlordane include impacts to the nervous system, respiratory system, reproductive system, liver, kidneys, blood, and thyroid, as well as an increased risk of cancer (USEPA 1994).

Chlordane is currently designated a s a pollutant of concern by the EPA's Great Waters Program because of its high toxicity to animals (particularly fish) and humans, persistence in the environment, and its ability to bioaccumulate in fatty tissues (Metcalf 2002).

For this study, analysis was conducted for the eight most common analytes of chlordane. The results for the eight analytes were added together for each sample and named Sum8Chlordane. The Sum8Chlordane concentrations for the Ruston Way caged mussel samples were all below the detection limit. The Sum8Chlordane concentrations for the Hylebos Waterway caged mussel samples ranged from 0.91 ppb to 1.2 ppb and the mean concentration was 0.96 ppb. The Hylebos Waterway caged mussel mean concentration was greater than the All Sites mean of 0.69 ppb. Chlordane was not detected in any of the Penn Cove baseline mussel samples. The Hylebos native mussel sample had a much higher Sum8Chlordane result, 2.0 ppb than any other sample, including the All Sites samples. Measureable concentrations of Chlordane were found in 21% of All Sites samples but there was no detectable levels found in the Penn Cove baseline samples.

Figure 15. DDT Concentrations (ppb, µg/kg) in Ruston and Hylebos Mussels



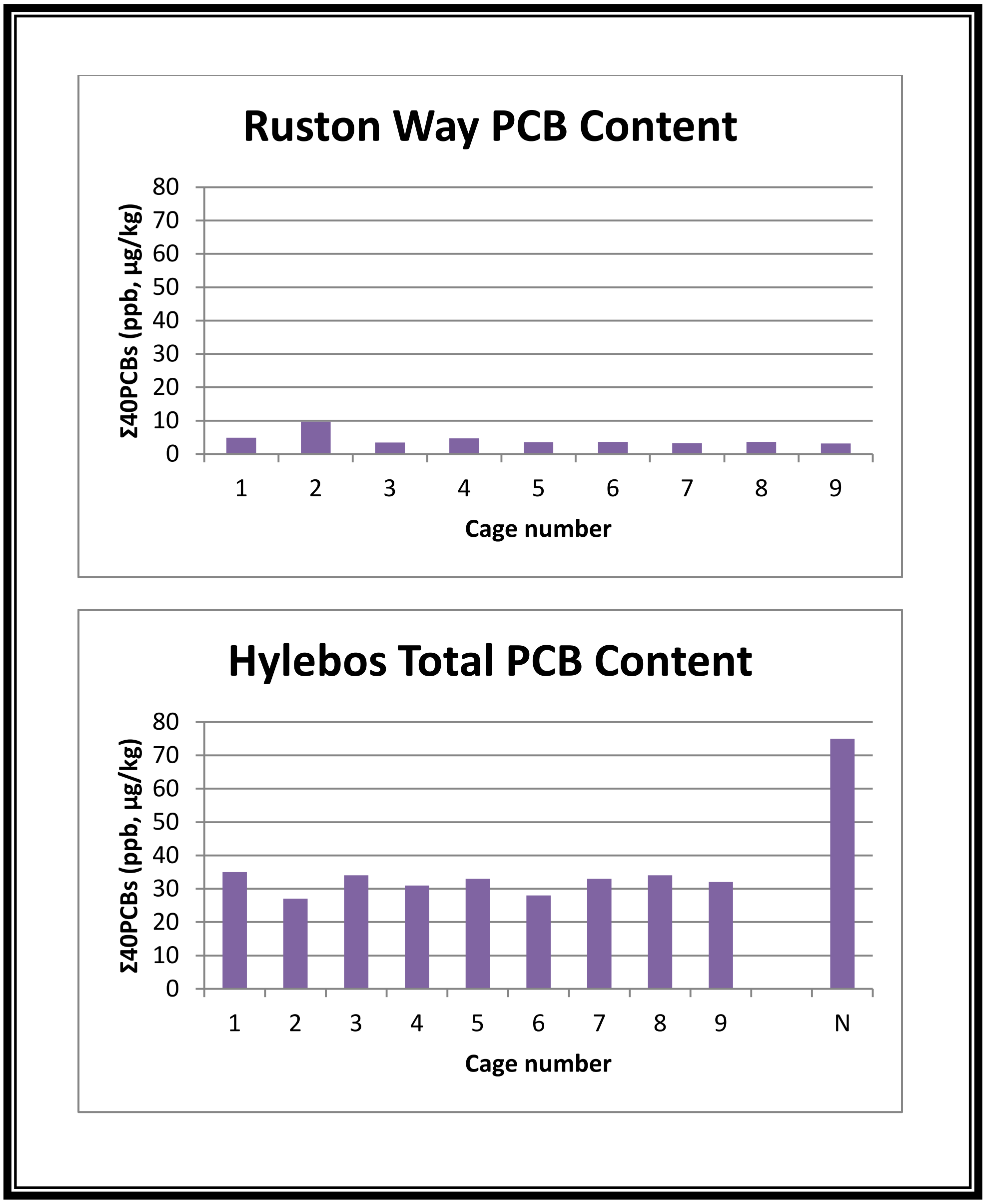
DDT (dichlorodiphenyltrichloroethane) is a colorless, crystalline, tasteless and almost odorless organochloride known for its insecticidal properties.

It is a persistent organic pollutant that is readily adsorbed to soils and sediments, which can act both as sinks and as long-term sources of exposure to terrestrial organisms (WHO 1989). Depending on conditions, its soil half life can range from 22 days to 30 years. Routes of loss and degradation include runoff, volatilization, photolysis and aerobic and anaerobic biodegradation. Due to hydrophobic properties, in aquatic ecosystems DDT and its metabolites are absorbed by aquatic organisms and adsorbed on suspended particles, leaving little DDT dissolved in the water itself. Its breakdown products and metabolites, DDE and DDD, are also highly persistent and have similar chemical and physical properties. DDT and its breakdown products are transported from warmer regions of the world to the Arctic by the phenomenon of global distillation, where they then accumulate in the region's food web (The Science and the Environment Bulletin 1999).

Because of its lipophilic properties, DDT has a high potential to bioaccumulate, especially in predatory birds (Connell, D., 1999). DDT, DDE, and DDD magnify through the food chain, with apex predators such as raptor birds concentrating more chemicals than other animals in the same environment. DDT compounds are very lipophilic and are stored mainly in body fat. DDT and DDE are very resistant to metabolism; in humans, their half-lives are 6 and up to 10 years, respectively. In the United States, these chemicals were detected in almost all human blood samples tested by the Centers for Disease Control in 2005, though their levels have sharply declined since most uses were banned in the US. Estimated dietary intake has also declined (Eskenazi, B. 2009) although FDA food tests commonly detect it.

This study analyzed for six analytes of DDT. These results were then summed for each sample to provide the Sum6DDT concentration. The Sum6DDT mean concentration for the Hylebos Waterway site was 6.8 ppb and exceeded the Ruston Way site mean of 0.33 ppb as well as the All Sites mean of 1.07 ppb and the Penn Cove baseline mean of 0.18 ppb. The Hylebos native mussel sample Sum6DDT concentration of 11 ppb was much higher than the caged mussel results. DDT was detected in 93% of the All Sites samples and 50% of the Penn Cove baseline samples.

Figure 16. Estimated Total PCB Concentrations (ppb, µg/kg) in Ruston and Hylebos Mussels



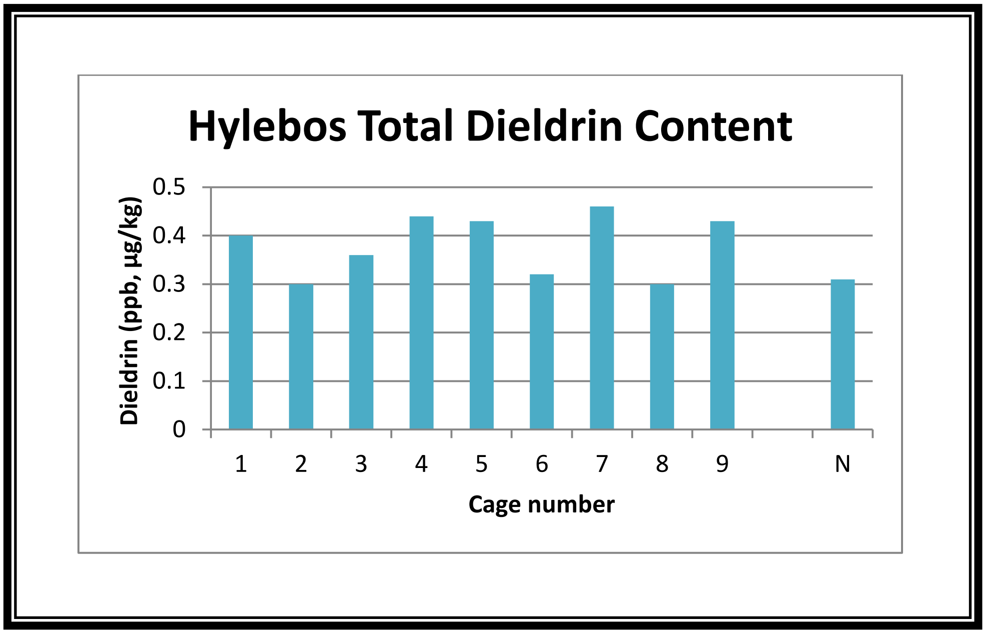
Polychlorinated biphenyls (PCBs) were widely used as coolant fluids in transformers, capacitors, and electric motors. Due to the environmental toxicity of PCBs and their classification as a persistent organic pollutant, PCB production was banned by the United States Congress in 1979 (Porta, M. 2002). PCBs are known to be carcinogenic, and are classified as endocrine disruptors and neurotoxins. They are very stable compounds and do not decompose readily. They have a half life of 8-15 years and are insoluble in water (US EPA, Jan 2013).

The manufacture of PCBs began in 1929. They were used extensively for commercial and industrial purposes because of their advantageous chemical properties (i.e.: non-flammability, chemical stability, high boiling point, water insolubility, and electrical insulation).

The environmental toxicity of PCBs is compounded by the fact that they do not readily degrade. Instead they are capable of cycling through the environment between the air, water, and soil for many years. They can also bioaccumulate in animal tissues, causing health issues with the immune system, reproductive system, nervous system, and endocrine system (US EPA, Jan 2013).

There are a large number of variants (congeners) of PCBs and this project analyzed for 40 congeners. The total concentration of PCBs is difficult to determine, given the very large number of congeners, but for this project we focused on the sum of the Estimated Total PCBs. The Estimated Total PCBs mean concentration for the Hylebos Waterway site was 32 ppb and exceeded the Ruston Way mean of 4.4 ppb and the All Sites mean of 7.47 ppb, as well as the Penn cove baseline mean of 4.0 ppb. The Estimated Total PCBs result for the Hylebos native mussel sample was much higher than any of the caged mussel results, with a concentration of 75 ppb. Estimated Total PCBs were found in 100% of the All Sites samples and Penn Cove baseline samples.

Figure 17. Total *Dieldrin Concentrations (ppb, µg/kg) in Hylebos Mussels
All Ruston Way mussel samples were non-detect for Dieldren



Dieldrin is a chlorinated hydrocarbon originally produced in 1948 as an insecticide. Dieldrin is closely related to aldrin, which reacts further to form dieldrin. Aldrin is not toxic to insects; it is oxidized in the insect to form dieldrin which is the active compound.

Originally developed as an alternative to DDT, dieldrin proved to be a highly effective insecticide and was very widely used during the 1950s to early 1970s.

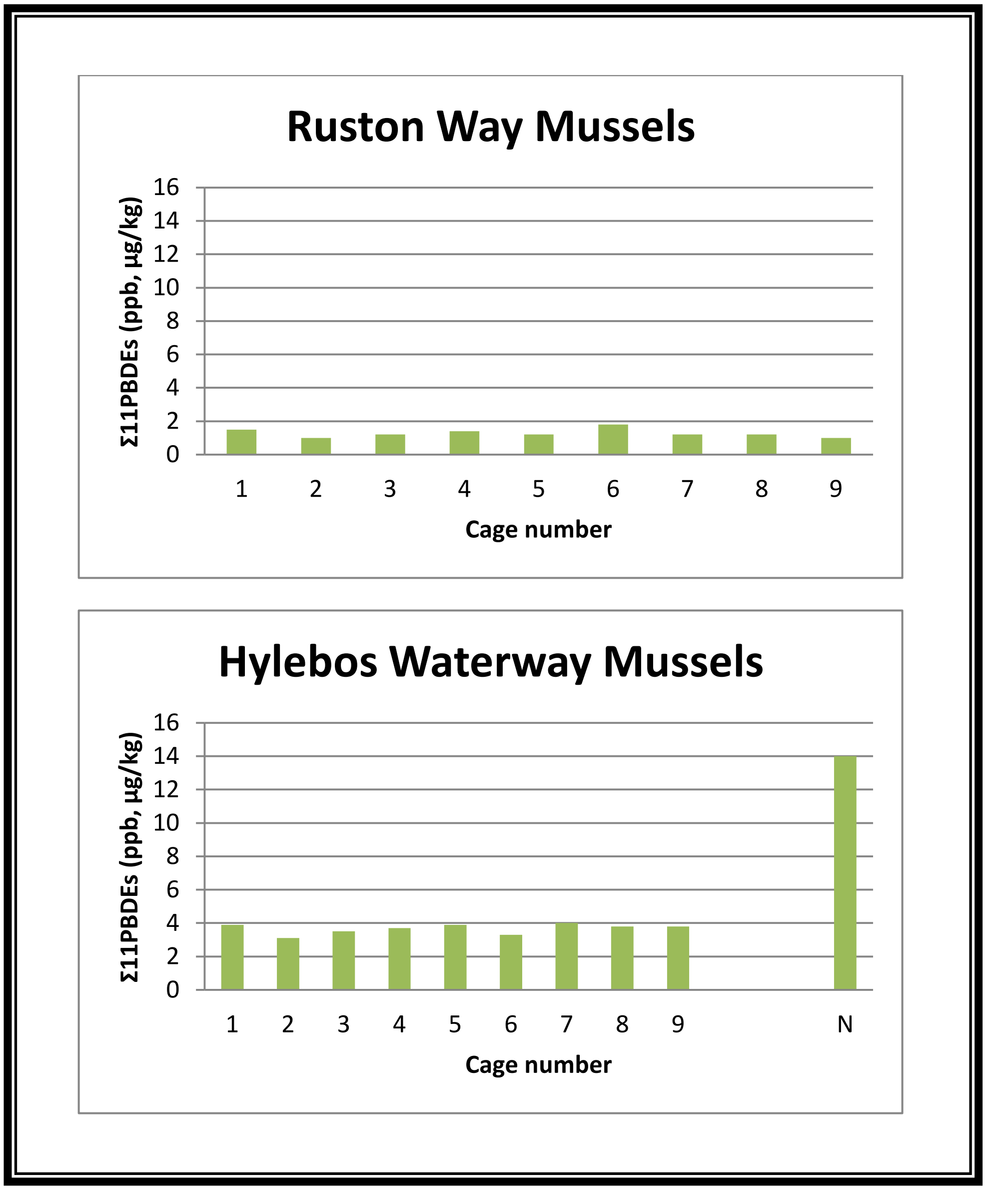
Dieldrin does not break down easily, making it an extremely persistent organic pollutant. It also tends to biomagnify as it is passed along the food chain. Long-term exposure has proven toxic to a very wide range of animals including humans, far greater than just the original insect targets. For this reason it is now banned in most of the world.

Health problems such as Parkinson's, breast cancer, and immune, reproductive, and nervous system damage, have been linked to dieldrin exposure (Jubb 1975; S. Kegley 2007).

All of the Ruston Way samples had Dieldrin concentrations below the detection limit. Results from the Hylebos Waterway caged mussel samples ranged from 0.3 ppb to 0.46 ppb, with a mean of 0.38 ppb. The Hylebos native mussel sample had a Dieldrin concentration of 0.31 ppb.

The Hylebos Waterway caged mussel results fall within the All Sites Dieldrin range of 0.13 ppb to 0.46 ppb. Dieldrin was detected in 17% of the All Sites samples and was not detected in any of the Penn Cove baseline samples.

Figure 18. Total PBDE Concentrations (ppb, µg/kg) in Ruston and Hylebos Mussels



Polybrominated diphenyl ethers or PBDEs, are organobromine compounds that are used as flame retardants. Like other brominated flame retardants, PBDEs have been used in a wide array of products, including building materials, electronics, furnishings, motor vehicles, airplanes, plastics, polyurethane foams, and textiles.

People are exposed to low levels of PBDEs through ingestion of food and by inhalation. PBDEs can also bioaccumulate in blood, breast milk, and fat tissues (Stapelton, Jun 15, 2011).

This study analyzed for 11 PBDE congeners. For this report, the 11 congeners for each sample were summed and reported as $\sum$11PBDEs. The Hylebos Waterway caged mussel samples had a mean $\sum$11 PBDEs concentration of 3.7 ppb, which was higher than the Ruston Way mean of 1.28 ppb, the All Sites mean of 1.38 ppb and the Penn Cove baseline mean of 0.44 ppb. The Hylebos native mussel sample had a $\sum$11PBDEs concentration of 14.0 ppb, which was much higher than all other samples. PBDEs were detected in 95% of the All Sites samples and 83% of the Penn Cove baseline samples.

Discussion

The concentrations of contaminants in the mussel tissue at both sites may be attributed in part to many of the current and historical activities adjacent to the water, and possibly to the discharge of contaminants from upland activities through stormwater outfalls.

Historically, the Hylebos Waterway was the site of an herbicide manufacturing plant which used arsenic. The area has undergone partial remediation for arsenic and a variety of other contaminants. The Ruston Way site is located less than a mile away from the former location of the Asarco Smelter. Many of the areas in and around the City of Tacoma are still contaminated with arsenic, lead, copper, zinc and cadmium. Expectations were that the Ruston Way site would follow suit.

Metals were detected in all samples, including the All Site samples and the Penn Cove baseline samples. Given the close proximity to the former Asarco Smelter some of the Ruston Way metals results were lower than originally anticipated. Although, the mean concentration for most metals was greater in mussels from the Hylebos Waterway site, with the exception of cadmium and lead. The Hylebos native mussel sample had metal concentrations higher than either the Hylebos or Ruston Way caged mussel samples, with the exception of arsenic and cadmium. The Hylebos native mussel concentrations for zinc and copper were almost twice as high as the mean concentrations for the Hylebos Waterway caged mussels.

When comparing the two sites, the Hylebos Waterway site had consistently higher organic contaminant results than the Ruston Way site and in some cases the results were two to ten times the concentrations of the Ruston Way caged mussels. For a few contaminants (HCH, Hexachlorobenzene, and Mirex), Ruston Way caged mussel results exceeded those from the Hylebos Waterway caged mussels, though they were still comparable or close to the Hylebos Waterway caged mussel results.

The organic contaminant results from the Hylebos native mussel sample in almost all cases exceeded the results for the entire study. Only two contaminants detected in the native mussels, Low Molecular Weight PAH (SUM22LMWPAH's) and HCH (SUM3HCHs), had results that fell within the same range of the caged Hylebos mussels.

The study results discussed in this report will be analyzed in combination with all other study sites by Washington State Department of Fish and Wildlife.

Conclusions

The results from this study showed a difference between the Hylebos Waterway site, which is in an industrial area, and the Ruston Way site, which is in a commercial/residential area. This indicates that land use may be an important influence on contaminant loading to mussels in the intertidal zone.

The results showed some variability in contaminant concentrations between cages within each site but this variability was more subtle than variability between the two sites.

In general the contaminant results for the Hylebos native mussel samples were higher than the results for the caged mussels for both the Hylebos Waterway and Ruston Way. Various reasons could account for the relatively high native mussel results and may have important implications if caged mussels are anticipated to be used for the assessment of land use impacts on near shore biota. Although these data are very limited, caged mussels may significantly underestimate contaminant loading to the near shore biota. Further monitoring with replicate cages and native mussels is recommended to better assess both intra-site variability and variability between caged mussels and native mussels.

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Appendix

Metals and Organics Results

Table A. Metals (wet weight) data

Wet Weight			Total Solids	Mercury, Total, CVAA	Arsenic, Total, ICP-MS	Cadmium, Total, ICP-MS	Copper, Total, ICP-MS	Lead, Total, ICP-MS	Zinc, Total, ICP-MS
LOCATOR	**PROJECT**	**SAMPLE**	**%**	**mg/Kg**	**mg/Kg**	**mg/Kg**	**mg/Kg**	**mg/Kg**	**mg/Kg**
13CPS_HYW1-MTW01B	421191-100	L57597-89	15.20	0.00	0.96	0.28	1.75	0.06	18.10
13CPS_HYW2-MTW01B	421191-100	L57597-90	14.00	0.00	0.84	0.30	1.08	0.04	15.10
13CPS_HYW3-MTW01B	421191-100	L57597-91	15.30	0.00	0.91	0.27	0.98	0.04	13.00
13CPS_HYW4-MTW01B	421191-100	L57597-92	15.80	0.01	1.12	0.29	1.28	0.04	15.70
13CPS_HYW5-MTW01B	421191-100	L57597-93	14.90	0.01	1.00	0.31	0.93	0.04	17.70
13CPS_HYW6-MTW01B	421191-100	L57597-94	14.10	0.00	0.90	0.33	1.02	0.06	18.40
13CPS_HYW7-MTW01B	421191-100	L57597-95	15.90	0.01	0.92	0.34	0.98	0.04	17.50
13CPS_HYW8-MTW01B	421191-100	L57597-96	14.70	0.01	0.94	0.30	0.96	0.04	14.90
13CPS_HYW9-MTW01B	421191-100	L57597-97	15.00	0.00	0.93	0.27	1.12	0.04	16.00
13CPS_RW1-MTW01B	421191-100	L57597-98	15.30	0.01	0.82	0.32	0.83	0.07	12.80
13CPS_RW2-MTW01B	421191-100	L57597-99	15.50	0.01	0.89	0.36	0.85	0.09	13.50
13CPS_RW3-MTW01B	421191-100	L57597-100	14.20	0.00	0.79	0.39	0.99	0.07	13.20
13CPS_RW4-MTW01B	421191-100	L57597-101	15.40	0.01	0.82	0.37	0.96	0.08	12.60
13CPS_RW5-MTW01B	421191-100	L57597-102	15.00	0.00	0.84	0.37	1.03	0.08	10.70
13CPS_RW6-MTW01B	421191-100	L57597-103	16.10	0.01	0.83	0.34	1.09	0.09	12.30
13CPS_RW7-MTW01B	421191-100	L57597-104	15.40	0.00	0.80	0.35	1.04	0.07	10.60
13CPS_RW8-MTW01B	421191-100	L57597-105	16.30	0.01	0.84	0.40	0.89	0.06	11.70
13CPS_RW9-MTW01B	421191-100	L57597-106	14.70	0.01	0.78	0.35	0.98	0.06	12.40
13CPS_HYW2-MXW01B	421191-100	L57597-111	11.80	0.01	0.82	0.23	1.64	0.09	31.80
* Not converted to dry weight basis									
If a parameter/analyze appears twice in the column header, it implies that they were analyzed by two different method codes									

Table B. Organics (wet weight) data

Wet Weight	Σ22LMWPAHs	Σ20HMWPAHs	Σ42PAHs	Hexachlorobenzene	Σ3HCHs	Σ8Chlordanes	Σ6DDTs	EstimatedTotalPCBs	Dieldrin	Mirex	ΣPBDEs
SiteID	µg/kg	µg/kg	µg/kg	µg/kg	µg/kg	µg/kg	µg/kg	µg/kg	µg/kg	µg/kg	µg/kg
CB_HYW1	82	137	219	0.27	< 0.22	0.91	6.3	35	0.4	< 0.22	3.9
CB_HYW2	78	118	196	<0.2	< 0.2	0.75	5.1	27	0.3	< 0.2	3.1
CB_HYW3	80	120	200	0.23	< 0.16	0.89	6.1	34	0.36	< 0.16	3.5
CB_HYW4	95	141	237	0.23	< 0.16	1	7.7	31	0.44	< 0.16	3.7
CB_HYW5	102	157	259	0.22	< 0.14	1.2	8.7	33	0.43	< 0.14	3.9
CB_HYW6	81	120	201	0.21	< 0.15	0.8	5.7	28	0.32	< 0.15	3.3
CB_HYW7	113	175	288	0.21	< 0.18	1	7.3	33	0.46	< 0.18	4
CB_HYW8	101	163	264	0.23	< 0.19	1	7.2	34	0.3	< 0.19	3.8
CB_HYW9	95	146	241	0.22	< 0.2	1.1	7	32	0.43	< 0.2	3.8
CB_HYWN	100	190	290	0.19	<0.14	2	11	75	0.31	0.61	14
CB_RW1	30	40	70	< 0.17	< 0.17	< 0.17	0.33	4.9	< 0.17	< 0.17	1.5
CB_RW2	46	55	101	< 0.24	< 0.24	< 0.25	0.33	9.7	< 0.24	< 0.24	1
CB_RW3	19	27	47	< 0.23	< 0.23	< 0.23	0.29	3.4	< 0.23	< 0.23	1.2
CB_RW4	33	43	76	< 0.19	< 0.19	< 0.19	0.34	4.7	< 0.18	< 0.19	1.4
CB_RW5	48	57	105	< 0.24	< 0.24	< 0.24	0.32	3.5	< 0.23	< 0.24	1.2
CB_RW6	63	71	134	< 0.26	< 0.26	0.29	0.41	3.6	< 0.26	< 0.26	1.8
CB_RW7	47	57	104	< 0.19	< 0.19	< 0.19	0.28	3.3	< 0.19	< 0.19	1.2
CB_RW8	40	44	84	< 0.21	< 0.21	< 0.21	0.32	3.6	< 0.21	< 0.21	1.2
CB_RW9	30	37	67	< 0.27	< 0.27	< 0.27	0.33	3.2	< 0.27	< 0.27	0.99

Values proceeded by < are less than the detection limit.