Puget Sound Ecosystem Monitoring Program (PSEMP)

Toxic Contaminants in Puget Sound's Nearshore Biota: A Large-Scale Synoptic Survey Using Transplanted Mussels (*Mytilus trossulus*)

Final Report
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Jennifer A. Lanksbury, Laurie A. Niewolny, Andrea J. Carey and James E. West

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EXECUTIVE SUMMARY

In the winter of 2012-13 the Washington Department of Fish and Wildlife, with the help of citizen science volunteers, other agencies, tribes, and non-governmental organizations, conducted the first synoptic, Puget Sound-wide assessment of toxic contaminants in nearshore biota. This project was funded by EPA's National Estuary Program (NEP) in support of Washington State's Action Agenda and their goal of restoring the health of Puget Sound. The Washington Department of Fish and Wildlife and Department of Natural Resources awarded this grant in their role as Lead Organization for NEP's Marine and Nearshore Protection and Restoration. This project was funded as a cross-cutting study, which drew together concepts related to three NEP-supported focal efforts in the Puget Sound: (1) Toxics and Nutrients, (2) Marine and Nearshore Protection and Restoration, and (3) Watershed Protection and Restoration. This study focused on toxic contaminants generated primarily from terrestrial sources, and conveyed to Puget Sound nearshore habitats via stormwater and other hydraulic watershed processes.

In this study we used native mussels (*Mytilus trossulus*) as indicators of the degree of contamination of nearshore habitats. We transplanted relatively uncontaminated mussels from an aquaculture source to 108 locations along the Salish Sea shoreline, covering a broad range of upland land-use types from rural to highly urban. At the end of the study we determined three biological endpoints (mortality, growth and condition index) and measured the concentration of several major contaminant classes in mussels: polycyclic aromatic hydrocarbons (PAHs), polychlorinated biphenyls (PCBs), polybrominated diphenyl ethers (PBDEs, or flame retardants), chlorinated pesticides (including dichlorodiphenyltrichloroethane compounds, or DDTs) and six metals (lead, copper, zinc, mercury, arsenic, cadmium).

Overall, PAHs, PCBs, PBDEs, and DDTs were the most abundant organic contaminants measured in this study. PAHs and PCBs were detected in mussels from every site, and highest concentrations were observed in four of Puget Sound's most urbanized embayments (466 - 5030 ng/g dry weight (dw) for Σ_{42}PAHs, and 38 - 216 ng/g dw for total PCBs in mussels from Elliott Bay, Salmon Bay, Commencement Bay, and Sinclair Inlet). Although lower in overall concentration, PBDEs and DDTs followed a similar pattern. In addition, although PCBs were elevated mainly along urbanized shorelines, PAHs were elevated in mussels from some non-urban shorelines (some near marinas or ferry terminals). The other organic contaminants were detected in mussels at fewer than 22% of study sites, and at low levels.

We observed significant positive correlations between both our proxies of nearshore watershed land development (impervious surface and road area), and levels of PAHs, PCBs, PBDEs, and DDTs. Variability in contaminant concentration increased exponentially with increasing impervious surface (or road area), suggesting other, unmeasured landscape factors may more fully explain the variation in mussel contaminant concentrations. These factors may include proximity to point sources (e.g., outfalls) or focal non-point sources (e.g., marinas or ferry terminals).

PAH analyte pattern analysis suggested the majority of mussel sites were dominated by pyrogenic (i.e. combustion) sources; however, atypical patterns at a few locations (Salmon Bay, Bremerton Shipyard-Charleston Beach, Hylebos Waterway, and the Thea Foss Waterway) suggested petroleum sources may be contributing a larger proportion of PAHs to the mussels in those areas. PCB congener-ratio analysis suggested

urban embayments in the Central Puget Sound (Elliott Bay, Commencement Bay, and Sinclair Inlets) are sources of PCBs for non-urban areas. The PCB pattern in mussels became “lighter” with distance from urban areas; that is, more highly chlorinated PCB congeners with greater molecular weight tended to be less abundant in mussel tissue with increasing distance from urban shorelines. Lighter congeners tend to migrate faster through the environment than heavier congeners.

Although the condition index of mussels declined at 72% of the study sites, condition index was not linked to impervious surface or road area; this decline was likely the result of natural processes related to normal declines in food supply and slowing of growth during winter months. However, there was a weak positive correlation between mortality and both impervious surface and road area, suggesting greater survival of mussels with decreasing contamination. Growth was not linked to either factor, however the short deployment time (60 days) and season (winter) probably hampered our ability to measure growth adequately.

All six metals were found in mussels from all the study sites, though their concentrations were relatively low and did not vary greatly from baseline (starting) values. There was a weak, positive relationship for lead, with impervious surface and road area, weaker relationships with copper, and a weak relationship between zinc and impervious surface. There was no link between mercury, arsenic, or cadmium with either factor, suggesting the concentration of these metals in mussels is not predictable from levels of impervious surface or road area.

Wild and transplanted mussels sampled simultaneously from six sites had similar concentrations of organic contaminants and metals, suggesting that caged mussels behaved similarly to wild-growing mussels. However this study was not designed to make such a comparison; sample size for these pairings was low and important factors such as tidal elevation were uncontrolled, so caution should be exercised when comparing contaminant levels between the two types.

These findings suggest toxic contaminants are entering the nearshore food web of the Salish Sea, especially along shorelines adjacent to highly urbanized areas. Some contaminants such as PAHs exhibited a wider, less predictable distribution, than the other organic chemicals, perhaps related to sources that may occur on rural or less developed landscapes (e.g., roadways, creosote pilings, marinas, and ferry terminals). We recommend that Washington State develop a long-term, regional, nearshore sampling program using caged mussels as a sentinel species to monitor status and trends of contaminants in nearshore biota. Success of such a large-scale field-intensive study is predicated on participation by citizen science volunteers to conduct the field work, and by partner groups interested in monitoring pollution in their nearshore areas to maximize spatial coverage in the Sound.

1 INTRODUCTION

Toxic contaminants enter Puget Sound from a variety of pathways including (a) non-point sources such as surface water runoff, groundwater releases, and air deposition, (b) focal non-point sources, such as marinas and ferry terminals, and (c) point sources such as discharges from stormwater outfalls (SWOs), wastewater treatment plants (WWTPs), combined sewer overflows (CSOs), and permitted industry, construction sites and boatyards. In addition, Puget Sound has been subject to contamination from a number of now-banned persistent bioaccumulative and toxic chemicals including polychlorinated biphenyls (PCBs) and dichlorodiphenyl-trichloroethanes (DDTs). A reservoir of these "legacy" contaminants persists in the sediments (Long et al., 2005) and the biota of Puget Sound (O'Neill and West, 2009; Ross et al., 2000; West et al., 2011a; West et al., 2011b; West et al., 2001; West et al., 2008). Although the manufacture of PCBs in the United States was banned in 1979, PCBs are still found in significant amounts in the Puget Sound basin (e.g. in building paints and caulks) and they continue to find their way into the stormwater (EnviroVision Corporation et al., 2008; Hart Crowser, 2007; Herrera Environmental Consultants Inc., 2009; Science Applications International Corporation, 2011) of the Puget Sound.

These toxic substances can cause harm to aquatic organisms and pose a risk to the people who consume them. Monitoring pollutants in Puget Sound is a critical component of tracking its recovery and informing best management practices for remediation efforts (Puget Sound Partnership, 2010; Puget Sound Partnership, 2012-14). However, an understanding of the extent and magnitude of contaminants in nearshore biota has long been recognized as an information gap in the Puget Sound. Understanding the sources, fate and transport of contaminants in the Puget Sound nearshore marine food web, and what impacts they have on biota, would improve our ability to make cost-effective decisions to mitigate the harm pollution causes in the nearshore environment of the greater Puget Sound.

The national Mussel Watch project, run by the National Oceanic and Atmospheric Administration's (NOAA) Coastal Ocean Assessments, Status, and Trends (COAST) program, has tracked chemical and biological contaminant trends in bivalves (mussels and oysters) across the U.S., using wild mussels (*Mytilus* spp.) in Washington State (Apeti et al., 2009a; Center for Coastal Monitoring and Assessment, 2014). Mussel Watch was designed on a national scale to monitor "the environmental quality of our nation's estuarine and coastal waters" and "provide coastal managers with national context" to measure local and regional environmental conditions (Center for Coastal Monitoring and Assessment, 2014). NOAA's historical Mussel Watch data from 1986 to 2007 (Kimbrough et al., 2008), and more recent Mussel Watch data (NOAA's Mussel Watch - unpublished data from 2009 - 2012), indicate a strong link between urbanization and certain persistent organic pollutants in nearshore areas of Puget Sound. These data have been useful for broadly characterizing ambient contaminant conditions in Puget Sound's nearshore biota (Puget Sound Action Team, 2007). However, NOAA's Mussel Watch program selected its original monitoring sites to characterize average conditions for the whole Puget Sound and as such avoided suspected point-sources or "hot spots" of toxic chemicals. Because of this study design, data from the Mussel Watch sites alone are insufficient to answer regional questions regarding the fate, transport, and effects of chemical contaminants in Puget Sound's nearshore urbanized waters (Lanksbury and West, 2011).

In 2009 NOAA's Mussel Watch program requested help in sampling their mussel monitoring sites in Washington State. In response, the Washington Department of Fish and Wildlife's (WDFW) Puget Sound Ecosystem Monitoring Program (PSEMP) teamed with the Snohomish County Marine Resources Committee (MRC), the Snohomish County Public Works-Surface Water Management, Washington Sea Grant, and citizen science volunteers to conduct field-sampling for the 2009/10 NOAA Mussel Watch season in Washington (Lanksbury et al., 2010). Three new "pilot" locations were added to the list of NOAA's Mussel Watch monitoring sites in Washington State that year, to evaluate contaminant loads in mussels from highly urbanized sites (Elliott Bay) and less contaminated reference sites (Nisqually Reach). These additional monitoring sites were sampled again in 2011/12 and have provided PSEMP with more detail about sources of nearshore contaminants on a regional/local scale.

Due to the success of the partnership between NOAA's Mussel Watch and Washington State, several state and county organizations responsible for managing regional stormwater and water quality (e.g. Washington Department of Ecology, various County water quality agencies, several tribes), as well as a number of volunteer groups (County MRCs, citizen science volunteers, and other non-governmental organizations (NGOs)), expressed the desire to see PSEMP build upon NOAA's Mussel Watch program and put together a larger network of mussel monitoring sites in Washington State, with the idea that monitoring could be accomplished through coordination of the various regional interest groups. Around this time the Washington Department of Ecology (Ecology) began to develop plans for a Regional Stormwater Monitoring Program in Puget Sound for permittees under the National Pollutant Discharge Elimination System (NPDES), to which they sought to include a mussel component (Washington State Department of Ecology, 2013). Thus the vision emerged of a Washington State mussel monitoring program similar to NOAA's Mussel Watch, but with a greater number of sites sponsored by a network of agencies and other groups interested in contaminant monitoring on a more local (or regional) level.

As a result of these interests, PSEMP sought a grant from the Environmental Protection Agency's National Estuary Program (NEP) for Puget Sound Recovery to fund a one-time, pilot project aimed at developing an expanded network of sites for evaluating toxics in nearshore biota (mussels). This *Mussel Watch Pilot Expansion* (MWPE) project was designed to provide a broad-scale, synoptic (one season) assessment of toxic contaminants in the nearshore biota of the greater Puget Sound, greatly expanding spatial coverage of previous mussel monitoring efforts, and testing the efficacy of using citizen science volunteers to conduct a large, spatially expansive field study in a short period of time.

1.1 Project Goals

The Mussel Watch Pilot Expansion project was designed to be a qualitative reconnaissance survey. Our goal was to provide data on the current extent and magnitude of contamination in the nearshore environment of the greater Puget Sound, across a wide range of upland land-use types (including rural, undeveloped, agricultural, urban, and industrial areas), and to provide recommendations towards developing a long-term, regional nearshore monitoring plan for Washington State. The objectives of this survey were to:

1. Evaluate the geographic extent of chemical contamination in shoreline biota, using Pacific blue mussels (*Mytilus trossulus*) as the primary indicator organism,
2. Measure the magnitude of contamination where it occurs,

3. Compare contamination patterns in mussels with adjacent shoreline land-use, covering a wide range of land-use types,
4. Analyze patterns of polycyclic aromatic hydrocarbon (PAHs) and PCBs to help infer potential sources, and
5. Provide recommendations for long-term status and trends monitoring.

A companion study, carried out by the Washington Department of Natural Resources (WDNR), investigated contaminants in eelgrass at a select number of the mussel sites used in this study (Gaeckle, 2013). One of the original goals of this study was to compare contaminant uptake between mussels and plants (eelgrass), using the results from the WDNR study. To date that study has not been completed. Thus a comparison between mussel and eelgrass contaminant concentrations will occur when eelgrass tissue analyses are completed and the final report for that study is made available.

1.2 Background

1.2.1 Mussels as Biomonitors

Mussels and other sessile, filter-feeding bivalves have been used to monitor contaminant conditions in nearshore biota since the late 1970s (Martin and Severeid, 1984; O'Connor and Lauenstein, 2006). One of the reasons mussels are ideal for contaminant monitoring is they are widespread and sedentary, which makes them easy to find and collect (ASTM International, 2007; Gosling, 1992). Mussels are exposed to both particulate and dissolved forms of pollution and accumulate chemical contaminants in their soft tissues via multiple pathways, including food, sediments and water (ASTM International, 2007). They absorb dissolved contaminants through ingestion of contaminated food and suspended sediments or directly across their gills. In fact, the gills and digestive gland are generally the most important target tissues for metal bioconcentration in mussels (Gagnon et al., 2006; Odźak et al., 1994; Roesijadi et al., 1984). Because they absorb chemicals from water and sediment, mussels are capable of integrating exposure from both the water column and benthic sources (ASTM International, 2007; Baumard et al., 1998).

One of the other benefits of using mussels for tracking pollutants is their low biotransformation abilities (Flemming et al., 2008). Mussel digestive systems are relatively primitive and lack a functioning liver. Their inability to metabolize most of the organic contaminants they absorb causes mussels to accumulate those contaminants in their tissues, so they reflect the profile of bioavailable pollution in their local environments (ASTM International, 2007; Baumard et al., 1998). Polycyclic aromatic hydrocarbon (PAH) profiles found in mussel and sediment samples taken from the same area are often similar (Guinan et al., 2001) and mussels have been used successfully to identify nearshore contamination resulting from major oil spills (Apeti et al., 2013; Babcock et al., 1996; Carls et al., 2001; Neff and Burns, 1996).

Because mussels concentrate environmental pollutants they can be used to measure contaminant conditions in areas where the pollutants are too low to measure in water. For instance, Sundt et al. (2011) demonstrated the effectiveness of mussels as sentinel organisms in monitoring of North Sea offshore oil drilling platforms, where PAH compounds present in small amounts in the seawater were bioaccumulated by the mussels. In other studies, polychlorinated biphenyls (PCBs) and PAHs were found in measurable amounts in the tissues of transplanted mussels when concentrations were below the limits of detection in local seawater (Green et al., 1986; Salazar et al., 1995; Salazar and Salazar, 1995; Short and Rounds, 1993). Generally speaking, contamination levels in mussel tissues are higher closer to pollution sources and decrease with distance from

those sources (Baumard et al., 1999a; Baumard et al., 1999b). This was the case in the North Sea, where mussel PAH concentration followed an increasing gradient approaching the oil drilling and petroleum production platforms (Sundt et al., 2011). For this reason mussels have successfully been employed in past gradient studies (ASTM International, 2007; Salazar et al., 1995).

As mussels are exposed to pollution the concentration of contaminants in their tissues varies until they reach a steady state with the environment. This environmental equilibrium is achieved through a balance of uptake (from intake of water, sediment and food) and depuration (excretion) of biological wastes, sediment and contaminants, and it fluctuates with the bioavailability of contaminants in the environment and direct exposure to chemicals (Baumard et al., 1998; Roesijadi et al., 1984). Because mussels are sedentary and they do not metabolize most organic contaminants they typically reflect local contaminant conditions. For example, mussels exposed to petroleum contamination in harbors have exhibited PAH profiles that clearly reflect that of petroleum (Baumard et al., 1999b), while mussels taken from creosote pilings have been shown to reflect a creosote-PAH signal (Dunn and Stich, 1975; Hyötyläinen et al., 2002). Baumard et al. (1999a) showed that mussels accumulate water-soluble, lower molecular weight PAHs to a greater extent when they are near the air-water interface in clear, low turbidity waters (where lower molecular weight PAHs are found), while mussels located close to the sediment or in highly turbid water tended to be enriched with less water soluble, higher molecular weight PAHs, a clear reflection of the sediment-associated PAH fraction. This distinction is not always clear however; storms can re-suspend benthic sediments, increasing the amount of organic contaminants and metals taken up by mussels and enriching them with high molecular weight PAHs which would otherwise have been sequestered in benthic sediment (Stella et al., 2002).

Finally, the use of transplanted mussels allows for the measurement of biological endpoints including survival, growth, and reproduction (gonad development) (ASTM International, 2007). Reduced mussel growth has been associated with a variety of contaminants in both laboratory and field studies (Salazar and Salazar, 1991; Stephenson et al., 1986; Strömgren, 1982; Strömgren, 1987; Valkirs et al., 1991; Widdows et al., 1995; Widdows et al., 2002; Widdows et al., 1997). In addition, biomarkers are utilized worldwide in biomonitoring programs using mussels (Dagnino et al., 2007). Some useful biomarkers include lysosomal membrane stability, neutral lipid and lipofuscin lysosomal content, DNA damage, catalase activity, metallothionein content, acetylcholinesterase and glutathione transferase activities, lysosome/cytoplasm volume ratio, and stress on stress response (e.g. reduction of survival in air), among others (ASTM International, 2007; Dagnino et al., 2007; Gagné et al., 2001; Solé et al., 1996; Viarengo et al., 1995). For instance, Gagnon et al. (2006) found an estrogenic response (higher vitellogenin-like proteins) in gonads from mussels living near a municipal wastewater plume. Interestingly, a number of studies examining biomarkers have shown that mussel populations can adapt to elevated levels of pollution (Acker et al., 2005; Da Ros and Nesto, 2005; Large et al., 2002; Rank et al., 2007; Regoli and Principato, 1995). When this is the case, the use of transplanted mussels over indigenous mussels, especially when measuring biological endpoints and biomarkers, may avoid bias resulting from adaptation related to previous contaminant exposures (Dagnino et al., 2007; Rank et al., 2007).

1.2.2 Other Mussel Monitoring Programs

There are a number of monitoring programs within the U.S. and around the world that have utilized mussels for contaminant monitoring. As mentioned earlier the NOAA's Mussel Watch relies on mussels and oysters to monitor spatial and temporal trends of contaminant concentrations in coastal and estuarine regions of the U.S.

(Center for Coastal Monitoring and Assessment, 2014). Begun in 1986, Mussel Watch is the longest running, continuous contaminant monitoring program in U.S. coastal waters (including the Great Lakes). The long-term data from this program have revealed national, regional and local trends in coastal contamination and have helped characterize the environmental impact of extreme events, including Hurricane Katrina and the Deepwater Horizon Oil Spill in the Gulf of Mexico (Apeti et al., 2013; Hunt and Slone, 2010; Kimbrough et al., 2009; Kimbrough et al., 2008; O'Connor and Lauenstein, 2006).

In 1995, Ecology began collecting bay mussels (*Mytilus trossulus*) as part of their yearly Washington State Pesticide Monitoring Program, started in 1992. They collected mussels from five sites in the Puget Sound and one site in the Columbia River and found 20 pesticides and PCBs in the mussels. DDTs and PCBs were found in mussels from all the sites, and the largest number and highest concentrations of contaminants occurred in mussels from the Commencement Bay's Hylebos Waterway, and the lowest levels occurred in Padilla Bay (Johnson and Davis 1996).

Beginning in 2007 the Snohomish County MRC built on NOAA's Mussel Watch monitoring framework in the Puget Sound to establish nine more sites in their county, two of which were funded and monitored by the Stillaguamish Tribe and one by the Tulalip Tribes (Snohomish County Marine Resources Committee, 2011). The Snohomish County MRC continues to monitor contaminants in indigenous mussels to determine whether concentrations in the County's nearshore merit action. Findings from their monitoring serve as the basis for recommendations to the Snohomish County Council and Executive Board (Whitney et al., 2011).

Also in Washington the ENVironmental inVESTment (ENVVEST) project, a cooperative partnership to improve the environmental quality of Sinclair and Dyes Inlets, is being conducted by the Puget Sound Naval Shipyard & Intermediate Maintenance Facility (PSNS&IMF), the Environmental Protection Agency, Ecology, and local stakeholders. As part of the studies conducted for ENVVEST, mussels were used to characterize nearshore contaminant levels in Sinclair and Dyes Inlets (ENVVEST, 2006; Johnston et al., 2007). In addition to collecting indigenous mussels from a number of sites in cooperation with NOAA's Mussel Watch Program, ENVVEST scientists also transplanted mussels (*Mytilus galloprovincialis*) to locations adjacent to the Bremerton Shipyard and at reference locations within Sinclair and Dyes Inlets in the summer of 2005 (Applied Biomonitoring, 2009; Johnston et al., 2007). Since the winter of 2009-2010, a network of 24 indigenous mussel monitoring stations located in Sinclair Inlet, Dyes Inlet, Port Orchard Passage, Rich Passage, Agate Passage, Keyport, and Liberty Bay have been monitored semi-annually to measure contaminant tissue residues in mussels. The data are being collected to assess the spatial distribution of contamination, evaluate temporal trends, determine whether exposure levels exceed screening benchmarks, and identify locations where corrective actions may be warranted (Johnston et al., 2011).

The California State Mussel Watch (SMW) program was initiated in 1977 and measures trace metals and legacy organic contaminants in the tissue of transplanted mussels (*Mytilus californianus*) at more than 20 stations along the California coastline (California Environmental Protection Agency State Water Resources Control Board, 2013). In addition, California supports a Regional Monitoring Program for Water Quality in the San Francisco Estuary (RMP). There are a total of 11 stations in the RMP where mussels, clams, and oysters are transplanted in marine, estuarine and freshwater (Sacramento River and San Joaquin River) sites for water quality monitoring purposes. Over the last several decades the California SMW and RMP have produced valuable long-term data on the abundance and distribution of select trace elements and organic contaminants in

California (Gunther et al., 1999; Martin and Severeid, 1984). In 2010, California partnered with NOAA's Mussel Watch to undertake a pilot study exploring the presence of compounds of emerging concern (CECs) in California's coastal waters, this time using resident and transplanted mussels as well as passive sampling devices (Maruya et al., in press as of 2014). This project, titled Mussel Watch California Pilot Study: Compounds of Emerging Concern, monitored 167 CECs in resident bivalves. They reported polybrominated diphenyl ethers (PBDEs-flame retardants), alkylphenols (fuel, detergent, and fragrance additives), and pharmaceutical and personal care products as most frequently detected in mussel tissue. CECs were detected more frequently and had higher concentrations along shorelines influenced by stormwater and treated municipal wastewater discharges. Mussels from urbanized shorelines generally had higher concentrations and detection frequencies of many CECs (perfluorinated compounds, alkylphenols, and PBDEs (California Environmental Protection Agency State Water Resources Control Board, 2013).

In Maine, Gulfwatch has been analyzing resident blue mussel populations since 1993 (Gulf of Maine Council on the Marine Environment, 2014). Their 38 sites around the Gulf of Maine include locations along the coast of Massachusetts, New Hampshire, Maine, New Brunswick, and Nova Scotia, Canada, which are sampled every one to three years. This program tracks spatial and temporal trends of contaminants in indigenous populations of blue mussels (*Mytilus edulis*) to assess the types and concentration of contaminants in coastal waters of the Gulf of Maine (Apeti et al., 2009b; Hunt and Slone, 2010).

The Massachusetts Water Resources Authority (MWRA) conducts mussel (*M. edulis*) monitoring as part of its NPDES permit program, to assess the bioaccumulation potential of sewage effluent discharge (Massachusetts Water Resources Authority, 2014). The MWRA deploys floating, transplanted mussels at mid-depth in the water column in Massachusetts and Cape Cod Bays and in Boston Harbor to evaluate the effectiveness of actions taken to reduce contaminant loading to their water bodies. Because of their mid-water column placement, the MWRA transplanted mussels tend to provide data on water column contaminants, rather than sediment-water interface contamination (Hunt and Slone, 2010).

The MYTILOS project conducted three years of interregional coastal water quality monitoring in the Mediterranean Sea, using a network of stations with transplanted, suspended mussels (*M. galloprovincialis*) (Ifremer, 2014). They placed mussel cages at sub-tidal sites from 2004 – 2006. Mussel transplantation was used to solve the problem of scarce natural mussel stocks in the Mediterranean Sea, and the authors reported that transplantation enabled for control of confounding factors such as the source, age and stage of sexual maturity of the mussels (Galgani et al., 2011). Each year of the study focused on a different part of the Mediterranean basin, for a total of 123 stations along the coasts of Spain, France, Italy, North Tunisia, Algeria and Morocco. Results from the MYTILOS project have been publish by many groups interested in contamination along the Mediterranean coast (Andral et al., 2011; Benedicto et al., 2011; Caixach et al., 2007; Galgani et al., 2011; Scarpato et al., 2010).

Related to MYTILOS was the Mediterranean Mussel Watch Program, a large-scale survey of radioactive and emerging contaminants in the Mediterranean and Black Seas, which took place in 2005. The program was primarily concerned with public health, with the objective of documenting baseline levels of radionuclides in Mediterranean and Black Seas coastal waters, specifically related to the aftermath of the Chernobyl accident. Both indigenous and transplanted *M. galloprovincialis* were sampled at more than 50 sites for this program. The Mediterranean Mussel Watch Program network produced the first regional distribution map of the

radioactive isotope caesium-137, showing the remaining impact of Chernobyl accident (CIESM: The Mediterranean Science Commission, 2012; Thébault and Rodriguez y Baena, 2007; Thébault et al., 2008).

The International Mussel Watch (IMW) Program assessed the extent of chemical contamination in the equatorial and subequatorial areas of the southern hemisphere, including South America, Central America, the Caribbean, and Mexico, in 1991-92 (Center for Coastal Monitoring and Assessment, 2011; Sericano et al., 1995). Seventy-six sites, including locations near known or suspected contamination sources as well as at non-contaminated sites, were sampled for this study.

From 1994–1999 scientists from the Asian Mussel Watch Program collected naturally occurring green mussels (*Perna viridis*) from a total of 48 locations in south and southeast Asia including India, Indonesia, Singapore, Malaysia, Thailand, Cambodia, Vietnam, and the Philippines. This study provided a "bench-mark for data on the distribution of anthropogenic contaminants in this region" (Isobe et al., 2007; Monirith et al., 2003; Sudaryanto et al., 2002; Tanabe, 1994). Asian Mussel Watch monitored a suite of organic contaminants, including PAHs, and phenolic endocrine disrupting compounds such as alkylphenols and bisphenol A (BPA). Results of this study showed extensive input of contamination from wastewater, with little or no treatment, to aquatic environments in South and Southeast Asia (Isobe et al., 2007).

Monitoring of mercury (Hg) levels in *M. edulis* was conducted by a biomonitoring program in the Ems Estuary, shared by Germany and the Netherlands, during the mid-1970s. For this program the mussels were initially collected in the intertidal zone (1972-74), then monitoring continued with transplanted mussels from 1974-1980. This monitoring program demonstrated the effectiveness of industrial emission reductions when Hg-concentrations in the mussels dropped off about four years after mercury abatement began (Kock, 1986). In Belgium, transplanted mussels were successfully used in exposure studies related to offshore oil and gas production water discharges. Mussels in these studies were shown to bioaccumulate PAH compounds and exhibit sensitive biological responses, which led to their continued use in the Norwegian offshore water column monitoring program (Brooks et al., 2012; Hylland et al., 2008; Thain et al., 2008).

Finally, the Budapest Water Works in Hungary uses an ingenious mussel monitoring system called The Mosselmonitor®, which is an online biological early warning system used to monitor water quality (Aquadect, 2014; Baretto, 2012). The system is based on the behavior of bivalves, which vary their valve movement-pattern (e.g. opening and closing) based on the amount of toxicants in the water (Kramer and Foekema, 2001; Sluyts et al., 1996). Under normal environmental conditions their valves remain open most of the time to accommodate filter-feeding and respiration, but the valve movement-pattern changes when pollutants are introduced into their water supply. If several bivalves close simultaneously for a prolonged period, this behavior is considered unusual and a reason for alarm (Delta Consult, 2012). The valve opening of mussels in the Mosselmonitor® are continuously monitored by sensors attached to their shells, which are connected to a computer monitor and alarm system with software to analyze the movement of the valves (Baretto, 2012; Delta Consult, 2012).

2 MATERIALS AND METHODS

Details of the design and methods for this study are described in the *Mussel Watch Pilot Expansion Project - Quality Assurance Project Plan* (Lanksbury et al., 2012). Our protocols were based on guidance from the

Standard Guide for Conducting In situ Field Bioassays with Caged Bivalves (ASTM International, 2007), with modifications to accommodate the specific needs of our study design. In summary, we transplanted cultivated native mussels (*Mytilus trossulus*) from a single source into anti-predator cages at 108 locations along the greater Puget Sound to synoptically evaluate the geographic extent and magnitude of contamination in the nearshore. The study occurred from mid-November, 2012 to mid-January, 2013, with a deployment (i.e. exposure) period of approximately two months. This sample window was selected to coincide with the period of maximum average rainfall in the Puget Sound, when the input of contaminants from stormwater runoff is potentially at its highest, and with the season when *M. trossulus* are reproductively quiescent, to avoid confounding factors associated with reproductive activities.

2.1 Study Area and Site Selection

The NEP funding for this study provided for 60 mussel sites (i.e. cages) distributed along the nearshore areas of the Puget Sound (north, central and south), the Whidbey Basin, and the Bellingham Basin. However, through sponsorship by a number of outside partner groups 48 extra sites were added to the shoreline, greatly expanding the scope of our study (see ACKNOWLEDGEMENTS). Thus, additional study areas incorporated through outside sponsorship included Admiralty Inlet, the San Juan Archipelago, and Hood Canal (Figure 1).

In total, 108 sites were identified for use in this study; see Appendix A for detailed information on the location of each mussel site. Within each basin sites were distributed widely to achieve the most extensive geographic coverage possible. Other factors considered when locating a site along a shoreline included ecological factors such as presence of eelgrass, forage fish spawning areas, and shellfish beds. Also considered was whether a site could be placed in areas with a history of contaminant monitoring (for data comparison) and/or a significant need for Natural Resource Damage Assessment (NRDA) baseline data in the area. All these factors influenced the placement of sites, with a preference to co-locate whenever possible.

One of the main goals of this study was to compare contaminants in shoreline biota with adjacent shoreline land-use. However, land-use patterns in the Puget Sound are highly heterogeneous, representing a wide range of legacy and current-use contamination sources and pathways, including current and former superfund cleanup sites, current permitted industrial outfalls, WWTP outfalls, CSOs, SWOs, failing septic systems, marinas, and ferry terminals, among others. The presence of these myriad contaminant sources precluded a balanced sample design based on point sources alone; therefore, we simplified the classification of the upland by using percent impervious surface (%IS) as an easily quantifiable proxy, as described in Lanksbury and West (2011).

We determined the mean %IS for predefined watershed catchment areas, called Assessment Units (AU), along the Puget Sound shorelines and then distributed our study sites among a range of %IS values. The predefined AUs were originally developed by Ecology (Stanley et al., 2012) and were determined to be of a size appropriate for this study (median area of 8.8 km^2 or 3.4 $mile^2$). We used "percent developed imperviousness" measures from the National Land Cover Database 2006 (Fry et al., 2011; Wickham et al., 2013), with a spatial resolution of 30 meters, to calculate the mean %IS within each AU along the greater Puget Sound shoreline. The mean %IS of the AUs used in our study area ranged from 0 to 94%. From this distribution we created four %IS categories ranging from mostly undeveloped to highly developed: 0-5%, >5- 5%, >15-50% and >50%. The final distribution of the 108 sites (i.e. cages) among the four categories of %IS was as follows: 26 sites in 0-5%, 23 sites in >5-15%, 42 sites in >15-50% and 17 sites in >50%.

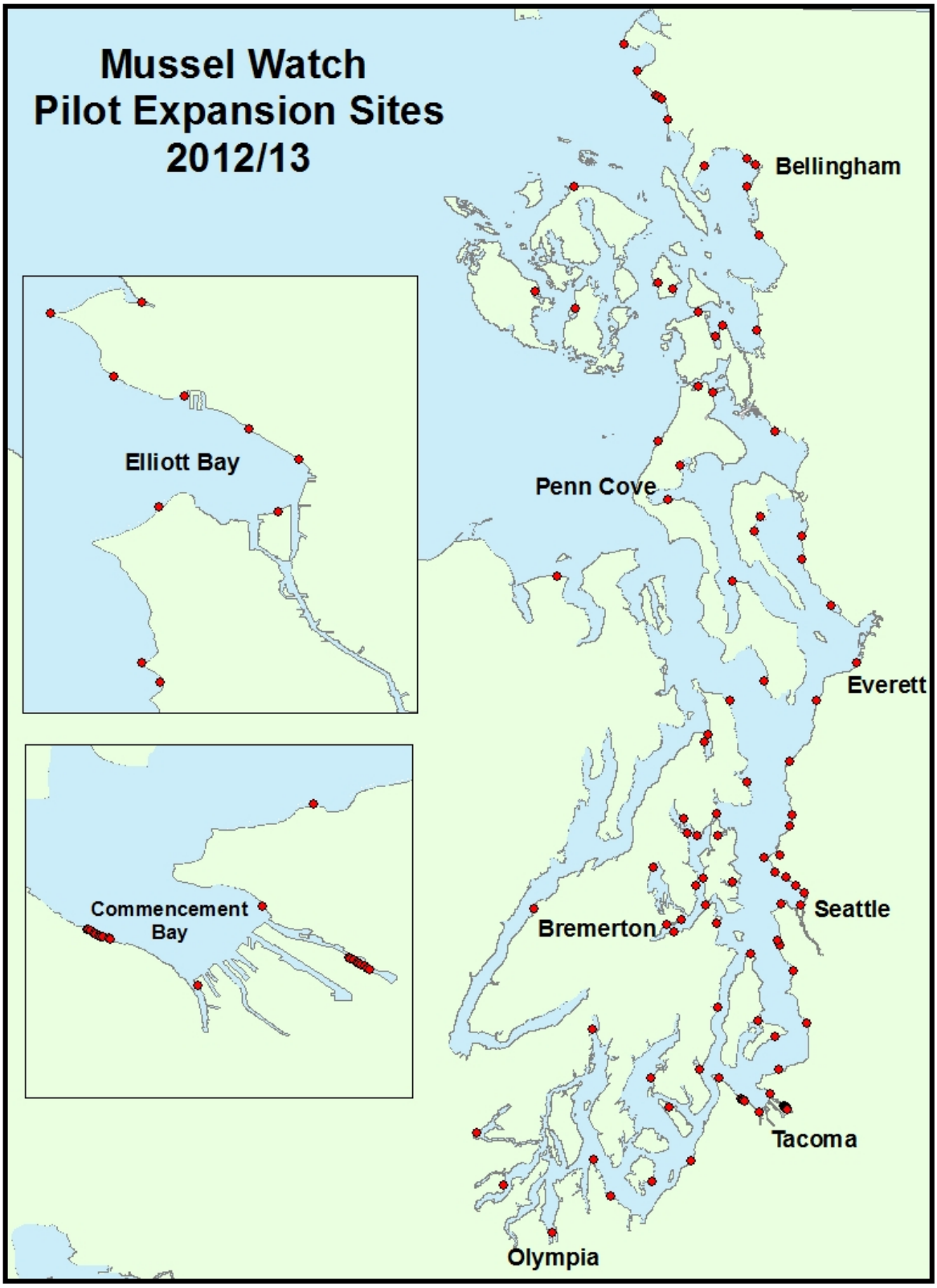

Figure 1. Map of 108 sites where transplanted (i.e. caged) mussels were placed for this study. See Appendix A for further information on the location of these sites.

2.2 Transplanted (i.e. Caged) Mussels

Success in this study was predicated on the availability of mussel populations along the shoreline. However, naturally occurring mussel populations were lacking at many of our desired sampling locations due to the wide variety of intertidal conditions, and the naturally unpredictable nature of mussel beds in Puget Sound. Because of this we chose an active biomonitoring technique wherein we transplanted mussels at desired locations under controlled conditions, rather than passive biomonitoring, which relies on collection of naturally occurring organisms. Active biomonitoring offered several advantages over passive biomonitoring, including:

1. the ability to place samples sites at almost any intertidal location,
2. greater statistical resolution by minimizing variability in contaminant metrics related to species, age, size, reproductive timing, and exposure history
3. a known, controlled exposure period,
4. reduced loss of samples from predation or population failure, and
5. the ability to measure initial population conditions to aid in calculating biological endpoints; specifically mortality, condition index, and growth.

2.3 Study specimen: *Mytilus trossulus*

Mussels are distributed widely on both coasts of North America (ASTM International, 2007; Gosling, 1992). Important both ecologically and economically, they are preyed upon by crabs, including young-of-the-year recruits of shore crabs that feed on post-larval mussels (Asmus and Asmus, 2011), sea stars, marine snails such as dog whelks and oyster drills, various shore birds including gulls, oystercatchers, eiders, scoters, and mammals such as sea otters and humans (Bustnes and Erikstad, 1990; Estes et al., 2003; Kitching et al., 1959; Marsh, 1986; Nyström and Pehrsson, 1988).

There are four distinct marine mussel taxa currently recognized under the genus *Mytilus*: *M. edulis* (Blue mussel), *M. trossulus* (Pacific blue mussel or bay mussel), *M. californianus* (California mussel), and *M. galloprovincialis* (Mediterranean mussel) (ASTM International, 2007; Gosling, 1992; Koehn, 1991). All four species have been used repeatedly in contaminant monitoring studies, sampled both as indigenous populations and in transplant (caged) studies (ASTM International, 2007). Although some mussel samples taken from Washington have been identified as *M. edulis*, there is no reliable evidence that this species occurs anywhere in the Pacific Ocean, except in Chile (Koehn, 1991).

Mytilus trossulus and *M. californianus* are both native to Washington state and easily distinguished from one another; *M. californianus* has distinct radiating ribs on its shell, its adult form is typically much larger than *M. trossulus*, and it tends to occur in more open coastal areas, such as the Pacific Coast of Washington. Although *M. galloprovincialis* (Mediterranean mussel) is not native to Washington State, it has been introduced through aquaculture and is now well established along the greater Puget Sound (Salish Sea) shoreline. Although slightly larger in adult form than *M. trossulus*, *M. galloprovincialis* is difficult, if not impossible, to distinguish from *M. trossulus* based on morphological characteristics. As is typical for species in this genus, hybridization occurs where *Mytilus* spp. occur together (Doherty et al., 2009). *M. trossulus* and *M. galloprovincialis* in Washington State often occur together and are known to hybridize as well (Elliott et al., 2008; Koehn, 1991). However, hybridization between these species is not uniform; rather hybrid zones are spatially complex with pure, mixed and hybrid populations occurring in a patchwork pattern (Elliott et al., 2008).

The temperature and salinity tolerance ranges of *M. trossulus* and *M. galloprovincialis*, both considered for this study, differ slightly, with *M. trossulus* (0-29ºC and 4-33 ppt) tolerating a wider range of conditions than *M. galloprovincialis* (8-25ºC and 10-33 ppt) (ASTM International, 2007; Elliott et al., 2008). The ability of *M. trossulus* to tolerate low salinity conditions (4-33 ppt) makes it a better candidate for transplantation into both marine and estuarine environments. The low temperature tolerance (down to 0 ºC) of this native Washington species also means it is better able to survive exposure to occasional freezing temperatures during winter low tide events in the Puget Sound. Thus *Mytilus trossulus* was chosen as the target species for this study because of its status as a native species, its well-defined, predictable peak spawn timing (see below), its tolerance for low temperature, and because it is readily available in large quantities via local aquaculture cultivation.

2.4 Exposure Timing

There were several factors taken into account when determining the timing of exposure for this study, including the spawning season for *M. trossulus* and average yearly rainfall patterns for the Puget Sound lowland. Also considered were various guides on the appropriate length of exposure for contaminant monitoring with mussels. In addition, because the durability of cages in high energy shoreline habitats was unknown, we sought the minimum exposure period that would satisfy the needs of the study. Deployment and retrieval timing also depended on extreme low tide events, which limited choices for timing of the field work.

2.4.1 Spawning

It is generally recommended that monitoring with bivalves be conducted with populations that will not spawn during the exposure period (ASTM International, 2007; Mourgaud et al., 2002). Losses of up to 50% of total body weight have been reported following bivalve spawning (Lachance et al., 2008). A large proportion of accumulated chemicals can be lost during spawning, which can complicate data interpretation. The reproductive timing of the various *Mytilus* species varies depending on their location. Past studies have indicated that mussels in the Puget Sound have similar spawn timing; mussels collected in September, 1992 from the north, central and south Puget Sound were all at a similar stage of gonadal development (Krishnakumar et al., 1994). Later, Kagley et al. (2003) showed that the peak spawning period for mussels from Coupeville and Seacrest occurred between April and May (Figure 2). *Mytilus galloprovincialis* in Penn Cove, Whidbey Island typically spawn in the early winter, while *M. trossulus* typically spawn in early spring (Penn Cove Shellfish LLC, 2012, pers. comm.).

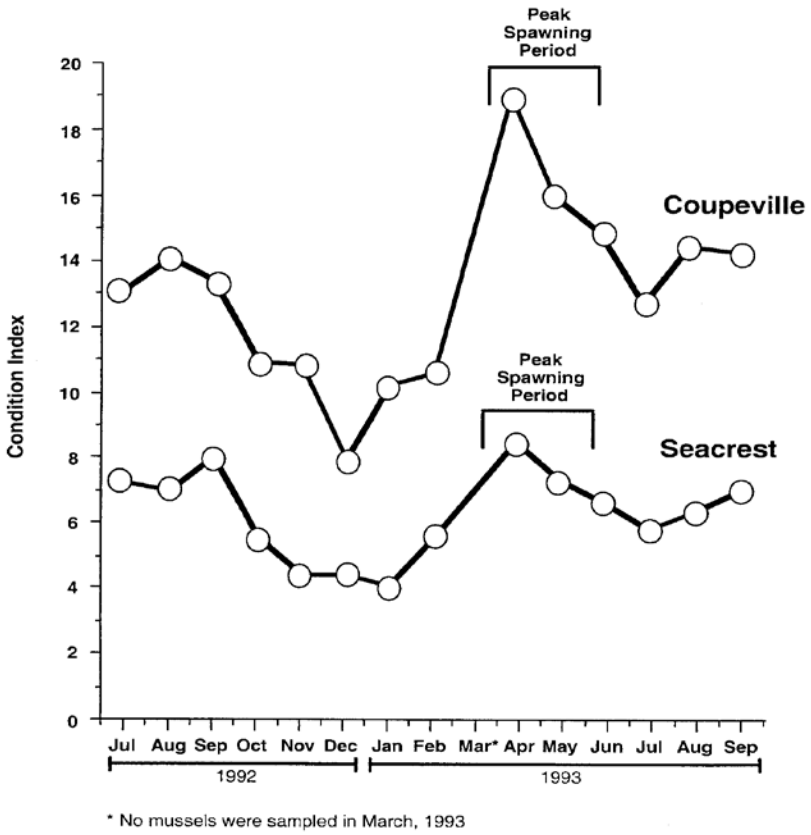

Figure 2. "Changes in the condition index of mussels (*Mytilus edulis* complex) from Coupeville and Seacrest (n = 25 per site, per month) from July 1992 to September 1993. The condition index is the somatic tissue wet weight (g)/(shell length [mm]) * 100" taken from Kagley et al. (2003). The "*M. edulis* complex" listed here is either *M. trossulus*, *M. galloprovincialis*, or a hybridization of the two species.

2.4.2 Rainfall

Data from previous mussel studies suggests winter is the best season to capture the signal of organic contaminants in Puget Sound, particularly for PAHs. As part of a study to compare seasonal differences in contaminants, the Snohomish County MRC collected wild mussels at seven NOAA Mussel Watch sites during the summer (dry) seasons of 2007, 2008 and 2009 and compared their contamination with mussels taken during the winter (wet) seasons of 2006, 2008, and 2009 (Whitney et al., 2011). They found that the concentrations of contaminants in mussels were higher during the winter as compared to the summer, especially for PCBs, DDTs and PAHs. Winter samples were somewhat elevated for chlordane as well.

Because we were particularly interested in contaminant input into the nearshore via watershed processes (e.g., stormwater), we timed our mussel deployments to match the period of maximum surface runoff into the Puget Sound. We examined a 50-year timeline of precipitation index data from the Puget Sound lowland, using data from the National Climatic Data Center (Figure 3) (National Oceanic and Atmospheric Administration, 2014a). From this data we observed that precipitation was lowest from June through September, and highest from November through January. Thus, to capture the seasonal maxima of surface water runoff, we targeted the months of November, December, and January.

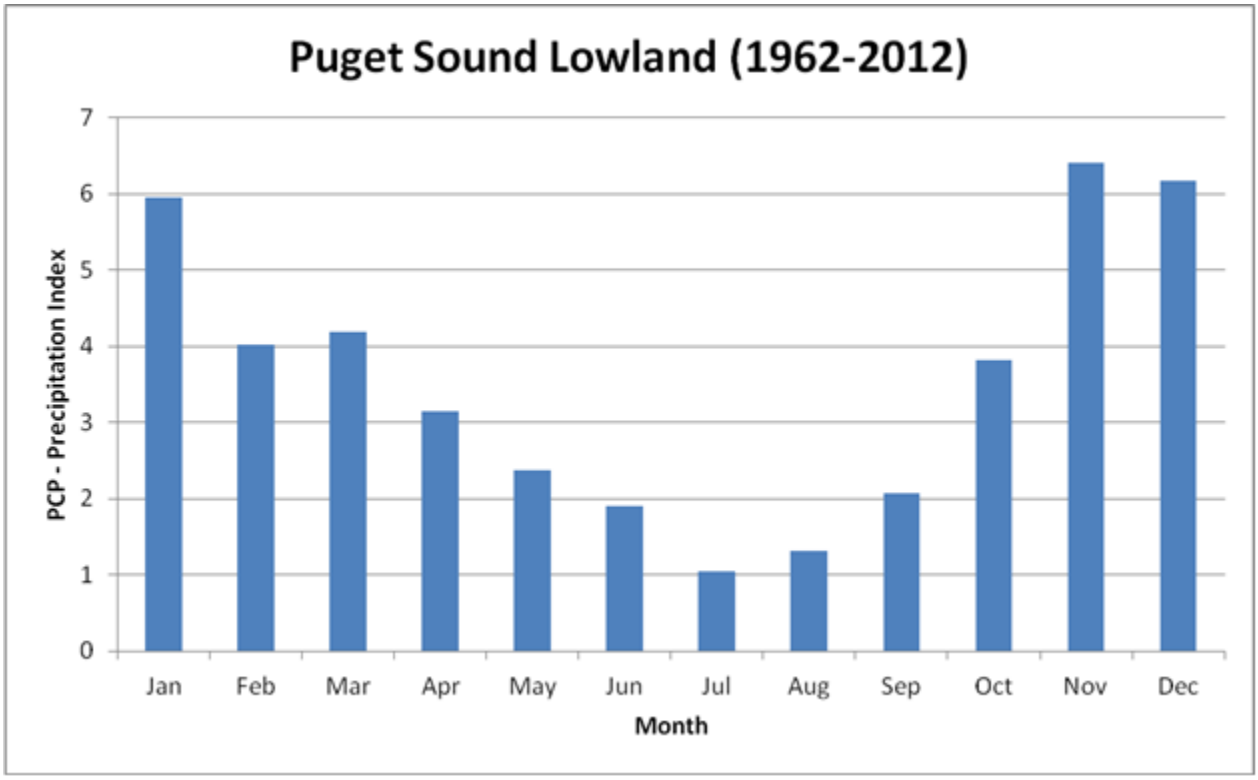

Figure 3. Fifty year timeline of precipitation data for the Puget Sound lowland. Data provided by the National Climate Data Center.

2.5 Length of Exposure

The duration of *in-situ* exposure for transplanted mussel varies depending on the goals of the study and the target contaminants being evaluated. The ASTM International (2007) guide to biomonitoring with bivalves suggests a minimum test period of 30 days "unless the chemicals of concern are low molecular weight organic compounds, such as some PAHs." However, it is generally agreed that 60 to 90 days is sufficient to ensure the mussels have sufficient time to "equilibrate" with their surroundings and the range of contaminants therein (ASTM International, 2007; Axelman et al., 1999; Baussant et al., 2001a; Baussant et al., 2001b; Durell et al., 2006; Neff and Burns, 1996; Peven et al., 1996; Prest et al., 1995; Richardson et al., 2003; Salazar and Salazar, 1995). The target duration of exposure of transplanted mussels in this study was 2 months (~60 days), from mid-November 2012 to mid-January 2013.

2.6 Sample Units - Mussel Cages

2.6.1 Preparation

We used cultured, pre-reproductive *M. trossulus* which were donated to the study from Penn Cove Shellfish, Inc., an aquaculture facility located in Penn Cove, Whidbey Island, Washington. Mussels used in this study were estimated to be 11 months old (Penn Cove Shellfish LLC, 2012, pers. comm.). Exposure to contaminants in Penn Cove was expected to be minimal, and because the animals had not yet reproduced we assumed no differences in initial contaminant load related to sex. A subset of 100 mussels was collected prior to transplantation to be analyzed as a baseline sample for this study, denoted hereafter as "Penn Cove, Baseline".

Sorting, measuring and bagging of mussels occurred from October 23 - 29, 2012, with volunteers providing considerable support to this effort. *M. trossulus* were taken directly from a Penn Cove Shellfish harvesting vessel, where an assembly of machines removed mussels from their aquaculture ropes, separated them from one another, cleaned them of sediment and other debris, and shaved off their byssal fibers (i.e. beards). Mussels taken from the harvesting vessel were held in ambient seawater kept within ±5° Celsius of Penn Cove surface water and changed as needed to maintain suitable water quality.

Only intact individuals that had no cracks in their shells and were responsive to physical stimulation (i.e. closed their shells when handled) were selected for use. Once sorted, acceptable mussels were measured using a digital caliper with measurement accuracy to a tenth of a millimeter (0.1 mm). Only mussels measuring 50 - 60 mm in shell length (as measured from umbo to farthest posterior margin) were included in this study. Mussels of that size were approximately 11 months old and had not yet spawned in their lifetime (Penn Cove Shellfish LLC, 2012, pers. comm.).

Once measured, 16 mussels of the appropriate length were placed into two separate pockets (eight per pocket) in heavy duty, extruded high density polyethylene (HDPE) mesh bags (Norplex, of the type used by mussel culturists) measuring approximately 20 inches in length (Figure 4). Nylon cable ties were used to secure the ends of the bags and to cinch down the center to create the two pockets. These pockets provided ample space for eight mussels to open and close their valves to filter feed, and to accommodate animal growth. The filled mussel bags were then placed into another holding cooler filled with ambient Penn Cove seawater, maintained in the same fashion as described above, until they could be re-hung from lines under the Penn Cove aquaculture raft #D-2. The bagged mussels remained hanging, undisturbed, at Penn Cove for 20 - 23 days, and then they were deployed to their individual study sites. This interim period was included to allow the mussels time to rest and re-cluster after handling and bagging (Andral et al., 2011; Benedicto et al., 2011; Galgani et al., 2011).

Figure 4. Photo of four mesh bags (each holding 16 mussels) secured into the upper section of an anti-predator cage. Each cage used in this study had a lid (not shown here) attached at the time of deployment.

2.6.2 Deployment and Retrieval

Plastic-coated, wire mesh cages with a mesh opening of 1.25 x 2.5cm were used in this study (McKay Shrimp & Crab Gear, 2014). These cages were designed to exclude large predators from reaching the mussels, while optimizing water flow. Each mussel cage had a stainless-steel identification plate attached to it that included the WDFW logo, study title, and contact information. The empty cages and all anchoring devices (bent-tip rebar stakes and helical "earth" anchors) used for the study were washed and then soaked in water for at least 24 hours in advance of mussel placement, to dissipate any potential surface contaminants.

PSEMP staff, sponsoring partners, and citizen volunteers (deployment teams) deployed a total of 108 cages to 108 individual sites during evening low tides from November 12 - 14, 2012 (Table 1). The mussel cages were anchored to intertidal substrate between 0 to -1.5 feet mean lower low water (MLLW), with mussels suspended approximately 35 cm above the substrate within the cage. This tidal elevation was selected to allow for occasional exposure to air during the tidal cycle, to simulate natural conditions experienced by mussels in the intertidal zone during the winter in Puget Sound, while keeping mussels submerged during daylight hours to minimize losses to vandalism or theft. The 105 cages that remained at the end of the study were retrieved over seven days, from January 7 – 14, 2013.

Table 1. Dates of mussel cage deployment and retrieval for Mussel Watch Pilot Expansion study.

Deployment		Retrieval	
November 2012	Number of Cages	January 2013	Number of Cages
12	22	7	12
13	46	8	29
14	40	9	48
		10	13
		11	4
		14	2

2.7 Biological Endpoints

2.7.1 Mortality Assessment

Individual mussels from each cage were assigned into one of three categories (1. healthy, 2. dead or moribund, and 3. missing) depending on their condition at the end of the study. Mussels were considered "healthy" when they were whole and in good condition, including some with shells that may have been cracked from handling . Only healthy, uncracked mussels were used for chemical analyses, while some of the mussels that may have been cracked during retrieval were used in the assessment of condition index. "Dead or moribund" included whole empty shells, matched broken shells and hinges, whole rotting mussels, or gaping mussels that would not close their shells. "Missing" mussels included mussels that were simply gone, which may have resulted from a miscount during the bagging phase, or could have occurred if a mussel became fragmented and its shell pieces fell through the cage mesh.

2.7.2 Growth

We measured the shell length of each mussel used in this study, from the umbo to the farthest posterior margin, to the nearest tenth of a millimeter (0.1 mm) using digital calipers. Shell length for each mussel was measured

both at the start and again at the end of the study, to investigate growth as a potential biological endpoint, however individual mussel lengths were not tracked. Because the starting shell lengths were measured during the bagging process, the lengths measured at the end of the study include growth during the 20 - 23 day resting period before mussels were deployed to their individual sites; thus the actual growth that occurred at each site is slightly overestimated. However, this bias was equal for all sites because the mussels all rested in the same location and for the same length of time prior to deployment. The shell growth rate (mm/day) for 44 mussels at each site (cage) was calculated using the following formula:

$$\text{Shell growth rate for cage X (mm/day)} = [\ \bar{x}(SL_{start} - SL_{end})] / \text{Days}$$

Where: SL_{start} = mean shell length (n = 64) of mussels from cage X at <u>start</u> of study (i.e. day of bagging)
SL_{end} = shell length (n = 44) of individual mussels from cage X at <u>end</u> of study (i.e. post-deployment)
Days = days of exposure at study site

2.7.3 Condition Index

To account for differences in growth related to food availability in this study, we calculated the Condition Index (CI) of mussels from each site. According to researchers from the MYTILOS mussel monitoring project, "although the concentrations measured in the tissues [were] a function of bioavailable pollutant levels, for some contaminants, the bioaccumulation factor depends on mussel growth in relation to the primary food production or trophic capacity of the environment (Nolan and Dahlgaard, 1991) or lipid content (Capuzzo et al., 1989)" (Galgani et al., 2011).

Condition indices function to normalize biological changes over time and can help assess the role of seasonal fluctuations in environmental factors (e.g., food availability, temperature), and serve as an indication of the impact of reproductive status on biological and chemical measurements in the mussels (Benedicto et al., 2011; Kagley et al., 2003; Roesijadi et al., 1984). We determined CI on twelve randomly selected mussels to represent each site according to a method reported by Kagley et al. (2003) as follows:

Condition Index (CI) = dry weight (g) of soft tissue/shell length (mm) X 100.

2.8 Chemical Analyses

2.8.1 Composite Sample Preparation

The soft tissue from approximately 32 mussels from each site was combined to create composites for chemical analysis. Frozen mussels were thawed and composited following a modification of the Field Procedure 11.7 from the Standard Guide for Conducting *In-situ* Field Bioassays with Caged Bivalves (ASTM International, 2007). Prior to shucking, the external byssal threads of the mussels and any sediment, biofouling, or barnacles were removed from the shells, then the shells were rinsed with deionized (DI) water. After this external cleaning, the mussels were opened by inserting a clean scalpel blade between the shells, severing the posterior and anterior adductor muscles. The shells were spread apart at the hinge and the remaining byssal fibers were trimmed from the byssal gland using scissors, then the soft tissue was gently rinsed clean of sediment and foreign material with DI water. Soft tissue (including the adductor muscle) from 32 mussels per site was scraped into a single pre-cleaned glass sample jar to create a site-composite sample. Composite samples were then frozen. Each composite sample was later homogenized; after partial thawing each composite was ground to a consistency resembling pudding using a hand mixer. Composites were then frozen to -20ºC until transfer to

the analytical lab. A more detailed description of this process is available in the study Quality Assurance Project Plan (QAPP; Lanksbury et al., 2012).

2.8.2 Analytical Methods

All samples were delivered frozen to analytical laboratories and thawed samples were stirred prior to extraction to ensure they were adequately homogenized. All sample data met QA/QC criteria as outlined in the study QAPP (Lanksbury et al., 2012), except for minor violations of holding time for mercury, which were considered inconsequential. Mussel samples were not analyzed for stable isotopes of nitrogen ($\delta^{15}N$) or carbon ($\delta^{13}C$), as originally proposed in the QAPP, to help contol expenses. Although the stable istotope data may have been useful in investigating differences in local food sources and trophic levels among mussels from the different study sites, the absence of this data was not a significant hinderance to the interpretation of the contaminant data.

The mussel soft tissue matrices were analyzed for concentrations of PCBs, PBDEs, organo-chlorinated pesticides (OCPs) and PAHs at NOAA's Northwest Fisheries Science Center (NWFSC) (National Oceanic and Atmospheric Administration, 2014b). After homogenization, all samples were analyzed for these persistent organic pollutants (POPs) using accelerated solvent extraction and gas chromatography/mass spectrometry according to Sloan et al. (2004). In brief, this method comprises three steps; 1) accelerated solvent extraction (ASE) of tissue using methylene chloride, 2) cleanup of the methylene chloride extract by silica/aluminum columns and size-exclusion high-performance liquid chromatography (SEC HPLC), and 3) quantitation of chlorinated hydrocarbons (CHs) and aromatic hydrocarbons (AHs) using gas chromatography/mass spectrometry (GC/MS) with selected-ion-monitoring (SIM). Extraction by ASE methods provided an extraction that was used for AH, CH recovery and gravimetric lipid evaluation. Alterations to the typical GC/MS methods were included in order to stabilize the instrument and improve accuracy, specifically chemical ionization filaments (used to increase source temperature) employed a cool on-column injection system in the GC, a guard column before the analytical column, and point-to-point calibration to improve data fit over the full range of GC/MS calibration standards (Sloan et al., 2004). Total solids (and % moisture) were analyzed gravimetrically according to Sloan et al. (2004) to allow reporting organics data in both dry and wet weight concentrations. Concentrations were reported as nanograms contaminant per gram mussel tissue (ng/g, equivalent to parts per billion).

Metals were analyzed using two methods. Mercury was analyzed via automated cold vapor atomic absorption spectrometry following King County Environmental Laboratory's (KCEL) Standard Operating Procedure (SOP) 604v6 (King County Water and Land Resources Division, 2014). This SOP incorporates elements of the following Environmental Protection Agency's (EPA) methods; 245.1 revision 3, SW-846 7470, 7471B and PSEP 1997. Arsenic, cadmium, copper, zinc and lead were analyzed via Thermo Elemental X Series II CCT (Collision Cell Technology) Inductively Coupled Plasma Mass Spectrometry (ICP-MA), following KCEL SOP 624v2. This SOP incorporates elements of EPA methods; 200.8 revision 5.4, SW-846 6020A February 2007, ILMO5.3 Exhibit D part B, and PSEP 1997. Total solids (and % moisture) were analyzed using KCEL SOP 307v3 to allow reporting metals data in both dry and wet weight concentrations. Concentrations were reported as microgram metal per gram mussel tissue (µg/g, equivalent to parts per million).

2.9 Data Analysis

2.9.1 Contaminant Concentrations

Mussel contaminant data are presented as summed concentrations (e.g., Σ_6DDTs) for analyte groups (Table 2), except in cases with fewer than two analytes per group. Summed analytes are the sum of all detected values, with zeroes substituted for non-detected analytes, within each group. In cases where *all* analytes in a group were not detected the greatest limit of quantitation (LOQ) for any single analyte in the group was used as the summation concentration, and the value was preceded by a "<" (less than) qualifier. An estimated total PCB (TPCB) concentration was calculated by summing the detected concentrations for 17 commonly detected congeners and multiplying the result by two[1], according to Lauenstein and Cantillo (1993). Summaries of the contaminant concentrations of mussel composites (n = 32 mussels) made for this study are provided in Appendices C - F. Individual results for each congener or analyte will be uploaded to Ecology's Environmental Information Management (EIM) database, and EPA's STORET database, where they will be available on-line. Though contaminant concentrations are reported in both wet and dry weight, all statistical tests were conducted using only dry weight (dw) contaminant concentrations. Appendices H – S include maps of the distributions of contaminant concentrations and cumulative frequency distribution plots for each contaminant type that was detected in mussels from at least 17% of the study sites.

Table 2. Analyte groups summed for the Mussel Watch Pilot Expansion study.

Sum 3 HCHs	Sum 8 Chlordanes	Estimated Total PCBs*	Sum 6 DDTs	Sum 11 PBDEs	Sum 42 PAHs	
					low molecular weight	*high molecular weight*
alpha hexachlorocyclohexane	alpha chlordane	PCB018	ppDDD	PBDE028	naphthalene	fluoranthene (FLA)
beta hexachlorocyclohexane	beta chlordane	PCB028	ppDDE	PBDE047	C1-naphthalenes	pyrene (PYR)
lindane	cis nonachlor	PCB044	ppDDT	PBDE049	C2-naphthalenes	C1-fluoranthenes/pyrenes
	heptachlor	PCB052	opDDD	PBDE066	C3-naphthalenes	C2-fluoranthenes/pyrenes
	heptachlor epoxide	PCB095	opDDE	PBDE085	C4-naphthalenes	C3-fluoranthenes/pyrenes
	nonachlor3	PCB101	opDDT	PBDE099	acenaphthylene (ACY)	C4-fluoranthenes/pyrenes
	Oxychlordane	PCB105		PBDE100	acenaphthene (ACE)	benz[*a*]anthracene (BAA)
	trans Nonachlor	PCB118		PBDE153	fluorene (FLU)	chrysene (CHR)[a]
		PCB128		PBDE154	C1-fluorenes	C1-benzanthracenes/chrysenes
		PCB138		PBDE155	C2-fluorenes	C2-benzanthracenes/chrysenes
		PCB153		PBDE183	C3-fluorenes	C3-benzanthracenes/chrysenes
		PCB170			dibenzothiophene (DBT)	C4-benzanthracenes/chrysenes
		PCB180			C1-dibenzothiophene	benzo[*b*]fluoranthene (BBF)
		PCB187			C2-dibenzothiophenes	benzo[*k*]fluoranthene (BKF)[b]
		PCB195			C3-dibenzothiophenes	benzo[*e*]pyrene (BEP)
		PCB206			C4-dibenzothiophenes	benzo[*a*]pyrene (BAP)
		PCB209			phenanthrene (PHN)	perylene (PER)
					anthracene (ANT)	indeno[*1,2,3-cd*]pyrene (IDP)
					C1-phenanthrenes/anthracene	dibenz[*a,h*]anthracene (DBA)[c]
					C2-phenanthrenes/anthracenes	benzo[g,h,i]perylene (BZP)
					C3-phenanthrenes/anthracenes	
					C4-phenanthrenes/anthracenes	
**Sum of 17 congeners, then multiplied by two*					[a] coelutes with triphenylene	
					[b] coelutes with benzo[j]flouranthene	
					[c] coelutes with dibenz[a,c]anthracene	

2.9.2 Impervious Surface and Road Area

We used impervious surface and road area as proxies for urbanization in this study. The metric utilized for impervious surface was calculated by determining the "percent developed imperviousness" (Fry et al., 2011; Wickham et al., 2013) within predefined watershed catchment areas called Assessment Units (AUs). The %IS values in our study ranged from 0 to 94% (see Study Area and Site Selection for details).

[a] originally calculated by Lauenstein and Cantillo (1993) as 2(Σ_{18} congeners).

Road area within each AU was also calculated to investigate whether vehicle traffic was correlated with, or could be used to predict, mussel contamination. Our road area metric was estimated by first determining the length of various classes of roadway, based on line data obtained from a 2003 Tele Atlas Dynamap Transportation, Version 5.2 digital dataset, (Spatial Insights Inc., 2012) within each AU, then matching each road class to a standard road width (25, 33, 52, 76, or 80 feet) based on data gleaned from the Washington State Highway Log, version 2013 (Washington State Department of Transportation, 2013). The road lengths and widths were then multiplied together to produce the estimated total road area within each AU. Then the total estimated road area was divided by the total area within each AU to give an estimated percent road area (%RA) for that AU. The estimated %RA in this study ranged from 0 to 26%. Because there was a high degree of correlation between %IS and estimated %RA (Pearson Correlation = 0.795), we did not include both factors in any regression or ANOVA model (GLM, see below), but instead investigated them separately.

2.9.3 Data Transformations and Statistical Analyses

All organic contaminants and metals were reported by the analytical labs on a wet weight basis, however to maintain consistency with the majority of published mussel contaminant studies we converted wet weight to dry weight using the %moisture value derived from the analytical process. In addition, all contaminant data were $\log_{10}$-transformed prior to analysis to achieve normality and equality of variances for statistical testing. Minor violations of the normality and equality of variances assumptions after transformation were ignored if they were near the acceptable threshold ($p = 0.05$). In a few cases transformation was not required to achieve normality or homoscedasticity; however we transformed all $\log_{10}$-contaminant data for consistency. All means, coefficients and confidence intervals generated via ANOVA and GLM were back-calculated and reported as geometric values.

We do not present lipid-adjusted concentrations by dividing wet or dry contaminant concentration by lipid% in this report; overall the lipid concentrations in our mussels were low and ranging narrowly from 0.66 - 1.34% wet weight (one outlier = 0.21%). This low and narrow range was not surprising considering mussels do not feed at maximum capacity during the winter and generally lose weight during this season (Kagley et al., 2003). Lipid concentrations below 1% are difficult to measure accurately, and very low lipid concentrations have a large effect when computing contaminant concentrations on a lipid basis. In addition, small inaccuracies in quantitation in the range we encountered can contribute to spurious conclusions. For these reasons we did not routinely lipid-normalize the mussel contaminant data in this study, but instead used lipid concentrations as a covariate in our statistical models. This approach follows protocols from other monitoring programs such as the Massachusetts Water Resource Authority (MWRA) mussel monitoring program, who originally normalized their mussel contaminant data with lipids through 1998, then dropped the practice after they discovered lipid normalization did not substantially alter the mussel contaminant trends when compared to non-lipid-normalized data (Hunt and Slone, 2010; Mitchell et al., 1998).

A simple first step in our analysis was to determine whether there were differences between urban growth areas (UGAs) and non-UGAs of the Puget Sound lowland, which was of concern to Ecology's Stormwater Work Group. Thus a two-sample *t*-test using UGA as the classification was run on each $\log_{10}$-transformed contaminant type to answer this question. Following the *t*-tests, stepwise general linear models (GLMs; Systat 12) were used to run stepwise multiple linear regressions to test hypotheses related to contaminant levels and site type. The goal of the GLMs was to construct predictive regression models between mussel contaminant concentration (and biological endpoints) and either %IS or %RA in the upland, while accounting and adjusting

for effects related to other factors including lipids, CI, and days of exposure. For each contaminant type we computed multiple linear regressions, removing or adding factors in a stepwise fashion, until we arrived at the most parsimonious model. In some cases where covariates were significant but contributed trivial additional explanatory power, they were noted but removed from the final model.

2.9.4 Pattern Analysis of PAHs

The chemical composition (analyte fingerprint) of PAHs has been used as a diagnostic tool to help infer sources of PAH pollution in air, water, sediment, and soil, and have more recently been applied to determine PAH sources in mussels (Amin et al., 2011; Francioni et al., 2007; Guinan et al., 2001; Maioli et al., 2010; Palma-Fleming et al., 2008; Palma-Fleming et al., 2012; Payne et al., 2008; Soriano et al., 2006; Tobiszewski and Namieśnik, 2012). PAH fingerprints can shed light on whether contamination in a sample came from petrogenic (related to unburned petroleum) or pyrogenic (generated by the combustion of fossil and other fuels, including coal and wood, or from creosote) sources. For instance, the fraction of parent PAHs (C_0) to their alkylated homologs (C_1, C_2, C_3, or C_4) is used extensively to infer sources in natural resource damage assessments for oil spills: petrogenic sources typically have a greater percentage of alkyl PAHs (C_1, C_2, C_3, or C_4) compared to their parent compounds (C_0), while pyrogenic sources, or highly weathered oil, tend to have a predominance of parent PAHs compared to their alkylated homologs (da Silva and Bícego, 2010; Lima et al., 2005; Payne et al., 2003; Tobiszewski and Namieśnik, 2012; Yunker et al., 2002). Using histogram plots we investigated and compared the concentration and overall percent of individual PAH analytes among the mussel sites (Appendix T). We also used histograms to compare mussels taken from Penn Cove, Shellfish Inc. shortly after an oil spill in Penn Cove (May of 2012) with the mussels we used from the same source several months later (November, 2012) as our baseline (i.e. control) mussels; see Penn Cove Oil Spill – Fingerprint Comparison section.

We further summarized and quantified the patterns of PAH analytes by examining the homolog series maximum (i.e. C_0 or C_1 or $\geq C_2$) for three of the most frequently detected analyte pairs, anthracene(ANT)/ phenanthrene(PHN), fluoranthene(FLA)/pyrene(PYR), and benz[*a*]anthracene(BAA)/chrysene(CHR). In addition, we calculated two commonly accepted ratios that have been used forensically to distinguish between petroleum and combustion PAH sources. We followed the methods used by Incardona et al. (2012), who used the ratio of the sum of alkylated PHNs to PHN (Σalkylated-PHNs/PHN) to distinguish petrogenic PAHs (from an oil spill) from background pyrogenic PAHs in Pacific herring (*Clupea pallasii*) embryos. Accordingly, we regarded mussels that exhibited a Σalkylated-PHNs/PHN ratio greater than 2 as indicative of petroleum exposure, and sites with a ratio less than 2 as indicative of pyrogenic PAH exposure. In addition, we used an increasing ratio of fluoranthene (FLU) plus pyrene (PYR) to the sum of C2- through C4-PHN (FLU+PYR/ Σalkylated-PHNs) to further distinguish between pyrogenic (low ratio) and petrogenic (high ratio) PAHs. All together the PAH fingerprint histograms, the homolog series maxima, and the PAH ratios were used in a "weight of evidence" approach to characterize the nearshore (mussel) sites as primarily exposed to pyrogenic or petrogenic PAH sources, or both.

2.9.5 PCB ratios

PCBs were produced as congener mixtures (e.g., Aroclors) comprising a wide range of congener profiles including mixtures dominated by less-chlorinated, or "lighter", PCB congeners and those dominated by more highly chlorinated, or "heavier", congeners. As PCBs move through the environment they spread at different speeds due to differences in their mass and lipophilicity, creating a gradient of PCB congeners whose relative abundance changes with distance from the source mixtures (Grant et al., 2011). Thus, when examining PCB profiles in biota one may observe loss of heavier congeners with increasing distance from the source, which may appear to be enriched with heavier PCB congeners. Lighter PCB congeners (which tend to travel faster) may be relatively more abundant at locations distant from the source (Ross et al., 2004). We used a simple ratio of two abundant congeners, one lighter (trichlorobiphenyl, or PCB28) and one heavier (heptachlorbiphenyl, or PCB187) in our mussel samples, with the formula PCB28/PCB28+187 for our comparisons to evaluate this differential movement of congeners relative to PCB source locations. We expected to see urban areas of the Puget Sound (like Elliott and Commencement Bays and Bremerton), which represent sources or hotspots of PCB contamination, to have lower PCB28:187 ratios compared to remote sites in non-urban areas.

2.9.6 Averaging of Hylebos Waterway and Ruston Waterfront Sites

One of our study partners, the Tacoma-Pierce County Health Department (TPCHD), sponsored an additional study along the Hylebos Waterway and the Tacoma Ruston Waterfront called the Mussel Watch Gradient Project. For that study the TPCHD placed nine cages, averaging about 100 meters apart from one another, along the length of the beach at each site (Figure 5). The high density of cages at these two sites was intended to help answer the question, "what is the length of shoreline that represents a site for mussel contamination sampling". Results from the TPCHD's analysis are available in a separate report (Callahan et al., 2014). Because the TPCHD's cage placement represented a much higher density compared to the rest of the study, here we used average values of contaminants from Hylebos Waterway sites 1-9 and Tacoma Ruston Waterfront sites 1-9 to represent two single points in our analyses, hereafter called the "Hylebos Waterway" and "Tacoma Ruston Waterfront" sites. Data from each of these averaged sites was assigned to the central-most position along the putative gradient for the two locations in that study.

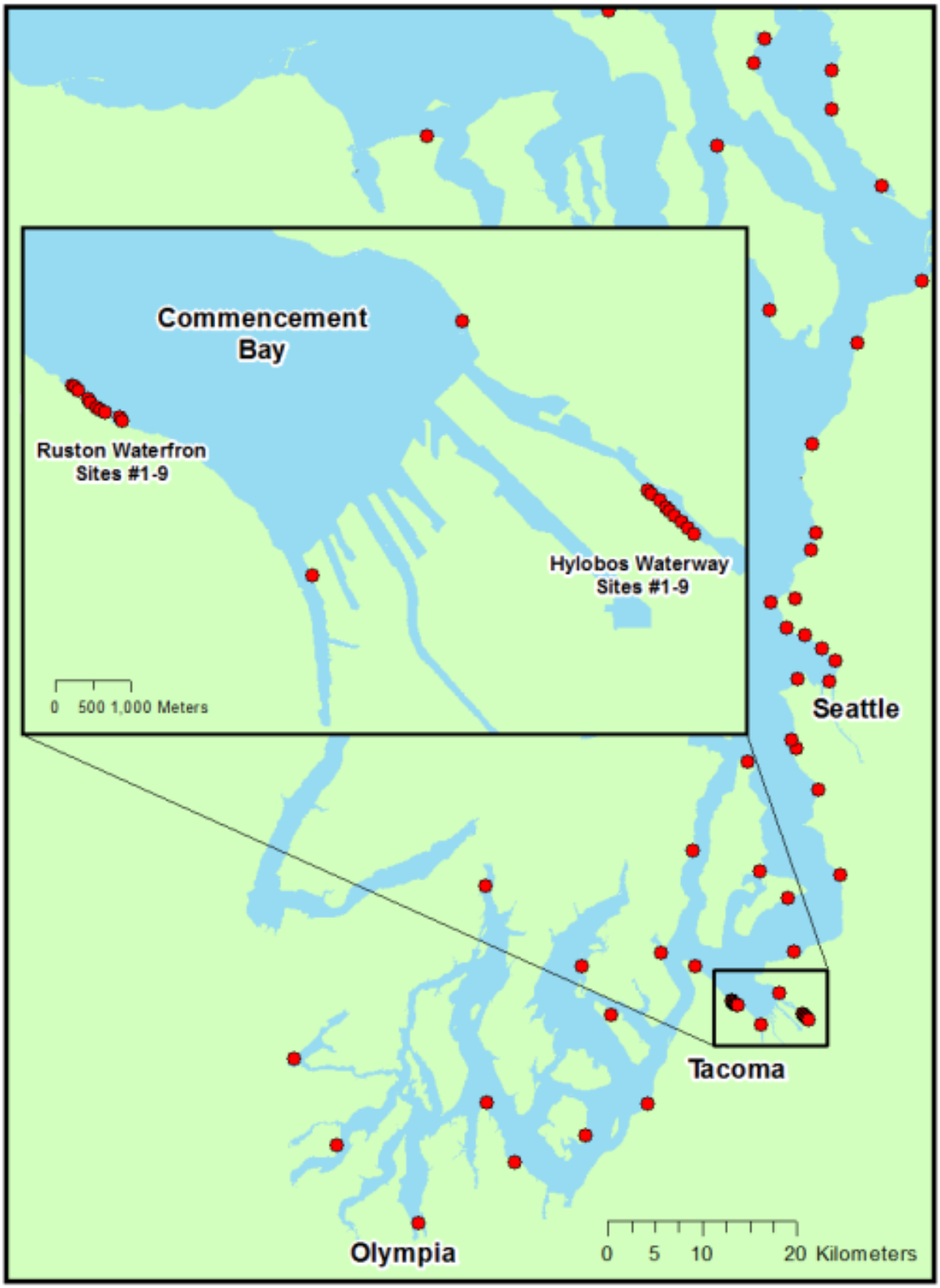

Figure 5. Map of transplanted (i.e. caged) mussel sites in Pierce County at the Hylebos Waterway and the Tacoma Ruston Waterfront.

2.9.7 Transplanted vs. Wild Mussels

At the end of the transplanted mussel deployment period, several of our study partners collected wild mussels near their transplanted mussel sites for comparison with the transplanted mussel data. Matching transplanted and wild mussel collection locations included Kayak Point, Cavalero Beach County Park, Hermosa Point, Edmonds Ferry and Everett Harbor (Snohomish County), and the Hylebos Waterway in Pierce County, where wild mussels were collected along a transect spanning the Hylebos Waterway sites #1 and 2 (Table 3). Native mussel samples were processed for CI and were made into composites using the same methods as for transplanted mussels. Although this study was not specifically designed for this purpose, we compared tissue contaminant concentrations between matching transplanted and wild mussels sites here. We used ANOVA with sample type (i.e. wild vs. transplanted) as the classification variable. As with previous analyses, the contaminant data were $\log_{10}$-transformed prior to analysis and lipid and condition index (CI) were included as covariates.

Table 3. Sites where wild mussels were collected near transplanted mussel counterparts.

Site Name		**Comment**
Transplanted	**Native**	
Cavalero Beach	Cavalero Beach Natives	Native mussels collected at the waterline, in cobble with sand substrate with a few small boulders around.
Kayak Point	Kayak Point Natives	Native mussels collected +5 to +7 feet above MLLW along a transect southeast of Kayak Point and in vicinity of (central to) deployed cage. This location is also a NOAA Mussel Watch site. Native mussels collected in cobble with sand substrate, feeder bluff above the beach.
Hermosa Point	Hermosa Point Natives	Native mussels collected at three stations at NOAA's Mussel Watch site near the tip of Hermosa Point. Substrate was cobble and sand.
Everett Harbor	Everett Harbor Natives	Native mussels collected along a transect located about +3 feet from MLLW. This location is also a NOAA Mussel Watch site. Substrate was boulders and cobble.
Edmonds Ferry	Edmonds Ferry Natives	Native mussels collected at three stations on the rock jetty by dive park. This location is also a NOAA Mussel Watch site. The transplanted mussels were deployed just seaward of the end of the rock jetty at zero tide height.
Hylebos Waterway 1 & 2	Hylebos Waterway Natives	Native mussels collected in close proximity to Hylebos Waterway sites #1 - #2.

3 RESULTS

3.1 Overview

Of the 108 cages deployed at the start of the study 105 were retrieved intact; three cages were lost or dislodged during the course of the study period. The first lost cage, from a site called Cherry Point Aquatic Reserve #2 - Alcoa-BP, was found washed up in the high intertidal area in December, a month after deployment. This cage was likely uprooted during a storm event in November. At the end of the study (i.e. January) volunteers discovered two more lost cages. The cage at the Smith and Minor Islands Aquatic Reserve - Joseph Whidbey State Park site was also found washed up in the high intertidal area, likely another storm event loss, and the cage at the site called Fauntleroy was found almost completely buried in sand, likely a result of a storm surge that moved sand along the shoreline in January (Lanksbury et al., 2013). We were unable to process mussels from these three cages.

Two sites received nine cages each, as part of a companion study on small-spatial scale distribution of toxics in mussels (see Averaging of Hylebos Waterway and Ruston Waterfront Sites and (Callahan et al., 2014). For the purposes of the current study however, we selected a central point from each of these locations to represent these sites; "Hylebos Waterway" (9 sites in the >50% %IS category) and the "Tacoma, Ruston Waterfront" (9 site in the >15-50% category). All biological and contaminant data were averaged within each of these sites and assigned to a central point along the 9-cage distribution.

3.2 Biological Endpoints

3.2.1 Survival and Mortality

Mussels survived the predeployment sorting and bagging proces well; only 5.36% (± 1.31 SE) died between the time they were sorted, measured and bagged and the time they were deployed. This resting phase also allowed mussels time to attach themselves to the deployment bag. In addition, on average over 80% of the mussels deployed at each site (i.e. cage) remained alive to the end of the study (Figure 6, Appendix B).

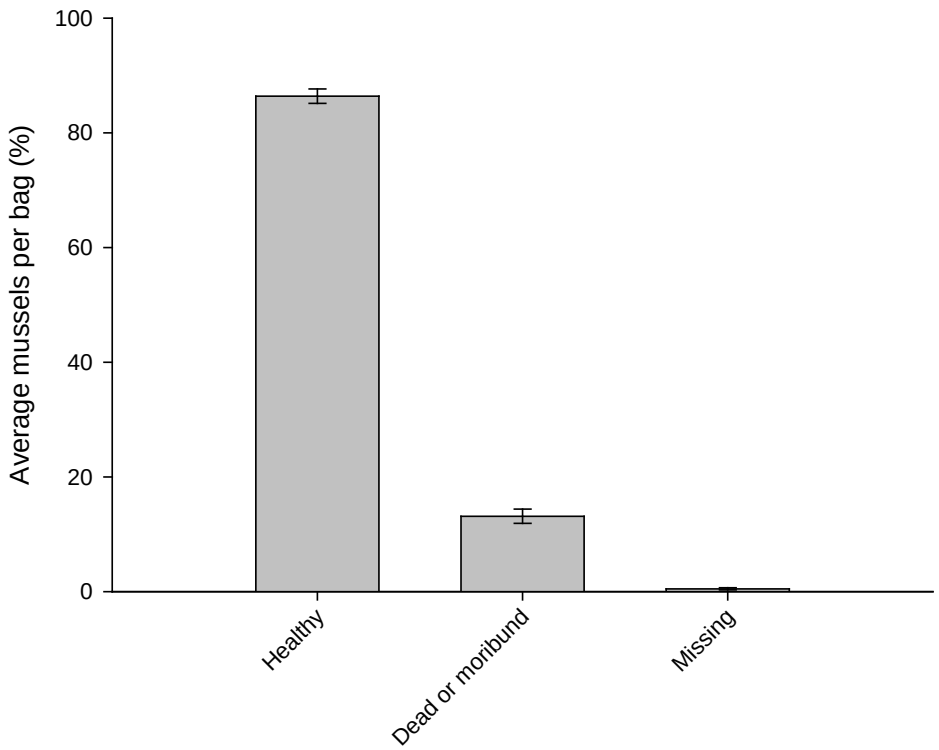

Figure 6. Average condition of bagged mussels deployed in 105 cages during study, n = 420 bags of mussels, 16 mussels per bag. Mean ± 95% confidence interval.

Mussel predators such as sea stars, including *Pycnopodia helianthoides*, and crabs, including *Pugettia producta*, were noted inside a few cages at the mid-point check and at the end of the study (Table 4). In addition, we noted drill holes in some of the empty mussel shells at the end of the study (Figure 7). From this latter evidence we assume that carnivorous snails, such as the dire whelk (*Lirabuccinum dirum*), wrinkled dogwinkle (*Nucella lamellose*) or Japanese oyster drill (*Ocinebrellus nornatus*), invaded some cages, though snails were not noted inside of any cages at the end of the study (Lanksbury et al., 2013).

Table 4. Mussel sites with predators found inside cages during the course of the study or with evidence of predation found on mussel shells (i.e. drill holes, crushed shells) during mortality assessment. Table reproduced from Lanksbury et al. (2013).

Site	**Predator found inside cage**	**Empty shells with drill holes**	**Mortality (%)**
Birch Point	-	1	10.9
Cherry Point Aquatic Reserve, 3 Alcoa-BP	sea star and crabs	-	6.3
Commencement Bay, Skookum Wuldge	-	1	28.1
Cypress Island Aquatic Reserve, Secret Harbor	-	1	12.5
Cypress Island Aquatic Reserve, Strawberry Bay	1 sea star	1	7.8
Des Moines Marina City Beach Park	*Pycnopodia helianthoides*	-	14.1
*Eagle Harbor, Bainbridge Ferry Terminal	*P. helianthoides*	>10	37.5
Gig Harbor, Narrows Passage	*P. helianthoides*	-	14.1
Hylebos Waterway 1	-	1	15.6
Johnson Point	3 – 4 *P. helianthoides*	-	18.8
Manchester, Stormwater Outfall	*Pugettia producta*	>1	6.3
Nisqually Reach Aquatic Reserve, Anderson Island	crabs	-	12.5
Suquamish, Stormwater Outfall	2 sea star, 1 crab	-	26.6
Tacoma Ruston Waterfront 1	2 sea star	-	26.6
Tacoma Ruston Waterfront 5	1 *P. producta*	-	10.9
Tacoma Ruston Waterfront 8	1 *P. producta*, 1 *P. helianthoides*	-	15.6
*Tolmie State Park	1 *P. helianthoides*	>4 (+12 crushed shells)	32.8

*Site removed from GLM assessment of relationship between mortality and degree of urbanization in the adjacent upland.

Figure 7. Eagle Harbor, Bainbridge Ferry Terminal cage at mid-point check (December 2, 2012). A hand-sized sunflower sea star (*Pycnopodia helianthoides*) was found inside, at the bottom of the cage. A kelp crab (*Pugettia producta*) was hanging on the outside of the cage. Figure from Lanksbury et al. (2013).

We did observed a weak positive relationship between mortality and both proxies for degree of urbanization in the adjacent uplands (Figure 8 and Figure 9). Mortality increased slightly but significantly with both impervious surface (%IS, $p = 0.003$, adjusted $r^2 = 0.087$) and road area (%RA, $p = 0.002$, adjusted $r^2 = 0.097$). Lipids, CI, and days of expsosure were not significant covariates in either of the models ($p>0.05$ for each when included in the stepwise multiple linear regression model). The regression analyses did not include the Eagle Harbor, Bainbridge Ferry Terminal and Tolmie State Park sites because a large amount of empty shells from those two sites contained drill holes or were crushed, both obvious signs of predation.

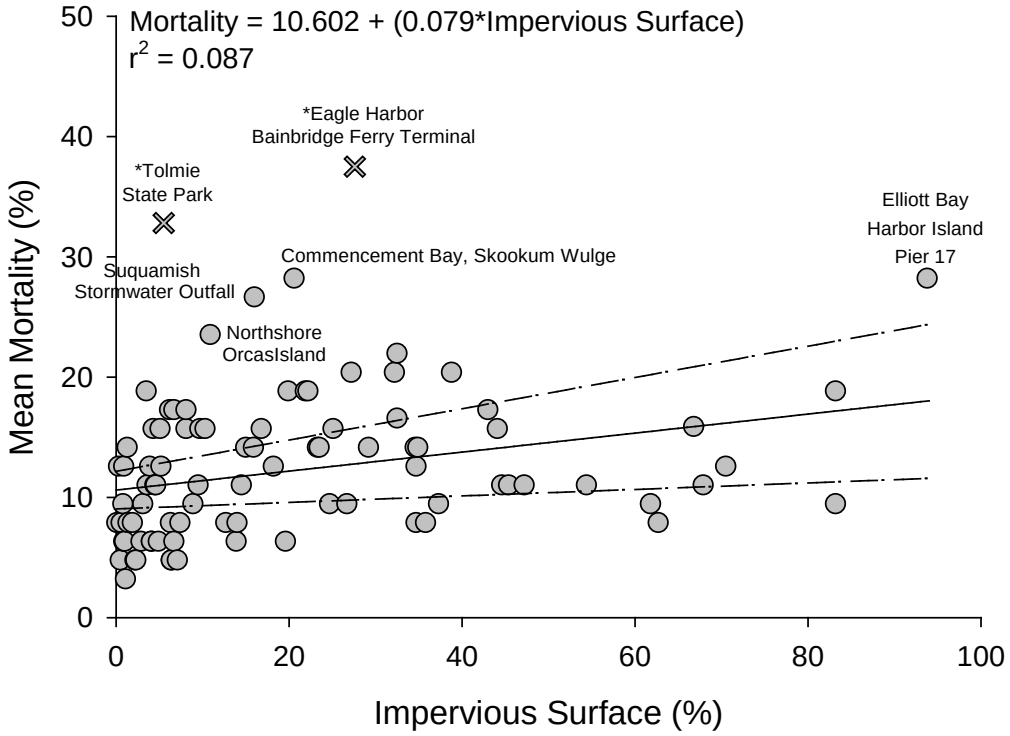

Figure 8. Mussel mortality increased with percent impervious surface (stepwise multiple linear regression of mortality versus Impervious Surface; $p = 0.003$, $r^2 = 0.087$). Each dot represents a transplanted (i.e. caged) mussel site; X's represent sites not included in analysis due to obvious signs of predation; solid black line is the predicted regression curve; dotted black lines are 95% confidence intervals.

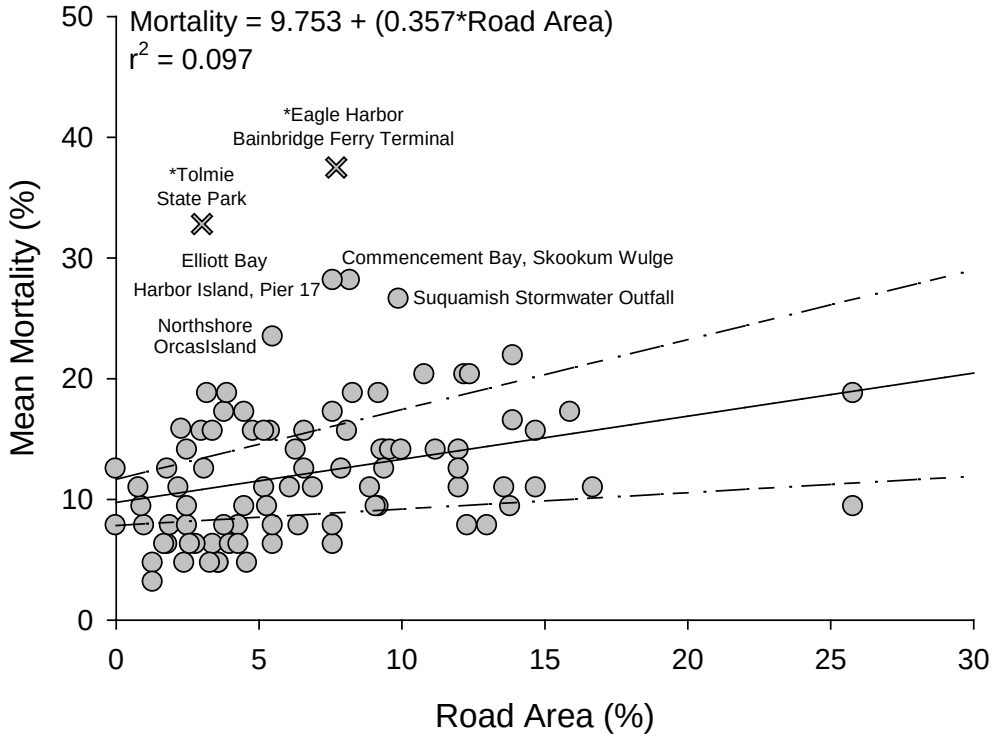

Figure 9. Mussel mortality increased with percent road area (stepwise multiple linear regression of mortality versus Road Area; $p = 0.002$, $r^2 = 0.097$). Each dot represents a transplanted (i.e. caged) mussel site; X's represent sites not included in analysis due to obvious signs of predation; solid black line is the predicted regression curve; dotted black lines are 95% confidence intervals.

3.2.2 Growth

Overall, mussels grew slightly during the 2 month deployment period, exhibiting an increase in shell length of approximately 0.8 mm, or 1.5%; Figure 10). Details for the average growth rate of mussels at each site are shown in Appendix B. Although the overall increase in shell length was significant (Mann-Whitney Rank Sum Test, $p < 0.001$), we observed no correlation between growth rate at the sites and the level of upland urbanization (linear regression of shell length increase versus %IS and versus %RA, $p > 0.05$ for both models).

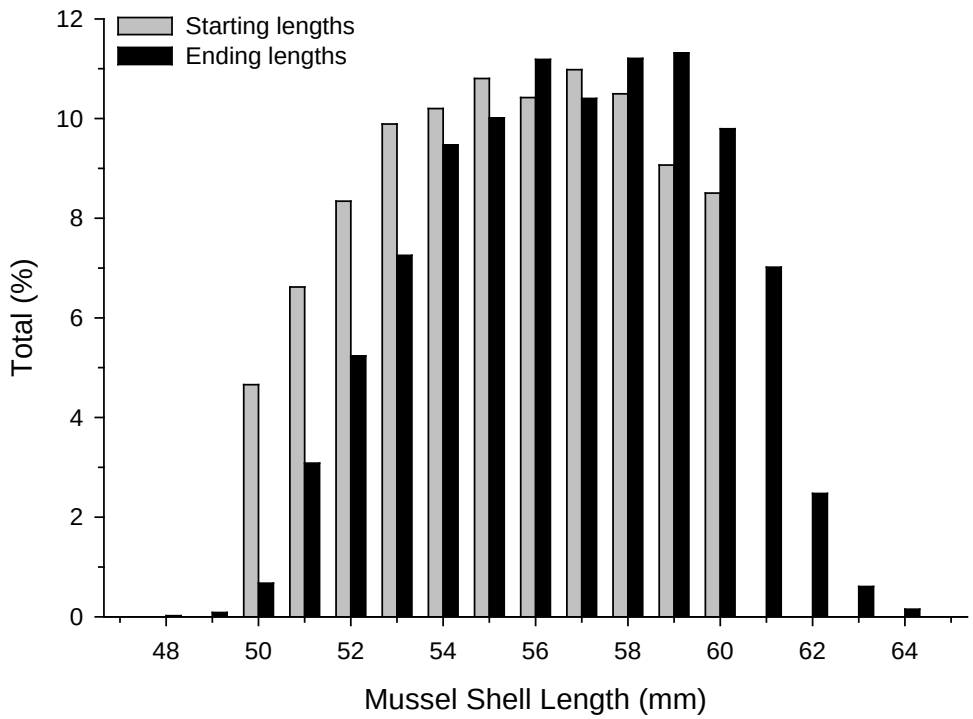

Figure 10. Distribution of starting (n = 6784) and ending (n = 4604) shell lengths of mussels deployed in cages for this study. Increase in shell length was significant (Mann-Whitney Rank Sum Test, p <0.001).

3.2.3 Condition Index

Although mussels grew in length overall, they exhibited a decline in CI, the measure of their mass of soft tissue, losing, on average, 0.4g dry mass/mm shell length. At the start of the study, the mean CI of mussels was 2.51 gm/mm ± 0.060 SE (Penn Cove, Baseline; n = 100 mussels). By the end of the study the average CI of the transplanted mussels from all the study sites (2.30 gm/mm ± 0.020 SE, n = 105 sites) was significantly lower than the starting CI ($t_{(203)} = 3.452$, $p<0.0001$, Figure 11). The control mussels held at Penn Cove under an aquaculture raft for the duration of the study (Penn Cove, Deployment Control; n = 101 mussels) also exhibited a significant drop in CI over the two months of the study (ending CI = 2.18 gm/mm dry ± 0.053 SE, $t_{(199)} = 4.118$, $p<0.0001$). Details for the CI of 12 mussels assessed from each site are shown in Appendix B. We note that the range of mussel CIs reported here (0.59 – 2.87 dw (g)/shell length (mm) are lower than those reported by Kagley et al. (2003): 4 –19 wet wt (g)/shell length (mm). This is because we used DRY tissue weight in our calculations, whereas Kagley et al. (2003) used WET tissue weight. When our CIs were recalculated using wet tissue weight, the range, 4.3 – 21.0 wet wt (g)/shell length (mm), matched those measured by Kagley et al. (2003) in 1992-1993.

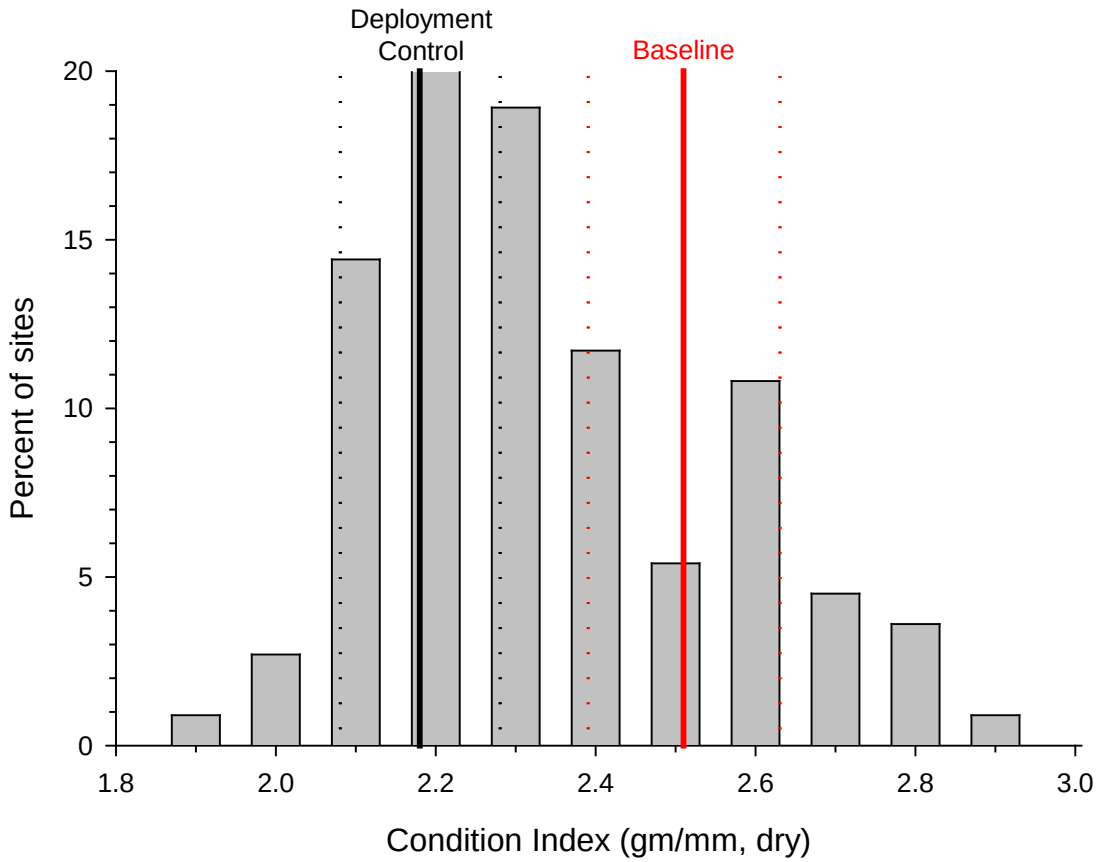

Figure 11. Frequency of condition index (CI) values exhibited by transplanted mussels at the end of the study (grey bars). Vertical solid and dotted lines indicate the mean ± 95% confidence intervals of Penn Cove Baseline mussels at the onset of the study (red) and Penn Cove Deployment Control mussels (black), measured at the termination of the study.

Although mussel CI declined at the majority of sites, the decline was not correlated with either of the two proxies for land development, impervious surface or road area (Figure 12).

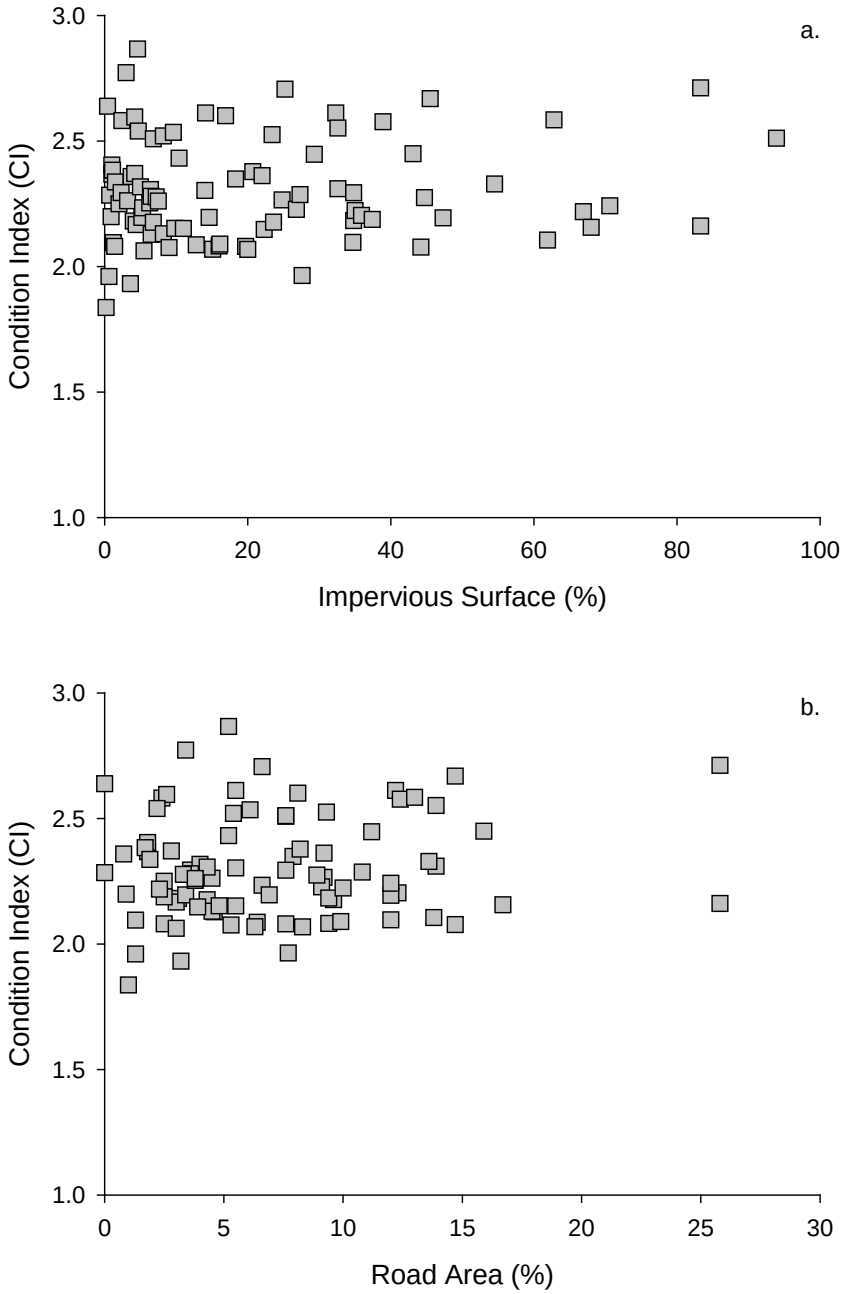

Figure 12. The condition index (CI) of transplanted mussels was not correlated with (a) upland impervious surface (stepwise multiple linear regression on CI data; p = 0.391) or (b) road area (p = 0.233). Each square represents a transplanted (i.e. caged) mussel site.

3.3 Overview of organic contaminant results

Overall, PAHs, PCBs, PBDEs, and DDTs were the most abundant organic contaminants measured in this study (Figure 13, Appendix C and Appendix D). PAHs and PCBs were detected in mussels from all 89 sites, PBDEs were detected at 84/89 sites, and DDTs at 82/89 sites. Chlordanes and dieldrin were less abundant, with Chlordanes detected at 14/89 sites and dieldrin detected at 10/89 sites. Two of the OCPs were rarely detected at the mussel transplant sites; 2/89 for hexachlorobenzene (HCB), 1/89 for Mirex, while the remaining three OCPs were not detected at any sites; 0/89 for aldrin, endosulfan 1, and hexachlorocyclohexanes (HCHs). Frequency

of detection for the eight detected organic contaminants ranged from 1 to 100% (Figure 13). A summary of the data quality review for the organic contaminants is available in Appendix G.

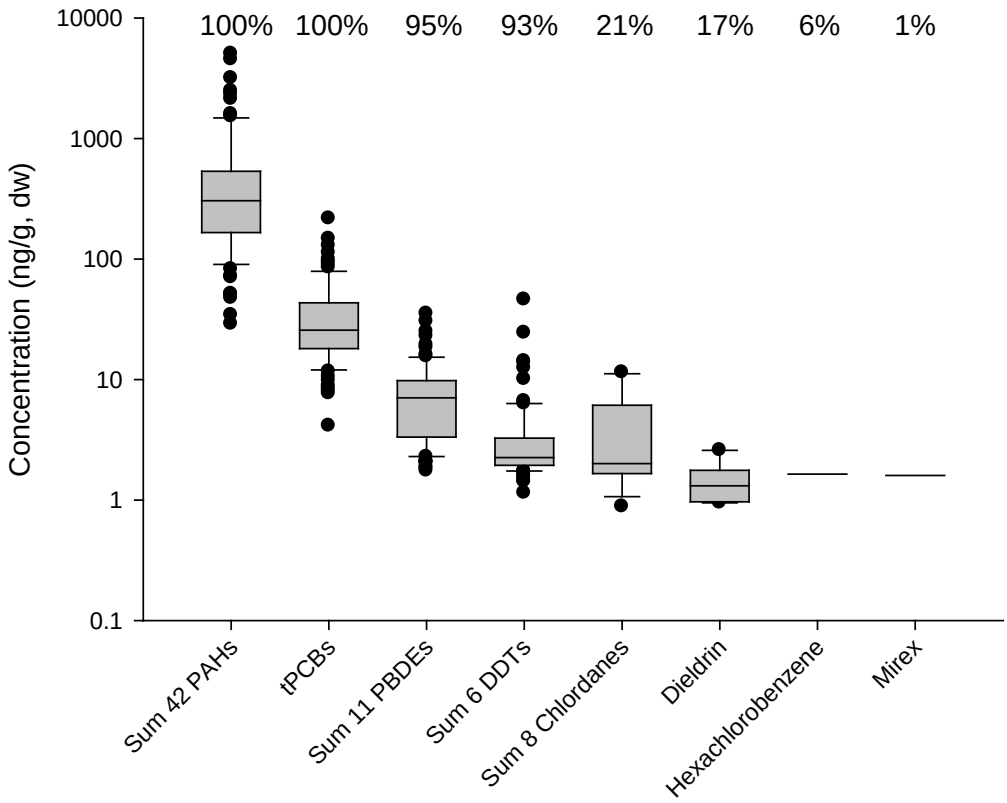

Figure 13. Range of concentrations for organic contaminants detected at transplanted mussel sites. Percent (%) of sites where contaminants were detected is indicated above each range.

We observed a statistically significant, positive correlation between the degree of upland urbanization and the concentration of Σ_{42}PAHs, TPCBs, Σ_{11}PBDEs, and Σ_{6}DDTs in mussels (Table 5 and Table 6). Details for each model are presented below, however in general, contaminants increased with both %IS and with Road Area (RA). Variability in contaminant concentration also increased substantially with increasing %IS or RA, such that while in most cases we observed the greatest contaminant concentration at the highest %IS or RA, we also observed low contaminant concentrations at some locations with high %IS or RA. We included CI and lipids[2] as covariates in the following multiple regression models. Due to the low number of detects, no linear regression analyses were performed on the $\sum_{8}$Chlordanes, dieldrin, HCB, Mirex, aldrin, endosulfan 1, or hexachlorocyclohexanes ($\sum_{3}$HCHs).

[2] These factor were not correlated with each other ($F_{(1, 87)} = 0.008$, $p = 0.927$), which satisfies the assumption of independence for factors in multiple linear regression.

Table 5. Results of the final regression models of the relationship between concentration (ng/g, dw) of organic contaminants in transplanted mussel tissue and the percent impervious surface (%IS) in adjacent upland watershed units. All chemical concentrations were log10 transformed for regression analyses.

Organic Contaminant	n	Slope		Y-intercept		Adj. r^2	ANOVA parameters	
		coefficient*	p-value	coefficient*	p-value		F-ratio (df = 1, 87)	p-value
$\sum_{42}$PAHs	89	1.03	<0.0001	166	<0.0001	0.372	53.035	<0.0001
TPCBs	89	1.01	<0.0001	20.7	<0.0001	0.193	21.979	<0.0001
$\sum_{11}$PBDEs	89	1.02	<0.0001	4.13	<0.0001	0.215	25.161	<0.0001
$\sum_{6}$DDTs	89	1.01	<0.0001	2.03	<0.0001	0.248	29.963	<0.0001

Chlordanes, dieldrin, HCB, Mirex, aldrin, endosulfan 1, and HCHs not analyzed due to low number of detects.

*Coefficients are back-transformed from $\log_{10}$ values.

Table 6. Results of the final regression models of the relationship between concentration (ng/g, dw) of organic contaminants in transplanted mussel tissue and the percent road area (%RA) in adjacent upland watershed units. All chemical concentrations were log10 transformed for regression analyses.

Organic Contaminant	n	Slope		Y-intercept		Adj. r^2	ANOVA parameters	
		coefficient*	p-value	coefficient*	p-value		F-ratio (df = 1, 87)	p-value
$\sum_{42}$PAHs	89	1.14	<0.0001	129	<0.0001	0.358	49.981	<0.0001
TPCBs	89	1.06	<0.0001	18.8	<0.0001	0.157	17.373	<0.0001
$\sum_{11}$PBDEs	89	1.08	<0.0001	3.43	<0.0001	0.254	30.971	<0.0001
$\sum_{6}$DDTs	89	1.06	<0.0001	1.87	<0.0001	0.187	21.257	<0.0001

Chlordanes, dieldrin, HCB, Mirex, aldrin, endosulfan 1, and HCHs not analyzed due to low number of detects.

*Coefficients are back-transformed from $\log_{10}$ values.

3.3.1 Total PAHs

$\sum_{42}$PAHs were detected in mussels from all of the transplanted sites, with concentrations ranging from 29 - 5030 ng/g dry weight (Figure 13, Appendix C and Appendix H). In addition, 93% of the sites had $\sum_{42}$PAH concentrations above the starting condition (Penn Cove, Baseline, n = 6: mean 71.36 ng/g dw, SD 20.385, shown in Figure 14 as a dotted line); see cumulative frequency distribution in Appendix H. PAHs declined from the initial condition at five locations.

Mussels placed inside UGAs (mean 857 ± 1065.7 ng/g, dw) accumulated significantly higher $\sum_{42}$PAH concentrations than mussels placed in non-UGAs (mean 285 ± 277.6 ng/g, dw) during the study; t-test of log-transformed $\sum_{42}$PAH concentration in mussels by UGA classification, $t_{(87)} = 4.991$, $p < 0.0001$. The concentrations of $\sum_{42}$PAHs were also positively correlated with %IS (Table 5 and Figure 14). In early stages of the GLM analyses with %IS several interaction terms emerged as significant, yet contributed only a trivial amount to the explanatory power of the model. Thus, for simplicity we retained only %IS in the final model (Figure 14). The concentrations of $\sum_{42}$PAHs were also positively correlated with %RA (Table 6 and Figure 15). A number of mussel sites exhibited $\sum_{42}$PAH concentrations well above the 95% prediction interval for both models (i.e. Elliott Bay, Myrtle Edwards; Elliott Bay, Four-Mile Rock; Smith Cove; Eagle Harbor, Bainbridge Ferry Terminal; Point No Point; and Salmon Bay among others), indicating the presence of other explanatory factors that were not measured in this study (see map in Appendix H).

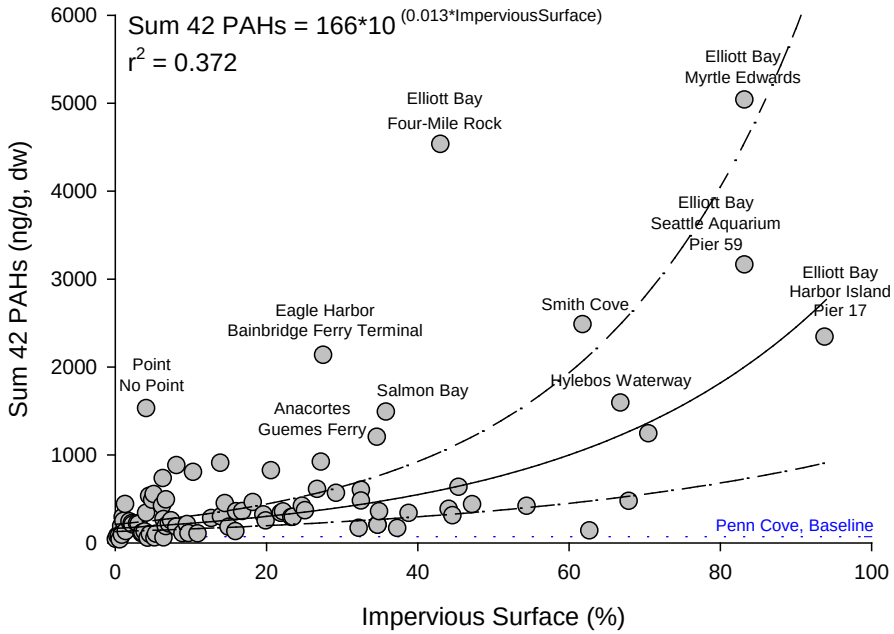

Figure 14. The concentration of $\sum_{42}$PAHs increased with percent impervious surface (stepwise multiple linear regression of log-transformed $\sum_{42}$PAHs versus Impervious Surface; $p < 0.0001$, $r^2 = 0.372$). Each dot represents a transplanted (i.e. caged) mussel site; solid black line is the predicted regression curve; dotted black lines are the 95% confidence intervals. Dotted blue line is the mean concentration of $\sum_{42}$PAHs in mussels at the start of the study (Penn Cove, Baseline).

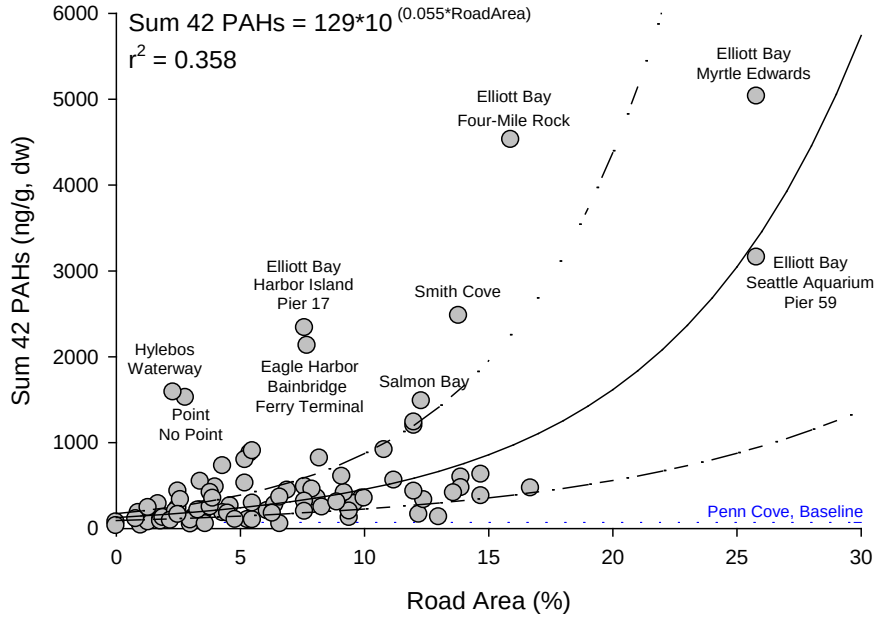

Figure 15. The concentration of $\sum_{42}$PAHs increased with percent road area (stepwise multiple linear regression of log-transformed $\sum_{42}$PAHs versus Road Area; $p < 0.0001$, $r^2 = 0.358$). Each dot represents a transplanted (i.e. caged) mussel site; solid black line is the predicted regression curve; dotted black lines are the 95% confidence intervals. Dotted blue line is the mean concentration of $\sum_{42}$PAHs in mussels at the start of the study (Penn Cove, Baseline).

3.3.2 Pattern Analysis of PAHs

The PAHs found in mussels from most of the study sites were dominated by three- and four-ring compounds; phenanthrene, pyrene, and their alkylated homologs. Four-ring (chrysene, benz[*a*]anthracene) and five-ring (benzo[*a*]pyrene and benzo[*e*]pyrene) compounds were also common, although less abundant, across most locations. Visual inspection of the PAH analyte histograms revealed a similar pattern across most of the mussel sites. Figure 16 shows three examples (reprinted from Appendix T) illustrating these typical PAH patterns. In general, for the most often detected homolog series the highest concentrations occurred at the parent analyte (i.e. C_0) with concentrations declining as degree of alkylation increased (i.e. $C_0 > C_1 > C_2 > C_3 > C_4$). The declining concentration of alkylated PAH analytes from C_0 through C_4 is often used as evidence for pyrogenic PAHs or highly weathered oil (Lima et al., 2005; Payne et al., 2003; Tobiszewski and Namieśnik, 2012) and was the pattern exhibited at the majority of our sites. The converse is true for petroleum sources, which was exhibited at four atypical sites. For three of the most frequently detected PAH analyte pairs at our typical locations (PHN/ANT, FLA/PYR, and BAA/CHR, Table 7), the parent compound (C_0) was detected at higher concentrations than any of their matching alkylated homologs at 96 - 99% of the mussel sites.

Table 7. Locations of the maximum analyte concentrations for the homolog series of three of the most frequently detected analytes. Percentages based on 96 total mussel sites.

	% of mussel sites with maximum at parent or alkylated homolog groups				
PAH homolog series	**C_0**	**C_1**	**C_2**	**C_3**	**C_4**
Phenanthrene (PHN)/anthracene (ANT)	97.9	0	0	0	2.1
fluoranthene (FLA)/pyrene (PYR)	99.0	0	0	0	0
benz[*a*]anthracene (BAA)/chrysene (CHR)	95.8	0	0	0	0

Four of the mussel sites; Salmon Bay, the Bremerton Shipyard-Charleston Beach, the Hylebos Waterway, and the Thea Foss Waterway exhibited atypical PAH analyte patterns; concentration across the PHN/ANT homolog series either increased, with maxima at C_4 for Salmon Bay and the Bremerton Shipyard-Charleston Beach (Figure 16, as compared with the Elliott Bay example), or remained relatively even across parent and alkyl groups (Hylebos and Thea Foss Waterways – Figure 17). In addition, these four atypical sites exhibited relatively high concentrations of dibenzothiophenes (DBTs) in relation to the rest of the PAH groups, with noticeable increases in concentrations as DBT homolog alkylation increased.

We further quantified these patterns by calculating two ratios that have been used forensically to distinguish between PAHs from petroleum and combustion sources. With a few exceptions[3] the ratios of Σalkylated-PHN/PHN[4] for the majority of our sites were well below 2, suggesting a dominance of pyrogenic PAHs. The Σalkylated-PHN/PHN ratios in mussels from the four atypical sites (Figure 17, bottom four sites) were among the highest in our study – all well above the 2 threshold: Thea Foss Waterway (2.5); Hylebos Waterway (3.3); Salmon Bay (4.7); and Bremerton Shipyard, Charleston Beach (5.5), suggesting petroleum sources of PAHs at these sites.

[3] Everett Harbor (2.3), Point Bolin and Shelton-Oak Bay Marina (2.5); Illahee Creek-Sinclair Inlet and Waterman Point (2.6)
[4] We only included sites where C_0 parent and C_{1-4} homolog analytes were detected above the LOQ.

We also used the ratio of FLU+PYR/Σalkylated-PHN to distinguish between pyrogenic (low ratio) and petrogenic (high ratio) PAHs. We observed lowest ratios from the Bremerton Shipyard-Charleston Beach (0.8), Salmon Bay (1.0), and Thea Foss Waterway (1.2) sites, adding weight to the evidence for petroleum as a source of PAHs in mussels from those locations. The majority of other sites exhibited higher ratios[5], ranging from 1.3 – 8.1, with the exception of Point Bolin (1.1) and the Protection Island Aquatic Reserve, Thompson Spit site (1.2).

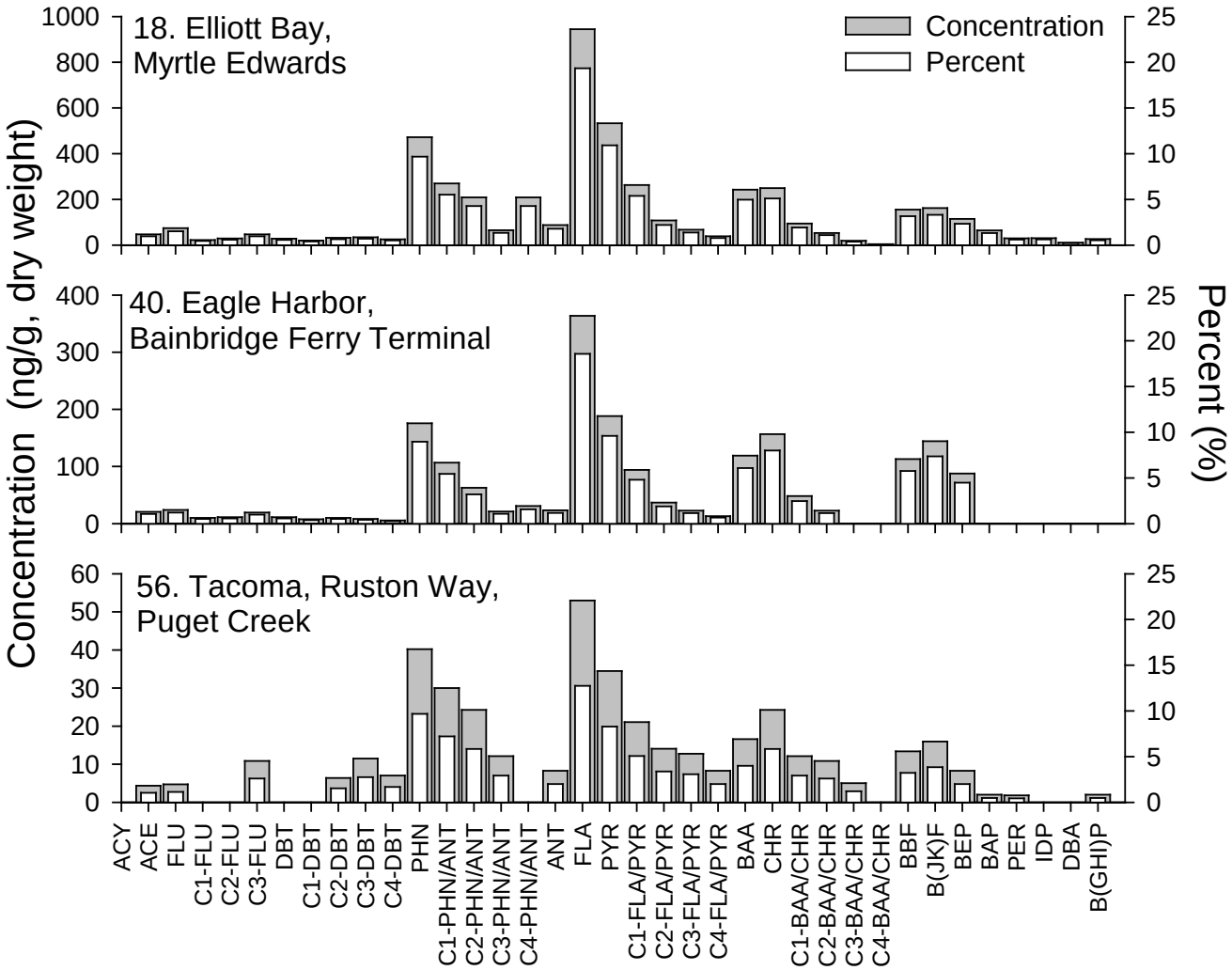

Figure 16. Histograms of PAH analytes detected in mussels from three sites in Puget Sound; these patterns were typical of those found at the majority of sites. See Table 2 for the names of the acronyms used along the X-axis. Histograms illustrating PAH fingerprints for the other mussel sites (96 total) are reported in Appendix T.

[5] We only included sites where C_0 parent and C_{1-4} homolog analytes were detected above the LOQ.

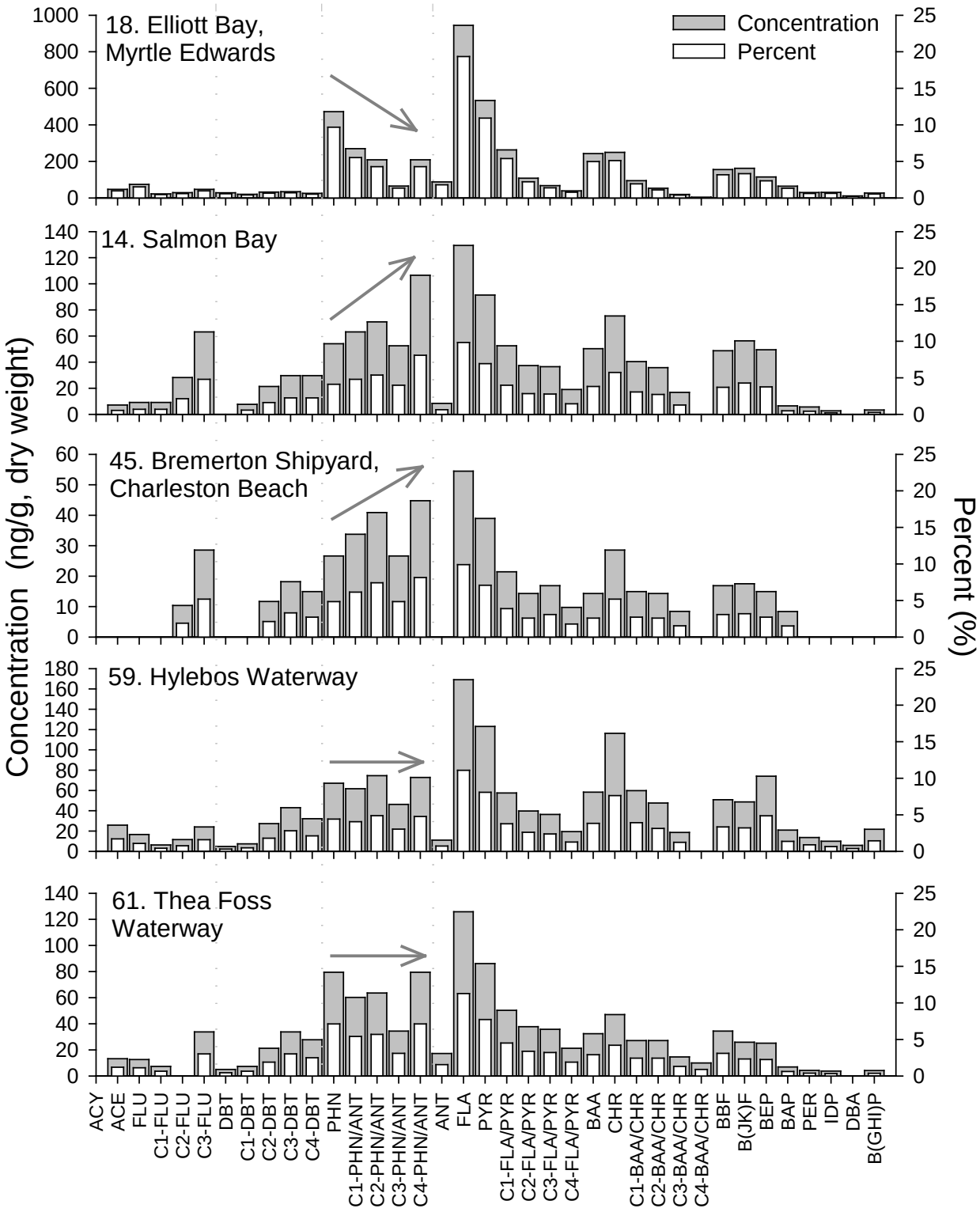

Figure 17. Histograms of PAH analytes detected in mussels from five sites in Puget Sound; the pattern at site 18 (Elliott Bay, Myrtle Edwards) is an example of that found at the majority of the study sites (see Figure 16). In contrast, atypical PAH analyte patterns were seen at sites 14, 45, 59 and 61. Note the change in direction of the phenanthrene/anthracene series (PHN, C1-C4-PHN/ANT; see arrows) and the higher relative abundance of dibenzothiophenes (DBTs) to the other analyte groups at the latter four sites, as compared to site 18. See Table 2 for the names of the acronyms used along the X-axis. Histograms illustrating PAH fingerprints for the other mussel sites (96 total) are reported in Appendix T.

3.3.3 PCBs

PCBs were detected in mussels from all of the transplanted sites, ranging in concentration from 4.1 ng/g dw to 216 ng/g dw (Figure 13, Appendix C and Appendix I). Seventy-one percent (71%) of the sites showed an increase in TPCB concentrations over the starting condition (Penn Cove, Baseline, n = 6: mean 19.27 ng/g dw, ±6.762 s.d.); see cumulative frequency distribution in Appendix I. In addition, mussels placed inside UGAs accumulated significantly higher TPCB concentrations (mean 60.3 ± 96.63 ng/g, dw) than mussels placed in non-UGAs during the study (mean 24.2 ± 12.32 ng/g, dw); t-test of log-transformed TPCB concentration in mussels by UGA classification, $t_{(87)} = 3.440$, $p = 0.001$.

There was a significant positive, but weak correlation between TPCB concentrations and IS% (Table 5). Lipids emerged as a significant covariate ($p = 0.027$) in the stepwise multiple linear regressions and removing it reduced some of the explanatory power of the final model; mixed model with Lipids $r^2 = 0.229$, final model without Lipids %IS $r^2 = 0.193$. Thus, by itself %IS accounted for about 19% of the variability in TPCB concentration (Figure 18). During analysis of %RA lipids again emerged as a significant covariate ($p = 0.04$, $r^2 = 0.188$), but as with the previous analysis that factor was removed from the final model for simplicity. %RA and IS% were similar in the degree to which they explained TPCB concentration in mussels (Table 6 and Figure 19).

A number of mussel sites exhibited TPCB concentrations outside the values predicted by the final regression model, including Hylebos Waterway; Sinclair Inlet, Sinclair Marina; Bremerton Shipyard, Charleston Beach; Salmon Bay; Smith Cove; and West Bainbridge, Westwood and others (see map in Appendix I). Mussels placed inside UGAs accumulated significantly higher TPCB concentrations (mean = 60.3 ± 96.63 ng/g, dw) than mussels placed in non-UGAs during the study (24.2 ± 12.32 ng/g, dw); t-test of TPCB concentrations in mussels from UGAs versus non-UGAs, $t_{(87)} = 3.440$, $p = 0.001$).

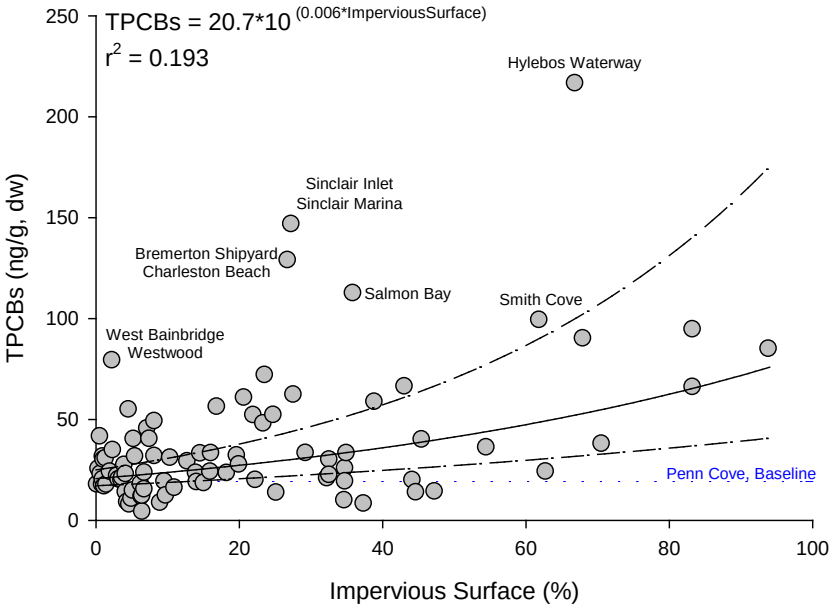

Figure 18. Estimated total PCB (TPCB) concentration increased with percent impervious surface (stepwise multiple linear regression of log-transformed TPCB versus Impervious Surface; $p < 0.0001$, $r^2 = 0.193$). Each dot represents a transplanted (i.e. caged) mussel site; solid black line is the predicted regression curve; dotted black lines are the 95% confidence intervals. Dotted blue line is the mean TPCB concentration in mussels at the start of the study (Penn Cove, Baseline).

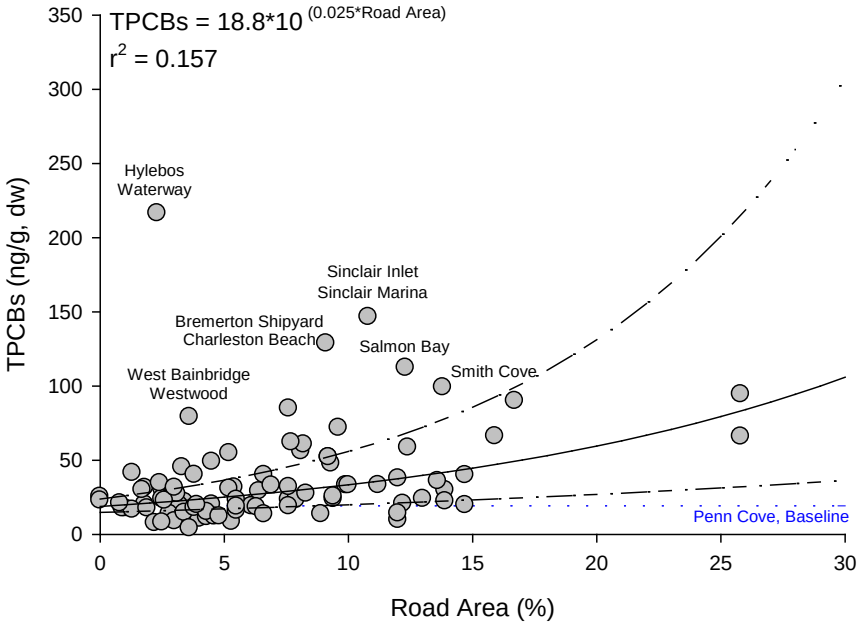

Figure 19. Estimated total PCB (TPCB) concentration increased with percent road area (stepwise multiple linear regression of log-transformed TPCB versus Road Area; $p < 0.0001$, $r^2 = 0.157$). Each dot represents a transplanted (i.e. caged) mussel site; solid black line is the predicted regression curve; dotted black lines are the 95% confidence intervals. Dotted blue line is the mean TPCB concentration in mussels at the start of the study (Penn Cove, Baseline).

3.3.4 PCB Ratios

Overall, the PCB28:(PCB28+PCB187) ratio was greatest in mussels from non-urbanized locations, and decreased with proximity to urban areas. Mussels from highly urbanized sites in Elliott Bay (Elliott Bay, Harbor Island, Pier 17; Elliott Bay, Four-Mile Rock; Elliott Bay, Myrtle Edwards), and two other urban embayments Sinclair Inlet (Sinclair Marina and Bremerton Shipyard Ferry Terminal), and Commencement Bay (Hylebos Waterway) exhibited the lowest ratio (i.e. had higher amounts of the heavier PCB187, relative to PCB28; Figure 20). Sites far removed from urban areas, including those in the northern Puget Sound area (San Juan, Whatcom County areas), Protection Island Aquatic Reserve in the Strait of Juan de Fuca, and a few sites in the south Puget Sound (Nisqually Reach Aquatic Reserve; Tolmie State Park; Totten Inlet) showed a "lighter" PCB signal characterized by a greater proportion of PCB28. The PCB pattern in mussels from the Penn Cove Baseline was similar to the other non-urban mussels, with a ratio of 0.79.

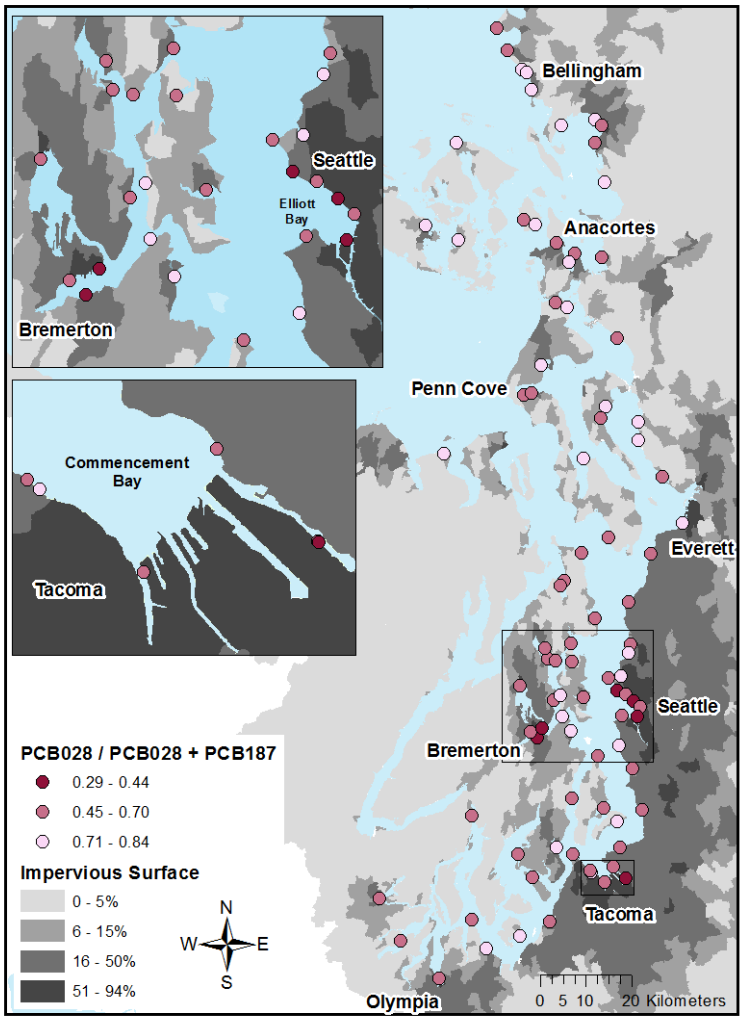

Figure 20. Map of the distribution of trichlorobiphenyl (PCB028) to heptachlorbiphenyl (PCB187) homolog ratios (PCB28:PCB28+PCB187) in transplanted mussels. Each dot represents a transplanted (i.e. caged) mussel site.

3.3.5 Total PBDEs

PBDEs were detected in mussels from all of the transplanted sites, ranging in concentration from 1.7 - 35 ng/g dw (Appendix C and Appendix J). Seventy-eight percent (78%) of the sites had $\sum_{11}$PBDE concentrations above

the starting condition (Penn Cove, Baseline, n = 6: mean 2.819 ng/g dw, ± 1.365 s.d.; see cumulative frequency distribution in Appendix J). In addition, mussels placed inside UGAs accumulated significantly higher $\sum_{11}$PBDE concentrations (mean 12.3 ± 17.66 ng/g, dw) than mussels placed in non-UGAs during the study (mean 5.85 ± 4.360 ng/g, dw); t-test of log-transformed $\sum_{11}$PBDE concentrations in mussels by UGA classification, $t_{(87)} = 3.554$, $p = 0.001$).

There was a significant positive relationship between the concentration of $\sum_{11}$PBDEs and %IS in adjacent upland watersheds, with no significant covariates (Table 5). Percent IS accounted for 21.5% of the variability in $\sum_{11}$PBDE concentrations (Figure 21). In a separate model, %RA explained slightly more of the variability in the $\sum_{11}$PBDE concentration (25.4%) than did %IS (Table 6 and Figure 22). As with the TPBCs, a number of mussel sites exhibited $\sum_{11}$PBDEs concentrations well above and below the 95% confidence intervals, with Bremerton Shipyard, Charleston Beach; Commencement Bay, Skookum Wuldge; Hylebos Waterway; and Salmon Bay well above the upper confidence interval (Figure 21, Figure 22, and Appendix J)

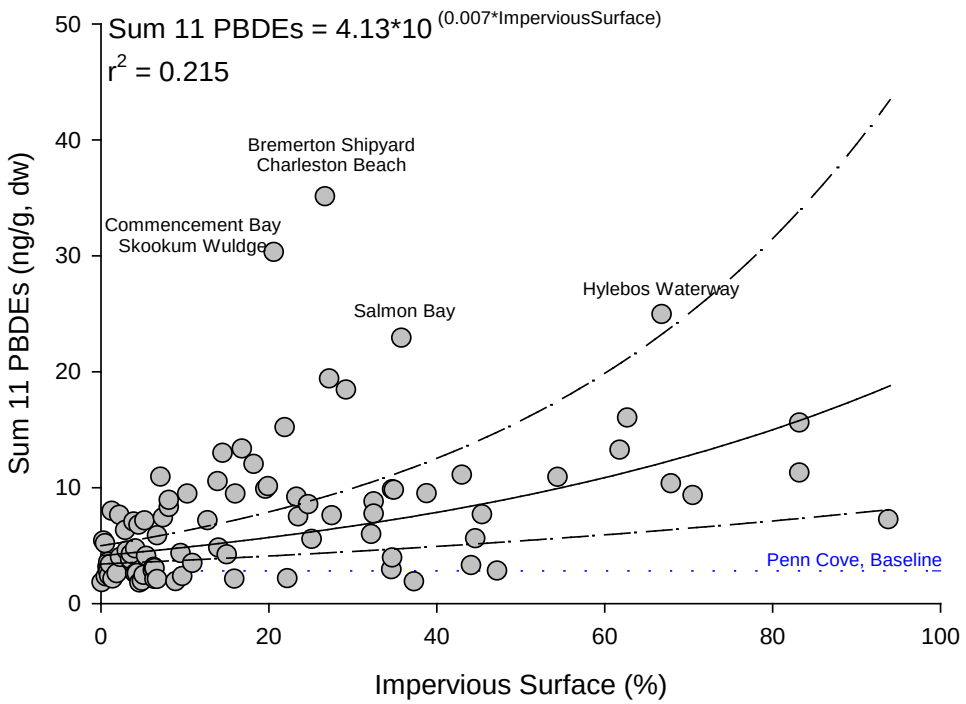

Figure 21. The concentration of $\sum_{11}$PBDEs increased with percent impervious surface (stepwise multiple linear regression of log-transformed $\sum_{11}$PBDE versus Impervious Surface; $p < 0.0001$, $r^2 = 0.215$). Each dot represents a transplanted (i.e. caged) mussel site; solid black line is the predicted regression curve; dotted black lines are the 95% confidence intervals. Dotted blue line is the mean $\sum_{11}$PBDE concentration in mussels at the start of the study (Penn Cove, Baseline).

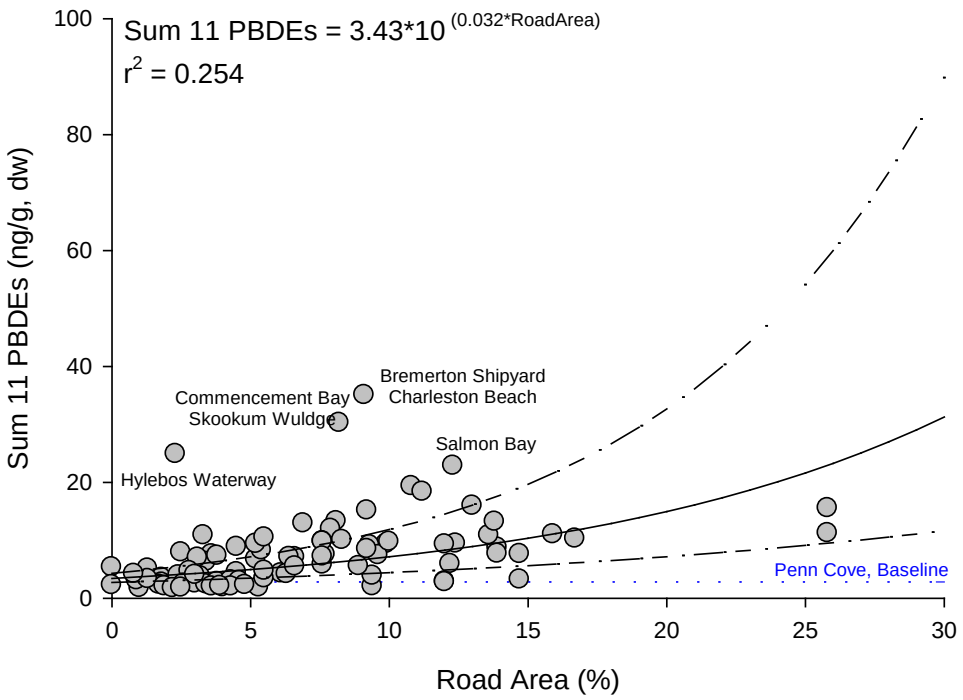

Figure 22. The concentration of ∑11PBDEs increased with percent road area stepwise multiple linear regression on log-transformed ∑11PBDE versus Road Area; $p < 0.0001$, $r^2 = 0.254$). Each dot represents a transplanted (i.e. caged) mussel site; solid black line is the predicted regression curve; dotted black lines are the 95% confidence intervals. Dotted blue line is the mean ∑11PBDE concentration in mussels at the start of the study (Penn Cove, Baseline).

3.3.6 Total DDTs

DDTs were detected in mussels from all of the transplanted sites, with concentrations ranging from 1.1 - 46 ng/g dw (Appendix C and Appendix K). All sites (100%) had $\sum_6$DDTs concentrations above the starting condition (Penn Cove, Baseline, n = 6: mean 1.12 ng/g dw, ±0.04749 s.d.; see cumulative frequency distribution in Appendix K). In addition, mussels placed inside UGAs accumulated significantly higher $\sum_6$DDT concentrations (mean 6.85 ± 14.96 ng/g, dw) than mussels placed in non-UGAs (mean 2.32 ± 1.006 ng/g, dw); t-test of log-transformed $\sum_6$DDT concentrations in mussels by UGA classification, $t_{(87)} = 3.426$, $p = 0.001$.

GLM analysis revealed a weak, significant positive relationship between IS% and $\sum_6$DDT concentrations (Table 5). During GLM analyses the interaction term Lipids*%IS ($p = 0.005$) emerged as a significant covariate. Although the multi-factor model had a somewhat higher r^2 (0.295) than the final, single-factor model (retaining only %IS; $r^2 = 0.248$), we omitted the interaction term from the final model for simplicity. Thus, %IS itself accounted for about 25% of the variability in $\sum_6$DDT concentrations (Figure 23). In a separate model the %RA was also a significant predictor of the concentration of $\sum_6$DDTs in mussels, however this model was not an improvement over the %IS model in explaining variability in $\sum_6$DDT concentrations (Table 6 and Figure 24).

As with the other organic contaminants already analyzed, a number of mussel sites exhibited $\sum_6$DDT concentrations above and below the 95% confidence intervals, suggesting the model could be improved with the addition of other explanatory factors not measured in this study. In particular, Hylebos Waterway; Salmon Bay;

Commencement Bay, Skookum Wuldge; and Elliott Bay, Four-Mile Rock exhibited DDT concentrations well above the upper confidence interval (Figure 23, Figure 24 and Appendix K).

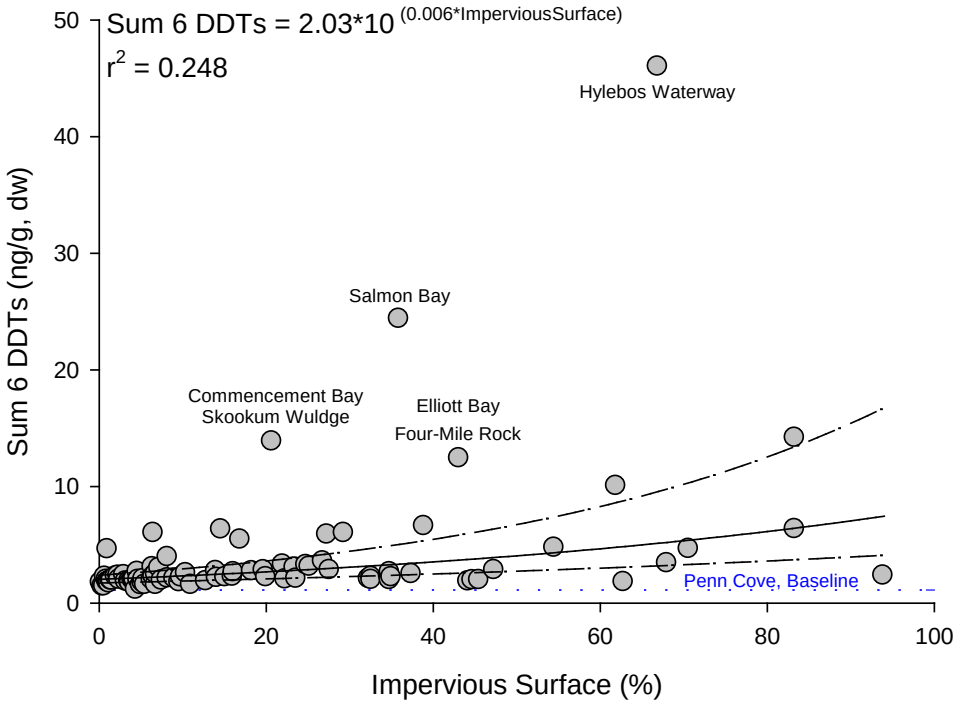

Figure 23. The concentration of $\sum_6$DDTs increased with percent impervious surface (stepwise multiple linear regression on log-transformed $\sum_6$DDT versus Impervious Surface; $p < 0.0001$, $r^2 = 0.248$). Each dot represents a transplanted (i.e. caged) mussel site; solid black line is the predicted regression curve; dotted black lines are the 95% confidence intervals. Dotted blue line is the mean $\sum_6$DDT concentration in mussels at the start of the study (Penn Cove, Baseline).

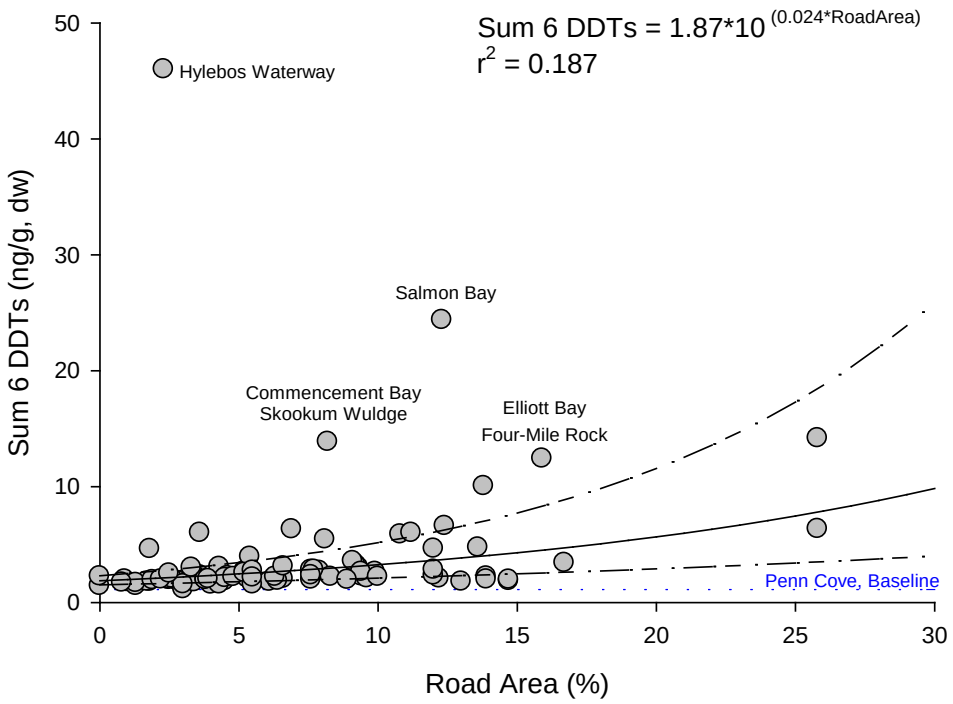

Figure 24. The concentration of ∑6DDTs increased with percent road area (stepwise multiple linear regression on log-transformed $\sum_6$DDT versus Road Area; $p < 0.0001$, $r^2 = 0.187$). Each dot represents a transplanted (i.e. caged) mussel site; solid black line is the predicted regression curve; dotted black lines are the 95% confidence intervals. Dotted blue line is the mean $\sum_6$DDT concentration in mussels at the start of the study (Penn Cove, Baseline).

3.3.7 Chlordanes

The $\sum_8$Chlordanes were detected above the limit of quantitation (LOQ) in mussels at 21% of the transplanted mussel sites and ranged in concentration from 0.88 to 11.42 ng/g dw (Appendix C and Appendix L). No $\sum_8$Chlordanes were detected in the Penn Cove, Baseline starting mussels (n = 6); all baseline concentrations were reported as below the LOQ (<2.1 ng/g dw). The LOQs for $\sum_8$Chlordanes ranged in concentration from 0.52 – 2.94 ng/g dw. Due to the low number of detects, many of which were near to the LOQ, no GLM analyses were performed on the $\sum_8$Chlordanes (Figure 25).

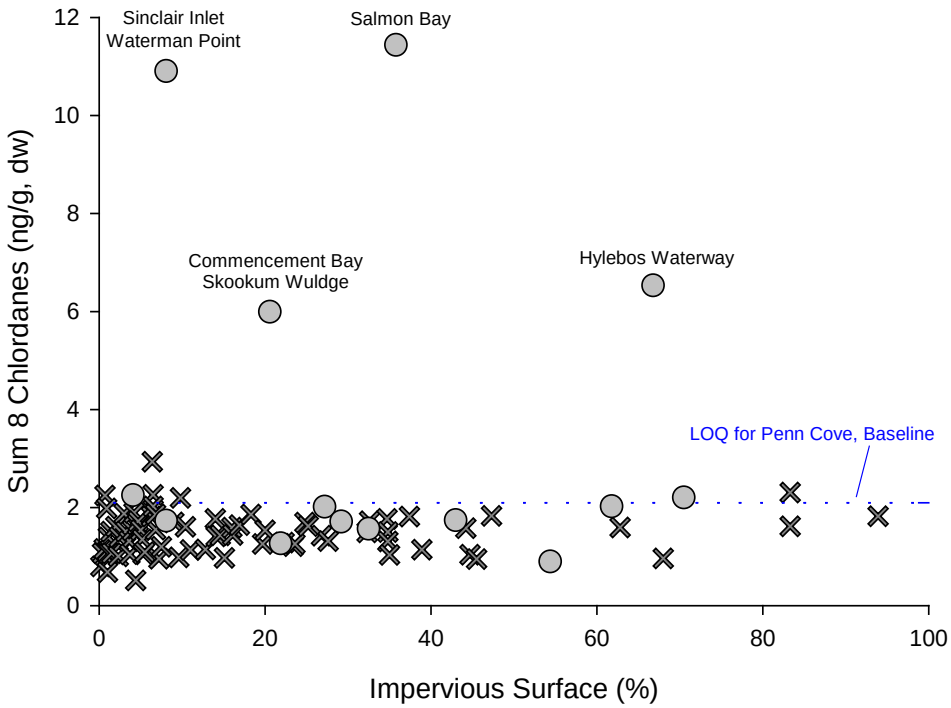

Figure 25. Concentration of $\sum_8$Chlordanes in relation to percent impervious surface. Each dot represents a transplanted (i.e. caged) mussel site; X's represent mussel cages where chlordane concentrations were detected below the limit of quantitation (<LOQ). $\sum_8$Chlordanes were not detected in any of the starting mussel samples (Penn Cove, Baseline, n = 6) and the dotted blue line indicates the $\sum_8$Chlordane LOQ for the baseline mussels.

3.3.8 Dieldrin

Minute amounts of dieldrin were detected at 17% of the transplanted mussel sites, and ranged in concentration from 0.95 to 2.59 ng/g dw (Appendix C and Appendix M). Dieldrin was not detected in any of the Penn Cove, Baseline starting mussels (n = 6), and all concentrations were reported as below the LOQ (<2.1 ng/g dw). Due to the low number of detected values and low range of detected values, dieldrin was not evaluated further in this study (Figure 26).

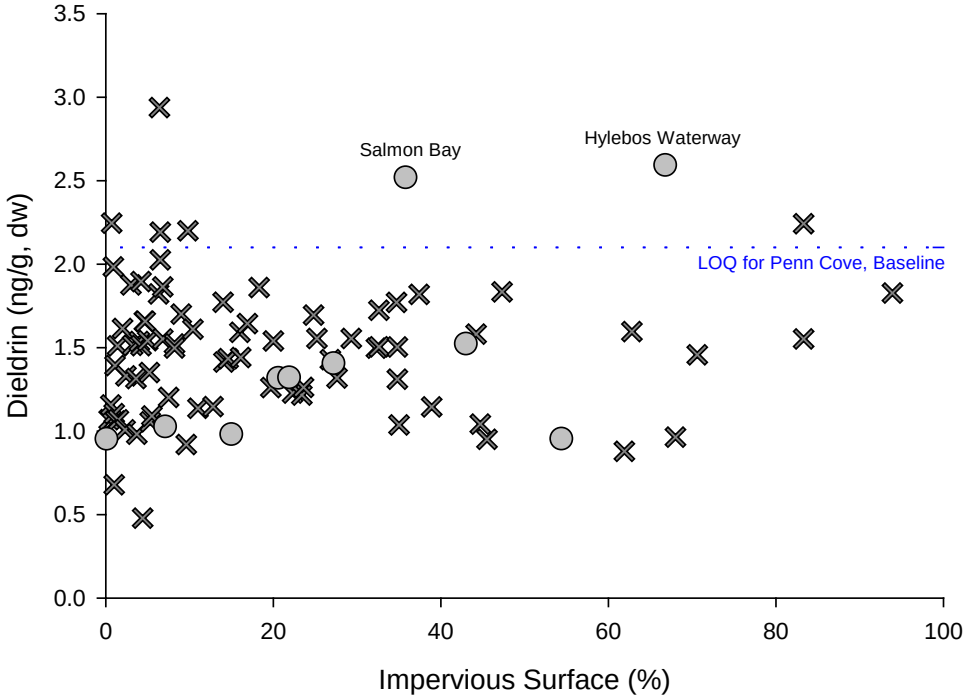

Figure 26. Concentration of dieldrin in relation to percent impervious surface. Each dot represents a transplanted (i.e. caged) mussel site; X's represent mussel cages where chlordane concentrations were detected below the limit of quantitation (<LOQ). No dieldrin was detected in any of the starting mussel samples (Penn Cove, Baseline, n = 6) and the dotted blue line indicates the dieldrin LOQ for the baseline mussels.

3.3.9 Hexachlorobenzene

Hexachlorobenzene was detected in minute quantities at two transplanted mussel sites (Manchester, Stormwater Outfall = 1.75 ng/g dw; Hylebos Waterway = 1.53 ng/g dw) (Appendix C). Hexachlorobenzene was not detected in any of the Penn Cove, Baseline starting mussels (n = 6); all baseline concentrations were reported as below the LOQ (<2.15 ng/g dw). Due to its absence from most of the study samples and the low detected concentrations, the hexachlorobenzene results were not evaluated further in this study.

3.3.10 Mirex

Though Mirex was detected at one transplanted mussel site (Sinclair Inlet, Sinclair Marina = 1.6 ng/g dw), the minute detected concentration was considered suspect because it fell within the range of LOQs for other mussel samples analyzed in the study (0.52 – 2.94 ng/g dw; Appendix C). Mirex was not detected in any of the Penn Cove, Baseline starting mussels (n = 6), and all concentrations were reported as below the LOQ (<2.1 ng/g dw).

3.3.11 Other Organohalogens

Aldrin, endosulfan 1, and hexachlorocyclohexanes (HCHs) were not detected above the LOQ in any of the transplanted mussels during this study. The LOQ for aldrin for all samples ranged in concentration from 0.48 – 2.94 ng/g dw, while the LOQs for endosulfan 1 and HCHs each ranged from 0.52 – 2.94 ng/g dw. Aldrin, endosulfan 1, and HCHs were not detected in any of the Penn Cove, Baseline starting mussels (n = 6), and all concentrations were reported as below the LOQ (<2.1 ng/g dw for all three contaminants).

3.4 Metals

3.4.1 Overview

The metals tested in this study were found at all 89 transplanted mussel sites (i.e. frequency of detection was 100%), with the majority of locations having concentrations above the starting condition (Figure 27, Appendix E and Appendix F). A summary of the data quality review for metals is available in Appendix G. Multiple linear regression analyses investigating the relationship between %IS and %RA and metal concentration in mussels revealed a weak, positive relationship with lead for both proxies of urbanization, weaker relationships with copper, and a weak relationship between zinc and %IS (Table 8 and Table 9). Lipid content, CI, and days of exposure were not significant covariates in any of the models. There was no significant relationship between mercury, arsenic or cadmium and either impervious surface or road area.

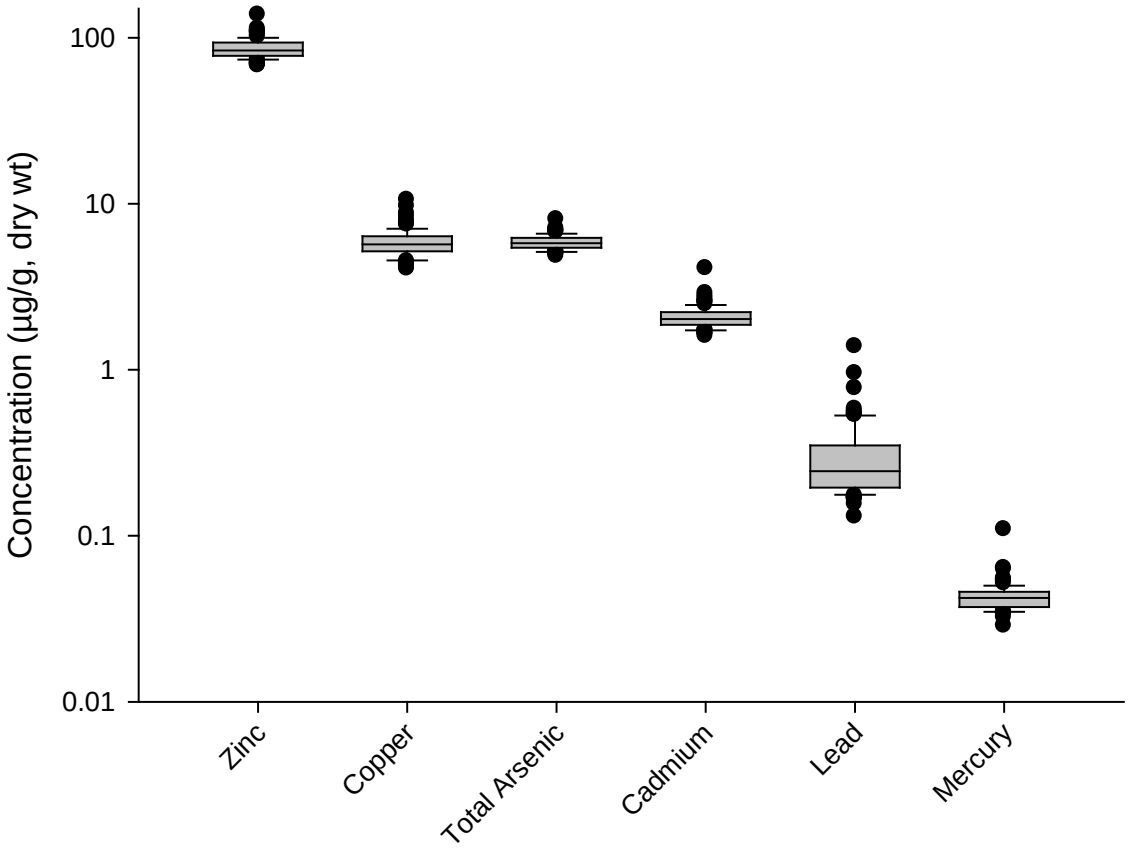

Figure 27. Range of concentrations for metals detected at transplanted mussel sites; all six metals were detected at 100% of sites.

Table 8. Results of the final multiple linear regression models of the relationship between concentration (µg/g, dw) of six metals in transplanted mussel tissue and the percent impervious surface (%IS) in adjacent upland watershed units. All metal concentrations were log10 transformed for regression analyses.

Metals	n	Slope		Y-intercept		Adj. r^2	ANOVA parameters	
		coefficient*	p-value	coefficient*	p-value		F-ratio (df = 1, 87)	p-value
Lead	89	1.01	<0.0001	0.223	<0.0001	0.198	22.749	<0.0001
Copper	89	1.002	0.002	5.46	<0.0001	0.098	10.603	<0.0001
Zinc	89	1.002	0.016	82.8	<0.0001	0.055	6.073	0.016
Mercury	89	-	-	-	-	-	-	NS
Arsenic	89	-	-	-	-	-	-	NS
Cadmium	89	-	-	-	-	-	-	NS

*Coefficients are back transformed from $\log_{10}$values. -

Table 9. Results of the final multiple linear regression models of the relationship between concentration (µg/g, dw) of six metals in transplanted mussel tissue and the percent road area (%RA) in adjacent upland watershed units. All metal concentrations were log10- transformed for regression analyses.

Metals	n	Slope		Y-intercept		Adj. r^2	ANOVA parameters	
		coefficient*	p-value	coefficient*	p-value		F-ratio (df = 1, 87)	p-value
Lead	89	1.05	<0.0001	0.196	<0.0001	0.274	34.224	<0.0001
Copper	89	1.009	0.016	5.42	<0.0001	0.054	6.026	0.016
Zinc	89	-	-	-	-	-	-	NS
Mercury	89	-	-	-	-	-	-	NS
Arsenic	89	-	-	-	-	-	-	NS
Cadmium	89	-	-	-	-	-	-	NS

*Coefficients are back transformed from $\log_{10}$ values.

3.4.2 Lead

Lead was detected in mussels from all of the transplanted sites, ranging in concentration from 0.13 - 1.38 µg/g dw (Appendix E and Appendix N). All sites (100%) had lead concentrations above the starting condition (Penn Cove, Baseline, n = 6: mean 0.1273 µg/g dw, ±0.02194 s.d.; see cumulative frequency distribution in Appendix N). Mussels placed inside UGAs accumulated significantly higher lead concentrations (mean 0.359 ± 0.2150 µg/g, dw) than mussels placed in non-UGAs (mean 0.280 ± 0.2081 µg/g, dw); t-test of log-transformed lead concentrations in mussels by UGA classification, $t_{(87)} = 4.008$, $p < 0.0001$. There was also a weak positive relationship between lead concentrations and IS%, with impervious surface accounting for only 19.8% of the variability in lead (Table 8 and Figure 28). However, in a separate model, %RA explained a slightly larger portion (27.4%) of the variability in lead, than did %IS (Table 9 and Figure 29).

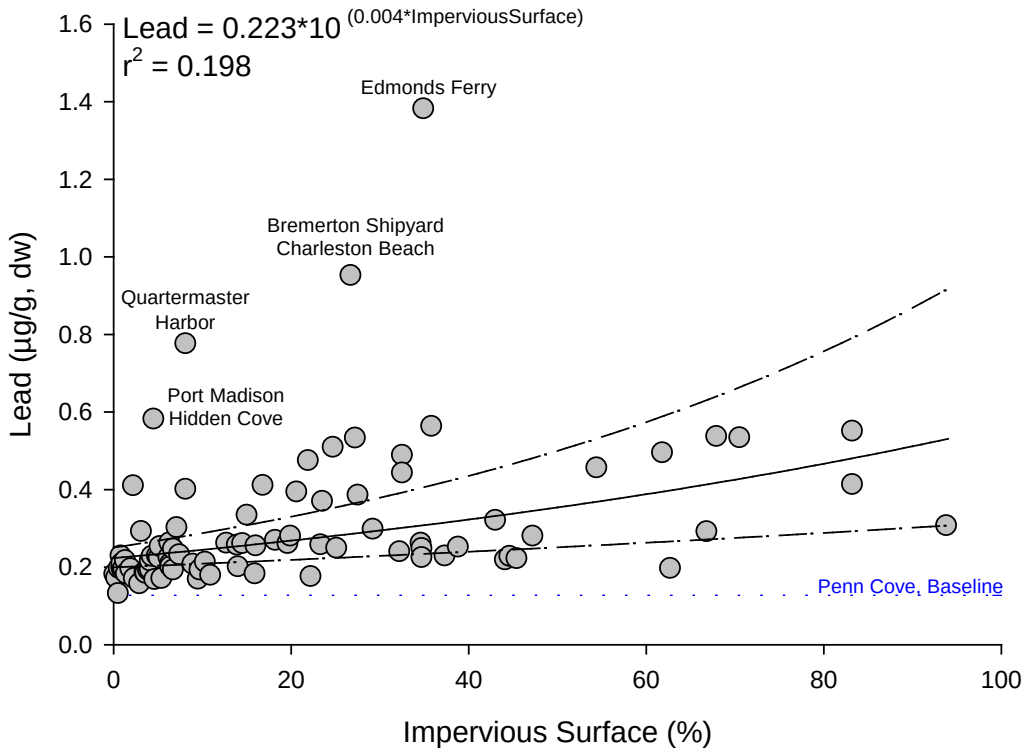

Figure 28. The concentration of lead increased with percent impervious surface (stepwise multiple linear regression on log-transformed lead versus Impervious Surface; $p < 0.0001$, $r^2 = 0.198$). Each dot represents a transplanted (i.e. caged) mussel site; solid black line is the linear regression; dotted black lines are the 95% confidence intervals. Dotted blue line is the mean lead concentration in mussels at the start of the study (Penn Cove, Baseline).

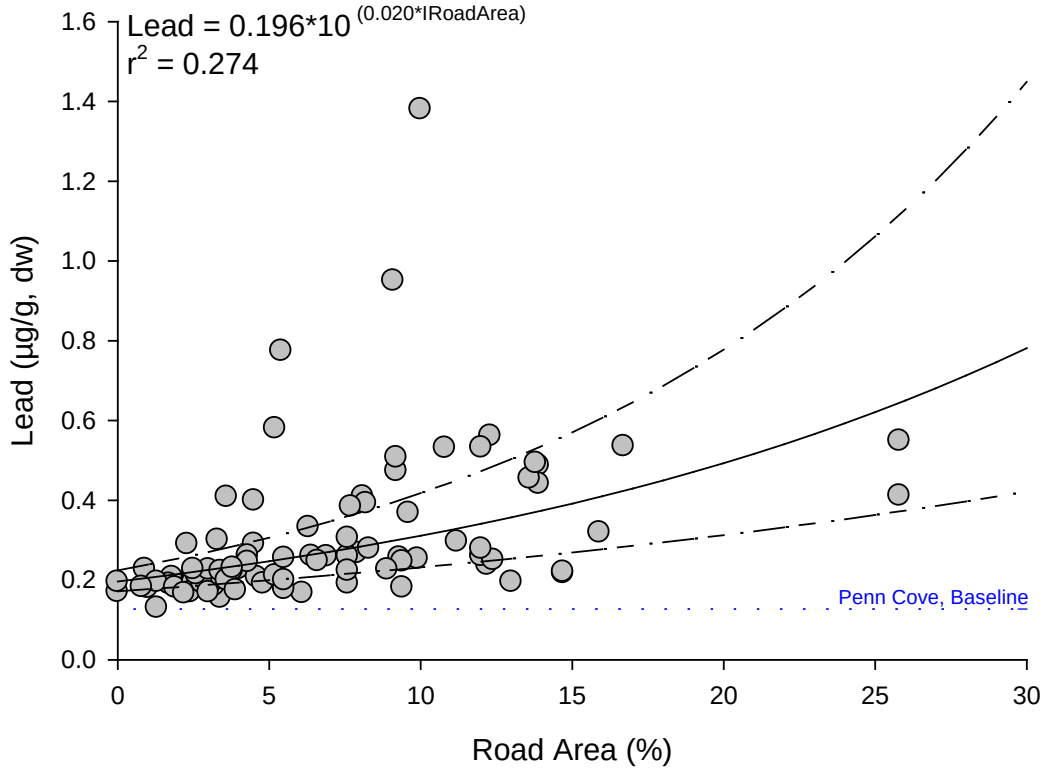

Figure 29. The concentration of lead increased with percent road area (stepwise multiple linear regression on log-transformed lead versus Road Area; $p < 0.0001$, $r^2 = 0.274$). Each dot represents a transplanted (i.e. caged) mussel site; solid black line is the predicted regression curve; dotted black lines are the 95% confidence intervals. Dotted blue line is the mean lead concentration in mussels at the start of the study (Penn Cove, Baseline).

3.4.3 Copper

Copper was detected in mussels from all of the transplanted sites, ranging in concentration from 4.05 – 10.5 µg/g dw (Appendix E and Appendix O). Copper concentrations were higher than the starting condition (Penn Cove, Baseline, n = 6: mean 4.977 µg/g dw, ± 0.5980 s.d.) at 84% of the sites; see cumulative frequency distribution in Appendix O. Mussels placed inside UGAs accumulated significantly higher copper concentrations (mean 6.32 ± 1.737 µg/g, dw) than mussels placed in non-UGAs (mean 5.88 ± 1.469 µg/g, dw); t-test of log-transformed copper concentrations in mussels by UGA classification, $t_{(87)} = 2.353$, $p = 0.021$. Although there was a weak positive relationship between copper concentration and %IS, impervious surface accounted for less than 10% of the variability in copper in this model (Table 8 and Figure 30). Although %RA was a significant predictor of copper concentration in mussels, unlike with lead the %RA model for copper was not an improvement over the %IS model for copper (Table 9 and Figure 31).

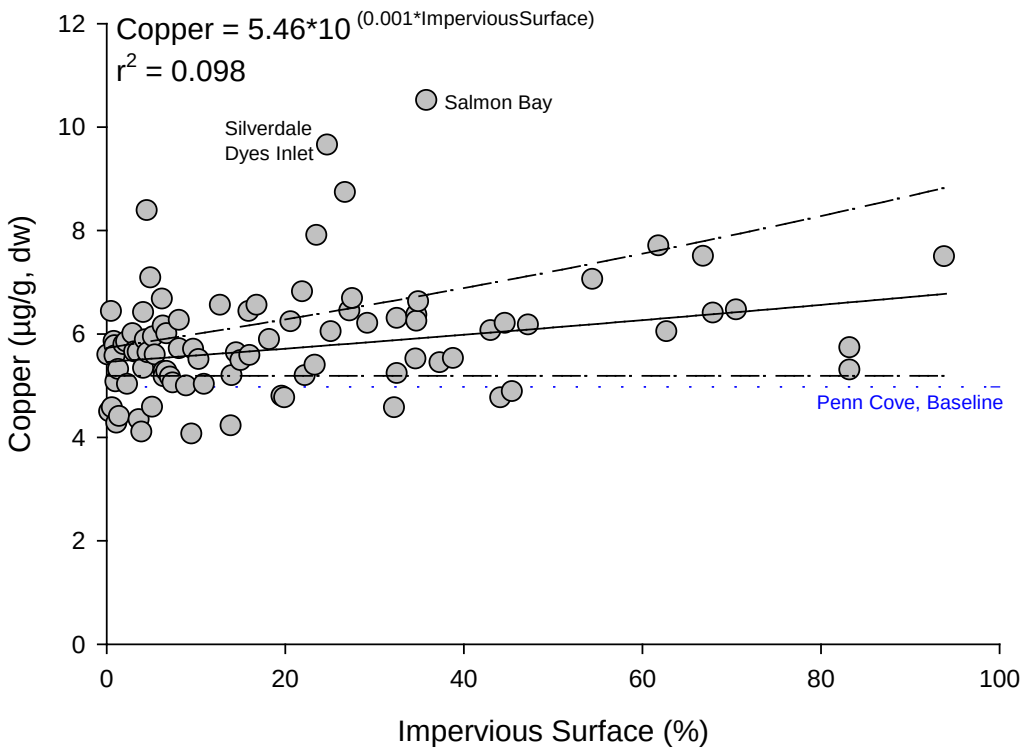

Figure 30. The concentration of copper increased with percent impervious surface (stepwise multiple linear regression on log-transformed copper versus Impervious Surface; $p < 0.0001$, $r^2 = 0.098$). Each dot represents a transplanted (i.e. caged) mussel site; solid black line is the predicted regression curve; dotted black lines are the 95% confidence intervals. Dotted blue line is the mean copper concentration in mussels at the start of the study (Penn Cove, Baseline).

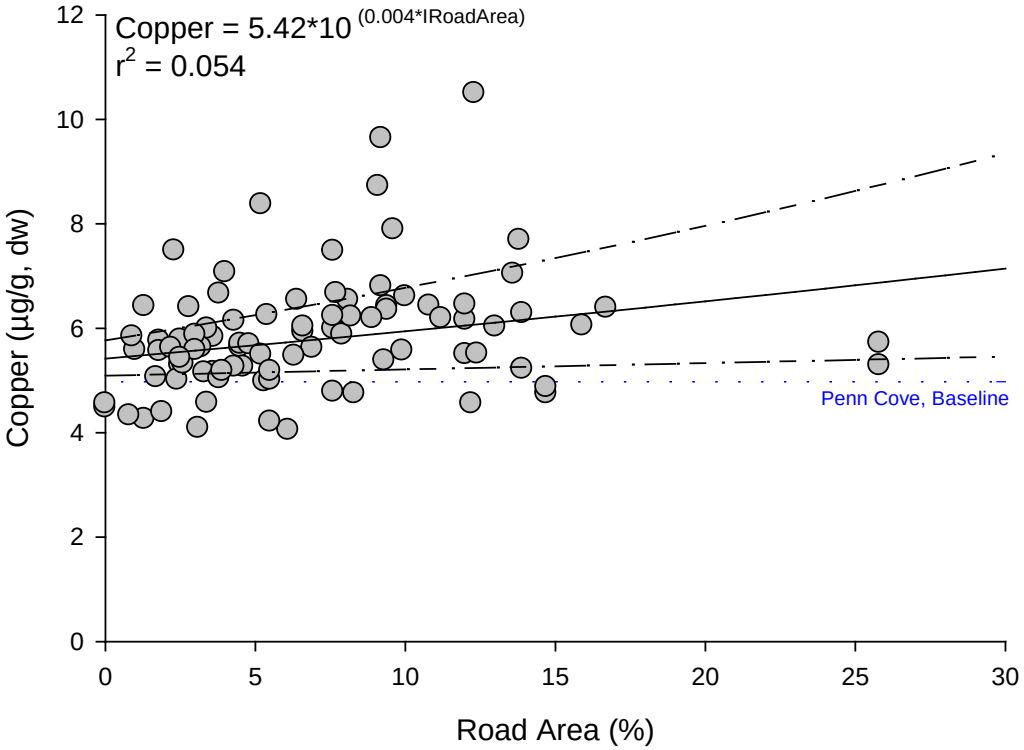

Figure 31. The concentration of copper increased with percent road area (stepwise multiple linear regression on log-transformed copper versus Road Area; $p = 0.016$, $r^2 = 0.054$). Each dot represents a transplanted (i.e. caged) mussel site; solid black line is the predicted regression curve; dotted black lines are the 95% confidence intervals. Dotted blue line is the mean copper concentration in mussels at the start of the study (Penn Cove, Baseline).

3.4.4 Zinc

Zinc was detected in mussels from all of the transplanted sites and ranged in concentration from 68 – 137 µg/g dw (Appendix E and Appendix P). Seventy-six percent (76%) of the sites had zinc concentrations above the starting condition (Penn Cove, Baseline, n = 6: mean 74.80 µg/g dw, ± 8.073 s.d.; see cumulative frequency distribution in Appendix P). Mussels placed inside UGAs accumulated significantly higher copper concentrations (mean 93.3 ± 27.71 µg/g, dw) than mussels placed in non-UGAs (mean 83.1 ± 10.83 µg/g, dw); t-test of log-transformed zinc concentrations in mussels by UGA classification, $t_{(87)} = 3.051$, $p = 0.003$. Similar to copper, zinc exhibited a very weak positive relationship with IS%, accounting for only 5.5% of the variability in the model (Table 8 and Figure 32). Road area was not a significant predictor of zinc concentration (Table 9).

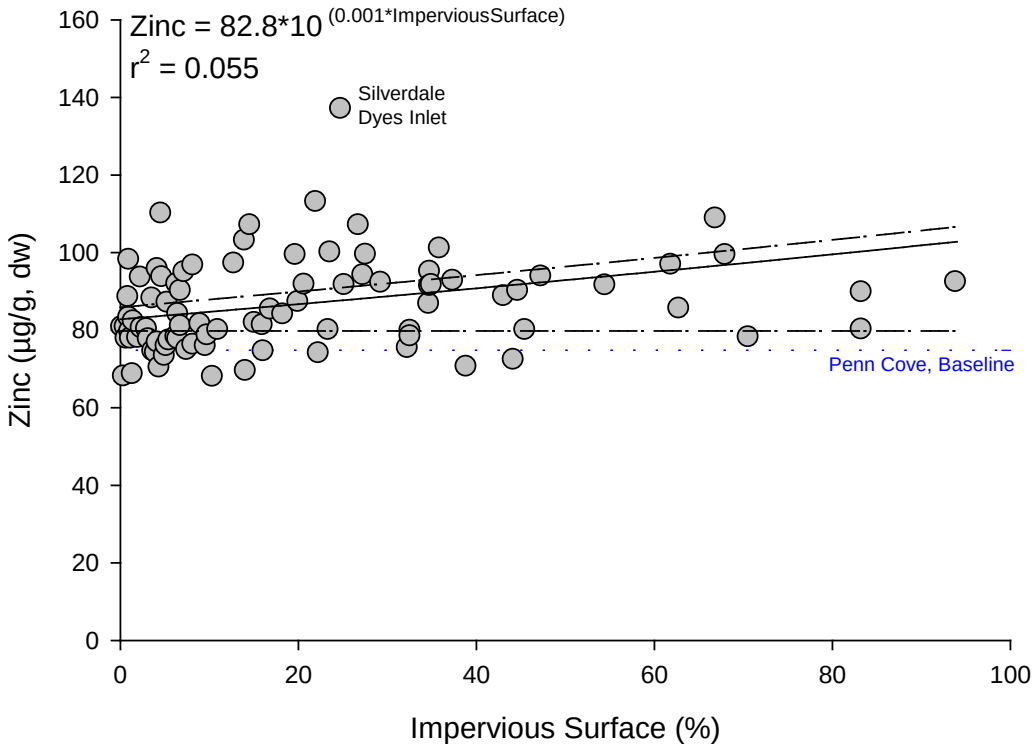

Figure 32. The concentration of zinc increased with percent impervious surface (stepwise multiple linear regression on log-transformed zinc versus Impervious Surface; $p = 0.016$, $r^2 = 0.055$). Each dot represents a transplanted (i.e. caged) mussel site; solid black line is the predicted regression curve; dotted black lines are the 95% confidence intervals. Dotted blue line is the mean zinc concentration in mussels at the start of the study (Penn Cove, Baseline).

3.4.5 Mercury

Mercury was detected in mussels from all of the transplanted sites and ranged in concentration from 0.03 - 0.11 µg/g dw (Appendix E and Appendix Q). Mercury concentrations at 99% of the sites were higher than the starting condition (Penn Cove, Baseline, n = 6: mean 0.0315 µg/g dw, ±0.0020 s.d.; see cumulative frequency distribution in Appendix Q). Unlike the previously mentioned metals, there was no significant difference in the concentration of mercury between mussels placed inside (mean 0.044 ± 0.0116 µg/g, dw) or outside (mean 0.049 ± 0.0322 µg/g, dw) the UGAs; t-test of log-transformed mercury concentrations in mussels by UGA classification, $t_{(87)} = 0.123$, $p = 0.902$.

%IS and %RA in the adjacent upland were not significant predictors of mercury concentrations in mussels (Table 8 and Table 9, Figure 33). The factor "days of exposure" (which varied from 54 - 62 days in this study) was the only significant covariate in early multi-factor models (model with %IS, days of exposure, CI and lipids; $F_{(4,84)} = 4.728$, $r^2 = 0.131$, $p = 0.000$). However, mercury concentrations declined (slightly) in mussels with days of exposure, both with and without including the unusually high Edmonds Ferry sample and the two Cypress Island Aquatic Reserve samples, which were the only ones collected at 62 days: model without those three high leverage sites had an $F(_{1,84}) = 7.648$, $r^2 = 0.073$, and $p = 0.007$ (Figure 34).

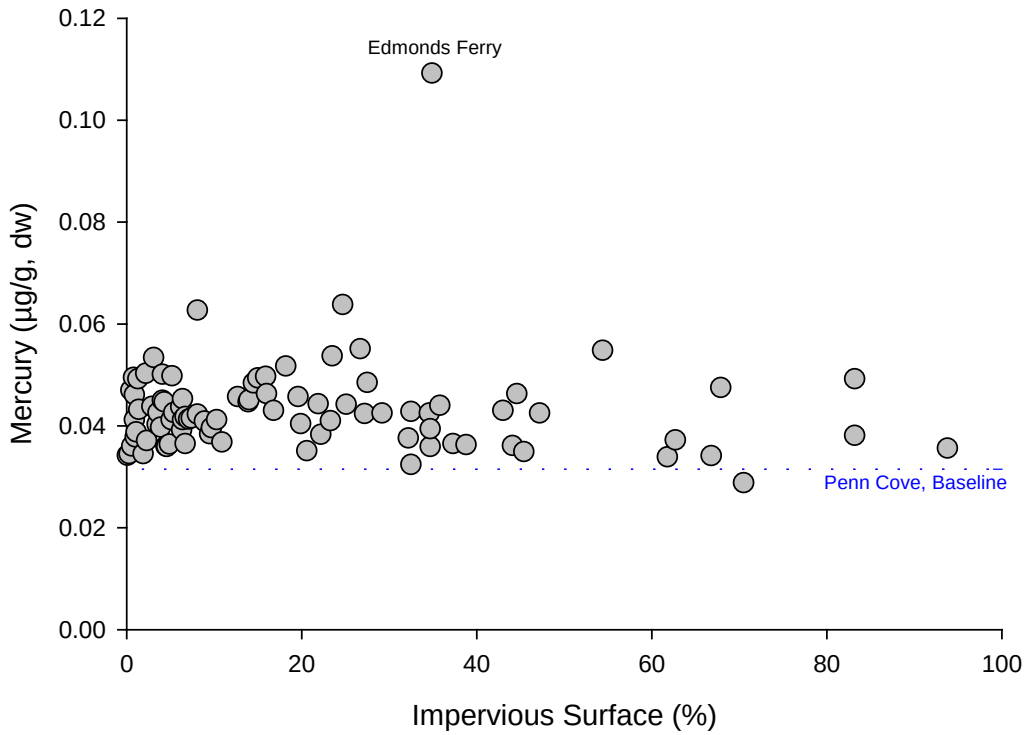

Figure 33. Mercury concentration in relation to percent impervious surface (no significant predictive relationship observed). Each dot represents a transplanted (i.e. caged) mussel site; dotted blue line is the mean mercury concentration in mussels at the start of the study (Penn Cove, Baseline).

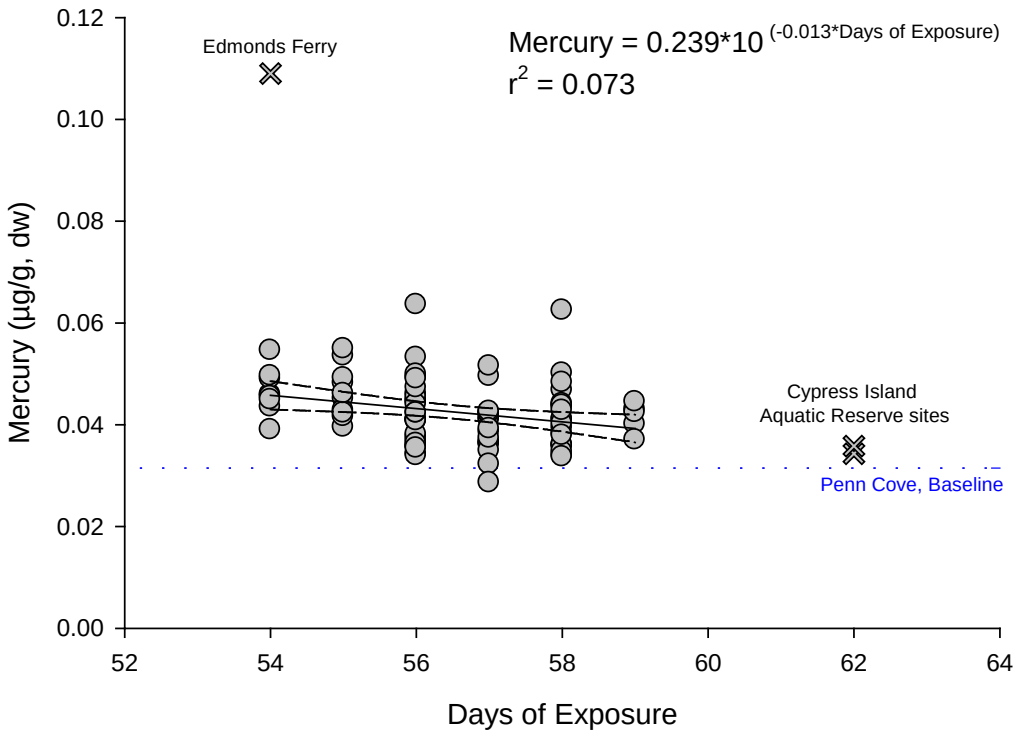

Figure 34. Mercury concentration decreased with days of exposure (stepwise multiple linear regression on log-transformed mercury versus Days of Exposure; $p = 0.007$, $r^2 = 0.073$). Each dot represents a transplanted (i.e. caged) mussel site; X's represent sites not included in analysis; solid black line is the predicted regression curve; dotted black lines are the 95% confidence intervals. Dotted blue line is the mean mercury concentration in mussels at the start of the study (Penn Cove, Baseline).

3.4.6 Arsenic and Cadmium

Total arsenic and cadmium were detected in mussels from all of the transplanted sites (Appendix E). Detected concentrations ranged from 4.83 - 8.02 µg/g dw for arsenic (Appendix R) and from 1.59 – 4.07 µg/g dw for cadmium (Appendix S). Eighty-three percent (83%) of the sites had higher arsenic concentrations over the starting condition (Penn Cove, Reference, n = 6: mean 5.28 µg/g dw, ±0.3396 s.d., see cumulative frequency distribution in Appendix R), but only 44% of the sites had higher cadmium concentrations (Penn Cove, Reference, n = 6: mean 2.04 µg/g dw, ± 0.2762 s.d., see cumulative frequency distribution in Appendix S). There was no significant difference in the concentration of arsenic in mussels placed inside (mean 5.86 ± 0.6188 µg/g, dw) or outside (mean 5.92 ± 0.5664 µg/g, dw) the UGAs; t-test of log-transformed arsenic concentrations in mussels by UGA classification, $t_{(87)} = -0.193$, $p = 0.848$. Likewise there was no significant difference in the concentration of cadmium in mussels placed inside (mean 2.10 ± 0.3798 µg/g, dw) or outside (mean 2.19 ± 0.6406 µg/g, dw) the UGAs; t-test of log-transformed cadmium concentrations in mussels by UGA classification, $t_{(87)} = 0.598$, $p = 0.551$. In addition, there was no correlation between the concentrations of arsenic or cadmium and upland impervious surface or road area in mussels from this study (Table 8 and Table 9 and Figure 35).

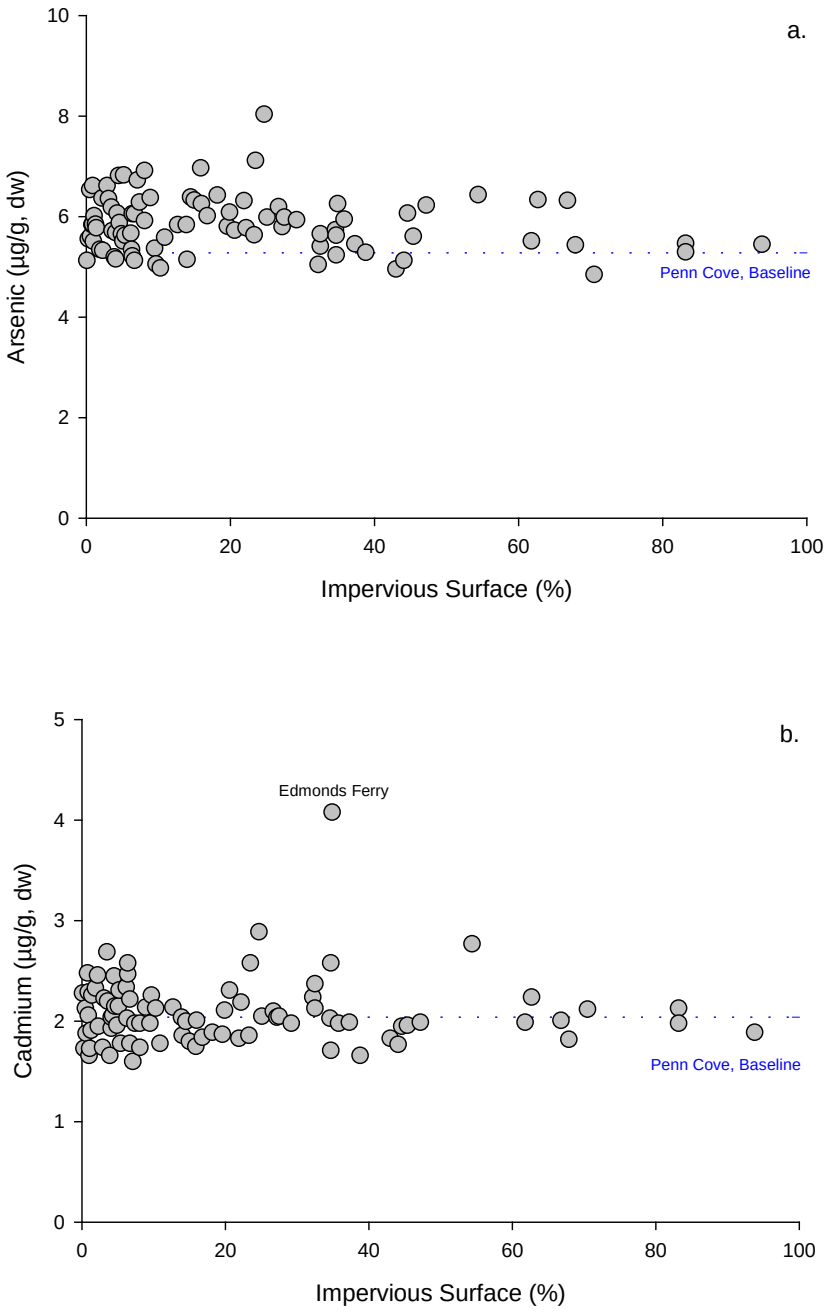

Figure 35. Concentrations of (a) arsenic and (b) cadmium in relation to percent impervious surface (no significant predictive relationships observed). Each dot represents a transplanted (i.e. caged) mussel site; dotted blue line is the mean concentration of (a) arsenic and (b) cadmium in mussels at the start of the study (Penn Cove, Baseline).

3.5 Comparison of Transplanted and Wild Mussels

At the end of the study exposure period, wild mussels were collected near the transplanted (i.e. caged) mussel sites during caged mussel retrieval at six locations; 1) Kayak Point, 2) Cavalero Beach County Park, 3) Hermosa Point, 4) Edmonds Ferry and 5) Everett Harbor in Snohomish County, and 6) in close proximity to Hylebos Waterway sites 1 and 2. Although this study was not designed to compare transplanted and wild mussels and the limited number of samples collected precluded a robust statistical analysis for contaminants, we

were able to perform statistical comparisons of the CIs. We graphically compared the ratios of contaminant concentrations from the tranplanted and wild mussels.

3.5.1 Condition Index

At each of the matching sites in Snohomish County the mean CIs of the wild mussels (n = 12) were significantly lower than the transplanted mussels (n = 12): Kayak Point (Mann-Whitney Rank Sum Test, U Statistic = 22.000, p = 0.004); Cavalero Beach County Park (Mann-Whitney Rank Sum Test, U Statistic = 0.000, p <0.001); Hermosa Point (t-test, $t_{(22)} = 4.388$, p <0.001); Edmonds Ferry (Mann-Whitney Rank Sum Test, U Statistic = 1.000); Everett Harbor (Mann-Whitney Rank Sum Test, U Statistic = 13.000, p <0.001). Similarly, at Tacoma's Hylebos Waterway the mean CI of wild mussels (n = 12) was significantly lower than the mean CI of the transplanted mussels from nearby Hylebos Waterway sites #1 and 2 (n = 24): t-test, $t_{(34)} = 4.274$, p <0.001 (Figure 36, Appendix B).

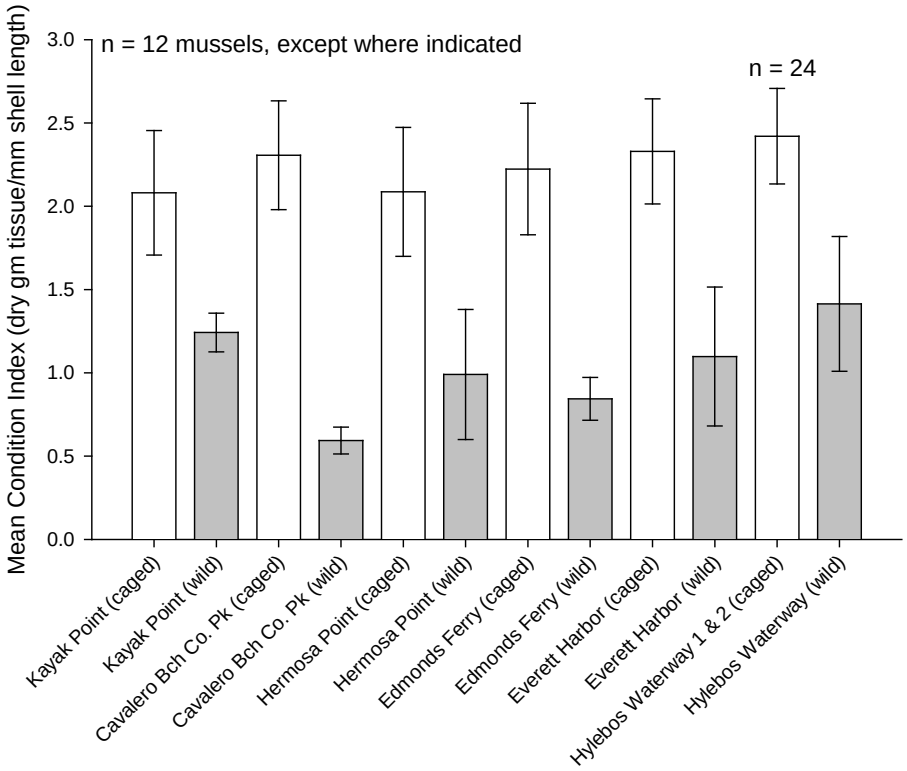

Figure 36. Condition index in transplanted/caged (C, white bars) and wild (N, solid grey bars) mussels collected at the same locations. Mean and 95% confidence intervals shown. All differences between matching sites were significant.

3.5.2 Contaminants

This study was not designed to compare the concentration of contaminants between transplanted and wild mussels, and wild mussels were only sampled from six sites, which precluded a robust statistical analysis. However a graphical comparison of the six paired samples shows many were near parity in their concentrations. The concentration ratios of PAHs, PCBs, PBDEs, DDTs, and all six metals were closely distributed around the

1:1 ratio line (Figure 37 and Figure 38). Contaminant data for the wild mussel collections are available in Appendices C - F.

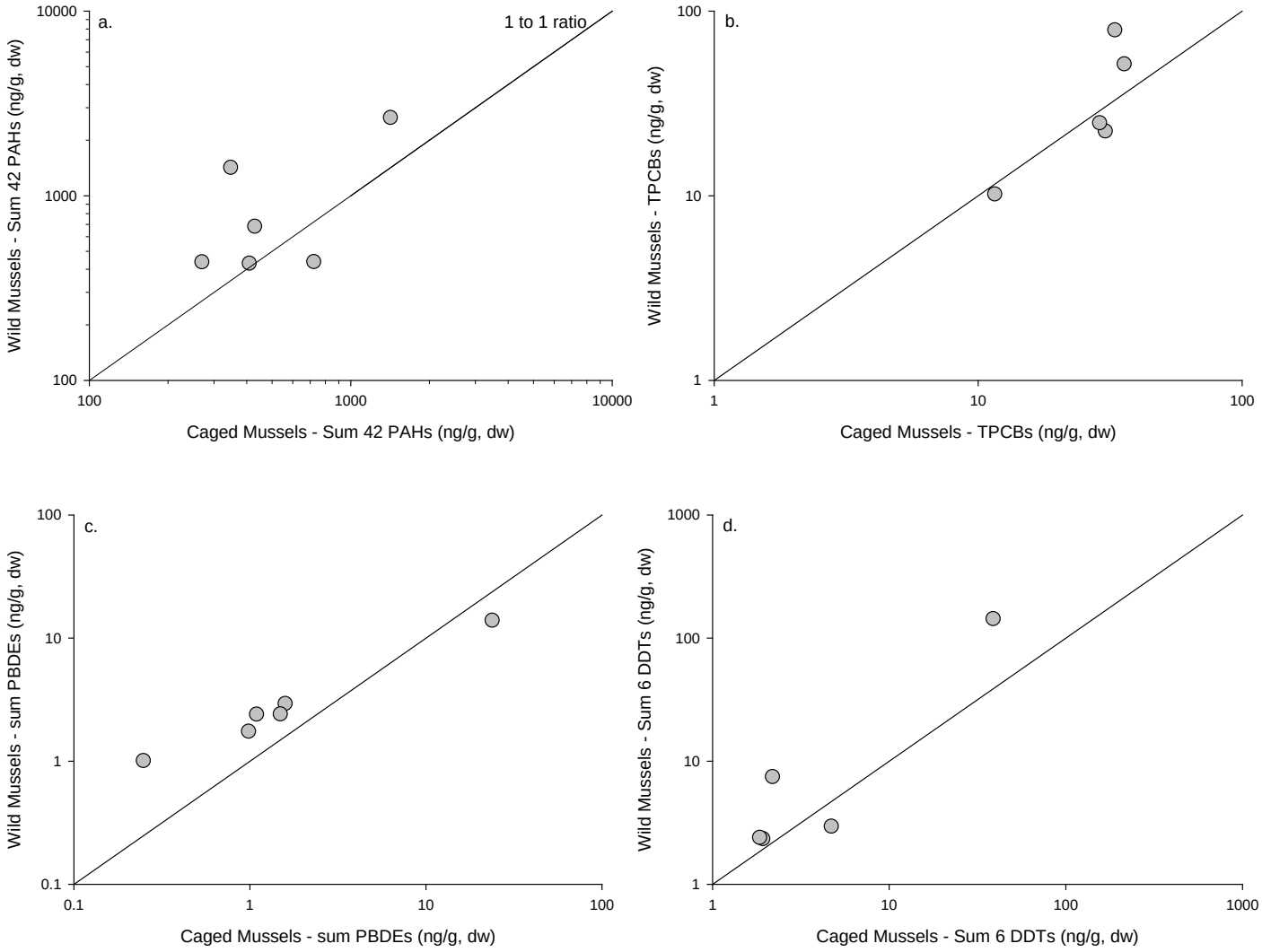

Figure 37. Ratio of ∑42PAH (a.), TPCB (b.), ∑11PBDE (c.) and ∑6DDT (d.) concentrations in transplanted (i.e. caged) and wild mussels collected at the same locations (represented by dots) during this study. Black line indicates one-to-one relationship

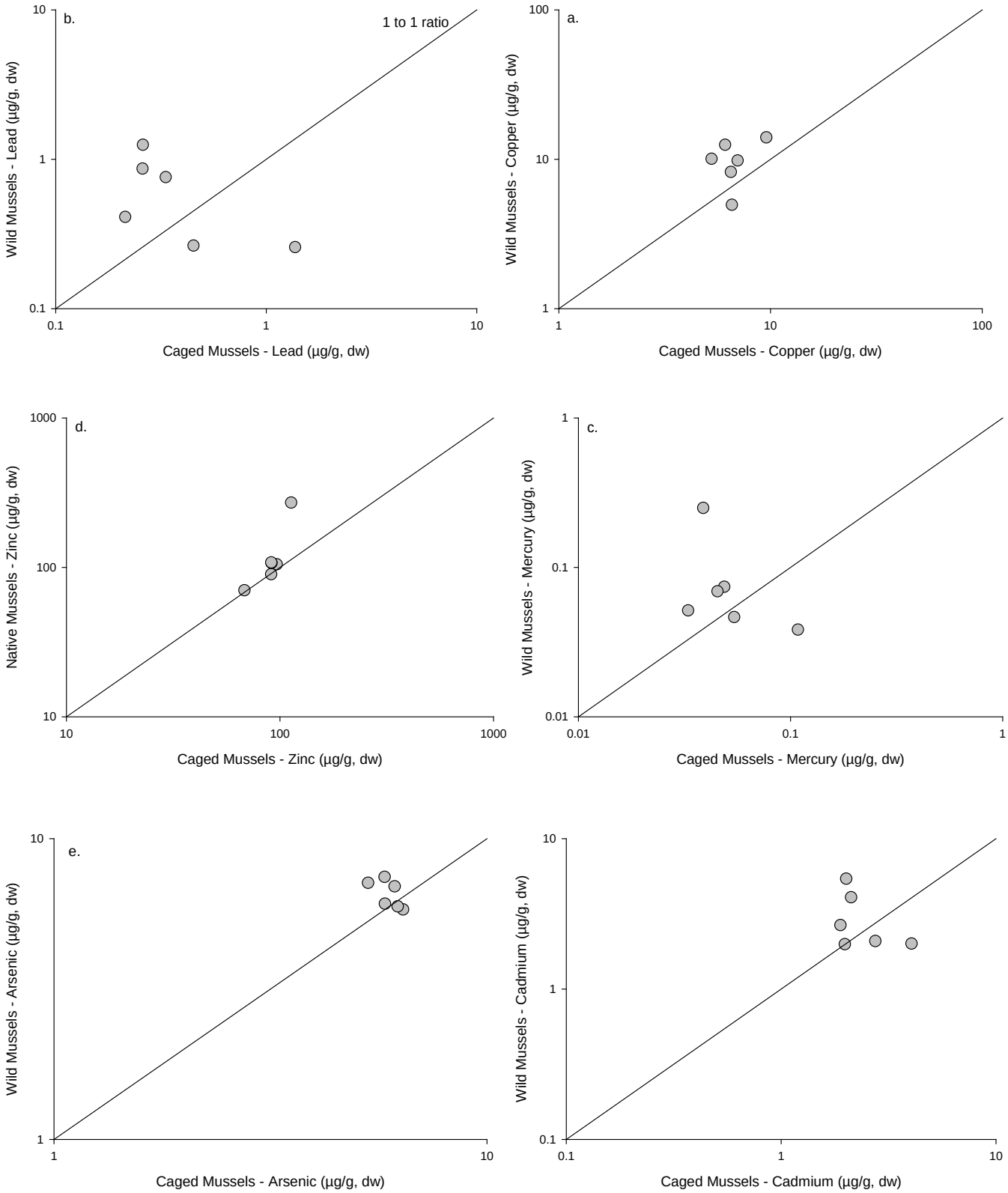

Figure 38. Ratio of copper (a.), lead (b.), mercury (c.), zinc (d.), arsenic (e.), and cadmium (f.) concentrations in wild and transplanted (i.e. caged) mussels collected at the same locations (represented by dots) during this study. Black line indicates one-to-one relationship

4 DISCUSSION

4.1 Overview

This study provides the first broad-scale, synoptic assessment of toxic contaminants in a native, nearshore-dwelling organism (the bay mussel, *Mytilus trossulus*) in Puget Sound. The deployment of transplanted native mussels from a single source over a uniform time period reduced the effects of unwanted covariates (e.g., species, size, age, and condition) on contaminant burdens, allowing a robust geographic comparison of winter contamination patterns throughout Puget Sound. The most significant observations from this study are the (a) disproportionate accumulation of several organic contaminants across the nearshore, including PAHs, PCBs, PBDEs, and DDTs, (b) moderate to weak correlation of contaminant concentrations with two measures of upland land use, percent impervious surface and road area, and (c) quantifiable contaminant patterns that help to elucidate contaminant sources.

Because mussels were only deployed for two months, they were not expected to represent contaminant conditions in wild mussels. The most appropriate use of the data herein is to compare the relative conditions of mussels from a single source exposed to variable environmental contaminants across the Puget Sound shoreline. Most of the mussels transplanted to sites used in this study accumulated relatively low concentrations of contaminants, except in highly urbanized areas where several classes of organic contaminants were relatively high. The greatest concentrations of organic contaminants detected in all samples (PAHs, PCBs, PBDEs, and DDTs) were observed in Elliott Bay and Salmon Bay, Commencement Bay (Hylebos Waterway) and in Sinclair Inlet (near Bremerton). This pattern of peak concentrations in the highly urbanized areas of Puget Sound was also observed with chlordane and dieldrin, which were not detected at all sites but were observed in samples from the most urban centers. In addition, PAH concentrations were elevated in mussels from some non-urban shorelines where there may have been other sources including marinas, ferry terminals, roadways, or other point sources.

4.2 Geographic Extent and Magnitude of Chemical Contamination in Shoreline Biota

The highest concentrations of organic contaminants (i.e. PAHs, PCBs, PBDEs, DDTs) were observed in the most urbanized embayments, including the Elliott Bay waterfront sites, Salmon Bay in the Lake Washington Ship Canal, Commencement Bay (especially in the Hylebos Waterway), and Sinclair Inlet (Appendices H - K). Elevated concentrations of organic contaminants have been found at these urban locations in the past. For instance, high concentrations of PCBs and DDTs were recorded in mussels taken from the Hylebos Waterway by Ecology in the mid-1990s (Johnson and Davis, 1996). A link between PAH and PCB contamination and the degree of urbanization in the upland has been apparent both on a national and regional scale since the 1990s. For instance, NOAA's historic Mussel Watch reported higher concentrations of PAHs and PCBs at central Puget Sound sites near Elliott Bay during that decade (Kimbrough et al., 2008; Mearns, 2001; Puget Sound Action Team, 2007). More recent data (2011-12) from NOAA's Mussel Watch indicates the concentrations of PAHs, PCBs, and DDTs in wild mussels continues to be high in the Elliott Bay area (see Comparison with NOAA's Mussel Watch below). This relatively high concentration of PCBs in Elliott Bay is consistent with previous sampling of finfish such as English sole (*Parophys vetulus*), and an invertebrate, Dungeness crab (*Metacarcinus magister*; (Carey et al., 2014) in that area. The Duwamish River and areas along the Seattle waterfront are historic PSEMP monitoring locations where English sole have consistently exhibited high PCB residues (West et al., 2001).

Interestingly, we found relatively low concentrations of PCBs in mussels taken from the Elliott Bay Alki-Duwamish Head site, on Alki Beach (Appendix I). This also supports previous findings by PSEMP, which has shown that English sole sampled along the southwestern shoreline of Elliott Bay (i.e. nearer to Alki Point) exhibited lower, near-background, levels of PCBs (West et al., 2001). This pattern of PCB distribution may result, in part, from a persistent anti-clockwise flow of water in Elliott Bay, which could move PCB-contaminated waters exiting from the Duwamish River towards the eastern and northern waterfronts of Seattle and away from its western shoreline (Alki Beach area). In contrast, the concentrations of PBDEs and DDTs were relatively low at the site directly in front of the mouth of the Duwamish River (Elliott Bay Harbor Island Pier 17 site).

The metals measured in this study were also found in mussels from all the study sites. Zinc had the highest concentration (max value 137 µg/g dw) with values decreasing ten-fold for copper (max 10.5 µg/g dw), arsenic (max 8.02 µg/g dw), cadmium (4.07 µg/g dw), and lead (max 1.38 µg/g dw) and another ten-fold lower for mercury (maximum 0.11 µg/g dw, Figure 27). Although visual examination of the geographic distributions of lead, copper and zinc concentrations supports a link with urbanization, the relationship was slight and less than it was for the organic contaminants, with the exception of lead (Appendix N). For some of the metals (mercury, arsenic, cadmium) there was no link with urbanization.

4.3 Contamination in Mussels and Adjacent Shoreline Land-use

With this project we sought to observe and describe a predictive relationship between land-use and contaminant load in a shoreline species, if one existed. We investigated the relationship between two specific land-use metrics, both simple proxies for the degree of land development, %IS and %RA, and compared them to nearshore contaminant patterns in mussels. Both metrics were positively correlated with the concentration of all the organic contaminants, as well as with lead and copper in mussels. In general %IS accounted for more variability in contaminant concentrations than %RA for most of the contaminants. The strongest relationships were between %IS and the concentration of organic contaminants, especially PAHs (Table 5). This is not surprising given that PAH concentrations tend to be higher in urban settings, which generally have both point sources (e.g. power plants and industries) and non-point sources (e.g. vehicle exhaust, road byproducts including tire treads, parking lot sealants, etc.) (Brette et al., 2014; Latimer and Zheng, 2003; West et al., 2014). Similar findings were recently reported in California, where significantly higher concentrations of PAHs and PCBs were detected in mussels from areas of high urban land use (Buchman, 2008). Correlations between the land-use metrics and metal concentrations were weaker, with the exception of lead (Table 6). In the case of lead the strongest relationship was with %RA, which accounted for 27% of the variability in mussel lead concentration (Figure 29).

Although these metrics explained some of the distribution of organic contaminants in mussels, other unknown factors not identified or included herein probably more fully explain the variation we observed in contaminant concentrations. For instance, there were a number of mussel sites placed in areas of moderate to low urbanization that exhibited relatively high PAH concentrations, including the Eagle Harbor Bainbridge Ferry Terminal, Anacortes Ferry Terminal, and Salmon Bay sites (Figure 14 and Appendix H). More perplexing was the relatively high concentration of PAHs found in the Point No Point mussels, which were deployed in an area of very low impervious surface and road area, with no obvious point sources within the immediate vicinity. These data suggest other unmeasured factors may account for the contaminant concentrations we observed in those areas.

The Puget Sound shoreline is extensive and heterogeneous; potential additional factors (sources of contaminants) include industrial outfalls, marinas, ferry terminals, CSOs, SWOs, sewage treatment plant outfalls, and failing septic systems, among others. In addition, rivers can serve as vectors for contaminants from a number of upstream point sources (e.g. industrial outfalls, marinas, septic systems) and non-point sources (e.g. agriculture, livestock, atmospheric deposition) into the Puget Sound. Although Ecology evaluated some of these contaminant sources from 2007 – 2011 as part of their Puget Sound toxics loading studies (EnviroVision Corporation et al., 2008; Hart Crowser, 2007; Herrera Environmental Consultants Inc., 2009), it was beyond the scope of this project to quantify the current flow volume, timing, or composition of all the potential contaminant sources in the study area. Further work is needed to investigate these and other source(s) of contamination in the nearshore areas of Puget Sound.

4.4 Patterns in PAHs and PCBs

Taken together in a weight-of-evidence approach the PAH pattern analysis (i.e. fingerprint histograms, homolog series maxima, and ratios) suggest the majority of mussel sites, regardless of their total $\sum_{42}$PAH concentration, were exposed primarily to a mixture of pyrogenic sources. Likely pyrogenic sources include atmospheric deposition and surface runoff from combustion of biomass (i.e. wood burning) and fossil fuels (i.e. diesel, gasoline, heating oil, natural gas, etc.), and dissolution of creosote pilings in the waters of the Puget Sound (Table 7 and Figure 16, Appendix T). High molecular weight PAHs generally indicative of combustion sources tend to dominate the PAH signature of marine sediments near urban areas (Erickson and Kaley, 2011; Latimer and Zheng, 2003; Wang et al., 1999; West et al., 2014). In a study of urban sediments from Boston Harbor, Voparil et al. (2004) reported that high molecular weight PAHs from diesel soot and tire tread materials are readily bioavailable to marine deposit feeders during digestion of contaminated sediments. Delivered via atmospheric deposition and surface-water runoff, soot can account for up to 30% of total organic carbon in coastal sediments near cities and tire tread debris can approach 15% of the total sediment by mass in areas prone to heavy automobile traffic (Voparil et al., 2004). In its 2012 Chemical Action Plan, Ecology reported wood burning stoves, creosote treated wood, and vehicle emissions (including tire wear, improper motor oil disposal and leaks) as the largest anthropogenic sources of PAHs to the Puget Sound (Davies et al., 2012). Kimbrough et al. (2008), citing the Puget Sound Action Team report from 2007, attributed the increase in PAH concentrations in the Puget Sound after the 1980s to increased vehicle traffic and urban sprawl. In addition, the volunteers who deployed mussel cages for this study reported the presence of creosote pilings near some of the study areas (e.g. Eagle Harbor-Bainbridge Ferry Terminal, Sinclair Inlet-Sinclair Marina, Hylebos Waterway, Salmon Bay, among others). Thus it is possible that contamination from creosote sources may also have contributed PAHs to the mussels at some of our study sites.

Although this pyrogenic PAH pattern appeared to dominate our mussel samples, we observed petroleum patterns at a few locations, including Salmon Bay, Bremerton Shipyard-Charleston Beach, Hylebos Waterway and the Thea Foss Waterway, suggesting petroleum sources (e.g. diesel, gasoline, motor oil, hydraulic fluids, etc.) in those areas (Figure 16, Appendix T). Oil sheens on the water are frequently spotted in Salmon Bay and Lake Union, to which Salmon Bay is connected, during monthly aerial surveys conducted by Ecology (pers. comm., C. Krembs, Environmental Assessment Program). Two aerial surveys that included flyovers of Salmon Bay and Lake Union during the course of this study documented oil sheens in those water bodies (Naidoo and Chirkoot, 2004; Short et al., 2007). Our data combined with Ecology's aerial evidence of oil spills suggests petroleum may play a significant role in the PAH contamination of nearshore organisms in Salmon Bay. We recommend further investigation to uncover potential source(s) of petrogenic PAHs in these areas.

Examination of the relative abundance of selected PCB congeners indicates a congener pattern of PCBs becoming "lighter" (i.e., the ratio of PCB028:PCB028+PCB187 increasing) with distance from Puget Sound's urban embayments, specifically Elliott Bay, Commencement Bay (especially the Hylebos Waterway), and Dyes Inlet (near Bremerton, Figure 20). These relatively high ratios suggest Puget Sound urban embayments are sources of PCBs for the rest of the Sound. Whether these PCBs originate from Puget Sound sediment or biotic reservoirs, or from new inputs from terrestrial or atmospheric sources (Erickson and Kaley, 2011; Grossman, 2013) is unknown. Recent studies suggest contemporary sources of PCBs continue to make their way into Elliott Bay (King County, 2013; Science Applications International Corporation, 2011). Higher PCB contamination in these locations is consistent with findings from previous PSEMP findings for English sole (West et al., 2001). In addition, a recent PSEMP survey of Dungeness crab in the Puget Sound indicated PCBs were highest in specimens taken from these urban areas (Carey et al., 2014). In general the PCBs congeners in our mussels appeared to follow a gradient of heavier-to-lighter from the Central Puget Sound area outward towards the Whidbey Basin, North Puget Sound, South Puget Sound, and Admiralty Inlet. To the north, Grant et al. (2011) reported high concentrations (i.e. hotspots) of PCBs in sediments from urban harbors in the Strait of Georgia, British Columbia, where patterns were consistent with historical point sources, with a change toward lighter profiles in more remote areas.

4.4.1 Penn Cove Oil Spill – Fingerprint Comparison

In mid-May of 2012 a derelict fishing vessel sank in Penn Cove, Whidbey Island, spilling approximately 1400 gallons of diesel fuel just 200 meters north of the outer edge of the Penn Cove Shellfish, Inc. aquaculture rafts. Aerial photos at the time of the spill showed a silver sheen of fuel passing through the commercial mussel culture floats in Penn Cove (Mearns et al., 2014). This oil spill was of concern to us since it had the potential to affect the source of our mussels prior to the beginning (mid-November, 2012) of our study. Mearns et al. (2014) tracked the contamination in these Penn Cove mussels after the vessel sank. On four occasions after the accident, they sampled *Mytilus trossulus* (40-60 mm in length; 30-50 mussels per sample) from near the surface at six locations across the full range of Penn Cove.

Here we compare the concentration of Σ_{43}PAHs of mussels taken from three of the six Penn Cove floats, sampled by Mearns et al. (2014) on November 7, 2012. These three floats (A-1, C-1 and F-4) were nearest to the mussel float (D-2) used as a source in our study on November 14, 2012. Their results indicate that the mean Σ_{43}PAH value from these three floats (2377.3 ng/g dw) was approximately 33 times higher than the mean Σ_{42}PAH measured in our mussels (71.36 ng/g dw, ± 20.385 s.d.) one week later (Table 10).

Table 10. Concentration of Σ_{43}PAHs of mussels (n = 1 composite per date/float) sampled off three Penn Cove Shellfish, Inc. aquaculture floats after a diesel spill in Penn Cove (Mearns et al., 2014), and Σ_{42}PAHs (mean ± standard deviation; n = 6 composites) from mussels taken as a baseline for this study (MWPE). All concentrations are in µ/kg (ng/g) dw.

Mussel Float:	**5/18/2012**	**7/4/2012**	**11/7/2012**	**11/14/2013**	**5/21/2013**
A-1 (Mearns)	7,108	7,135	**2,438**		1,591
C-1 (Mearns)	7,447	5,921	**2,874**		114
F-4 (Mearns)	6,593	2,190	**1,820**		772
D-2 (MWPE)				**71.36 (±20.385)**	

Although the Mearns et al. (2014) mussel samples and our samples were analyzed at two different laboratories, using slightly different methods, and the PAH concentrations were summed slightly differently, a comparison of lab techniques revealed nothing noteworthy that would have accounted for the large differences in total PAH concentrations. However, there are several significant differences between the collection methods used by Mearns et al. (2014) and those used in this study. Although the timing and geographic locations were very similar (Table 10) the depth from which the mussels were sampled were different, which likely explained the disparity in the PAH concentrations we observed.

Mearns et al. (2014) sampled mussels near the surface (up to one meter in depth), while our mussels were taken from the entire length of aquaculture line on which the mussels were growing (up to 8 meters in depth). Thus mussels from our study (MWPE) came from a much greater depth range. A comparison of the PAH fingerprints for MWPE mussels with mussels taken from the surface (Mearns et al., 2014) in November of 2013 reveals disparate patterns supporting a conclusion that exposure of the mussels we used in our study was negligible (Figure 39). The Mearns et al. profile showed a pattern typical of petroleum (diesel oil) contamination, with increasing dibenzothiophene and phenanthrene alkylated homologs (C_1-C_3) relative to the parent C_0 compounds, while the mussels from this study showed a pattern more typical of combustion sources in the phenanthrene/anthracene homolog series (decreasing concentrations from C_0 through C_4 homologs), and a lack of dibenzothiophenes overall. These differences in PAH concentrations, fingerprints, and sample depths support the conclusion that these two studies collected and used different populations of mussels exposed to different PAH conditions, and that mussels used for the MWPE were largely unexposed to the nearby fuel spilled in May 2012.

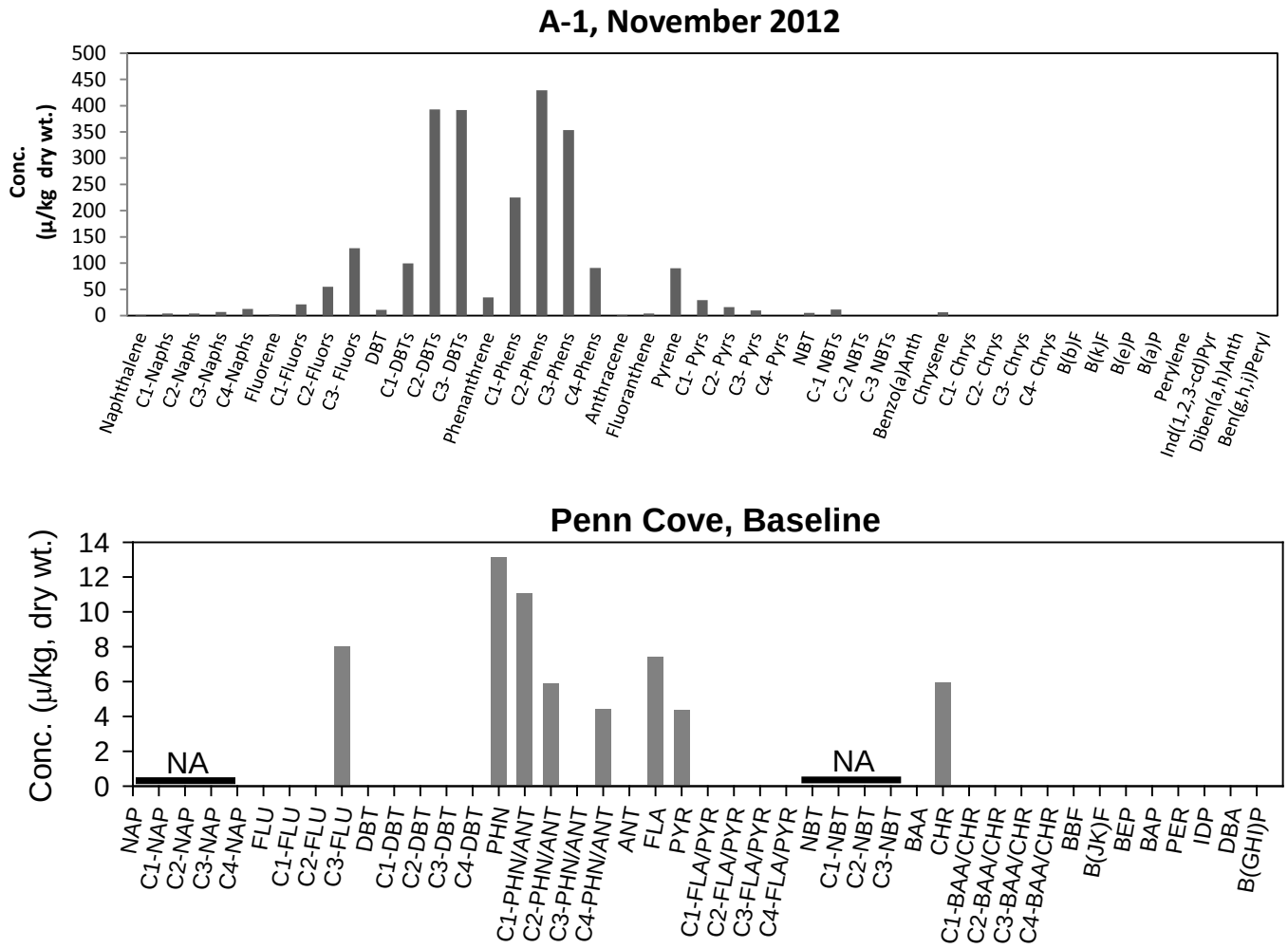

Figure 39. A comparison of the PAH analyte histograms of mussels collected from Penn Cove A-1 float on November 7, 2014 (top graph, courtesy of Mearns et al. 2014) and the Penn Cove, Baseline mussels collected for the MWPE study from the D-2 float on November 14, 2014 (bottom graph).

4.5 Biological Endpoints

Although mussels are known to survive in relatively contaminated environments (ASTM International, 2007), we found weak positive relationships between mortality and the degree of urbanization, both in relation to %IS and %RA (Figure 8 and Figure 9). Although it is impossible to fully control all potential sources of mortality related to water quality in a field study such as this, it is not unreasonable to assume that toxic contaminants represent a significant potential source of stress. The increase in mortality of mussels along the contaminant gradient from urban to rural shorelines suggests causation – that mussels in highly contaminated habitats had a lowered fitness. This is congruent with results from previous studies that showed decreasing fitness with increasing exposure to contaminants (ASTM International, 2007; Gagné et al., 2001; Salazar and Salazar, 1991; Solé et al., 1996; Stephenson et al., 1986; Strömgren, 1982; Strömgren, 1987; Valkirs et al., 1991; Widdows et al., 1995; Widdows et al., 2002; Widdows et al., 1997).

The overall decline in CI of mussels at most (72%) of our deployment sites and at the Penn Cove aquaculture (mussel source) reference area over the course of the study (Figure 11) was likely a normal response of mussels

to winter conditions. The decline occurred across the full range of land-use types, and was not correlated with %IS or %RA (Figure 12). Kagley et al. (2003) reported a reduction in CI of wild mussels during the winter months in Puget Sound (Figure 2). During the winter in Puget Sound phytoplankton growth (i.e. primary production) declines due to limitations in sunlight and photosynthesis. Several researchers have noted that mussels stop feeding at minimal algal concentrations; for instance, Riisgård and Larsen (2000) demonstrated that mussels can alter their clearance rates, via valve closure, in response to either extremely low or very high algal concentrations. Low feeding thresholds for mussels have been reported at chlorophyll concentrations (a measure of phytoplankton abundance) of between 0.4 - 0.5 µg Chl-*a* dm^{-3} (Dolmer, 2000; Noren et al., 1999). Though we did not measure chlorophyll concentrations at our study sites, we assume food during our winter study was less abundant than the spring and summer season, when most somatic growth and reproduction occur.

Although the CI of mussels at most of the study sites declined, there was a small but significant increase in shell length (1.5%) over the course of this study (Figure 10). Thus it appears that the mussels' shells grew during the winter, at the expense of body mass. This conclusion is supported by a study of mussel growth during the winter months in Nova Scotia, Canada, where Johnson and Bustin (2006) found that although soft tissue growth was food-limited in areas, shell growth (length and weight) was similar among sites. Thus it is likely that shell growth is not as sensitive a biological endpoint as CI, especially when using adult mussels. If growth is a desired biological endpoint in future mussel studies in the Puget Sound, we recommend selecting pre-reproductive, juvenile mussels (which grow faster), deploying cages during the spring or summer months, when food is more abundant, and extending the deployment period.

We noted a number of predators, including sea stars and crabs inside a few cages (Table 4; taken from Lanksbury et al., 2013). Kelp crabs (*Pugettia producta*), specifically identified in some of the cages, are primarily herbivores that consume algae though they also eat barnacles, mussels, hydroids, and bryozoans when their primary food source is scarce (Rudy and Rudy, 1987). Sunflower sea stars (*Pycnopodia helianthoides*) were identified in several cages as well (Figure 7). Individuals from this species are flexible and fleshy and use hydrostatic pressure to maintain their body form. Anecdotal evidence suggests sunflower sea stars can reduce their internal hydrostatic pressure (i.e. drastically reduce the amount of water in their bodies) to squeeze through small spaces. We assume this is how hand-sized individuals were able to pass through the 1 x 1.5 inch mesh into our cages. We think that hanging the mussels in bags in the upper portion of the cages further protected mussels and mitigated mortality from sunflower sea stars, because the sea stars appeared to have a difficult time crawling up the sides of the cages.

4.6 Transplanted vs. Wild Mussels

Although this study was not specifically designed to compare contaminant concentrations between transplanted and wild mussels, we were able to do some limited qualitative comparisons between the two groups based on wild mussel samples taken by our partners in Snohomish County (five sites), and one additional wild mussel sample collected in the Hylebos Waterway in Commencement Bay (Table 3). Thus, the following comparisons are limited in scope to those areas where matching samples were collected.

Graphical comparison of the six paired samples suggested the wild and transplanted mussels were nearly equivalent in terms of their concentrations of PCBs, PBDEs, DDTs, PAHs, and all six metals (Figure 37 and Figure 38). We saw a signficant difference in the CI of the two groups, with wild mussels having lower CIs than their transplanted counterparts (Figure 36). This difference in CI can likely be explained by the fact that our starting mussels were grown in Penn Cove, which is known for its ideal conditions for raising bivalves

(Penn Cove Shellfish LLC, 2014). During the study the transplanted mussels were placed at the zero tide level, while the wild mussels were sampled from the middle to high intertidal area (Table 3).

A survey of studies that compared contaminant concentrations in transplanted versus wild mussels showed disparate conclusions. For instance, a Dutch coast study revealed two to ten-fold decreases in the concentrations PCBs and PBDEs in transplanted over wild mussels (Gustafsson et al., 1999), though later a study of freshwater Zebra mussels (*Dreissena polymorpha*) in Belgium showed high correlations between organic contaminants in transplanted and resident mussels (Bervoets et al., 2004). Other researchers suggest transplanted mussels need adequate time to attain "equilibrium" with the environment, the length of which depends on the contaminant of interest (ASTM International, 2007; Booij et al., 2002; Kock, 1986; Oros and Ross, 2005; Roesijadi et al., 1984; Salazar and Salazar, 1995; Widdows and Donkin, 1992).

When comparing long-term mussel monitoring data between programs that collected wild mussels, like NOAA's Mussel Watch (Center for Coastal Monitoring and Assessment, 2014) and Maine's Gulf Watch (Gulf of Maine Council on the Marine Environment, 2014), versus programs that transplanted mussels, like the Massachusetts Water Resources Authority (2014), Hunt and Slone (2010) warned that although either approach can reveal trends in environmental contamination, care should be taken when interpreting data in terms of deployment methodology (e.g. mid-water transplanted mussels vs. intertidal resident populations). In a study off the coast of Italy, wild mussels sampled from the intertidal zone showed higher concentrations of PAHs than transplanted mussels placed at 2 - 4 m under water, leading the authors to conclude that PAHs could be investigated in wild and transplanted mussels from the same location, but only if they were sampled at the same depth (Piccardo et al., 2001). Differences in physiological adaptation to environmental stressors (both natural and anthropogenic) between wild and transplanted mussels may also be an important factor to consider (Acker et al., 2005; Nigro et al., 2006).

4.7 Comparison with NOAA's Mussel Watch

As part of its long-term monitoring efforts, the NOAA's Mussel Watch project has monitored contaminants in wild Washington State mussels since 1986. In their assessment of two decades (1986-2005) of contaminant monitoring data, NOAA's Mussel Watch highlighted the Puget Sound as a region of concern especially for PAH contamination (Kimbrough et al., 2008). Based on their PAH data from 2004/05, NOAA's Mussel Watch characterized seven locations in the Puget Sound as "medium" in contamination on both a regional (Northwest Region) and national scale, and characterized one site (Elliott Bay-Four-Mile Rock) as "high" in contamination on both scales (Kimbrough et al., 2008). In addition, they concluded that sites in Puget Sound had PAH concentrations among the highest in the nation, nearly double what was observed nationally at that time. In this report mussels from the Elliott Bay-Four-Mile Rock site (from 2004/05) also fell within the high range (regional scale) for PCB concentrations. In a later report summarizing PBDE data (2004-2007), NOAA's Mussel Watch characterized five Puget Sound sites into the medium and seven sites into the high concentration categories (Kimbrough et al., 2009). NOAA's Mussel Watch also reported medium to high range concentrations for lead (Puget Sound-Everett Harbor), copper, mercury (Sinclair Inlet-Waterman Point), cadmium, and zinc at multiple mussel monitoring sites around the Puget Sound (Kimbrough et al., 2008).

To this background of information we add a summary of the most recent NOAA Mussel Watch results for Puget Sound, based on analysis of historical wild mussel stations plus results from three new locations (two in South Puget Sound and one in Elliott Bay) collected in the winter of 2011/12. Here we used the same summations

described in in Table 2. The goal of this summary was to allow comparison of recent broad-scale patterns in the distribution of contaminants in wild mussels with regional patterns seen in our transplanted mussels. Though the distribution patterns of contaminant concentrations appear similar between projects, the overall scale of contaminant concentrations differs, in some cases (i.e. PAHs) by ten-fold. Because of these discrepancies, likely due to differences between study methods (listed below), we do not make direct comparisons between contaminant concentrations seen in the 2011/12 NOAA Mussel Watch data and our 2012/13 MWPE data. Some of the major differences between methods include:

1. Study organism and exposure period - NOAA Mussel Watch collected wild (*Mytilus* sp.) mussels that could have included several species; we used transplanted mussels of a single species (*Mytilus trossulus*) exposed on site for 60 days,
2. Mussel size/age - NOAA's Mussel Watch sampled a range of mussel sizes/ages; we used mussels of a uniform size/age,
3. Tidal height sampled - NOAA's Mussel Watch collected mussels in the medium - high intertidal zone (+3 to +6 feet MLLW); we located our mussels at zero (0) MLLW,
4. Proximity to substrate – some of NOAA's Mussel Watch mussels are taken from cobble or boulders (above the substrate) and some are taken from the sediment (i.e. partially buried); our mussels were consistently placed 35 cm above the substrate.
5. Laboratories - different analytical labs were used in each study, though chemical analysis techniques were similar.

The highest $\sum_{42}$PAHs concentrations in wild mussels from the 2011/12 NOAA Mussel Watch data occurred in the central and northern areas of Elliott Bay (Figure 40 – left map). This data set included a new site for NOAA's Mussel Watch, "Elliott Bay, Myrtle Edwards" (Figure 40 – left map inset), which was added to the Puget Sound site list in 2009; prior to that the NOAA Mussel Watch sites nearest Elliott Bay were "Elliott Bay-Four-Mile Rock" to the north, and "Elliott Bay-Duwamish Head" to the south, at the tip of Alki Point. Though the NOAA Mussel Watch sites are relatively far apart, their data suggest a hotspot for PAHs in Elliott Bay, with medium levels of contamination in the central Puget Sound and lower contamination in the north and south Puget Sound. This broad-scale pattern was supported by the results of our 2012/13 study, where we found highest $\sum_{42}$PAHs concentrations within Elliott Bay, medium concentrations in the central Puget Sound (including Everett Harbor), and relatively low concentrations at rural sites in north and south Puget Sound (Figure 40 – right map). Moreover, our data supplies additional information about regional patterns of contamination, filling in the gaps not covered by the NOAA Mussel Watch monitoring sites. This pattern of higher organic contaminants in the highly urbanized bays, medium concentrations in the central Puget Sound, and lowest concentrations in the rural north and south was mirrored in the TPCB (Figure 41 – left map) and $\sum_{6}$DDT data (Figure 42) for both projects. Although the absolute concentrations of contaminants differ between projects, the general patterns of contaminant distributions are in agreement.

Mearns (2001) used NOAA Mussel Watch data to compare geographic areas and conditions in Puget Sound with the rest of the U.S. coast. Through his work he confirmed that PCB concentrations in the Puget Sound have been declining over the past three to four decades, and that concentrations of PAHs in this area were high. He also showed that mussels from sites within the Puget Sound, the Strait of Juan de Fuca, and the Strait of Georgia contained similar concentrations of most metals, when compared to other Pacific coastal sites, but lower concentrations of arsenic and cadmium. Mearns and other researchers frequently referred to Washington

State data gathered by the NOAA's Mussel Watch program as good background information for potential oil spills and other hazardous materials spills (Mearns, 2001).

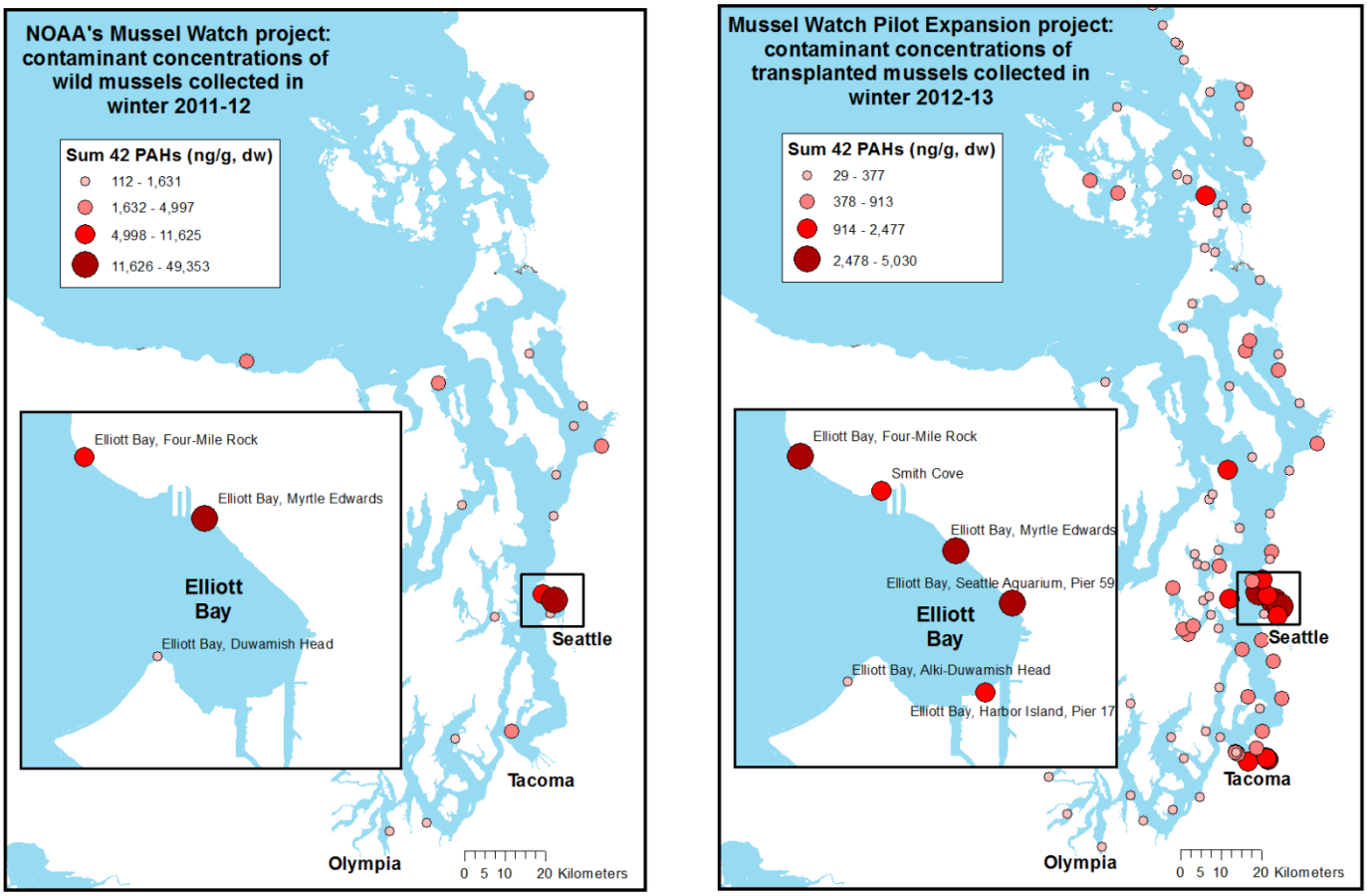

Figure 40. Concentration of $\sum_{42}$PAHs in wild mussels taken as part of NOAA's Mussel Watch project in 2011-12 (left), and in transplanted (i.e. caged) mussels taken from our Mussel Watch Pilot Expansion project in 2012-13 (right). Note scales on maps are different, as concentrations were different; figures show relative values.

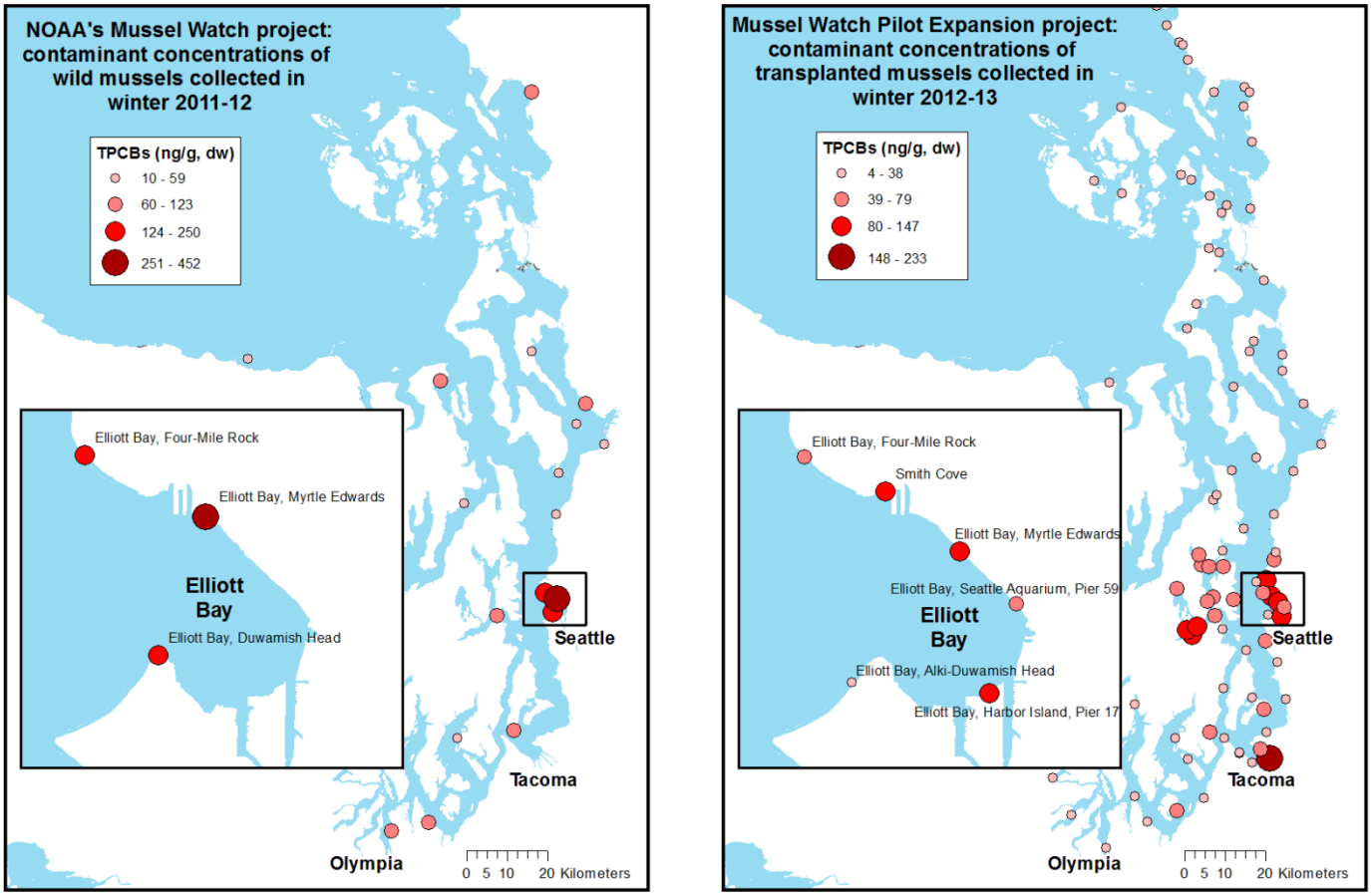

Figure 41. Concentration of estimated total PCBs (TPCBs) in wild mussels taken as part of NOAA's Mussel Watch project in 2011-12 (left), and in transplanted (i.e. caged) mussels taken from our Mussel Watch Pilot Expansion project in 2012-13 (right). Note scales on maps are different, as concentrations were different; figures show relative values.

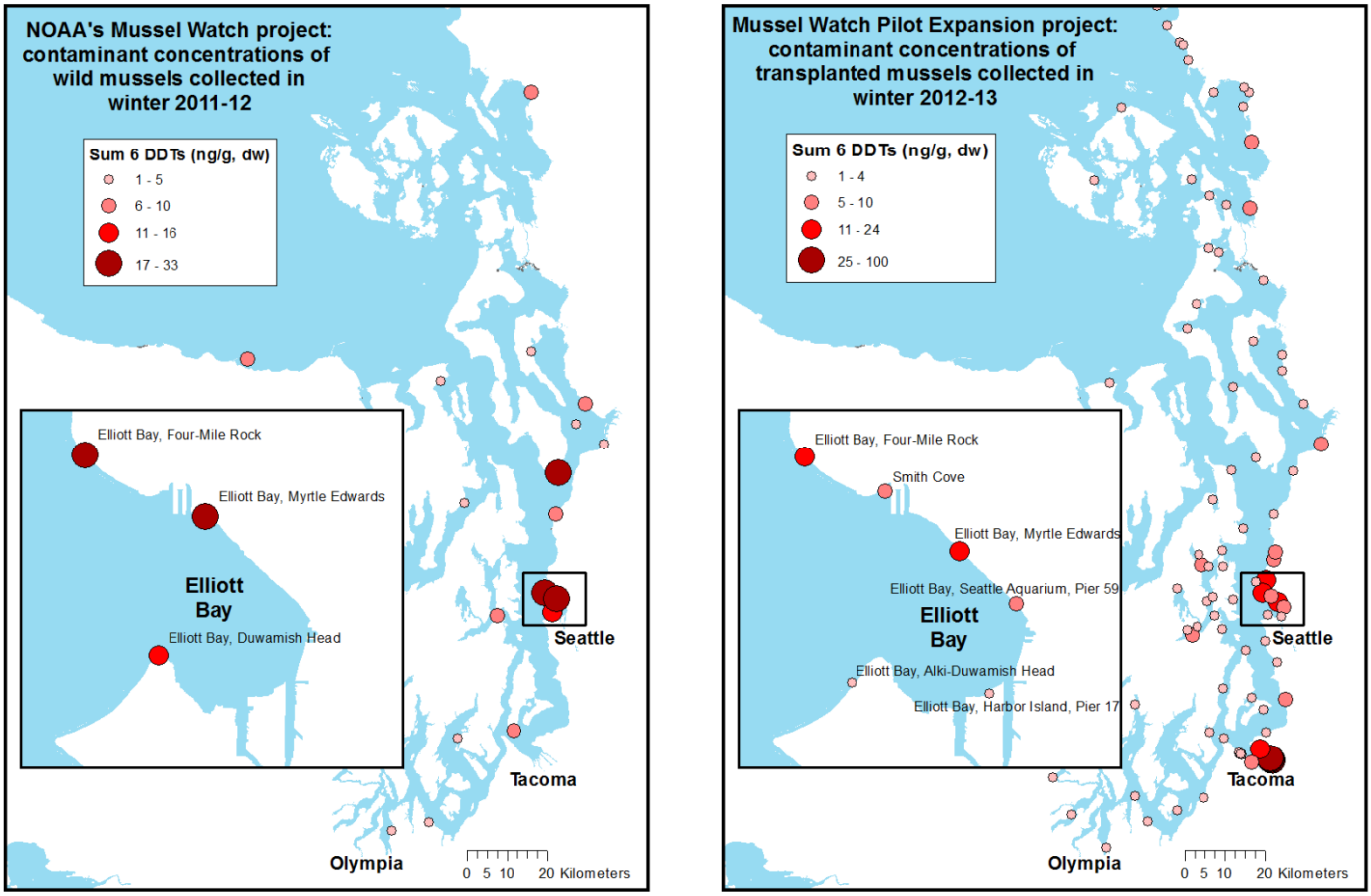

Figure 42. Concentration of $\sum_6$DDTs in wild mussels taken as part of NOAA's Mussel Watch project in 2011-12 (left), and in transplanted (i.e. caged) mussels taken from our Mussel Watch Pilot Expansion project in 2012-13 (right). Note scales on maps are different, as concentrations were different; figures show relative values.

4.8 Recommendations for Long-term, Nearshore Status and Trends Monitoring

After realizing the overall success of completing this one-time, synoptic pilot project, we recommend the development of a long-term network of mussel monitoring sites in Washington State, including sites along both the Washington Salish Sea and the outer Pacific coast. This "Washington State Mussel Watch" (WSMW) program would expand the spatial coverage of NOAA's previous mussel monitoring efforts in Washington and provide valuable information about where contaminants may be entering Puget Sound. Moreover, combining mussel results with existing PSEMP contaminant assessment and monitoring would provide important information regarding the fate and transport of contaminants in the Puget Sound food web over time. Having data on the current levels of pollutants in the nearshore environment would improve our ability to make cost-effective decisions to mitigate the harm pollution causes in the nearshore environments of Washington State, and would help us to gauge the recovery of the Puget Sound into the future (Puget Sound Partnership, 2010; Puget Sound Partnership, 2012-14).

We envision a WSMW program that includes voluntary participation by partner groups (e.g. state, county, and city agencies, county MRCs, tribes, WWTPs, other permitted dischargers, NGOs, private companies, volunteer groups, etc.) interested in monitoring pollution in their nearshore areas. Partnerships could include monetary support and sponsorship of sites and/or lending of resources and staff or volunteer time to help organize and

execute the surveys. Through this network of partnerships with other agencies and the use of citizen volunteers, efficiencies could be realized and the cost for the monitoring program could be reduced. Experiences and data gained from the monitoring program would be shared and disseminated by all the partners, and the public education opportunities afforded would be supportive of community stewardship in the Puget Sound. However, a monitoring program of this size would require an organizational platform, which could be provided by the WDFW's PSEMP Unit.

Depending on the scale (i.e. number) and desired locations of the mussel monitoring sites, either wild mussels or transplanted mussels could be utilized. Considering the need for regional/local data and the desire from several outside groups to monitor in their specific areas of interest, we recommend the continued use of transplanted mussels for status and trends monitoring in the future for Washington State. However, we also recommend that in the future transplanted mussels be left on site for approximately 90 days (October – January) to be sure they are given adequate time to equilibrate to the contaminants in their environment. We also suggest a comparison study of contaminant concentrations in wild versus transplanted mussels, to evaluate the latter as a predictive tool for the former.

There are a wide range of contaminants of emerging concern (CECs) that are expected to increase in production and usage in the United States in the years to come. These CECs many of which may originate from municipal, agricultural, and industrial wastewater sources and pathways, include pharmaceuticals and personal care products (PPCPs), perfluorinated compounds (PFCs), detergents, disinfectants, plasticizers, preservatives, contemporary use pesticides, and food additives (e.g. caffeine) among others. Many of these compounds are not currently regulated and/or routinely monitored, have likely been present in the aquatic ecosystems of the Puget Sound for years, and may already be having a negative impact on the Sound's marine life (Johnson et al., 2008). Some of these CECs are likely persistent in the environment and their potential for impacts to ecological and human health remains largely unknown. Recently the State of California, in partnership with NOAA's Mussel Watch, carried out a pilot study using mussels to assess the presence of CECs along the California coast. They found the greatest number of CECs detected and the highest concentrations were associated with urban areas and locations influenced by storm water discharge (Voparil et al., 2004; Yunker et al., 2012). Considering the rising concern about CECs in the environment, we suggest evaluating and potentially adding a range of CECs to the list of contaminants assessed through mussel monitoring in Washington State.

Finally, a number of biomarkers that are utilized worldwide in mussel monitoring programs may be useful in helping to characterize mussel health in the Puget Sound. Some examples of biomarkers that have been used successfully on mussels in the past include lysosomal membrane stability, neutral lipid and lipofuscin lysosomal content, DNA damage, catalase activity, metallothionein content, acetylcholinesterase and glutathione transferase activities, vitellogenin and estrogen receptor 2 mRNA expression, lysosome/cytoplasm volume ratio, and stress on stress response (ASTM International, 2007; Dagnino et al., 2007; Gagné et al., 2001; Gagnon et al., 2006; Solé et al., 1996; Viarengo et al., 1995). We suggest exploring the usefulness of coupling tissue contaminant concentrations with select biomarker status in future Puget Sound mussel monitoring programs, to help better answer questions regarding mussel health and exposure to toxics in the nearshore (Dagnino et al., 2007).

In summary, we recommend a long-term Washington State mussel monitoring program that includes;

1) a network of mussel sites monitored by a number of contributing partners and volunteers and managed under WDFW's PSEMP,

2) a focus on answering science questions that can increase the utility of monitoring, including:
   - How does contamination and health of wild vs. transplanted (i.e. caged) mussels compare?,
3) the evaluation and addition of contaminants of emerging concern,
4) an evaluation of the efficacy of using mussels for targeted effectiveness monitoring (e.g., stormwater or CSO retrofits),
5) identification of biomarkers that can help better answer questions regarding mussel health and exposure to toxics,
6) *post-hoc* analyses to explore factors that may further explain variability in the contaminant data, and
7) splitting samples between laboratories to facilitate comparison of historical (NOAA Mussel Watch) to present and future mussel contaminant data.

4.9 Conclusions

The Mussel Watch Pilot Expansion study demonstrated that transplanted mussels can be used successfully on a large scale to characterize patterns of nearshore contamination in the greater Puget Sound. The transplanted mussels provided data on the current geographic extent and magnitude of contamination in our nearshore environments, and offer insight into how contamination in nearshore biota may be related to upland land-use patterns. The regional and local-scale contaminant patterns observed in this study support the broader-scale patterns observed in previous mussel monitoring studies in the Puget Sound, where higher concentrations ("hotspots") were found in areas of high urbanization (Elliott Bay, Commencement Bay, Sinclair Inlet/Bremerton). This pattern is seen in other studies of benthic and pelagic fish and shellfish of the Puget Sound, confirming the role of urbanization as a major source of pollution to our marine waters, while adding data relevant to our nearshore environments.

While the mussels used in this study provided valuable information on the extent and magnitude of contamination in the Puget Sound, they were also useful for inferring sources of PAHs to the nearshore. The majority of mussel sites show patterns consistent with pyrogenic sources, a few locations were atypical, exhibiting characteristics of petrogenic PAH contamination. Further investigation into potential sources of PAHs in those atypical areas is warranted. In the future, mussels may prove useful in identifying other nearshore areas affected by oil leaks from point or focal-point sources. In addition, PAH data from mussel sites can be used as baseline for assessing damage from future oil spills in the Puget Sound. Mussels have been used to help understand the nearshore impacts of the Exxon Valdez and Deepwater Horizon Oil Spills (Apeti et al., 2013; Babcock et al., 1996; Carls et al., 2001; Carls et al., 1996; Neff and Burns, 1996; Short and Rounds, 1993; Thomas et al., 1999). Soriano et al. (2006) showed that mussels taken from the 2002 *Prestige* oil spill area, along the coasts of Spain, Portugal and France, had a Fl/Fl+Pyr ratio (0.18) similar to the oil itself (0.22), helping to track the nearshore impacts of that disaster.

In terms of preparation and implementation, this study also demonstrated how citizen volunteers can be utilized to help execute a major, large-scale monitoring effort. This study could not have been accomplished without the help of the many partnering organizations and volunteers who provided their money, time and efforts to help prepare mussels for the study, and deploy and retrieve the mussels in cages at the 108 study sites around the Puget Sound. In addition, the extent of this study would have been much reduced without the sponsorship and staff time provided by the 13 additional organizations that signed on to participate in this project (see Appendix A for fund sources). These partners greatly expanded the scope and geographic coverage of this study, enhancing the overall results for everyone involved. This network of sponsoring groups and helpful volunteers

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8 APPENDIX A: Study Site Details

Table includes information on mussel study site location, source of funding for individual sites, deployment and retrieval dates of transplanted/caged mussels, whether a cage was lost (i.e. site failed), and when a wild mussel sample was collected near a transplanted mussel site. Site order is arranged alphabetically by county and then from north to south within each county.

Site Order	County	Site Name	Latitude	Longitude	Fund Source	Deployment Date	Retrieval Date	Cage Lost	Native Mussel Sample
1	Clallam	Protection Island Aquatic Reserve, Thompson Spit	48.097	-122.939	DNR Aquatic Reserves Program	11/13/2012	1/9/2013		
2	Island	Deception Pass State Park, Cornet Bay	48.402	-122.621	EPA - NEP grant	11/12/2012	1/10/2013		
3	Island	Ala Spit County Park	48.393	-122.587	EPA - NEP grant	11/12/2012	1/10/2013		
NA	Island	Smith and Minor Islands Aquatic Reserve, Joseph Whidbey State Park	48.314	-122.711	DNR Aquatic Reserves Program	11/13/2012	NA	X	
4	Island	Oak Harbor, Crescent Harbor	48.278	-122.660	EPA - NEP grant	11/14/2012	1/8/2013		
5	Island	Coupeville Wharf, Toby's Tavern	48.222	-122.686	EPA - NEP grant	11/14/2012	1/8/2013		
6	Island	PENN COVE, BASELINE	48.218	-122.709	EPA - NEP grant	NA	*11/14/2012		
7	Island	Triangle Cove	48.199	-122.465	Stillaguamish Tribe	11/14/2012	1/7/2013		
8	Island	Cavalero Beach County Park	48.175	-122.478	Stillaguamish Tribe	11/14/2012	1/7/2013		
9	Island	Cavalero Beach Natives	48.175	-122.478	Stillaguamish Tribe of Indians	NA	*1/7/2013		X
10	Island	Holmes Harbor, Rocky Point, Baby Island	48.096	-122.527	EPA - NEP grant	11/13/2012	1/10/2013		
11	Island	Maxwelton, Dave Mackie County Park	47.940	-122.447	EPA - NEP grant	11/12/2012	1/10/2013		
12	King	Richmond Highlands Beach	47.729	-122.374	EPA - NEP grant	11/15/2012	1/9/2013		
13	King	Carkeek Park	47.713	-122.381	Puget Soundkeeper Alliance	11/12/2012	1/9/2013		
14	King	Salmon Bay	47.666	-122.402	Puget Soundkeeper Alliance	11/12/2012	1/9/2013		

Site Order	County	Site Name	Latitude	Longitude	Fund Source	Deployment Date	Retrieval Date	Cage Lost	Native Mussel Sample
15	King	Discovery Park, West Point	47.662	-122.436	EPA - NEP grant	11/12/2012	1/9/2013		
16	King	Elliott Bay, Four-Mile Rock	47.638	-122.412	EPA - NEP grant	11/12/2012	1/9/2013		
17	King	Smith Cove	47.631	-122.386	Puget Soundkeeper Alliance	11/12/2012	1/9/2013		
18	King	Elliott Bay, Myrtle Edwards	47.619	-122.361	EPA - NEP grant	11/12/2012	1/7/2013		
19	King	Elliott Bay, Seattle Aquarium, Pier 59	47.607	-122.342	EPA - NEP grant	11/12/2012	1/9/2013		
20	King	Elliott Bay, Harbor Island, Pier 17	47.588	-122.351	EPA - NEP grant	11/12/2012	1/7/2013		
21	King	Elliott Bay, Alki-Duwamish Head	47.589	-122.395	EPA - NEP grant	11/12/2012	1/9/2013		
22	King	Lincoln Park	47.531	-122.401	King County DNRP	11/12/2012	1/9/2013		
NA	King	Fauntleroy	47.524	-122.395	King County DNRP	11/12/2012	NA	X	
23	King	Vashon Ferry, N. End Boat Ramp	47.509	-122.463	EPA - NEP grant	11/13/2012	1/9/2013		
24	King	Seahurst County Park	47.484	-122.362	EPA - NEP grant	11/14/2012	1/9/2013		
25	King	Quartermaster Harbor	47.405	-122.441	EPA - NEP grant	11/12/2012	1/9/2013		
26	King	Des Moines Marina City Beach Park	47.403	-122.329	EPA - NEP grant	11/14/2012	1/9/2013		
27	King	Maury Island Aquatic Reserve, Old Marine Park	47.380	-122.402	DNR Aquatic Reserves Program	11/14/2012	1/10/2013		
28	King	Dumas Bay	47.329	-122.390	EPA - NEP grant	11/14/2012	1/10/2013		
29	Kitsap	Point No Point	47.909	-122.527	EPA - NEP grant	11/13/2012	1/8/2013		
30	Kitsap	Port Gamble, Point Julia	47.853	-122.574	Port Gamble S'Klallam Tribe	11/13/2012	1/8/2013		
31	Kitsap	Port Gamble, West	47.842	-122.585	Port Gamble S'Klallam Tribe	11/13/2012	1/8/2013		
32	Kitsap	Point Jefferson	47.780	-122.482	EPA - NEP grant	11/13/2012	1/10/2013		
33	Kitsap	Suquamish, Stormwater Outfall	47.730	-122.551	Kitsap County Public Works	11/13/2012	1/8/2013		
34	Kitsap	Liberty Bay, Poulsbo	47.719	-122.627	EPA - NEP grant	11/13/2012	1/10/2013		
35	Kitsap	Liberty Bay, Keyport	47.697	-122.617	EPA - NEP grant	11/14/2012	1/8/2013		
36	Kitsap	Point Bolin	47.694	-122.595	EPA - NEP grant	11/13/2012	1/10/2013		

Site Order	County	Site Name	Latitude	Longitude	Fund Source	Deployment Date	Retrieval Date	Cage Lost	Native Mussel Sample
37	Kitsap	Port Madison, Hidden Cove	47.694	-122.545	EPA - NEP grant	11/13/2012	1/10/2013		
38	Kitsap	Silverdale, Dyes Inlet	47.643	-122.697	Kitsap County Public Works	11/13/2012	1/8/2013		
39	Kitsap	West Bainbridge, Westwood	47.627	-122.578	EPA - NEP grant	11/13/2012	1/10/2013		
40	Kitsap	Eagle Harbor, Bainbridge Ferry Terminal	47.623	-122.510	EPA - NEP grant	11/13/2012	1/10/2013		
41	Kitsap	Illahee Creek	47.616	-122.595	Kitsap County Public Works	11/13/2012	1/8/2013		
42	Kitsap	Sinclair Inlet, Waterman Point	47.585	-122.571	EPA - NEP grant	11/13/2012	1/9/2013		
43	Kitsap	Hood Canal, Holly	47.571	-122.972	Kitsap County Public Works	11/14/2012	1/11/2013		
44	Kitsap	Bremerton Shipyard, Ferry Terminal	47.560	-122.628	EPA - NEP grant	11/14/2012	1/9/2013		
45	Kitsap	Bremerton Shipyard, Charleston Beach	47.552	-122.661	EPA - NEP grant	11/14/2012	1/8/2013		
46	Kitsap	Manchester, Stormwater Outfall	47.556	-122.543	Kitsap County Public Works	11/13/2012	1/8/2013		
47	Kitsap	Sinclair Inlet, Sinclair Marina	47.541	-122.642	EPA - NEP grant	11/13/2012	1/9/2013		
48	Kitsap	Colvos Passage, Olalla, Prospect Point Beach	47.423	-122.537	EPA - NEP grant	11/14/2012	1/8/2013		
49	Mason	Case Inlet, Allyn	47.384	-122.826	EPA - NEP grant	11/14/2012	1/10/2013		
50	Mason	Shelton, Oak Bay Marina	47.214	-123.086	EPA - NEP grant	11/12/2012	1/7/2013		
51	Mason	Totten Inlet	47.133	-123.022	EPA - NEP grant	11/14/2012	1/7/2013		
52	Pierce	Gig Harbor, Narrows Passage	47.326	-122.576	EPA - NEP grant	11/14/2012	1/8/2013		
53	Pierce	Point Defiance Park	47.313	-122.528	EPA - NEP grant	11/12/2012	1/9/2013		
54	Pierce	Kopachuck State Park	47.310	-122.688	EPA - NEP grant	11/14/2012	1/8/2013		
55	Pierce	Commencement Bay, Skookum Wuldge	47.290	-122.410	EPA - NEP grant	11/13/2012	1/9/2013		
56	Pierce	Tacoma, Ruston Way, Puget Creek	47.281	-122.477	EPA - NEP grant	11/13/2012	1/9/2013		
57^	Pierce	Tacoma Ruston Waterfront 1	47.281	-122.477	TPCHD	11/13/2012	1/9/2013		
NA	Pierce	Tacoma Ruston Waterfront 2	47.281	-122.476	TPCHD	11/14/2012	1/9/2013		

Site Order	County	Site Name	Latitude	Longitude	Fund Source	Deployment Date	Retrieval Date	Cage Lost	Native Mussel Sample
NA	Pierce	Tacoma Ruston Waterfront 3	47.280	-122.474	TPCHD	11/13/2012	1/9/2013		
NA	Pierce	Tacoma Ruston Waterfront 4	47.279	-122.474	TPCHD	11/13/2012	1/9/2013		
NA	Pierce	Tacoma Ruston Waterfront 5	47.279	-122.473	TPCHD	11/13/2012	1/9/2013		
NA	Pierce	Tacoma Ruston Waterfront 6	47.278	-122.472	TPCHD	11/13/2012	1/9/2013		
NA	Pierce	Tacoma Ruston Waterfront 7	47.278	-122.471	TPCHD	11/13/2012	1/9/2013		
NA	Pierce	Tacoma Ruston Waterfront 8	47.277	-122.469	TPCHD	11/13/2012	1/9/2013		
NA	Pierce	Tacoma Ruston Waterfront 9	47.277	-122.468	TPCHD	11/13/2012	1/9/2013		
58	Pierce	Fox Island, Tanglewood Island	47.265	-122.644	EPA - NEP grant	11/12/2012	1/9/2013		
59^^	Pierce	Hylebos Waterway 1	47.270	-122.377	TPCHD	11/14/2012	1/9/2013		
NA	Pierce	Hylebos Waterway 2	47.270	-122.376	TPCHD	11/14/2012	1/9/2013		
NA	Pierce	Hylebos Waterway 3	47.269	-122.375	TPCHD	11/14/2012	1/9/2013		
NA	Pierce	Hylebos Waterway 4	47.268	-122.374	TPCHD	11/14/2012	1/9/2013		
NA	Pierce	Hylebos Waterway 5	47.268	-122.373	TPCHD	11/14/2012	1/9/2013		
NA	Pierce	Hylebos Waterway 6	47.267	-122.372	TPCHD	11/14/2012	1/9/2013		
NA	Pierce	Hylebos Waterway 7	47.267	-122.371	TPCHD	11/14/2012	1/9/2013		
NA	Pierce	Hylebos Waterway 8	47.266	-122.370	TPCHD	11/14/2012	1/9/2013		
NA	Pierce	Hylebos Waterway 9	47.265	-122.369	TPCHD	11/14/2012	1/9/2013		
60	Pierce	Hylebos Waterway Natives	47.268	-122.373	TPCHD	NA	*1/9/2013		X
61	Pierce	Thea Foss Waterway	47.259	-122.435	EPA - NEP grant	11/13/2012	1/9/2013		
62	Pierce	Steilacoom, Sunnyside Beach Park	47.179	-122.590	EPA - NEP grant	11/12/2012	1/8/2013		
63	Pierce	Nisqually Reach Aquatic Reserve, Anderson Island, Sandy Bay	47.149	-122.676	DNR Aquatic Reserves Program	11/14/2012	1/7/2013		
64	San Juan	North Shore, Orcas Island	48.711	-122.929	San Juan County MRC	11/13/2012	1/9/2013		
65	San Juan	Friday Harbor Labs, San Juan Island	48.545	-123.013	San Juan County MRC	11/13/2012	1/9/2013		
66	San Juan	Fisherman's Bay, Weeks Wetland, Lopez Island	48.519	-122.917	San Juan County MRC	11/13/2012	1/9/2013		

Site Order	County	Site Name	Latitude	Longitude	Fund Source	Deployment Date	Retrieval Date	Cage Lost	Native Mussel Sample
67	Skagit	Larrabee State Park	48.642	-122.486	EPA - NEP grant	11/13/2012	1/8/2013		
68	Skagit	Cypress Island Aquatic Reserve, Strawberry Bay	48.564	-122.722	DNR Aquatic Reserves Program	11/13/2012	1/14/2013		
69	Skagit	Cypress Island Aquatic Reserve, Secret Harbor	48.554	-122.688	DNR Aquatic Reserves Program	11/13/2012	1/14/2013		
70	Skagit	Anacortes, Guemes Ferry	48.519	-122.624	EPA - NEP grant	11/14/2012	1/8/2013		
71	Skagit	March Point	48.500	-122.567	EPA - NEP grant	11/13/2012	1/8/2013		
72	Skagit	Padilla Bay	48.492	-122.487	EPA - NEP grant	11/13/2012	1/8/2013		
73	Skagit	Fidalgo Bay Aquatic Reserve, Weaverling Spit	48.482	-122.584	DNR Aquatic Reserves Program	11/14/2012	1/8/2013		
74	Skagit	Skagit River Delta	48.334	-122.437	EPA - NEP grant	11/12/2012	1/9/2013		
75	Snohomish	Port Susan, Warm Beach	48.171	-122.367	Stillaguamish River Clean Water District	11/14/2012	1/7/2013		
76	Snohomish	Kayak Point	48.134	-122.366	EPA - NEP grant	11/14/2012	1/7/2013		
77	Snohomish	Kayak Point Natives	48.134	-122.366	Snohomish County MRC	NA	*1/7/2013		X
78	Snohomish	Hermosa Point	48.062	-122.293	EPA - NEP grant	11/14/2012	1/7/2013		
79	Snohomish	Hermosa Point Natives	48.062	-122.293	Tulalip Tribes	NA	*1/7/2013		X
80	Snohomish	Everett Harbor	47.972	-122.232	EPA - NEP grant	11/14/2012	1/7/2013		
81	Snohomish	Everett Harbor Natives	47.972	-122.232	Snohomish County MRC	NA	*1/7/2013		X
82	Snohomish	Mukilteo WWTP, Big Gulch	47.911	-122.322	EPA - NEP grant	11/12/2012	1/8/2013		
83	Snohomish	Edmonds Ferry	47.814	-122.382	EPA - NEP grant	11/14/2012	1/7/2013		
84	Snohomish	Edmonds Ferry Natives	47.814	-122.382	Snohomish County MRC	NA	*1/7/2013		X
85	Thurston	Johnson Point	47.178	-122.816	EPA - NEP grant	11/13/2012	1/11/2013		
86	Thurston	Tolmie State Park	47.122	-122.773	EPA - NEP grant	11/12/2012	1/8/2013		
87	Thurston	Olympia, Budd Inlet, North Point	47.061	-122.905	EPA - NEP grant	11/13/2012	1/11/2013		
88	Whatcom	Birch Point	48.939	-122.820	EPA - NEP grant	11/13/2012	1/8/2013		
89	Whatcom	Cherry Point Aquatic Reserve, Birch Bay South	48.896	-122.785	EPA - NEP grant	11/13/2012	1/8/2013		

Site Order	County	Site Name	Latitude	Longitude	Fund Source	Deployment Date	Retrieval Date	Cage Lost	Native Mussel Sample
90	Whatcom	Cherry Point Aquatic Reserve, 1 Alcoa-BP	48.858	-122.741	SSA Marine	11/14/2012	1/9/2013		
NA	Whatcom	Cherry Point Aquatic Reserve, 2 Alcoa-BP	48.857	-122.736	SSA Marine	11/14/2012	NA	X	
91	Whatcom	Cherry Point Aquatic Reserve, 3 Alcoa-BP	48.855	-122.727	SSA Marine	11/14/2012	1/9/2013		
92	Whatcom	Cherry Point Aquatic Reserve, 4 Conoco Phillips	48.821	-122.710	DNR Aquatic Reserves Program	11/14/2012	1/9/2013		
93	Whatcom	West Bellingham Bay, Lummi Nation	48.751	-122.619	EPA - NEP grant	11/13/2012	1/8/2013		
94	Whatcom	Bellingham Bay, Little Squalicum Creek	48.764	-122.518	City of Bellingham	11/13/2012	1/8/2013		
95	Whatcom	Bellingham Bay, Squalicum Harbor	48.753	-122.499	EPA - NEP grant	11/13/2012	1/8/2013		
96	Whatcom	Bellingham Bay, Post Point	48.719	-122.517	EPA - NEP grant	11/13/2012	1/8/2013		

EPA - NEP grant = Environmental Protection Agency - National Estuary Program grant

NA = not applicable

* indicates the date mussels were collected from that location

^ data for Tacoma Ruston Waterfront sites 1-9 averaged in subsequent tables

^^ data for Hylebos Waterway sites 1-9 averaged in subsequent tables

DNR = Washington Department of Natural Resources

MRC = Marine Resources Committee

WWTP = wastewater treatment plant

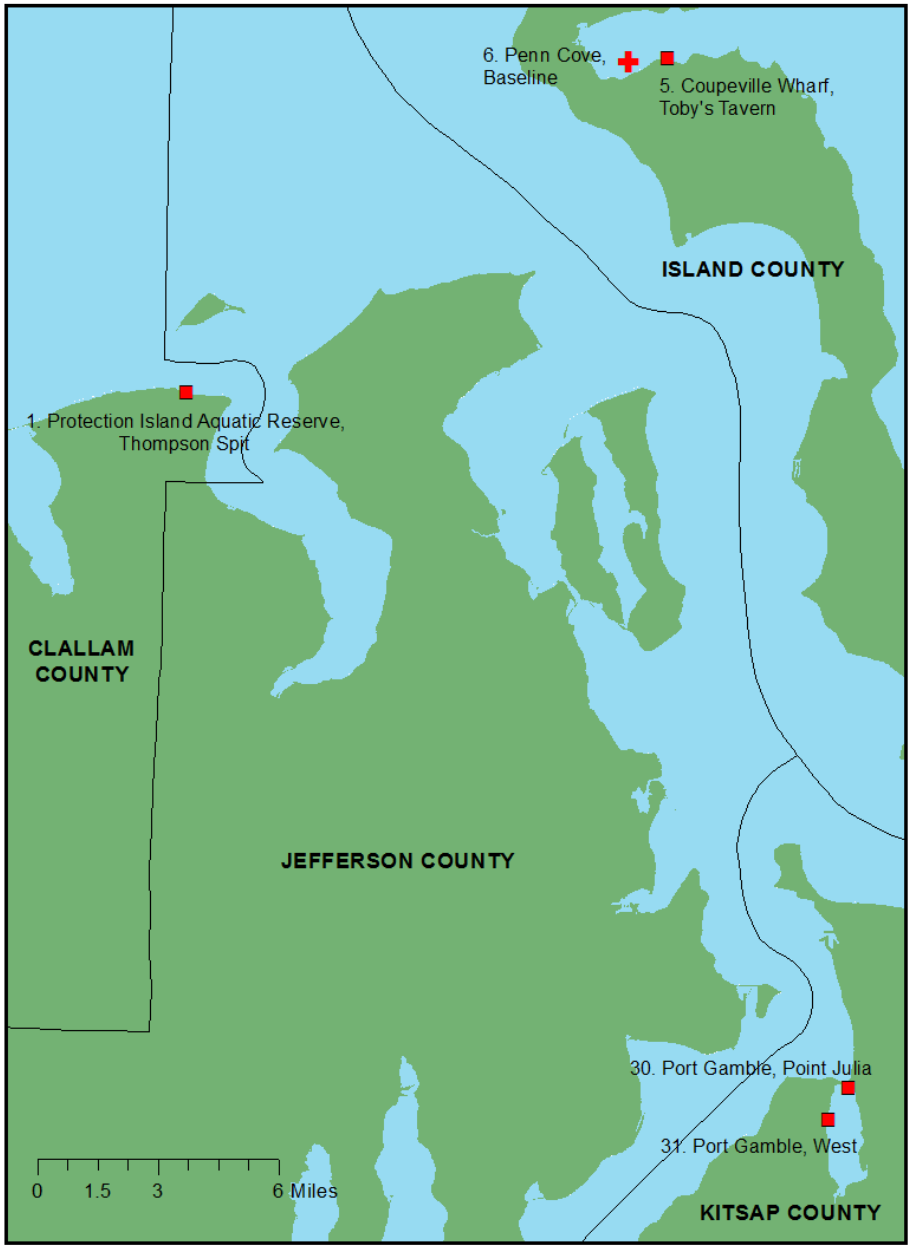
DNRP = Department of Natural Resources and Parks

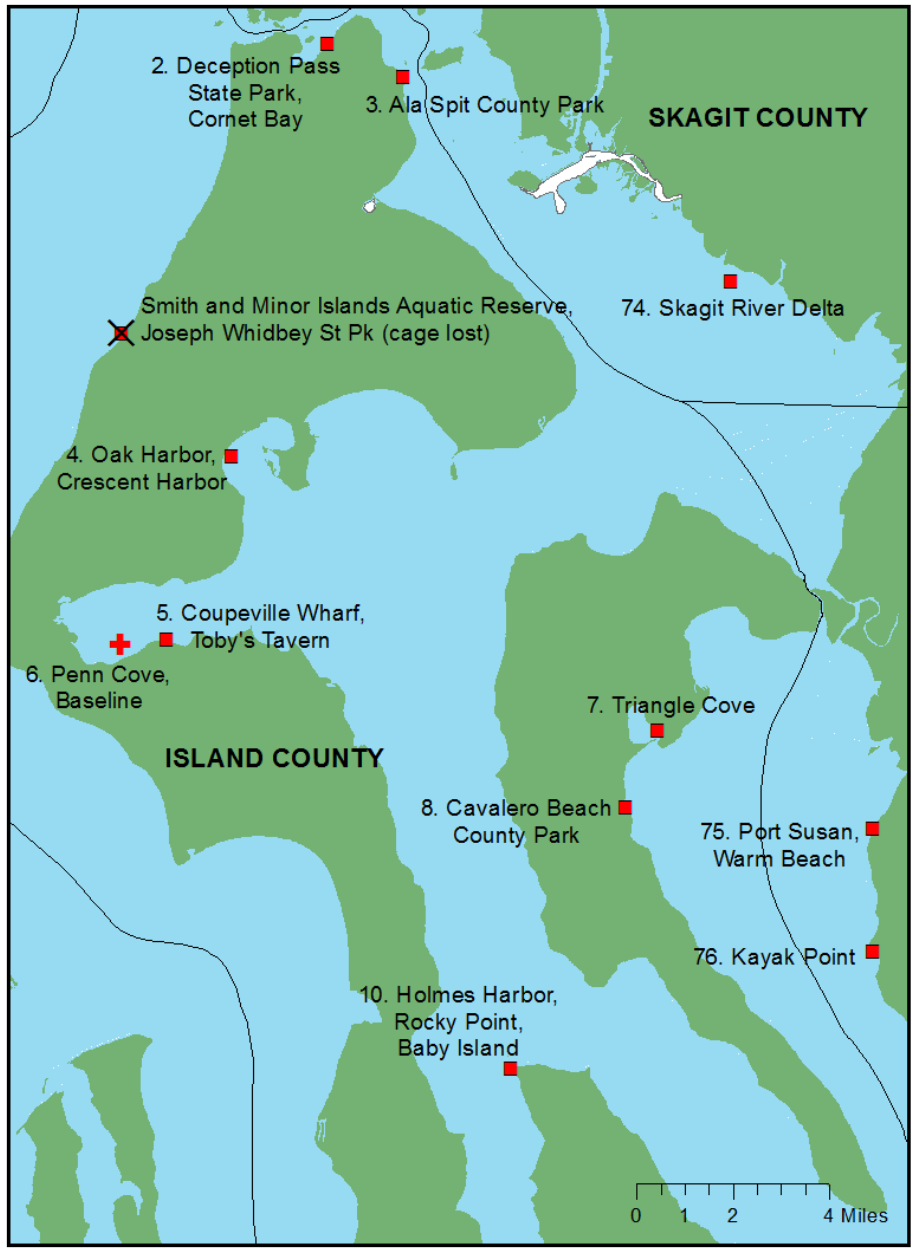
† biological and chemical data for these nine consecutively placed sites are averaged in subsequent tables under the site name “Hylebos Waterway”

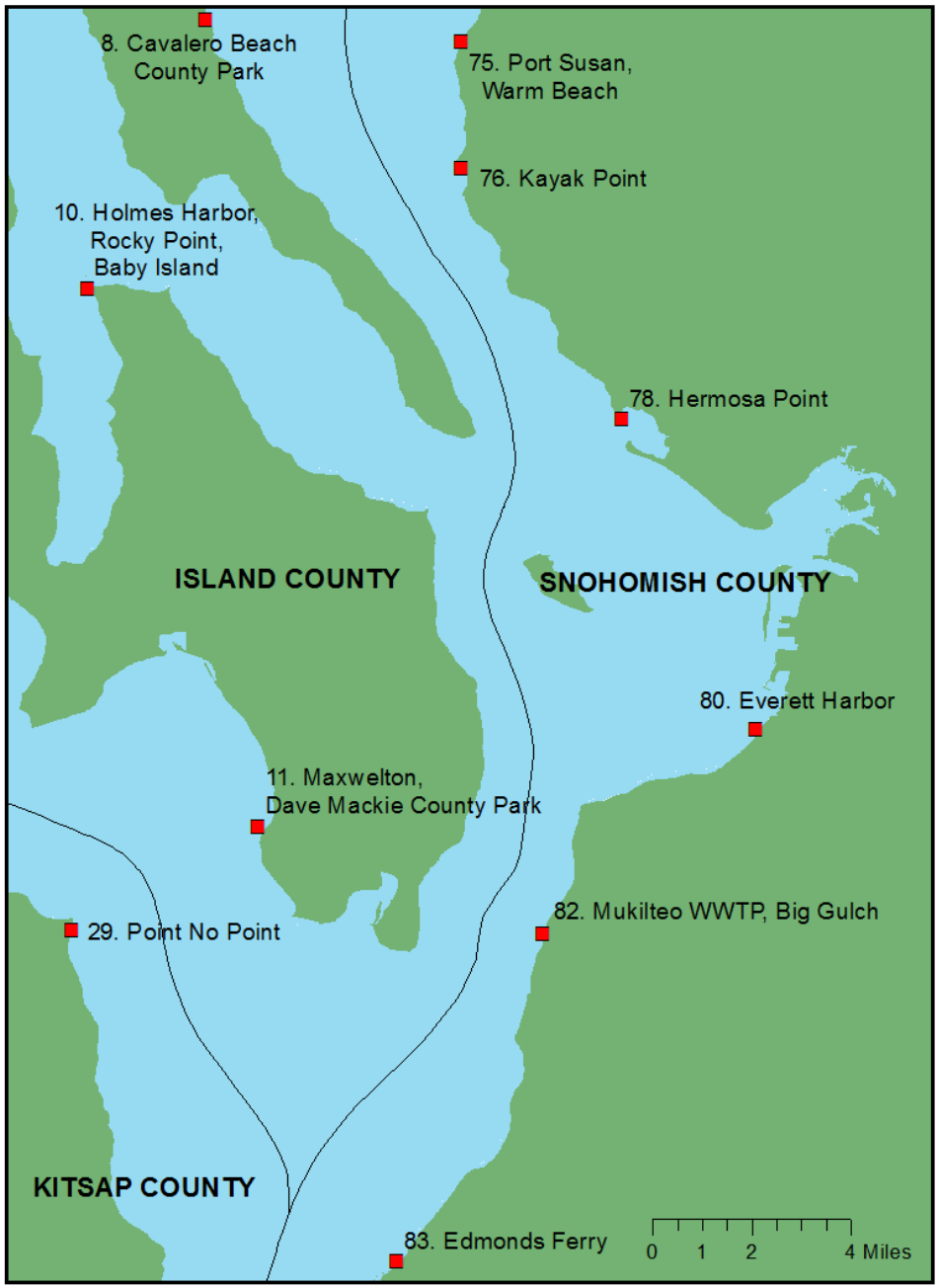
TPCHD = Tacoma-Pierce County Health Department

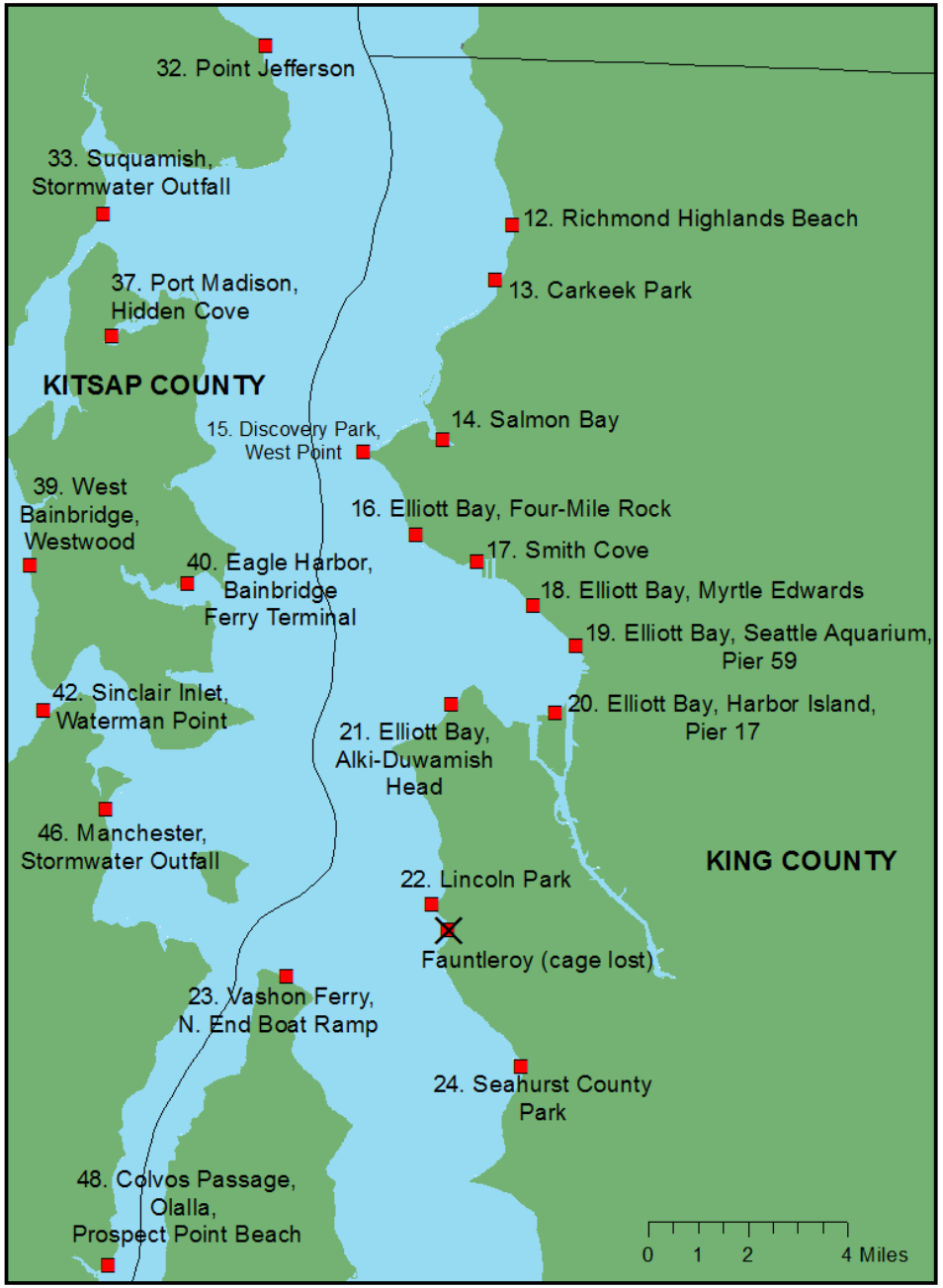
‡ biological and chemical data for these nine consecutively placed sites are averaged in subsequent tables under the site name “Tacoma Ruston Waterfront”

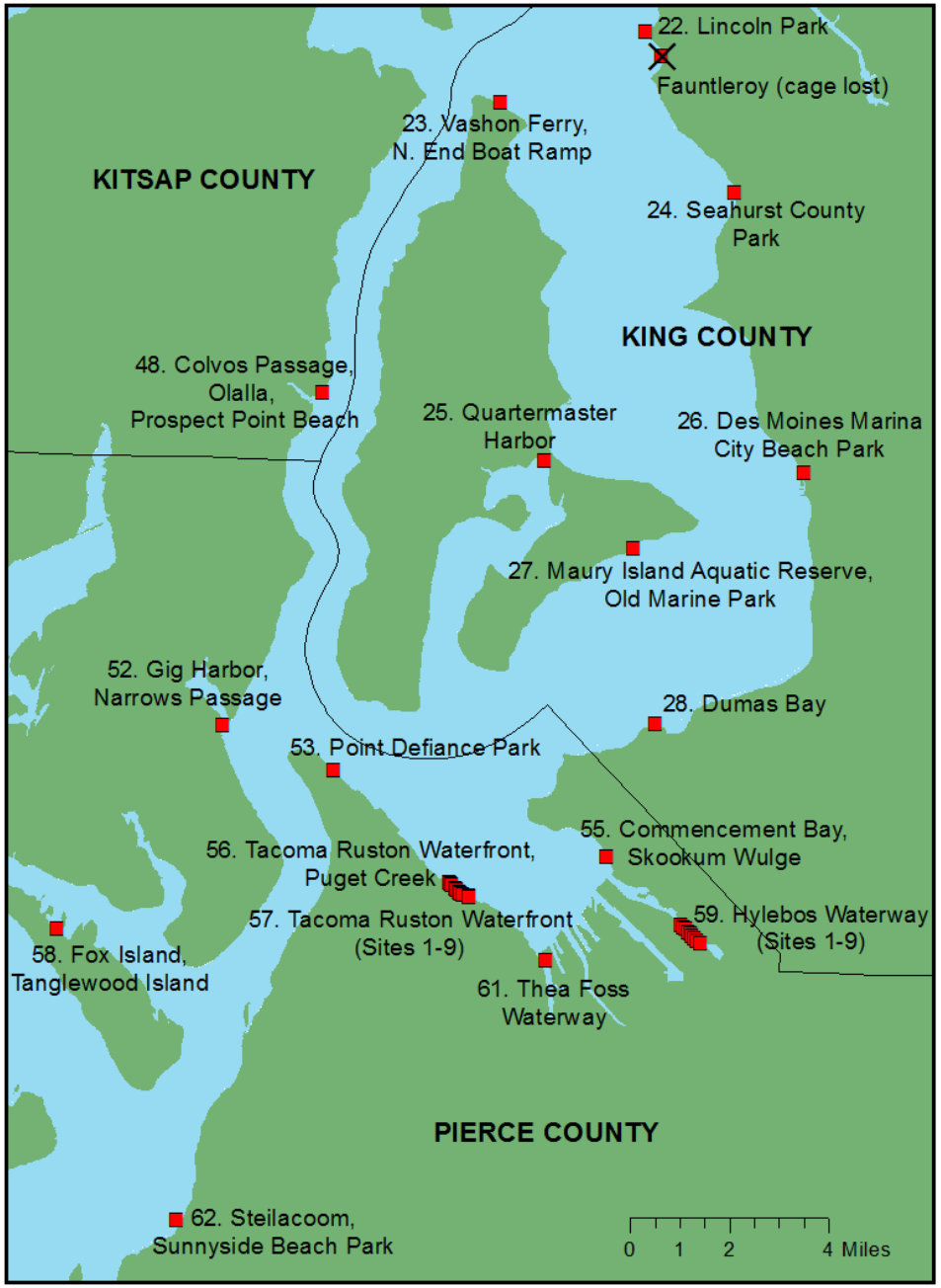
8.1 Maps of Transplanted (i.e. Caged) Mussel Sites by County

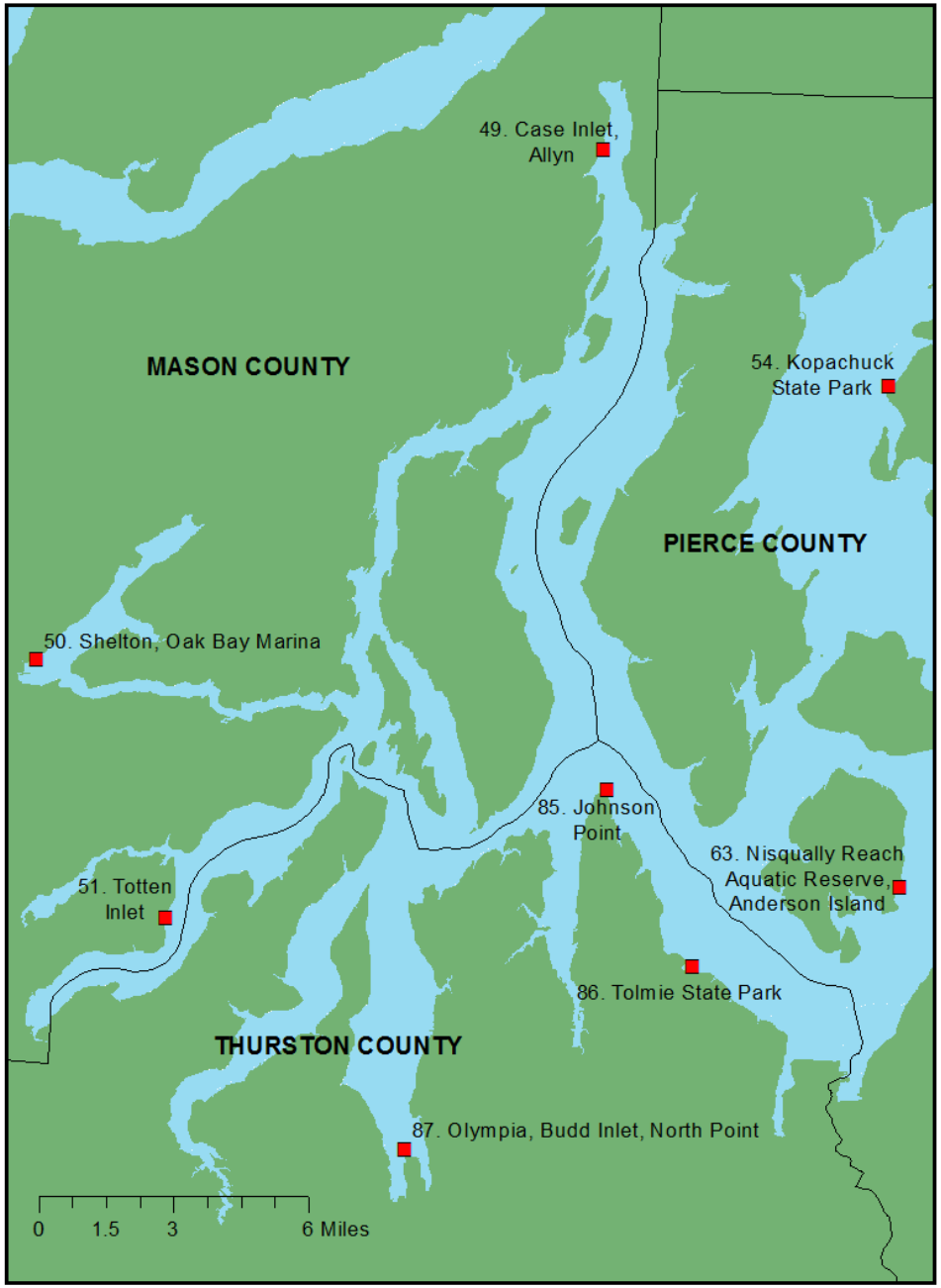


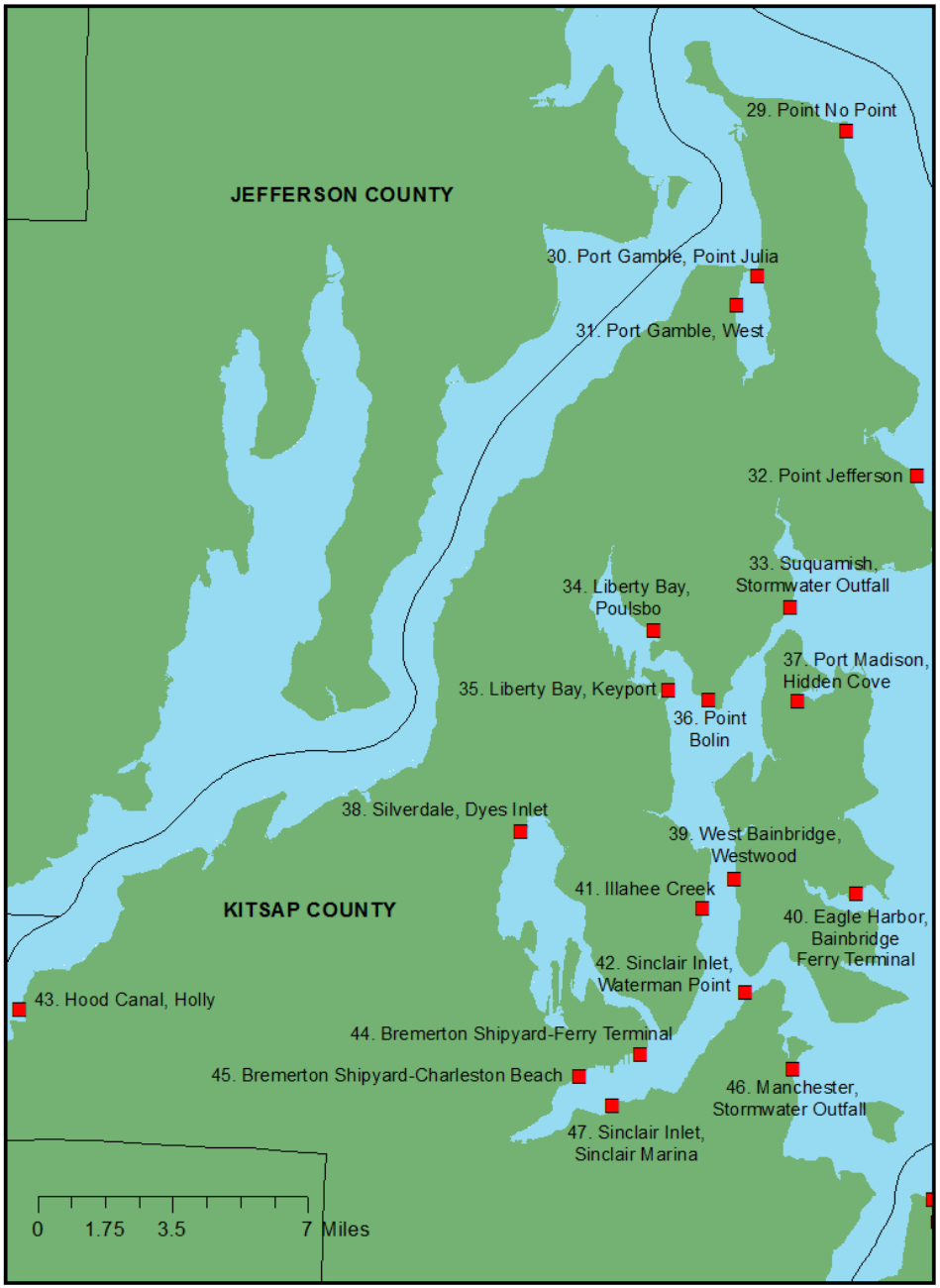


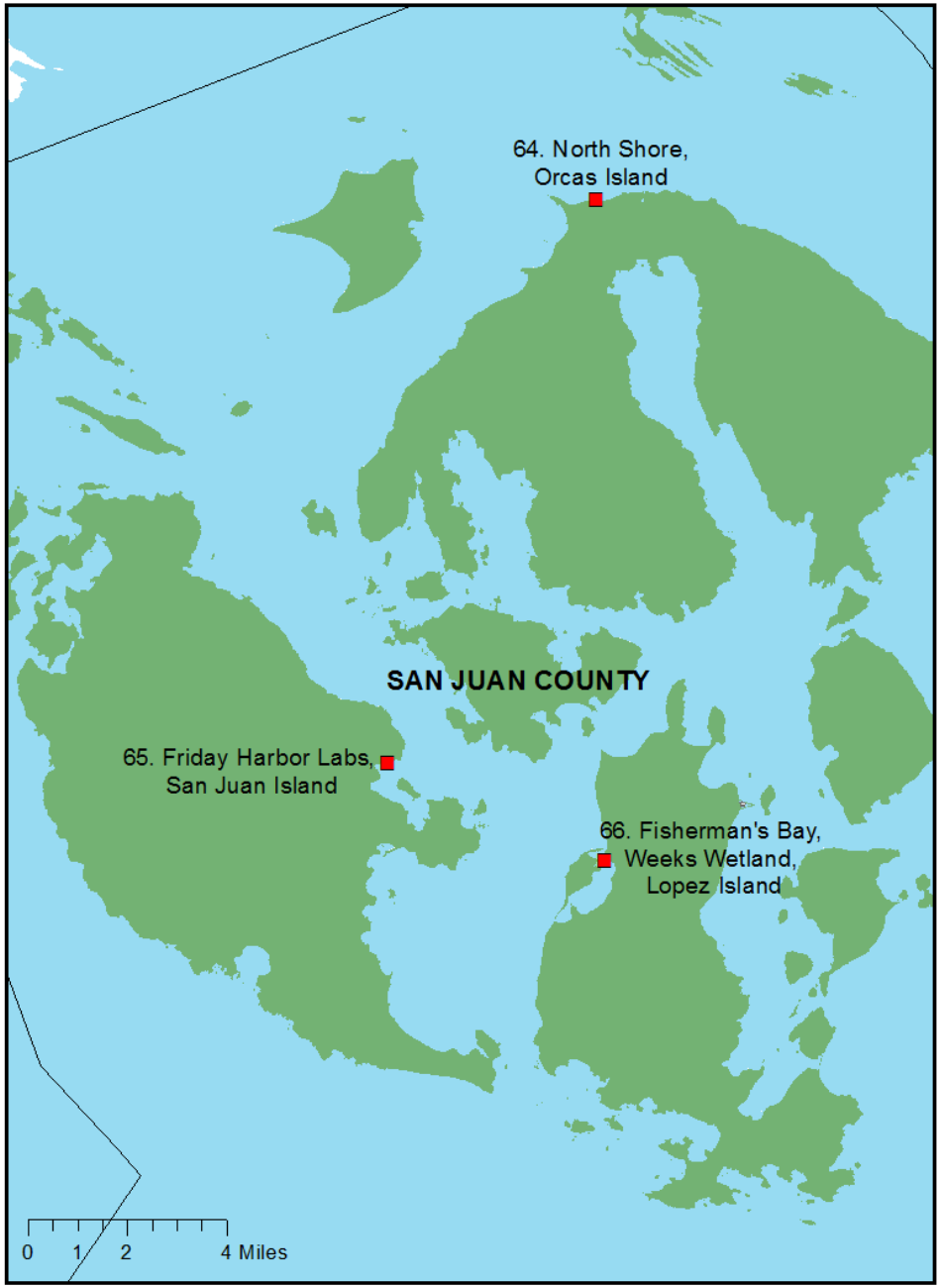

8. Cavalero Beach County Park
75. Port Susan, Warm Beach
76. Kayak Point
10. Holmes Harbor, Rocky Point, Baby Island
78. Hermosa Point
ISLAND COUNTY
SNOHOMISH COUNTY
80. Everett Harbor
11. Maxwelton, Dave Mackie County Park
82. Mukilteo WWTP, Big Gulch
29. Point No Point
KITSAP COUNTY
83. Edmonds Ferry
0 1 2 4 Miles

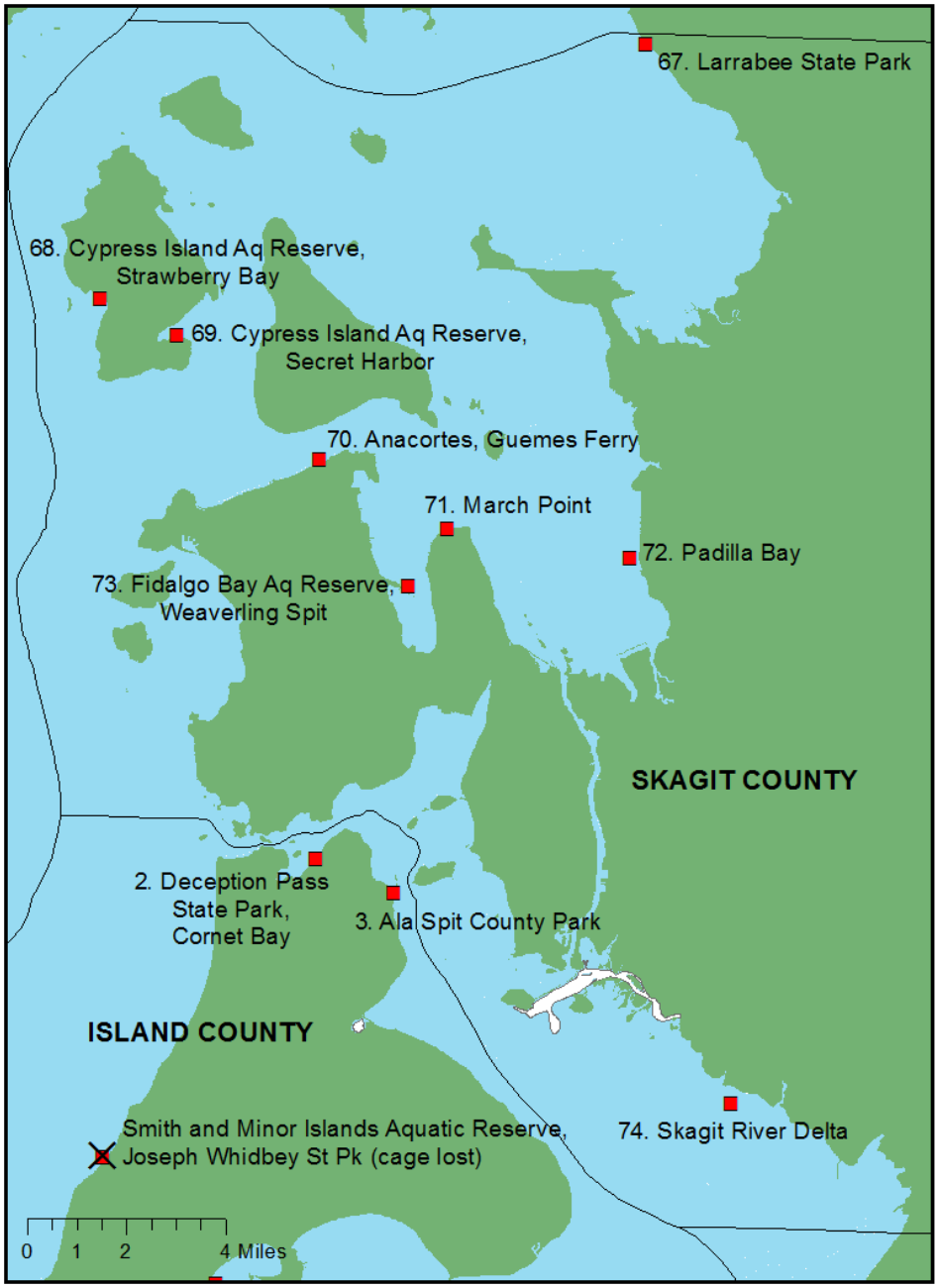

32. Point Jefferson
33. Suquamish, Stormwater Outfall
12. Richmond Highlands Beach
37. Port Madison, Hidden Cove
13. Carkeek Park
KITSAP COUNTY
14. Salmon Bay
15. Discovery Park, West Point
39. West Bainbridge, Westwood
16. Elliott Bay, Four-Mile Rock
17. Smith Cove
40. Eagle Harbor, Bainbridge Ferry Terminal
18. Elliott Bay, Myrtle Edwards
19. Elliott Bay, Seattle Aquarium, Pier 59
42. Sinclair Inlet, Waterman Point
20. Elliott Bay, Harbor Island, Pier 17
21. Elliott Bay, Alki-Duwamish Head
46. Manchester, Stormwater Outfall
KING COUNTY
22. Lincoln Park
Fauntleroy (cage lost)
23. Vashon Ferry, N. End Boat Ramp
24. Seahurst County Park
48. Colvos Passage, Olalla, Prospect Point Beach
0 1 2 4 Miles

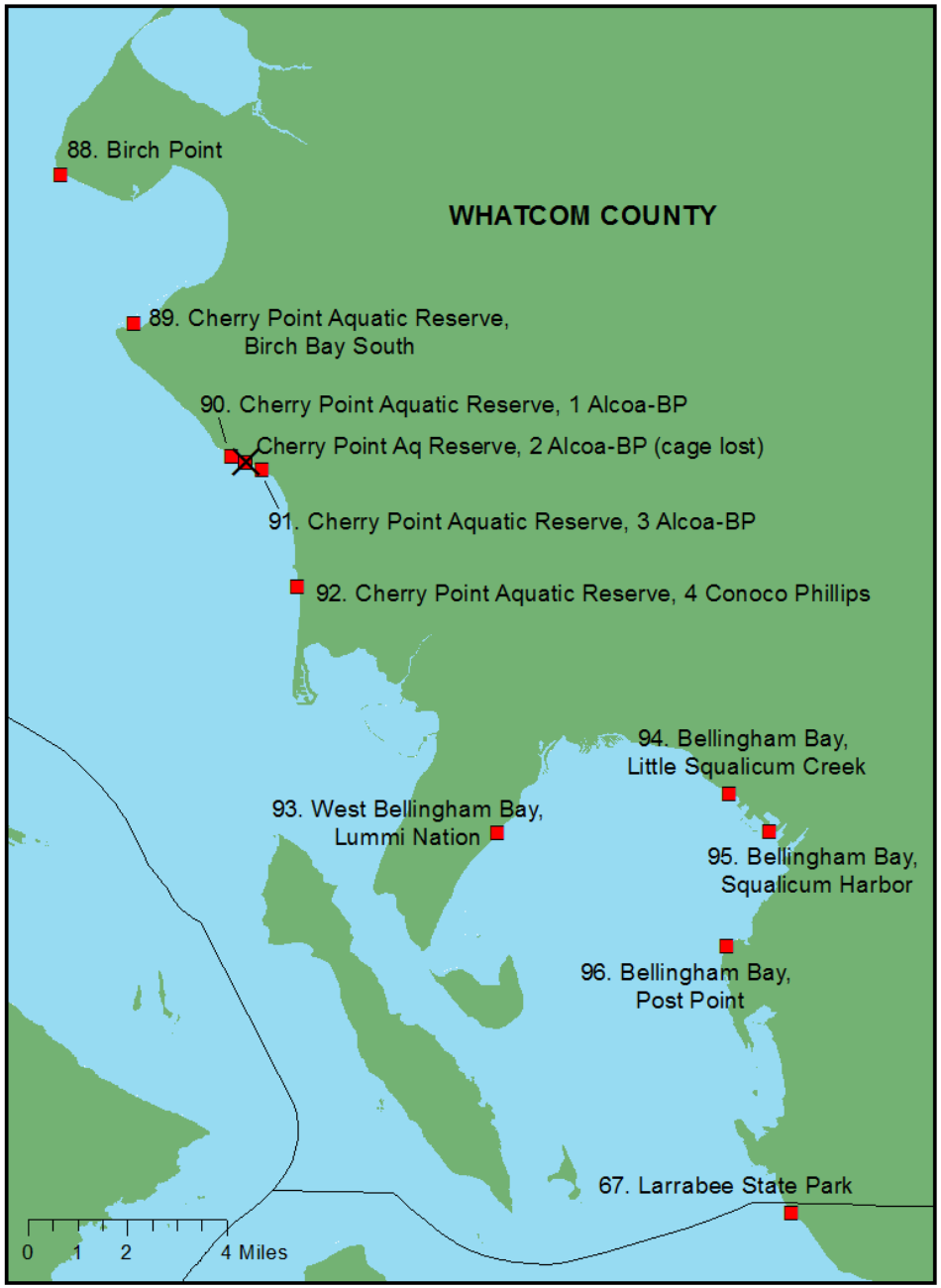

22. Lincoln Park
Fauntleroy (cage lost)
23. Vashon Ferry, N. End Boat Ramp
KITSAP COUNTY
24. Seahurst County Park
KING COUNTY
48. Colvos Passage, Olalla, Prospect Point Beach
25. Quartermaster Harbor
26. Des Moines Marina City Beach Park
27. Maury Island Aquatic Reserve, Old Marine Park
52. Gig Harbor, Narrows Passage
28. Dumas Bay
53. Point Defiance Park
55. Commencement Bay, Skookum Wulge
56. Tacoma Ruston Waterfront, Puget Creek
57. Tacoma Ruston Waterfront (Sites 1-9)
59. Hylebos Waterway (Sites 1-9)
58. Fox Island, Tanglewood Island
61. Thea Foss Waterway
PIERCE COUNTY
62. Steilacoom, Sunnyside Beach Park
0 1 2 4 Miles


49. Case Inlet, Allyn
MASON COUNTY
54. Kopachuck State Park
PIERCE COUNTY
50. Shelton, Oak Bay Marina
85. Johnson Point
63. Nisqually Reach Aquatic Reserve, Anderson Island
51. Totten Inlet
86. Tolmie State Park
THURSTON COUNTY
87. Olympia, Budd Inlet, North Point
0 1.5 3 6 Miles


JEFFERSON COUNTY
29. Point No Point
30. Port Gamble, Point Julia
31. Port Gamble, West
32. Point Jefferson
33. Suquamish, Stormwater Outfall
34. Liberty Bay, Poulsbo
35. Liberty Bay, Keyport
36. Point Bolin
37. Port Madison, Hidden Cove
38. Silverdale, Dyes Inlet
39. West Bainbridge, Westwood
40. Eagle Harbor, Bainbridge Ferry Terminal
41. Illahee Creek
42. Sinclair Inlet, Waterman Point
43. Hood Canal, Holly
44. Bremerton Shipyard-Ferry Terminal
45. Bremerton Shipyard-Charleston Beach
46. Manchester, Stormwater Outfall
47. Sinclair Inlet, Sinclair Marina
KITSAP COUNTY
0 1.75 3.5 7 Miles


64. North Shore, Orcas Island
SAN JUAN COUNTY
65. Friday Harbor Labs, San Juan Island
66. Fisherman's Bay, Weeks Wetland, Lopez Island
0 1 2 4 Miles


67. Larrabee State Park
68. Cypress Island Aq Reserve, Strawberry Bay
69. Cypress Island Aq Reserve, Secret Harbor
70. Anacortes, Guemes Ferry
71. March Point
72. Padilla Bay
73. Fidalgo Bay Aq Reserve, Weaverling Spit
SKAGIT COUNTY
2. Deception Pass State Park, Cornet Bay
3. Ala Spit County Park
ISLAND COUNTY
Smith and Minor Islands Aquatic Reserve, Joseph Whidbey St Pk (cage lost)
74. Skagit River Delta
0 1 2 4 Miles


88. Birch Point
WHATCOM COUNTY
89. Cherry Point Aquatic Reserve, Birch Bay South
90. Cherry Point Aquatic Reserve, 1 Alcoa-BP
Cherry Point Aq Reserve, 2 Alcoa-BP (cage lost)
91. Cherry Point Aquatic Reserve, 3 Alcoa-BP
92. Cherry Point Aquatic Reserve, 4 Conoco Phillips
94. Bellingham Bay, Little Squalicum Creek
93. West Bellingham Bay, Lummi Nation
95. Bellingham Bay, Squalicum Harbor
96. Bellingham Bay, Post Point
67. Larrabee State Park
0 1 2 4 Miles

9 APPENDIX B: Biological Effects Data

Site order is arranged alphabetically by county and then from north to south within each county.

Site Order	County	Site Name	Condition Index (CI) n = 12 (unless otherwise indicated)	Growth Rate (mm/day) n = 44	Mortality (%) n = 64
1	Clallam	Protection Island Aquatic Reserve, Thompson Spit	2.10	0.01	3.13
2	Island	Deception Pass State Park, Cornet Bay	2.34	0.02	7.81
3	Island	Ala Spit County Park	2.17	0.01	15.63
4	Island	Oak Harbor, Crescent Harbor	2.27	0.02	10.94
5	Island	Coupeville Wharf, Toby's Tavern	2.07	0.03	14.06
6	Island	PENN COVE, BASELINE	2.51 (n = 100)	NA	NA
7	Island	Triangle Cove	2.25	0.01	17.19
8	Island	Cavalero Beach Co. Park	2.31	0.02	7.81
9	Island	Cavalero Beach Natives	0.59	NA	NA
10	Island	Holmes Harbor, Rocky Point, Baby Island	2.77	0.04	6.25
11	Island	Maxwelton, Dave Mackie County Park	2.36	0.02	10.94
12	King	Richmond Highlands Beach	2.20	0.03	10.94
13	King	Carkeek Park	2.58	0.03	20.31
14	King	Salmon Bay	2.20	0.03	7.81
15	King	Discovery Park, West Point	2.43	0.02	15.63
16	King	Elliott Bay, Four-Mile Rock	2.45	0.02	17.19
17	King	Smith Cove	2.11	0.01	9.38
18	King	Elliott Bay, Myrtle Edwards	2.16	0.01	9.38
19	King	Elliott Bay, Seattle Aquarium, Pier 59	2.71	0.02	18.75
20	King	Elliott Bay, Harbor Island, Pier 17	2.51	0.01	28.13
21	King	Elliott Bay, Alki-Duwamish Head	2.08	0.02	15.63
22	King	Lincoln Park	2.67	0.01	10.94
23	King	Vashon Ferry, N. End Boat Ramp	2.51	0.01	17.19

Site Order	County	Site Name	Condition Index (CI) n = 12 (unless otherwise indicated)	Growth Rate (mm/day) n = 44	Mortality (%) n = 64
24	King	Seahurst County Park	2.30	0.03	6.25
25	King	Quartermaster Harbor	2.52	0.04	15.63
26	King	Des Moines Marina City Beach Park	2.45	0.02	14.06
27	King	Maury Island Aquatic Reserve, Old Marine Park	2.26	0.02	7.81
28	King	Dumas Bay	2.35	0.03	12.50
29	Kitsap	Point No Point	2.37	0.01	6.25
30	Kitsap	Port Gamble, Point Julia	2.20	0.02	9.38
31	Kitsap	Port Gamble, West	2.26	0.02	9.38
32	Kitsap	Point Jefferson	2.53	0.01	10.94
33	Kitsap	Suquamish, Stormwater Outfall	2.09	0.00	26.56
34	Kitsap	Liberty Bay, Poulsbo	2.36	0.04	18.75
35	Kitsap	Liberty Bay, Keyport	2.60	0.03	15.63
36	Kitsap	Point Bolin	2.28	0.03	4.69
37	Kitsap	Port Madison, Hidden Cove	2.87	0.02	10.94
38	Kitsap	Silverdale, Dyes Inlet	2.27	0.02	9.38
39	Kitsap	West Bainbridge, Westwood	2.29	0.01	4.69
40	Kitsap	Eagle Harbor, Bainbridge Ferry Terminal	1.96 (n = 8)	0.01	37.50
41	Kitsap	Illahee Creek	2.53 (n = 9)	0.01	14.06
42	Kitsap	Sinclair Inlet, Waterman Point	2.13	0.01	17.19
43	Kitsap	Hood Canal, Holly	1.96	0.02	4.69
44	Kitsap	Bremerton Shipyard, Ferry Terminal	2.16	0.01	10.94
45	Kitsap	Bremerton Shipyard, Charleston Beach	2.23	0.02	9.38
46	Kitsap	Manchester, Stormwater Outfall	2.08	0.01	6.25
47	Kitsap	Sinclair Inlet, Sinclair Marina	2.29	0.02	20.31
48	Kitsap	Colvos Passage, Olalla, Prospect Point Beach	2.18	0.01	12.50
49	Mason	Case Inlet, Allyn	2.08	0.03	14.06
50	Mason	Shelton, Oak Bay Marina	2.60	0.02	6.25
51	Mason	Totten Inlet	2.40	0.02	6.25
52	Pierce	Gig Harbor, Narrows Passage	2.18	0.01	14.06

Site Order	County	Site Name	Condition Index (CI) n = 12 (unless otherwise indicated)	Growth Rate (mm/day) n = 44	Mortality (%) n = 64
53	Pierce	Point Defiance Park	2.07	0.01	18.75
54	Pierce	Kopachuck State Park	2.13	0.01	4.69
55	Pierce	Commencement Bay, Skookum Wuldge	2.38	0.01	28.13
56	Pierce	Tacoma, Ruston Way, Puget Creek	2.55	0.01	21.88
57	Pierce	Tacoma Ruston Waterfront	2.31 (n = 108)	0.01	16.49
58	Pierce	Fox Island, Tanglewood Island	2.15	0.01	15.63
59	Pierce	Hylebos Waterway	2.22 (n = 100)	0.01	15.80
60	Pierce	Hylebos Waterway Native	1.41	NA	NA
61	Pierce	Thea Foss Waterway	2.24	0.01	12.50
62	Pierce	Steilacoom, Sunnyside Beach Park	2.61	0.02	20.31
63	Pierce	Nisqually Reach Aq Reserve, Anderson Island, Sandy Bay	2.23	0.01	12.50
64	San Juan	North Shore, Orcas Island	2.15	0.01	23.44
65	San Juan	Friday Harbor Labs, San Juan Island	2.20	0.01	15.63
66	San Juan	Fisherman's Bay, Weeks Wetland, Lopez Island	2.32	0.01	6.25
67	Skagit	Larrabee State Park	2.37	0.00	12.50
68	Skagit	Cypress Island Aquatic Reserve, Strawberry Bay	2.28	0.01	7.81
69	Skagit	Cypress Island Aquatic Reserve, Secret Harbor	2.64	0.01	12.50
70	Skagit	Anacortes, Guemes Ferry	2.10	0.00	14.06
71	Skagit	March Point	2.19	0.00	9.38
72	Skagit	Padilla Bay	2.28	0.01	4.69
73	Skagit	Fidalgo Bay Aquatic Reserve, Weaverling Spit	2.18	0.02	6.25
74	Skagit	Skagit River Delta	1.84	0.00	7.81
75	Snohomish	Port Susan, Warm Beach	2.61	0.01	7.81
76	Snohomish	Kayak Point	2.08	0.02	14.06
77	Snohomish	Kayak Point Natives	1.24	NA	NA
78	Snohomish	Hermosa Point	2.09	0.01	7.81
79	Snohomish	Hermosa Point Natives	0.99	NA	NA
80	Snohomish	Everett Harbor	2.33	0.02	10.94

Site Order	County	Site Name	Condition Index (CI) n = 12 (unless otherwise indicated)	Growth Rate (mm/day) n = 44	Mortality (%) n = 64
81	Snohomish	Everett Harbor Natives	1.10	NA	NA
82	Snohomish	Mukilteo WWTP, Big Gulch	2.29	0.02	7.81
83	Snohomish	Edmonds Ferry	2.22	0.00	14.06
84	Snohomish	Edmonds Ferry Natives	0.84	NA	NA
85	Thurston	Johnson Point	1.93	0.02	18.75
86	Thurston	Tolmie State Park	2.06 (n = 11)	0.02	32.81
87	Thurston	Olympia, Budd Inlet, North Point	2.58	0.04	7.81
88	Whatcom	Birch Point	2.54	0.01	10.94
89	Whatcom	Cherry Point Aquatic Reserve, Birch Bay South	2.08	0.02	9.38
90	Whatcom	Cherry Point Aquatic Reserve, 1 Alcoa-BP	2.58	0.01	4.69
91	Whatcom	Cherry Point Aquatic Reserve, 3 Alcoa-BP	2.38	0.01	6.25
92	Whatcom	Cherry Point Aquatic Reserve, 4 Conoco Phillips	2.15	0.00	18.75
93	Whatcom	West Bellingham Bay, Lummi Nation	2.25	0.00	7.81
94	Whatcom	Bellingham Bay, Little Squalicum Creek	2.18	0.01	12.50
95	Whatcom	Bellingham Bay, Squalicum Harbor	2.19	0.00	10.94
96	Whatcom	Bellingham Bay, Post Point	2.71	0.02	15.63

NA = data not available

* biological data is an average of mussels taken from nine consecutively placed sites (see Appendix A)

WWTP = wastewater treatment plant

10 APPENDIX C: Dry Weight Organic Tissue Chemistry Data

Organic contaminants that were not detected above the limit of quantitation (LOQ) at any of the sample sites (i.e. aldrin, endosulfan 1, hexachlorocyclohexane) were not included in this table. Site order is arranged alphabetically by county and then from north to south within each county.

		Concentrations in ng/g, dry weight								
Site Order	Site Name	$\sum_{42}$PAHs	HCB	Dieldrin	Mirex	$\sum_{8}$Chlordanes	$\sum_{6}$DDTs	$\sum_{40}$PCBs	TPCBs	$\sum_{11}$PBDEs
1	Protection Island Aquatic Reserve, Thompson Spit	239.2	<1.08	<1.08	<1.08	<1.08	1.7	7.6	16.7	3.3
2	Deception Pass State Park, Cornet Bay	123.1	<1.03	<1.03	<1.03	<1.03	1.9	11.4	17.5	2.1
3	Ala Spit County Park	47.3	<0.95	<0.88	<0.95	<0.95	1.1	7.7	8.8	2.5
4	Oak Harbor, Crescent Harbor	304.2	<1.06	<1.06	<1.06	<1.06	2.0	11.1	13.7	5.5
5	Coupeville Wharf, Toby's Tavern	171.5	<0.97	1.0	<0.97	<0.97	2.2	11.9	18.3	4.2
6	PENN COVE, BASELINE	71.36	<1.85	<1.79	<1.85	<1.85	1.12	9.79	19.27	2.82
7	Triangle Cove	414.8	<1.73	<1.73	<1.73	<1.73	2.0	4.6	17.8	2.8
8	Cavalero Beach Co. Park	726.3	<1.63	<1.63	<1.63	<1.63	<1.70	<1.70	11.6	<1.70
9	Cavalero Beach Natives	436.2	<1.83	<1.83	<1.83	<1.83	1.6	7.2	10.1	6.6
10	Holmes Harbor, Rocky Point, Baby Island	211.3	<1.85	<1.85	<1.85	<1.85	2.4	5.2	21.8	6.3
11	Maxwelton, Dave Mackie County Park	108.1	<1.05	<0.98	<1.05	<1.05	1.7	15.0	20.9	4.2
12	Richmond Highlands Beach	441.5	<1.46	<1.46	<1.46	<1.46	6.3	22.9	33.0	12.9
13	Carkeek Park	329.9	<1.09	<1.09	<1.09	<1.09	6.6	40.5	58.5	9.4
14	Salmon Bay	1481.9	<1.16	2.5	<1.16	11.4	24.4	82.7	112.4	22.8
15	Discovery Park, West Point	796.4	<1.54	<1.54	<1.54	<1.54	2.5	20.1	30.8	9.4
16	Elliott Bay, Four-Mile Rock	4525.7	<1.33	1.5	<1.33	1.7	12.4	44.8	66.2	11.0
17	Smith Cove	2476.6	<0.9	<0.9	<0.9	2.0	10.0	74.1	99.1	13.2
18	Elliott Bay, Myrtle Edwards	5030.1	<1.48	<1.48	<1.55	<1.55	14.2	74.2	94.4	15.5
19	Elliott Bay, Seattle Aquarium, Pier 59	3154.9	<2.32	<2.25	<2.32	<2.32	6.3	46.8	65.9	11.2
20	Elliott Bay, Harbor Island, Pier 17	2333.3	<1.81	<1.81	<1.81	<1.81	2.3	59.4	84.8	7.2
21	Elliott Bay, Alki-Duwamish Head	376.5	<1.55	<1.55	<1.55	<1.55	1.8	13.2	19.8	3.2
22	Lincoln Park	627.2	<0.94	<0.94	<0.94	<0.94	2.0	25.0	39.9	7.6

Site Order	Site Name	$\sum_{42}$PAHs	HCB	Dieldrin	Mirex	$\sum_{8}$Chlordanes	$\sum_{6}$DDTs	$\sum_{40}$PCBs	TPCBs	$\sum_{11}$PBDEs
		Concentrations in ng/g, dry weight								
23	Vashon Ferry, N. End Boat Ramp	484.5	<1.78	<1.78	<1.78	<1.78	2.5	15.9	23.5	5.8
24	Seahurst County Park	901.6	<1.51	<1.51	<1.51	<1.51	2.7	15.3	23.3	10.5
25	Quartermaster Harbor	873.5	<1.43	<1.43	<1.43	1.7	3.9	22.2	31.7	8.2
26	Des Moines Marina City Beach Park	557.5	<1.43	<1.43	<1.43	1.7	6.0	22.6	33.2	18.4
27	Maury Island Aquatic Reserve, Old Marine Park	246.3	<1.22	<1.22	<1.22	<1.22	1.9	25.6	40.2	7.3
28	Dumas Bay	453.9	<1.79	<1.79	<1.79	<1.79	2.7	15.9	23.2	12.0
29	Point No Point	1521.4	<1.48	<1.48	<1.48	2.2	1.8	6.6	13.8	4.7
30	Port Gamble, Point Julia	180.3	<1.96	<1.96	<1.96	<1.96	<1.96	10.9	17.5	3.1
31	Port Gamble, West	205.2	<1.56	<1.56	<1.56	<1.56	1.8	11.3	20.1	4.5
32	Point Jefferson	203.5	<0.92	<0.92	<0.98	<0.98	1.8	12.5	19.0	4.3
33	Suquamish, Stormwater Outfall	348.3	<1.39	<1.39	<1.39	<1.39	2.6	24.3	33.1	9.4
34	Liberty Bay, Poulsbo	329.4	<1.05	1.3	<1.05	1.2	3.3	36.8	51.9	15.1
35	Liberty Bay, Keyport	351.1	<1.67	<1.67	<1.67	<1.67	5.4	40.3	56.1	13.3
36	Point Bolin	209.2	<0.95	1.0	<0.95	<0.95	3.0	33.2	45.3	10.8
37	Port Madison, Hidden Cove	524.1	<1.56	<1.56	<1.56	<1.56	2.7	36.0	54.8	6.7
38	Silverdale, Dyes Inlet	411.7	<2.18	<2.18	<2.18	<2.18	3.2	36.6	52.0	8.5
39	West Bainbridge, Westwood	213.1	<1.04	<1.04	<1.04	<1.04	2.3	52.0	79.1	7.6
40	Eagle Harbor, Bainbridge Ferry Terminal	2128.1	<1.32	<1.32	<1.32	<1.32	2.8	42.6	62.1	7.5
41	Illahee Creek	283.1	<1.21	<1.21	<1.21	<1.21	3.0	34.2	47.8	9.1
42	Sinclair Inlet, Waterman Point	174.6	<1.48	<1.48	<1.48	10.9	2.1	29.3	48.9	8.8
43	Hood Canal, Holly	70.3	<1.13	<1.13	<1.13	<1.13	1.4	24.5	41.4	5.1
44	Bremerton Shipyard, Ferry Terminal	466.5	<0.98	<0.98	<0.98	<0.98	3.4	70.7	90.0	10.3
45	Bremerton Shipyard, Charleston Beach	602.6	<1.56	<1.56	<1.56	<1.56	3.6	92.6	128.8	35.0
46	Manchester, Stormwater Outfall	312.3	1.8	<1.22	<1.22	<1.22	2.8	24.0	31.9	9.8
47	Sinclair Inlet, Sinclair Marina	912.8	<0.66	1.4	1.6	2.0	5.9	113.3	146.6	19.3
48	Colvos Passage, Olalla, Prospect Point Beach	119.6	<1.5	<1.5	<1.5	<1.5	2.0	18.9	27.2	7.0
49	Case Inlet, Allyn	122.0	<1.56	<1.56	<1.56	<1.56	2.3	15.2	23.9	2.1
50	Shelton, Oak Bay Marina	331.7	<1.77	<1.77	<1.77	<1.77	2.0	11.4	22.7	2.5
51	Totten Inlet	82.0	<0.67	<0.66	<0.67	<0.67	1.8	31.5	31.4	2.7

Site Order	Site Name	$\sum_{42}$PAHs	HCB	Dieldrin	Mirex	$\sum_{8}$Chlordanes	$\sum_{6}$DDTs	$\sum_{40}$PCBs	TPCBs	$\sum_{11}$PBDEs
		Concentrations in ng/g, dry weight								
52	Gig Harbor, Narrows Passage	285.2	<1.55	<1.55	<1.55	<1.55	2.1	46.2	71.8	7.4
53	Point Defiance Park	243.4	<1.47	<1.47	<1.47	<1.47	2.2	17.4	27.4	10.0
54	Kopachuck State Park	259.9	<2.32	<2.25	<2.32	<2.32	<2.32	6.1	12.0	3.0
55	Commencement Bay, Skookum Wuldge	814.5	<1.13	1.3	<1.13	6.0	13.8	51.0	60.6	30.2
56	Tacoma, Ruston Way, Puget Creek	470.5	<1.82	<1.82	<1.82	<1.82	2.0	15.3	22.3	7.7
57	Tacoma Ruston Waterfront	594.19	<1.76		<1.76	1.94	2.24	21.61	29.88	8.72
58	Fox Island, Tanglewood Island	100.9	<2.07	<2.07	<2.07	<2.07	<2.07	6.1	12.1	2.3
59	Hylebos Waterway	1584.95	1.53	2.59	<1.47	6.51	46.00	177.89	216.43	24.88
60	Hylebos Waterway Natives	2632.8	1.7	2.8	5.5	18.2	99.9	533.0	682.8	125.3
61	Thea Foss Waterway	1235.6	<1.42	<1.42	<1.42	2.2	4.6	25.8	37.7	9.3
62	Steilacoom, Sunnyside Beach Park	159.7	<1.44	<1.44	<1.44	<1.44	2.1	14.2	20.7	5.9
63	Nisqually Reach Aquatic Reserve, Anderson Island, Sandy Bay	51.2	<1.22	<1.22	<1.22	<1.22	2.0	25.9	40.1	7.1
64	North Shore, Orcas Island	96.0	<1.15	<1.15	<1.15	<1.15	1.6	4.5	16.0	3.4
65	Friday Harbor Labs, San Juan Island	543.8	<1.37	<1.37	<1.37	<1.37	1.7	3.8	14.5	2.4
66	Fisherman's Bay, Weeks Wetland, Lopez Island	478.8	<1.53	<1.53	<1.53	<1.53	<1.53	1.9	10.5	1.9
67	Larrabee State Park	100.6	<1.09	<1.09	<1.09	<1.09	4.6	12.3	20.1	3.5
68	Cypress Island Aquatic Reserve, Strawberry Bay	28.9	<2.29	<2.29	<2.29	<2.29	<2.29	8.0	22.8	<2.29
69	Cypress Island Aquatic Reserve, Secret Harbor	71.5	<1.07	<1.07	<1.07	<1.07	1.4	12.6	25.3	5.3
70	Anacortes, Guemes Ferry	1195.0	<1.75	<1.75	<1.75	<1.75	2.3	4.8	9.8	2.9
71	March Point	158.3	<1.8	<1.8	<1.8	<1.8	2.5	4.0	8.1	<1.8
72	Padilla Bay	50.6	<1.96	<1.96	<1.96	<1.96	6.0	2.1	4.1	<1.96
73	Fidalgo Bay Aquatic Reserve, Weaverling Spit	178.9	<1.41	<1.41	<1.41	<1.41	<1.41	3.6	15.2	2.0
74	Skagit River Delta	34.2	<0.76	0.9	<0.76	<0.76	1.7	12.1	17.6	1.7
75	Port Susan, Warm Beach	290.2	<1.46	<1.46	<1.46	<1.46	2.2	7.8	18.7	4.7
76	Kayak Point	431.6	<1.45	<1.45	<1.45	<1.45	1.9	13.5	30.5	7.9
77	Kayak Point Natives	677.9	<0.98	<0.98	<0.98	1.1	2.3	16.8	22.2	19.0
78	Hermosa Point	271.1	<1.15	<1.15	<1.15	<1.15	1.9	15.9	29.0	7.1
79	Hermosa Point Natives	434.8	1.9	<1.61	<1.61	<1.61	2.4	16.5	24.7	19.6

Site Order	Site Name	$\sum_{42}$PAHs	HCB	Dieldrin	Mirex	$\sum_{8}$Chlordanes	$\sum_{6}$DDTs	$\sum_{40}$PCBs	TPCBs	$\sum_{11}$PBDEs
		Concentrations in ng/g, dry weight								
80	Everett Harbor	411.4	<1.11	0.9	<1.11	0.9	4.7	23.2	36.0	10.8
81	Everett Harbor Natives	427.9	<1.0	<1.0	<1.0	<1.0	2.9	36.2	51.4	25.1
82	Mukilteo WWTP, Big Gulch	194.4	<1.2	<1.2	<1.2	<1.2	2.0	14.6	19.1	9.8
83	Edmonds Ferry	349.1	<1.54	<1.54	<1.54	<1.54	2.2	20.0	33.1	9.7
84	Edmonds Ferry Natives	1414.8	<0.88	<0.88	<0.88	1.4	7.4	57.3	78.4	22.7
85	Johnson Point	98.0	<1.32	<1.32	<1.32	<1.32	1.8	13.1	19.7	3.7
86	Tolmie State Park	94.1	<1.07	<1.07	<1.07	<1.07	1.6	17.1	31.4	4.0
87	Olympia, Budd Inlet, North Point	130.9	<1.66	<1.66	<1.66	<1.66	1.8	16.0	23.9	16.0
88	Birch Point	90.1	<1.66	<1.66	<1.66	<1.72	2.0	4.0	7.7	<1.72
89	Cherry Point Aquatic Reserve, Birch Bay South	99.4	<1.72	<1.72	<1.72	<1.72	2.1	4.3	8.5	1.8
90	Cherry Point Aquatic Reserve, 1 Alcoa-BP	198.4	<1.36	<1.3	<1.36	<1.36	2.0	19.4	34.6	3.9
91	Cherry Point Aquatic Reserve, 3 Alcoa-BP	283.9	<1.35	<1.35	<1.35	<1.35	1.8	16.8	29.9	2.3
92	Cherry Point Aquatic Reserve, 4 Conoco Phillips	348.0	<1.4	<1.33	<1.4	<1.4	2.0	8.7	19.8	2.1
93	West Bellingham Bay, Lummi Nation	234.3	<1.62	<1.62	<1.62	<1.62	2.3	9.9	23.9	2.5
94	Bellingham Bay, Little Squalicum Creek	198.6	<1.51	<1.51	<1.51	<1.51	2.6	11.0	25.6	3.9
95	Bellingham Bay, Squalicum Harbor	429.5	<1.79	<1.79	<1.79	<1.79	2.8	7.1	14.1	2.7
96	Bellingham Bay, Post Point	360.9	<1.56	<1.49	<1.56	<1.56	3.1	6.8	13.5	5.5

$\sum_x$ = summed value, number of analytes in sum indicated by x

PAHs = polycyclic aromatic hydrocarbons (summation of low and high molecular weight PAHs)

HCB = hexachlorobenzene

DDTs = dichlorodiphenyltrichloroethanes

TPCBs = estimated total polychlorinated biphenyls

PBDEs = polybrominated diphenyl ethers

< X.XX = the limit of quantitation (LOQ) for that analyte, i.e. analyte was not detected above the LOQ in that sample

WWTP = wastewater treatment plant

11 APPENDIX D: Wet Weight Organic Tissue Chemistry Data

Organic contaminants that were not detected above the limit of quantitation (LOQ) at any of the sample sites (i.e. aldrin, endosulfan 1, hexachlorocyclohexane) were not included in this table. Site order is arranged alphabetically by county and then from north to south within each county.

		Concentrations in ng/g, wet weight							
Site Order	Site Name	$\sum_{42}$PAHs	HCB	Dieldrin	Mirex	$\sum_{8}$Chlordanes	$\sum_{6}$DDTs	TPCBs	$\sum_{11}$PBDEs
1	Protection Island Aquatic Reserve, Thompson Spit	38.33	< 0.17	< 0.17	< 0.17	< 0.17	0.27	1.2	2.67
2	Deception Pass State Park, Cornet Bay	17.3	<0.15	<0.15	<0.15	<0.15	0.27	1.6	2.5
3	Ala Spit County Park	12.85	<0.14	<0.13	<0.14	<0.14	0.31	2.1	2.4
4	Oak Harbor, Crescent Harbor	46.72	<0.16	<0.16	<0.16	<0.16	0.3	1.7	2.1
5	Coupeville Wharf, Toby's Tavern	26.4	<0.15	0.15	<0.15	<0.15	0.34	1.8	2.8
6	PENN COVE, BASELINE	10.98	< 0.3	< 0.29	< 0.3	< 0.3	0.18	1.54	3.00
7	Triangle Cove	52.37	< 0.23	< 0.23	< 0.23	< 0.23	0.25	0.58	2.25
8	Cavalero Beach Co. Park	59.34	< 0.24	< 0.24	< 0.24	< 0.24	< 0.25	< 0.25	0.95
9	Cavalero Beach Natives	66.21	<0.15	<0.15	<0.15	<0.15	0.24	1.1	1.54
10	Holmes Harbor, Rocky Point, Baby Island	33.8	< 0.3	< 0.3	< 0.3	< 0.3	0.38	0.83	3.49
11	Maxwelton, Dave Mackie County Park	16.53	<0.16	<0.15	<0.16	<0.16	0.26	2.3	3.2
12	Richmond Highlands Beach	61.58	< 0.2	< 0.2	< 0.2	< 0.2	0.88	3.2	4.6
13	Carkeek Park	49	< 0.17	< 0.17	< 0.17	< 0.17	0.98	6.0	8.69
14	Salmon Bay	194.64	< 0.16	0.33	< 0.16	1.5	3.2	11	14.76
15	Discovery Park, West Point	118.78	< 0.24	< 0.24	< 0.24	< 0.24	0.38	3	4.6
16	Elliott Bay, Four-Mile Rock	656.65	< 0.2	0.22	< 0.2	0.25	1.8	6.5	9.6
17	Smith Cove	394.6	< 0.14	< 0.14	< 0.14	0.32	1.6	12	15.79
18	Elliott Bay, Myrtle Edwards	745.63	< 0.23	< 0.23	< 0.24	< 0.24	2.1	11	14
19	Elliott Bay, Seattle Aquarium, Pier 59	478.53	< 0.35	< 0.34	< 0.35	< 0.35	0.96	7.1	10
20	Elliott Bay, Harbor Island, Pier 17	357.7	< 0.28	< 0.28	< 0.28	< 0.28	0.36	9.1	13
21	Elliott Bay, Alki-Duwamish Head	57.16	< 0.24	< 0.24	< 0.24	< 0.24	0.28	2	3
22	Lincoln Park	99.02	< 0.15	< 0.15	< 0.15	< 0.15	0.31	4.0	6.3

Site Order	Site Name	$\sum_{42}$PAHs	HCB	Dieldrin	Mirex	$\sum_{8}$Chlordanes	$\sum_{6}$DDTs	TPCBs	$\sum_{11}$PBDEs
		Concentrations in ng/g, wet weight							
23	Vashon Ferry, N. End Boat Ramp	70.2	< 0.27	< 0.27	< 0.27	< 0.27	0.36	2.3	3.4
24	Seahurst County Park	111.99	< 0.22	< 0.22	< 0.22	< 0.22	0.34	1.9	2.9
25	Quartermaster Harbor	137.83	< 0.24	< 0.24	< 0.24	0.27	0.62	3.5	5
26	Des Moines Marina City Beach Park	78.9	< 0.22	< 0.22	< 0.22	0.24	0.85	3.2	4.7
27	Maury Island Aquatic Reserve, Old Marine Park	36.9	< 0.18	< 0.18	< 0.18	< 0.18	0.29	3.8	6.02
28	Dumas Bay	68.34	< 0.28	< 0.28	< 0.28	< 0.28	0.41	2.4	3.5
29	Point No Point	231.49	< 0.23	< 0.23	< 0.23	0.34	0.28	1	2.1
30	Port Gamble, Point Julia	26.36	< 0.29	< 0.29	< 0.29	< 0.29	< 0.29	1.6	2.56
31	Port Gamble, West	31.18	< 0.23	< 0.23	< 0.23	< 0.23	0.28	1.7	3.06
32	Point Jefferson	31	<0.14	<0.14	<0.15	<0.15	0.27	1.9	2.9
33	Suquamish, Stormwater Outfall	55.59	< 0.23	< 0.23	< 0.23	< 0.23	0.42	3.9	5.29
34	Liberty Bay, Poulsbo	50.1	<0.16	0.2	<0.16	0.19	0.5	5.6	7.9
35	Liberty Bay, Keyport	55.54	< 0.26	< 0.26	< 0.26	< 0.26	0.86	6.4	8.87
36	Point Bolin	32.8	<0.15	0.16	<0.15	<0.15	0.47	5.2	7.1
37	Port Madison, Hidden Cove	72.72	< 0.23	< 0.23	< 0.23	< 0.23	0.37	5	7.6
38	Silverdale, Dyes Inlet	58.3	< 0.24	< 0.24	< 0.24	< 0.24	0.46	5.2	7.37
39	West Bainbridge, Westwood	33.83	< 0.16	< 0.16	< 0.16	< 0.16	0.37	8.3	12.56
40	Eagle Harbor, Bainbridge Ferry Terminal	339.33	< 0.21	< 0.21	< 0.21	< 0.21	0.45	6.8	9.9
41	Illahee Creek	46.56	< 0.2	< 0.2	< 0.2	< 0.2	0.5	5.6	7.86
42	Sinclair Inlet, Waterman Point	25.66	< 0.22	< 0.22	< 0.22	1.6	0.31	4.3	7.19
43	Hood Canal, Holly	10.34	< 0.17	< 0.17	< 0.17	< 0.17	0.21	3.6	6.09
44	Bremerton Shipyard, Ferry Terminal	72.6	<0.15	<0.15	<0.15	<0.15	0.53	11.0	14
45	Bremerton Shipyard, Charleston Beach	92.89	< 0.22	< 0.22	< 0.22	< 0.22	0.55	14.3	19.85
46	Manchester, Stormwater Outfall	44.6	0.25	< 0.18	< 0.18	< 0.18	0.4	3.4	4.56
47	Sinclair Inlet, Sinclair Marina	137	<0.1	0.21	0.24	0.3	0.88	17.0	22
48	Colvos Passage, Olalla, Prospect Point Beach	17.12	< 0.22	< 0.22	< 0.22	< 0.22	0.28	2.7	3.9
49	Case Inlet, Allyn	18.4	< 0.24	< 0.24	< 0.24	< 0.24	0.34	2.3	3.6
50	Shelton, Oak Bay Marina	43.76	< 0.25	< 0.25	< 0.25	< 0.25	0.27	1.5	3
51	Totten Inlet	11.97	<0.1	<0.099	<0.1	<0.1	0.26	4.6	4.6

Site Order	Site Name	$\sum_{42}$PAHs	HCB	Dieldrin	Mirex	$\sum_{8}$Chlordanes	$\sum_{6}$DDTs	TPCBs	$\sum_{11}$PBDEs
		Concentrations in ng/g, wet weight							
52	Gig Harbor, Narrows Passage	38.44	< 0.17	< 0.17	< 0.17	< 0.17	0.28	6.2	9.68
53	Point Defiance Park	36.38	< 0.23	< 0.23	< 0.23	< 0.23	0.33	2.6	4.1
54	Kopachuck State Park	36.77	< 0.32	< 0.31	< 0.32	< 0.32	< 0.32	0.86	1.7
55	Commencement Bay, Skookum Wuldge	111.78	<0.15	0.18	<0.15	0.82	1.9	7	8.32
56	Tacoma, Ruston Way, Puget Creek	73.7	< 0.27	< 0.27	< 0.27	< 0.27	0.31	2.4	3.5
57	Tacoma Ruston Waterfront	87.51	< 0.27	< 0.27	< 0.27	0.29	0.33	3.23	4.43
58	Fox Island, Tanglewood Island	13.3	< 0.29	< 0.29	< 0.29	< 0.29	< 0.29	0.8	1.6
59	Hylebos Waterway	233.83	0.23	0.38	< 0.22	0.96	6.79	26.22	31.89
60	Hylebos Waterway Natives	289.82	0.19	0.31	0.61	2	11	58.67	75.16
61	Thea Foss Waterway	186.64	< 0.22	< 0.22	< 0.22	0.33	0.7	3.9	5.7
62	Steilacoom, Sunnyside Beach Park	22.41	< 0.21	< 0.21	< 0.21	< 0.21	0.29	2	2.9
63	Nisqually Reach Aq Reserve, Anderson Island, Sandy Bay	6.8	< 0.14	< 0.14	< 0.14	< 0.14	0.27	3.4	5.32
64	North Shore, Orcas Island	16.05	< 0.19	< 0.19	< 0.19	< 0.19	0.26	0.75	2.67
65	Friday Harbor Labs, San Juan Island	84.55	< 0.21	< 0.21	< 0.21	< 0.21	0.27	0.59	2.26
66	Fisherman's Bay, Weeks Wetland, Lopez Island	71.45	< 0.23	< 0.23	< 0.23	< 0.23	< 0.23	0.28	1.57
67	Larrabee State Park	15.49	< 0.17	< 0.17	< 0.17	< 0.17	0.71	1.9	3.1
68	Cypress Island Aquatic Reserve, Strawberry Bay	4.5	< 0.35	< 0.35	< 0.35	< 0.35	< 0.35	1.3	3.56
69	Cypress Island Aquatic Reserve, Secret Harbor	12.05	< 0.18	< 0.18	< 0.18	< 0.18	0.24	2.1	4.27
70	Anacortes, Guemes Ferry	195.7	< 0.29	< 0.29	< 0.29	< 0.29	0.38	0.78	1.6
71	March Point	23.5	< 0.27	< 0.27	< 0.27	< 0.27	0.37	0.59	1.2
72	Padilla Bay	7	< 0.28	< 0.28	< 0.28	< 0.28	0.83	0.29	0.57
73	Fidalgo Bay Aquatic Reserve, Weaverling Spit	26.53	< 0.23	< 0.23	< 0.23	< 0.23	< 0.23	0.53	2.25
74	Skagit River Delta	4.7	<0.11	0.13	<0.11	<0.11	0.24	1.7	2.4
75	Port Susan, Warm Beach	47.22	< 0.23	< 0.23	< 0.23	< 0.23	0.35	1.3	3.05
76	Kayak Point	60	< 0.21	< 0.21	< 0.21	< 0.21	0.27	1.9	4.24
77	Kayak Point Natives	84.75	<0.13	<0.13	<0.13	0.14	0.29	2.1	2.78
78	Hermosa Point	37.84	< 0.16	< 0.16	< 0.16	< 0.16	0.26	2.2	4.05
79	Hermosa Point Natives	38.44	0.17	<0.16	<0.16	<0.16	0.21	1.46	2.18

Site Order	Site Name	$\sum_{42}$PAHs	HCB	Dieldrin	Mirex	$\sum_{8}$Chlordanes	$\sum_{6}$DDTs	TPCBs	$\sum_{11}$PBDEs
		Concentrations in ng/g, wet weight							
80	Everett Harbor	60.76	< 0.13	0.14	< 0.13	0.13	0.7	3.4	5.31
81	Everett Harbor Natives	49.64	<0.15	<0.15	<0.15	<0.15	0.34	4.2	5.96
82	Mukilteo WWTP, Big Gulch	26.7	<0.18	<0.18	<0.18	<0.18	0.27	2.0	2.6
83	Edmonds Ferry	53.86	< 0.16	< 0.16	< 0.16	< 0.16	0.34	3.1	5.11
84	Edmonds Ferry Natives	149.02	<0.14	<0.14	<0.14	0.15	0.78	6.04	8.26
85	Johnson Point	14.9	< 0.2	< 0.2	< 0.2	< 0.2	0.28	2	3
86	Tolmie State Park	13.8	< 0.16	< 0.16	< 0.16	< 0.16	0.23	2.5	4.61
87	Olympia, Budd Inlet, North Point	19.69	< 0.24	< 0.24	< 0.24	< 0.24	0.27	2.4	3.6
88	Birch Point	14.13	< 0.26	< 0.26	< 0.26	< 0.27	0.31	0.62	1.2
89	Cherry Point Aquatic Reserve, Birch Bay South	15.2	< 0.26	< 0.26	< 0.26	< 0.26	0.32	0.66	1.3
90	Cherry Point Aquatic Reserve, 1 Alcoa-BP	29.82	< 0.21	< 0.2	< 0.21	< 0.21	0.3	2.9	5.2
91	Cherry Point Aquatic Reserve, 3 Alcoa-BP	42.82	< 0.21	< 0.21	< 0.21	< 0.21	0.27	2.5	4.51
92	Cherry Point Aquatic Reserve, 4 Conoco Phillips	56.71	< 0.21	< 0.2	< 0.21	< 0.21	0.33	1.4	3.23
93	West Bellingham Bay, Lummi Nation	36.28	< 0.25	< 0.25	< 0.25	< 0.25	0.35	1.5	3.7
94	Bellingham Bay, Little Squalicum Creek	31.7	< 0.24	< 0.24	< 0.24	< 0.24	0.42	1.8	4.09
95	Bellingham Bay, Squalicum Harbor	60.92	< 0.26	< 0.26	< 0.26	< 0.26	0.4	1	2
96	Bellingham Bay, Post Point	53.4	< 0.24	< 0.23	< 0.24	< 0.24	0.46	1	2

$\sum_x$ = summed value, number of analytes in sum indicated by x

PAHs = polycyclic aromatic hydrocarbons (summation of low and high molecular weight PAHs)

HCB = hexachlorobenzene

DDTs = dichlorodiphenyltrichloroethanes

TPCBs = estimated total polychlorinated biphenyls

PBDEs = polybrominated diphenyl ethers

< X.XX = the limit of quantitation (LOQ) for that analyte, i.e. analyte was not detected above the LOQ in that sample

WWTP = wastewater treatment plant

12 APPENDIX E: Dry Weight Metal Tissue Chemistry Data

Site order is arranged alphabetically by county and then from north to south within each county.

		Concentrations in µg/g, dry weight					
Site Order	Site Name	Arsenic	Cadmium	Copper	Lead	Mercury	Zinc
1	Protection Island Aquatic Reserve, Thompson Spit	6	1.72	4.26	0.2	0.0386	77.8
2	Deception Pass State Park, Cornet Bay	5.76	2.25	4.39	0.18	0.043	82.2
3	Ala Spit County Park	6.05	2.05	5.87	0.23	0.0445	70.3
4	Oak Harbor, Crescent Harbor	6.05	1.94	6.19	0.23	0.0461	90.1
5	Coupeville Wharf, Toby's Tavern	6.31	1.79	5.47	0.33	0.0492	81.8
6	PENN COVE, BASELINE	5.28	2.04	4.98	0.13	0.03	74.80
7	Triangle Cove	5.65	2.33	6.66	0.22	0.0435	78.2
8	Cavalero Beach Co. Park	5.33	2.02	6.14	0.26	0.039	91.8
9	Cavalero Beach Natives	7.1	5.37	12.4	1.24	0.248	106
10	Holmes Harbor, Rocky Point, Baby Island	6.6	1.73	5.99	0.16	0.0436	80.2
11	Maxwelton, Dave Mackie County Park	5.7	2.19	4.33	0.18	0.0424	74.5
12	Richmond Highlands Beach	6.37	1.99	5.62	0.26	0.0482	107
13	Carkeek Park	5.27	1.65	5.51	0.25	0.0361	70.5
14	Salmon Bay	5.93	1.97	10.5	0.56	0.0438	101
15	Discovery Park, West Point	4.96	2.12	5.49	0.21	0.041	67.9
16	Elliott Bay, Four-Mile Rock	4.94	1.82	6.05	0.32	0.0428	88.7
17	Smith Cove	5.5	1.98	7.69	0.49	0.0337	96.8
18	Elliott Bay, Myrtle Edwards	5.45	2.12	5.72	0.55	0.049	89.7
19	Elliott Bay, Seattle Aquarium, Pier 59	5.28	1.97	5.29	0.41	0.0379	80.1
20	Elliott Bay, Harbor Island, Pier 17	5.43	1.88	7.48	0.31	0.0354	92.3
21	Elliott Bay, Alki-Duwamish Head	5.11	1.76	4.75	0.22	0.0359	72.3
22	Lincoln Park	5.59	1.95	4.87	0.22	0.0347	80
23	Vashon Ferry, N. End Boat Ramp	5.11	2.21	5.99	0.19	0.0363	90.1
24	Seahurst County Park	5.82	2.03	4.21	0.26	0.0445	103

Site Order	Site Name	Arsenic	Cadmium	Copper	Lead	Mercury	Zinc
		Concentrations in µg/g, dry weight					
25	Quartermaster Harbor	6.9	1.73	6.25	0.77	0.0625	76.2
26	Des Moines Marina City Beach Park	5.92	1.97	6.19	0.3	0.0423	92.2
27	Maury Island Aquatic Reserve, Old Marine Park	6.27	1.97	5.04	0.23	0.0413	74.8
28	Dumas Bay	6.41	1.88	5.88	0.27	0.0515	84
29	Point No Point	5.66	1.92	6.4	0.19	0.0448	76.8
30	Port Gamble, Point Julia	5.82	2.47	5.84	0.23	0.0493	88.5
31	Port Gamble, West	6.34	2.22	5.64	0.29	0.0532	77.6
32	Point Jefferson	5.35	1.97	4.05	0.17	0.0382	75.8
33	Suquamish, Stormwater Outfall	6.24	2	5.57	0.25	0.0461	74.5
34	Liberty Bay, Poulsbo	6.3	1.82	6.8	0.47	0.0441	113
35	Liberty Bay, Keyport	6	1.83	6.54	0.41	0.0428	85.3
36	Point Bolin	6.71	1.59	5.15	0.3	0.0411	94.9
37	Port Madison, Hidden Cove	6.8	2.44	8.37	0.58	0.0358	110
38	Silverdale, Dyes Inlet	8.02	2.88	9.64	0.51	0.0636	137
39	West Bainbridge, Westwood	6.35	2.45	5.83	0.41	0.0501	93.5
40	Eagle Harbor, Bainbridge Ferry Terminal	5.97	2.04	6.67	0.38	0.0483	99.4
41	Illahee Creek	5.62	1.85	5.38	0.26	0.0408	80
42	Sinclair Inlet, Waterman Point	5.9	1.97	5.7	0.4	0.0421	96.6
43	Hood Canal, Holly	6.52	2.12	6.42	0.13	0.0468	80.7
44	Bremerton Shipyard, Ferry Terminal	5.42	1.81	6.39	0.54	0.0473	99.3
45	Bremerton Shipyard, Charleston Beach	6.18	2.09	8.72	0.95	0.0549	107
46	Manchester, Stormwater Outfall	5.79	1.86	4.78	0.26	0.0455	99.3
47	Sinclair Inlet, Sinclair Marina	5.78	2.03	6.43	0.53	0.0422	94.1
48	Colvos Passage, Olalla, Prospect Point Beach	5.18	1.65	4.09	0.19	0.0395	74.1
49	Case Inlet, Allyn	6.95	1.74	6.42	0.18	0.0495	81.2
50	Shelton, Oak Bay Marina	5.14	2.03	5.32	0.21	0.0499	95.7
51	Totten Inlet	6.6	2.05	5.56	0.21	0.0459	83.2
52	Gig Harbor, Narrows Passage	7.1	2.57	7.89	0.37	0.0535	100
53	Point Defiance Park	6.07	2.1	4.75	0.28	0.0402	87.2

Site Order	Site Name	Arsenic	Cadmium	Copper	Lead	Mercury	Zinc
		Concentrations in µg/g, dry weight					
54	Kopachuck State Park	6.04	2.46	5.26	0.21	0.0451	84.1
55	Commencement Bay, Skookum Wuldge	5.71	2.3	6.22	0.39	0.0349	91.7
56	Tacoma, Ruston Way, Puget Creek	5.64	2.12	5.22	0.44	0.0426	78.4
57	Tacoma Ruston Waterfront	5.39	2.36	6.29	0.49	0.03	79.81
58	Fox Island, Tanglewood Island	5.04	2.25	5.69	0.19	0.0394	78.6
59	Hylebos Waterway	6.30	2.00	7.49	0.29	0.03	108.71
60	Hylebos Waterway Natives	6.91	1.97	13.9	0.75	0.0511	269
61	Thea Foss Waterway	4.83	2.11	6.45	0.53	0.0286	78.1
62	Steilacoom, Sunnyside Beach Park	5.03	2.23	4.56	0.24	0.0374	75.3
63	Nisqually Reach Aquatic Reserve, Anderson Island, Sandy Bay	6.81	2.3	5.93	0.25	0.0496	87
64	North Shore, Orcas Island	5.57	1.77	5.01	0.18	0.0366	80
65	Friday Harbor Labs, San Juan Island	5.5	2.14	4.57	0.22	0.041	75.8
66	Fisherman's Bay, Weeks Wetland, Lopez Island	5.64	1.95	7.07	0.23	0.0362	73.3
67	Larrabee State Park	5.83	2.28	5.76	0.2	0.041	98.1
68	Cypress Island Aquatic Reserve, Strawberry Bay	5.59	1.87	4.56	0.2	0.0358	77.8
69	Cypress Island Aquatic Reserve, Secret Harbor	5.54	1.72	4.48	0.17	0.0343	68
70	Anacortes, Guemes Ferry	5.72	2.02	5.5	0.26	0.0423	86.7
71	March Point	5.44	1.98	5.43	0.23	0.0363	92.7
72	Padilla Bay	5.2	2.57	5.16	0.2	0.041	77.6
73	Fidalgo Bay Aquatic Reserve, Weaverling Spit	6.04	1.77	5.26	0.25	0.0416	81
74	Skagit River Delta	5.11	2.27	5.58	0.18	0.034	80.7
75	Port Susan, Warm Beach	5.13	1.85	5.18	0.2	0.0449	69.4
76	Kayak Point	5.83	1.9	5.3	0.22	0.049	68.6
77	Kayak Point Natives	6.05	2.64	10	0.41	0.0735	69.7
78	Hermosa Point	5.82	2.13	6.54	0.26	0.0455	97.1
79	Hermosa Point Natives	7.43	4.04	8.16	0.86	0.0686	104
80	Everett Harbor	6.42	2.76	7.04	0.45	0.0546	91.5
81	Everett Harbor Natives	5.79	2.07	9.73	0.26	0.0461	89.3
82	Mukilteo WWTP, Big Gulch	5.22	2.57	6.23	0.22	0.0392	91.3

Site Order	Site Name	Arsenic	Cadmium	Copper	Lead	Mercury	Zinc
		Concentrations in µg/g, dry weight					
83	Edmonds Ferry	6.24	4.07	6.61	1.38	0.109	91.6
84	Edmonds Ferry Natives	5.93	1.99	4.92	0.26	0.038	107
85	Johnson Point	6.17	2.68	5.63	0.19	0.0401	88.1
86	Tolmie State Park	5.61	1.77	5.58	0.17	0.0424	77.3
87	Olympia, Budd Inlet, North Point	6.32	2.23	6.03	0.2	0.037	85.5
88	Birch Point	5.86	2.14	5.62	0.17	0.0357	93.6
89	Cherry Point Aquatic Reserve, Birch Bay South	6.36	2.13	4.98	0.21	0.0407	81.5
90	Cherry Point Aquatic Reserve, 1 Alcoa-BP	5.31	1.94	5.01	0.17	0.0369	80.5
91	Cherry Point Aquatic Reserve, 3 Alcoa-BP	5.49	1.65	5.06	0.19	0.0376	79.5
92	Cherry Point Aquatic Reserve, 4 Conoco Phillips	5.76	2.18	5.18	0.17	0.0381	74
93	West Bellingham Bay, Lummi Nation	5.33	2.32	5.78	0.19	0.0343	77.9
94	Bellingham Bay, Little Squalicum Creek	5.61	1.7	6.35	0.25	0.0357	95
95	Bellingham Bay, Squalicum Harbor	6.21	1.98	6.16	0.28	0.0423	93.8
96	Bellingham Bay, Post Point	5.97	2.04	6.03	0.25	0.044	91.6

WWTP = wastewater treatment plant

13 APPENDIX F: Wet Weight Metal Tissue Chemistry Data

Site order is arranged alphabetically by county and then from north to south within each county.

		Concentrations in µg/g, wet weight					
Site Order	Site Name	Arsenic	Cadmium	Copper	Lead	Mercury	Zinc
1	Protection Island Aquatic Reserve, Thompson Spit	0.948	0.272	0.673	0.031	0.0061	12.3
2	Deception Pass State Park, Cornet Bay	0.841	0.329	0.641	0.026	0.00628	12
3	Ala Spit County Park	0.895	0.303	0.869	0.033	0.00658	10.4
4	Oak Harbor, Crescent Harbor	0.913	0.293	0.934	0.034	0.00696	13.6
5	Coupeville Wharf, Toby's Tavern	0.972	0.276	0.842	0.051	0.00757	12.6
6	PENN COVE, BASELINE	0.85	0.33	0.80	0.02	0.01	12.07
7	Triangle Cove	0.752	0.31	0.886	0.03	0.00579	10.4
8	Cavalero Beach Co. Park	0.783	0.297	0.903	0.038	0.00573	13.5
9	Cavalero Beach Natives	0.583	0.441	1.02	0.102	0.0204	8.73
10	Holmes Harbor, Rocky Point, Baby Island	1.07	0.281	0.971	0.025	0.00706	13
11	Maxwelton, Dave Mackie County Park	0.872	0.335	0.663	0.028	0.00648	11.4
12	Richmond Highlands Beach	0.873	0.273	0.77	0.036	0.0066	14.6
13	Carkeek Park	0.822	0.258	0.859	0.039	0.00563	11
14	Salmon Bay	0.818	0.272	1.45	0.077	0.00604	14
15	Discovery Park, West Point	0.773	0.331	0.856	0.033	0.0064	10.6
16	Elliott Bay, Four-Mile Rock	0.741	0.273	0.908	0.048	0.00642	13.3
17	Smith Cove	0.858	0.309	1.2	0.077	0.00525	15.1
18	Elliott Bay, Myrtle Edwards	0.845	0.328	0.886	0.085	0.00759	13.9
19	Elliott Bay, Seattle Aquarium, Pier 59	0.798	0.298	0.799	0.062	0.00572	12.1
20	Elliott Bay, Harbor Island, Pier 17	0.841	0.292	1.16	0.047	0.00548	14.3
21	Elliott Bay, Alki-Duwamish Head	0.792	0.273	0.736	0.034	0.00556	11.2
22	Lincoln Park	0.894	0.312	0.779	0.035	0.00555	12.8
23	Vashon Ferry, N. End Boat Ramp	0.777	0.336	0.91	0.029	0.00551	13.7
24	Seahurst County Park	0.849	0.296	0.614	0.037	0.00649	15.1

Site Order	Site Name	Arsenic	Cadmium	Copper	Lead	Mercury	Zinc
		Concentrations in µg/g, wet weight					
25	Quartermaster Harbor	1.16	0.291	1.05	0.13	0.0105	12.8
26	Des Moines Marina City Beach Park	0.912	0.304	0.953	0.046	0.00652	14.2
27	Maury Island Aquatic Reserve, Old Marine Park	0.921	0.289	0.741	0.034	0.00607	11
28	Dumas Bay	1	0.293	0.918	0.042	0.00803	13.1
29	Point No Point	0.877	0.297	0.992	0.03	0.00694	11.9
30	Port Gamble, Point Julia	0.861	0.365	0.864	0.034	0.0073	13.1
31	Port Gamble, West	0.932	0.326	0.829	0.043	0.00782	11.4
32	Point Jefferson	0.819	0.301	0.619	0.026	0.00584	11.6
33	Suquamish, Stormwater Outfall	1.03	0.33	0.919	0.042	0.0076	12.3
34	Liberty Bay, Poulsbo	0.964	0.278	1.04	0.072	0.00675	17.3
35	Liberty Bay, Keyport	0.936	0.285	1.02	0.064	0.00667	13.3
36	Point Bolin	1.06	0.252	0.813	0.047	0.0065	15
37	Port Madison, Hidden Cove	1	0.358	1.23	0.085	0.00526	16.1
38	Silverdale, Dyes Inlet	0.882	0.317	1.06	0.056	0.007	15.1
39	West Bainbridge, Westwood	0.978	0.377	0.898	0.063	0.00772	14.4
40	Eagle Harbor, Bainbridge Ferry Terminal	0.949	0.324	1.06	0.061	0.00768	15.8
41	Illahee Creek	0.927	0.305	0.888	0.042	0.00673	13.2
42	Sinclair Inlet, Waterman Point	0.879	0.294	0.85	0.06	0.00628	14.4
43	Hood Canal, Holly	0.978	0.318	0.963	0.02	0.00702	12.1
44	Bremerton Shipyard, Ferry Terminal	0.829	0.277	0.978	0.082	0.00723	15.2
45	Bremerton Shipyard, Charleston Beach	0.872	0.295	1.23	0.134	0.00774	15.1
46	Manchester, Stormwater Outfall	0.857	0.275	0.707	0.038	0.00673	14.7
47	Sinclair Inlet, Sinclair Marina	0.879	0.309	0.977	0.081	0.00642	14.3
48	Colvos Passage, Olalla, Prospect Point Beach	0.762	0.243	0.601	0.028	0.0058	10.9
49	Case Inlet, Allyn	1.07	0.268	0.989	0.028	0.00762	12.5
50	Shelton, Oak Bay Marina	0.725	0.286	0.75	0.03	0.00704	13.5
51	Totten Inlet	0.984	0.305	0.828	0.031	0.00684	12.4
52	Gig Harbor, Narrows Passage	0.781	0.283	0.868	0.041	0.00589	11
53	Point Defiance Park	0.947	0.328	0.741	0.043	0.00627	13.6

Site Order	Site Name	Arsenic	Cadmium	Copper	Lead	Mercury	Zinc
		Concentrations in µg/g, wet weight					
54	Kopachuck State Park	0.834	0.339	0.726	0.029	0.00623	11.6
55	Commencement Bay, Skookum Wuldge	0.759	0.306	0.827	0.052	0.00464	12.2
56	Tacoma, Ruston Way, Puget Creek	0.835	0.314	0.773	0.065	0.00631	11.6
57	Tacoma Ruston Waterfront	0.83	0.36	0.96	0.07	0.00	12.20
58	Fox Island, Tanglewood Island	0.706	0.315	0.796	0.027	0.00552	11
59	Hylebos Waterway	0.95	0.30	1.12	0.04	0.01	16.27
60	Hylebos Waterway Natives	0.815	0.233	1.64	0.089	0.00603	31.8
61	Thea Foss Waterway	0.749	0.327	1	0.083	0.00444	12.1
62	Steilacoom, Sunnyside Beach Park	0.734	0.326	0.666	0.035	0.00546	11
63	Nisqually Reach Aq Reserve, Anderson Island, Sandy Bay	0.783	0.264	0.682	0.029	0.0057	10
64	North Shore, Orcas Island	0.919	0.292	0.827	0.029	0.00604	13.2
65	Friday Harbor Labs, San Juan Island	0.842	0.328	0.699	0.034	0.00627	11.6
66	Fisherman's Bay, Weeks Wetland, Lopez Island	0.846	0.293	1.06	0.034	0.00543	11
67	Larrabee State Park	0.91	0.356	0.899	0.031	0.0064	15.3
68	Cypress Island Aquatic Reserve, Strawberry Bay	0.856	0.286	0.698	0.03	0.00547	11.9
69	Cypress Island Aquatic Reserve, Secret Harbor	0.936	0.291	0.757	0.029	0.0058	11.5
70	Anacortes, Guemes Ferry	0.949	0.336	0.913	0.043	0.00703	14.4
71	March Point	0.816	0.297	0.815	0.034	0.00544	13.9
72	Padilla Bay	0.744	0.367	0.738	0.029	0.00587	11.1
73	Fidalgo Bay Aquatic Reserve, Weaverling Spit	0.985	0.288	0.858	0.04	0.00678	13.2
74	Skagit River Delta	0.741	0.329	0.809	0.026	0.00493	11.7
75	Port Susan, Warm Beach	0.805	0.29	0.813	0.031	0.00705	10.9
76	Kayak Point	0.846	0.276	0.769	0.031	0.00711	9.95
77	Kayak Point Natives	0.805	0.351	1.33	0.054	0.00978	9.27
78	Hermosa Point	0.809	0.296	0.909	0.036	0.00633	13.5
79	Hermosa Point Natives	0.739	0.402	0.811	0.085	0.00682	10.3
80	Everett Harbor	0.751	0.323	0.824	0.053	0.00639	10.7
81	Everett Harbor Natives	0.869	0.31	1.46	0.039	0.00691	13.4
82	Mukilteo WWTP, Big Gulch	0.783	0.386	0.934	0.034	0.00588	13.7

Site Order	Site Name	Arsenic	Cadmium	Copper	Lead	Mercury	Zinc
		Concentrations in µg/g, wet weight					
83	Edmonds Ferry	0.649	0.423	0.687	0.143	0.0113	9.53
84	Edmonds Ferry Natives	0.949	0.319	0.787	0.041	0.00608	17.1
85	Johnson Point	0.931	0.404	0.85	0.028	0.00606	13.3
86	Tolmie State Park	0.842	0.265	0.837	0.025	0.00636	11.6
87	Olympia, Budd Inlet, North Point	0.916	0.323	0.875	0.028	0.00536	12.4
88	Birch Point	0.92	0.336	0.883	0.026	0.00561	14.7
89	Cherry Point Aquatic Reserve, Birch Bay South	0.96	0.322	0.752	0.031	0.00614	12.3
90	Cherry Point Aquatic Reserve, 1 Alcoa-BP	0.817	0.299	0.772	0.026	0.00568	12.4
91	Cherry Point Aquatic Reserve, 3 Alcoa-BP	0.856	0.257	0.789	0.03	0.00586	12.4
92	Cherry Point Aquatic Reserve, 4 Conoco Phillips	0.864	0.327	0.777	0.026	0.00572	11.1
93	West Bellingham Bay, Lummi Nation	0.821	0.358	0.89	0.03	0.00528	12
94	Bellingham Bay, Little Squalicum Creek	0.892	0.271	1.01	0.039	0.00568	15.1
95	Bellingham Bay, Squalicum Harbor	0.9	0.287	0.893	0.04	0.00614	13.6
96	Bellingham Bay, Post Point	0.919	0.314	0.928	0.038	0.00678	14.1

WWTP = wastewater treatment plant

14 APPENDIX G: Summary of Laboratory Data Quality Review

The limit of quantitation (LOQ) for most organic contaminants fell within expected ranges (Table 11); LOQs were slightly higher than anticipated (up to 2.3 ng/g wet weight) for some PAH analytes. All metals in all samples were detected above method detection limits (MDL). As mentioned in Section 2.9.1, the summed analytes used in this study are the sum of all *detected* values, with zeroes substituted for non-detected (<LOQ) analytes, within each group. In cases where *all* analytes in a group were not detected the greatest LOQ for all the analytes in the group was used as the summation concentration, and the value was preceded by a "<" (less than) qualifier to indicate it was not detected. In most cases summed totals were dominated by substantial concentrations of a number of individual analytes; substituting zero or a nominal, low value for non-detects would not have substantially altered comparison results for the summed analytes.

Table 11. Limit of quantitation (LOQ) ranges for analytes or analyte groups (see Table 2 for groupings) analyzed in this study. LOQs for groups are the range of values for individual analytes within the group. Original LOQs reported in wet weight; dry weight LOQs calculated using percent moisture measurements for each sample.

	Range of LOQs (ng/g)	
Analyte or Group	**Wet weight**	**Dry weight**
$\sum_{42}$PAHs	0.13 – 2.3	0.79 – 15
total PCBs	0.099 – 0.35	0.68 – 2.3
$\sum_{11}$PBDEs	0.099 – 0.35	0.67 – 2.3
$\sum_{6}$DDTs	0.098 – 0.35	0.67 – 2.3
$\sum_{8}$Chlordanes	0.099 – 0.35	0.68 – 2.3
$\sum_{3}$HCHs	0.098 – 0.35	0.67 – 2.3
Aldrin	0.099 – 0.35	0.68 – 2.3
Dieldrin	0.099 – 0.35	0.69 – 2.2
HCB	0.10 – 0.35	0.68 – 2.3
Mirex	0.10 – 0.35	0.68 – 2.3
Endosulfan 1	0.10 – 0.35	0.68 – 2.3

During data quality review of the 105 samples analyzed and 115 analytes measured (i.e. 12,075 values) a number of values were censored (Table 12) with standard qualifiers as follows:

- "B" indicated values where the analyte was detected in the method blank and the value was less than three times the blank value.
- "A" indicated an estimated value resulting from the analyte response exceeding the response in the highest calibration standard.
- "i" indicated a value that was suspect because of interference; it included peaks with retention times that did not match those of known analytes.

- "J" indicated an analyte that was positively identified, but the quantitation was estimated because the analyte was not present in the calibration standards; the analyte value was quantitated using the response factor of another, closely related compound.

The values qualified with a "B" flag (4.4% of all study values) were treated as non-detects in summations; in other words zeroes were substituted for "B" qualified data. These values included primarily PAH analytes, especially the naphthalenes, and a few PCB congeners. Potential for contamination of the naphthalene-compound data was mentioned as a possibility in the QAPP for this study (Lanksbury et al., 2012), and was thus not unexpected. The remaining data with qualifiers were used "as is" (i.e. not censored or modified) for all summations and analyses in this study.

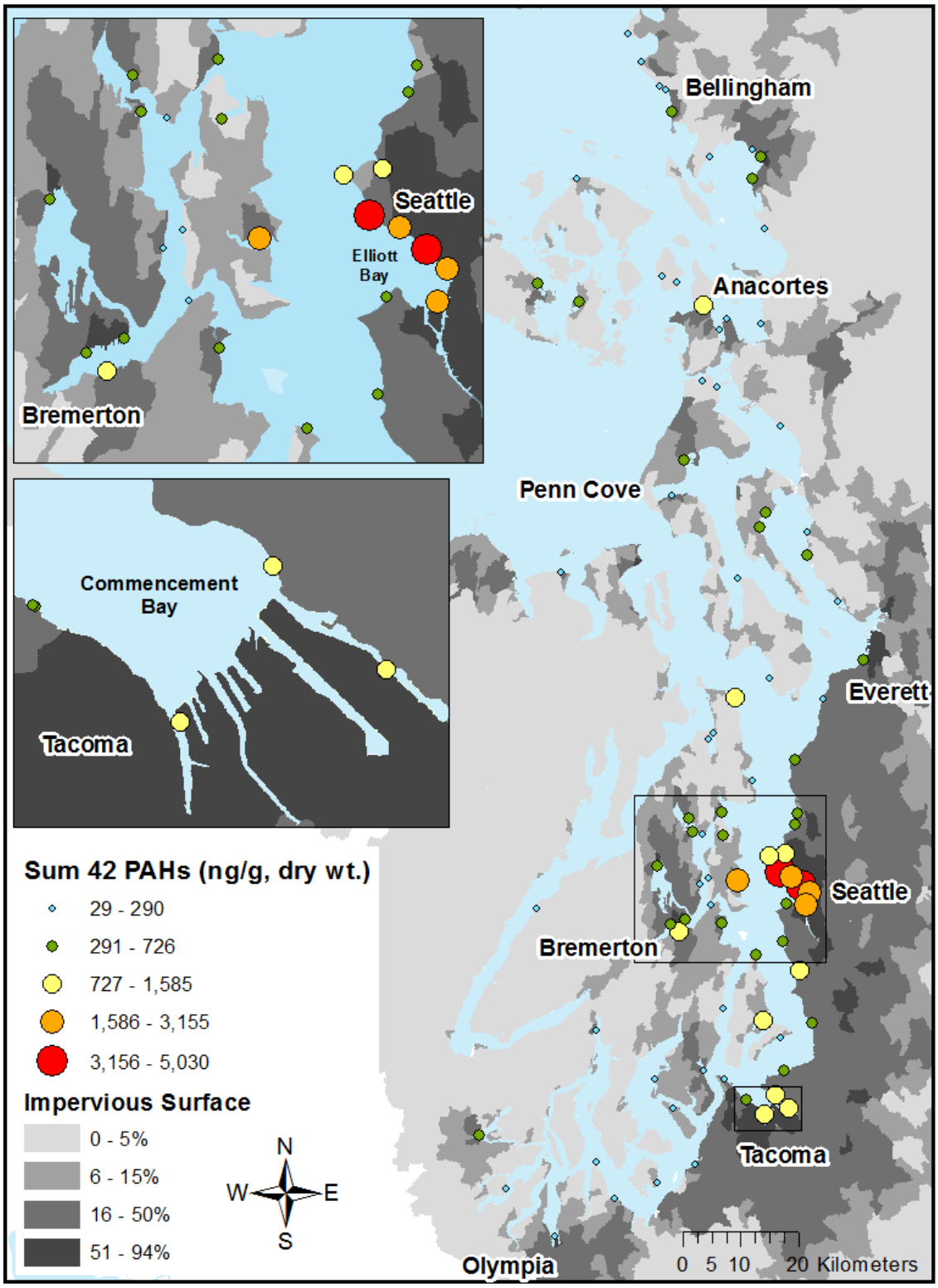
Table 12. Number of analyte values censored with qualifiers in this study. Any <LOQ value or "B" qualified value that also received another qualifier was considered already censored and not included in the counts of "A", "i", and "J" qualified data below.

	Qualifiers			
Analyte/congener name	**"B"**	**"A"**	**"i"**	**"J"**
naphthalene	38			
C1-naphthalenes	18			
C2-naphthalenes	96			21
C3-naphthalenes	40		9	76
C4-naphthalenes	48		32	67
C1-fluorenes				32
C2-fluorenes	11		2	46
C3-fluorenes	5		61	76
C1-dibenzothiophenes				25
C2-dibenzothiophenes				45
C3-dibenzothiophenes	8		24	45
C4-dibenzothiophenes			2	40
phenanthrene		3		
C1-phenanthrenes/anthracenes		2		115
C2-phenanthrenes/anthracenes		1		107
C3-phenanthrenes/anthracenes			2	71
C4-phenanthrenes/anthracenes	20	1	84	85
fluoranthene		5		
pyrene		3		
C1-fluoranthenes/pyrenes		2		92
C2-fluoranthenes/pyrenes				66
C3-fluoranthenes/pyrenes				57
C4-fluoranthenes/pyrenes				43
benz(a)anthracene		3		
C1-benzanthracenes/chrysenes			66	79
C2-benzanthracenes/chrysenes				61
C3-benzanthracenes/chrysenes				35
C4-benzanthracenes/chrysenes				4
chrysene		3		

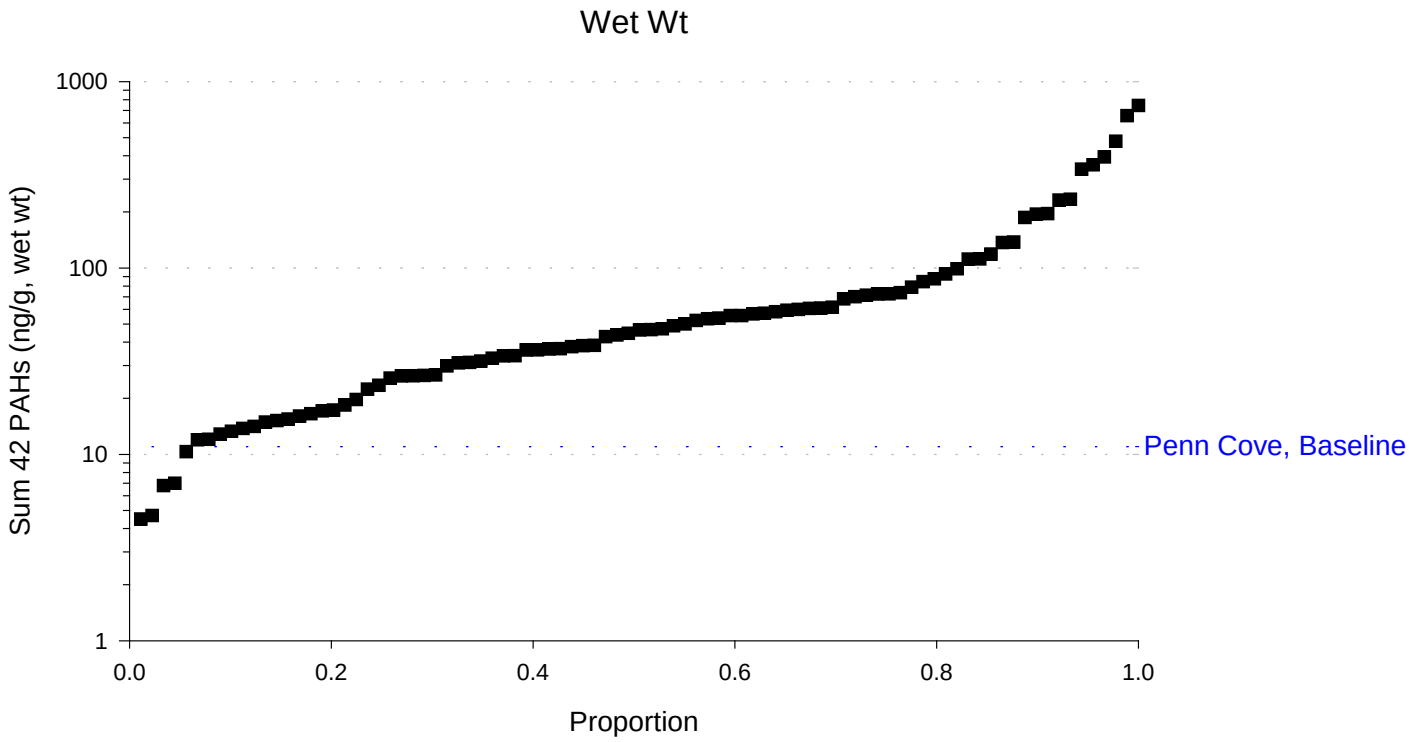
	Qualifiers			
Analyte/congener name	**"B"**	**"A"**	**"i"**	**"J"**
benzo(b)fluoranthene		1		
benzo(j,k)fluoranthene		1		
perylene	5			
PCB017	7			
PCB018	48			
PCB028	55			
PCB031	55			
PCB033	48			
PCB052	8			
PCB101	5			
PCB110	7			
PBDE049	9			
Total	531	25	282	1288

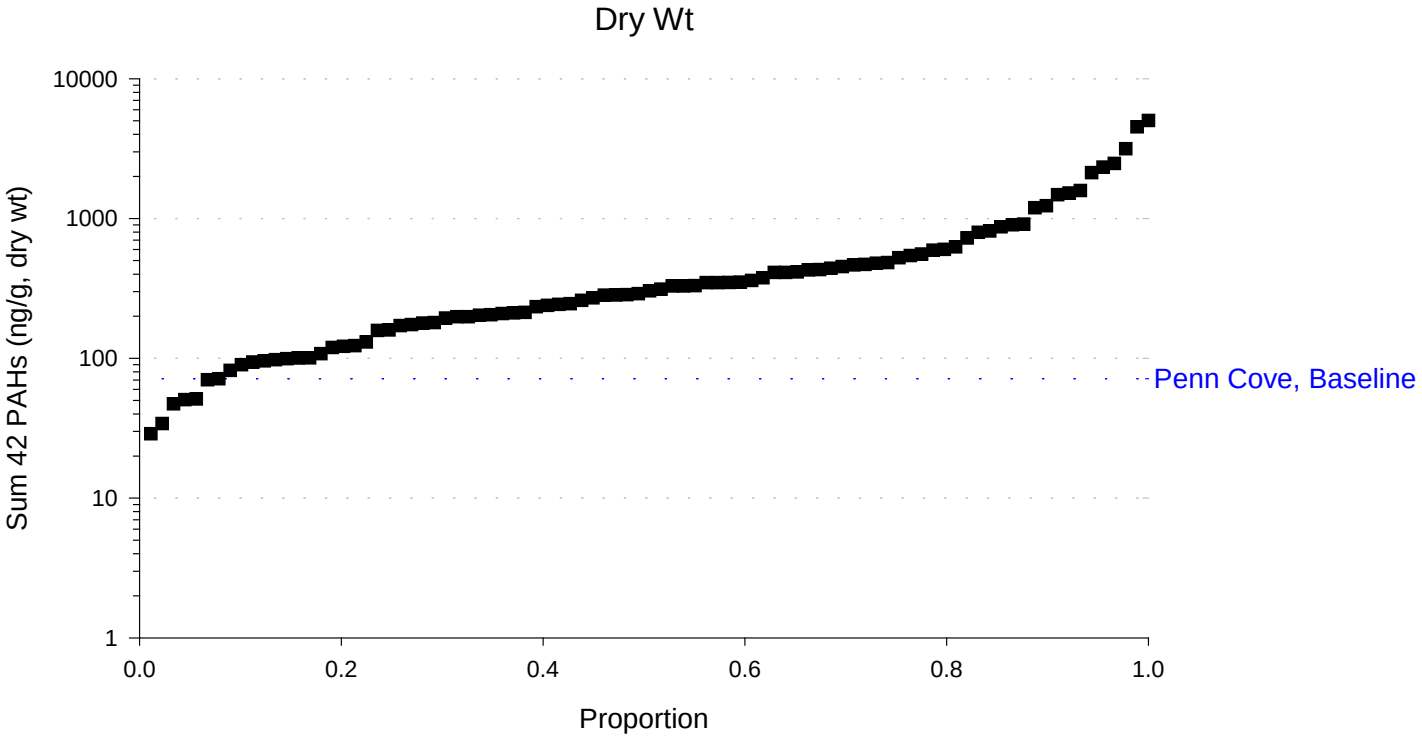
15 APPENDIX H: Details of PAH Findings at Transplanted (i.e. Caged) Mussel Sites

15.1 Map of $\sum_{42}$PAH concentrations



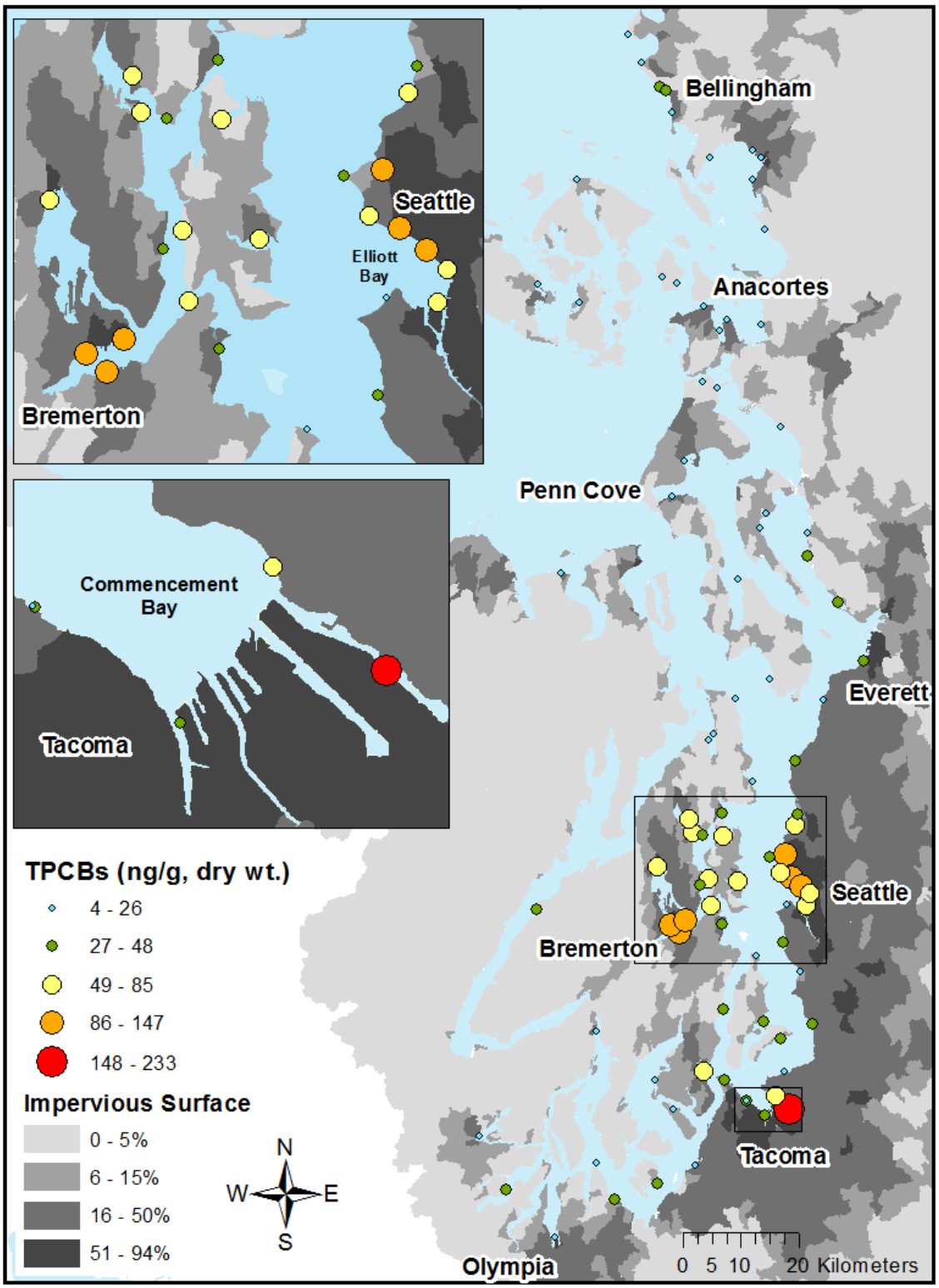
15.2 Cumulative Frequency Distribution of $\sum_{42}$PAH Concentrations



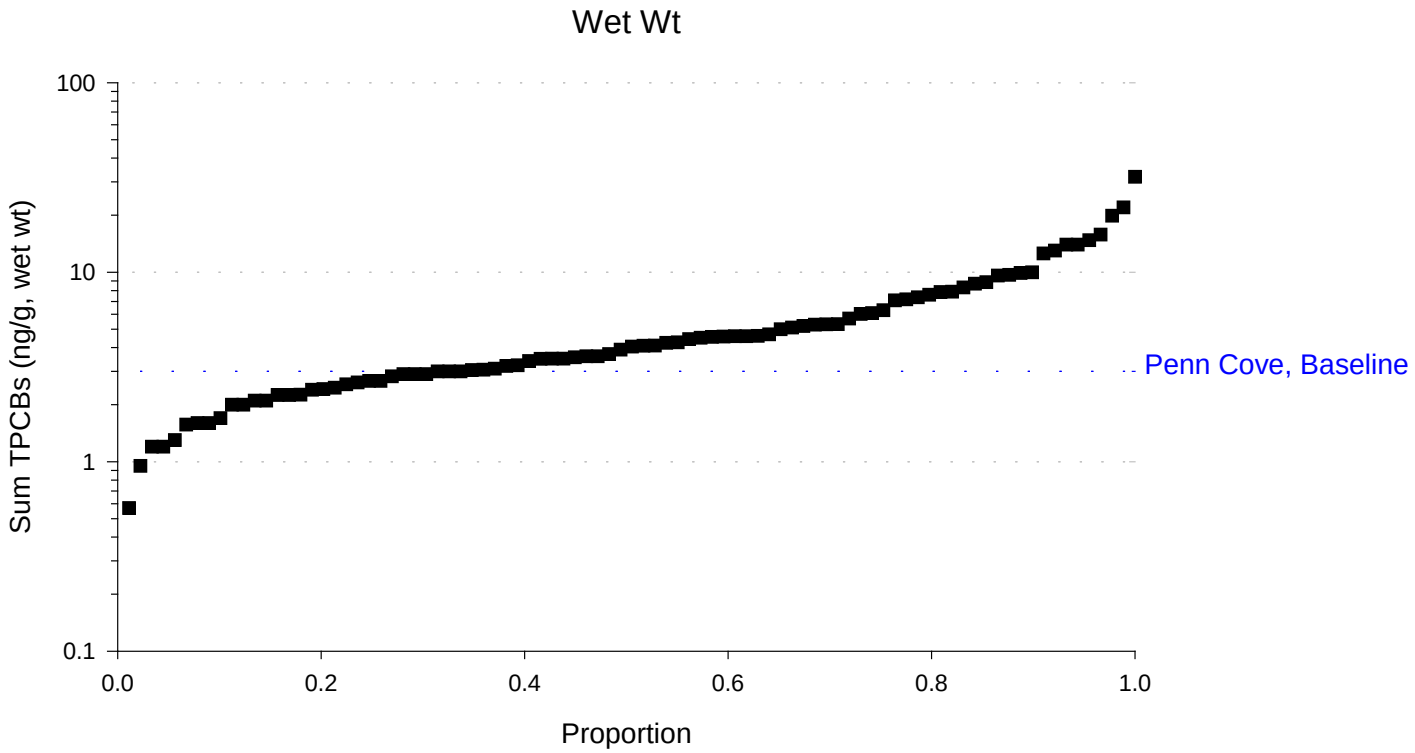


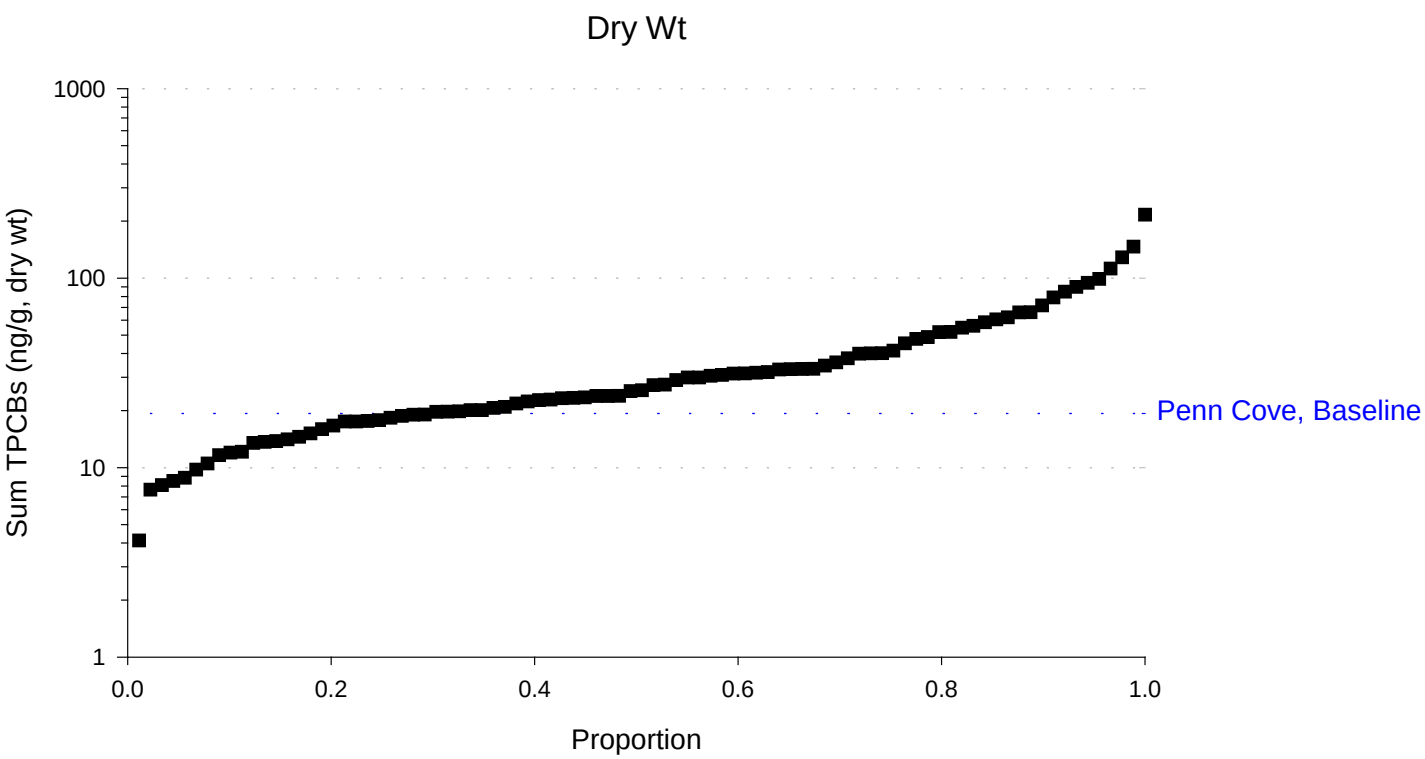
16 APPENDIX I: Details of PCB Findings at Transplanted (i.e. Caged) Mussel Sites

16.1 Map of Estimated Total PCB (TPCB) Concentrations



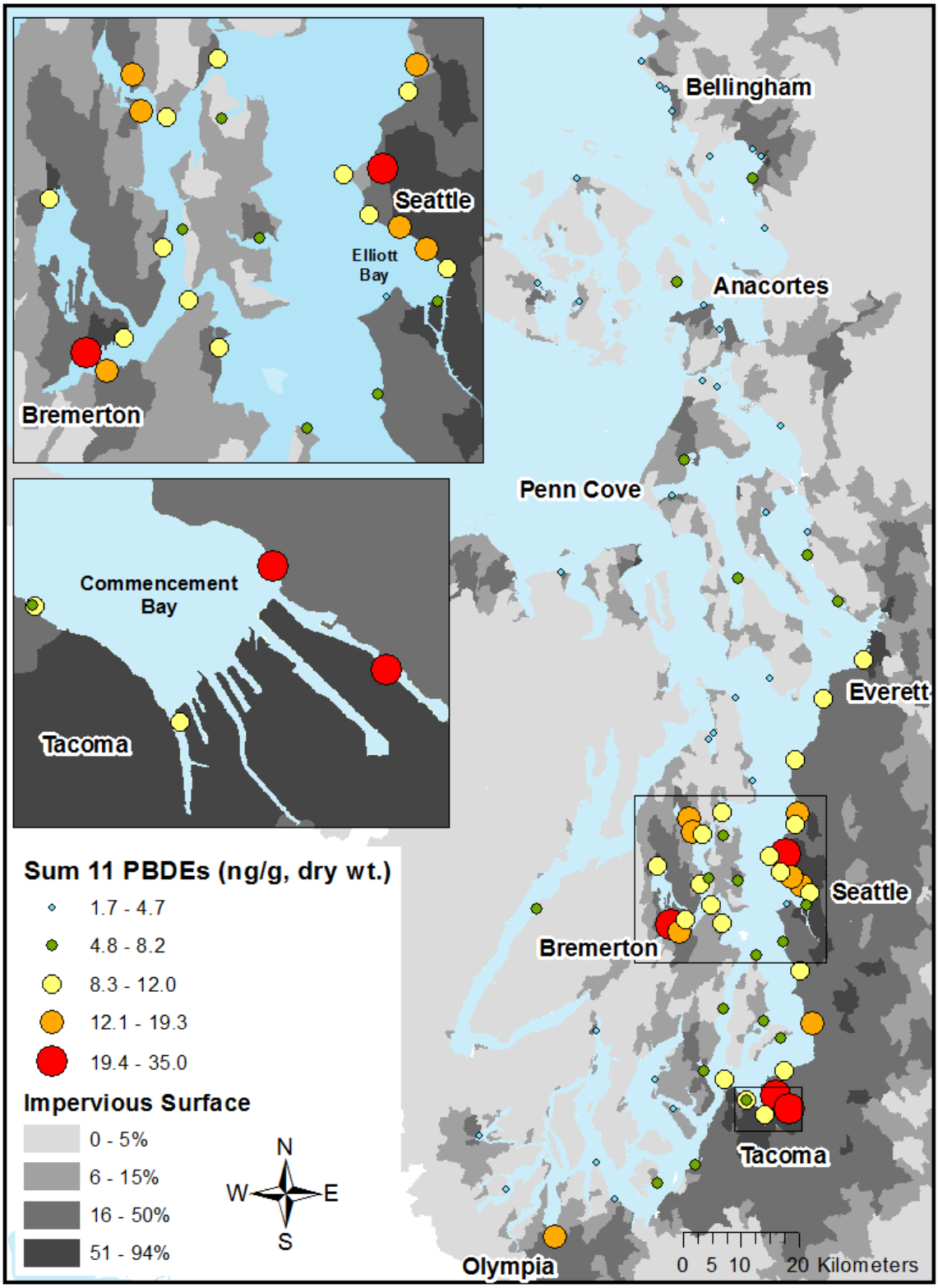
16.2 Cumulative Frequency Distribution of Estimated Total PCB (TPCB) Concentrations



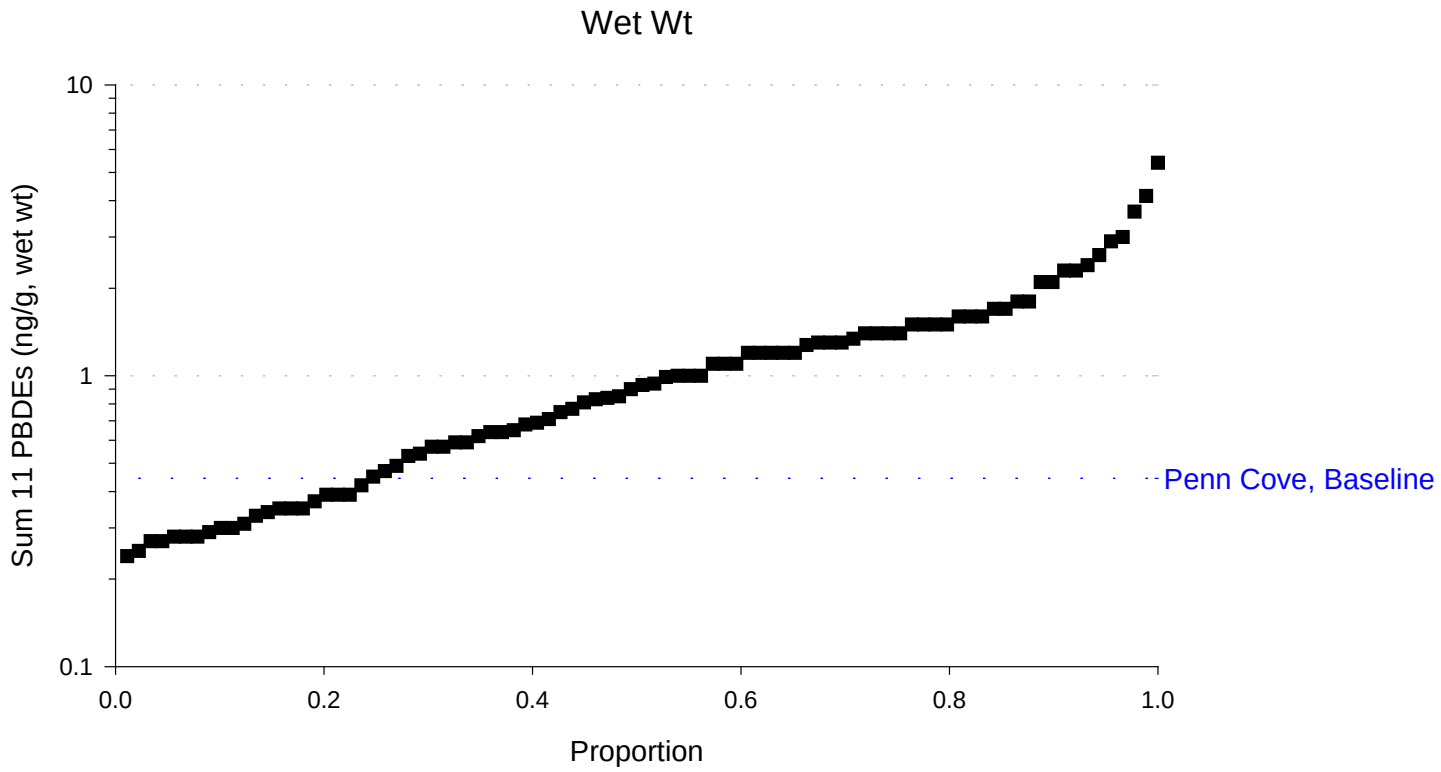


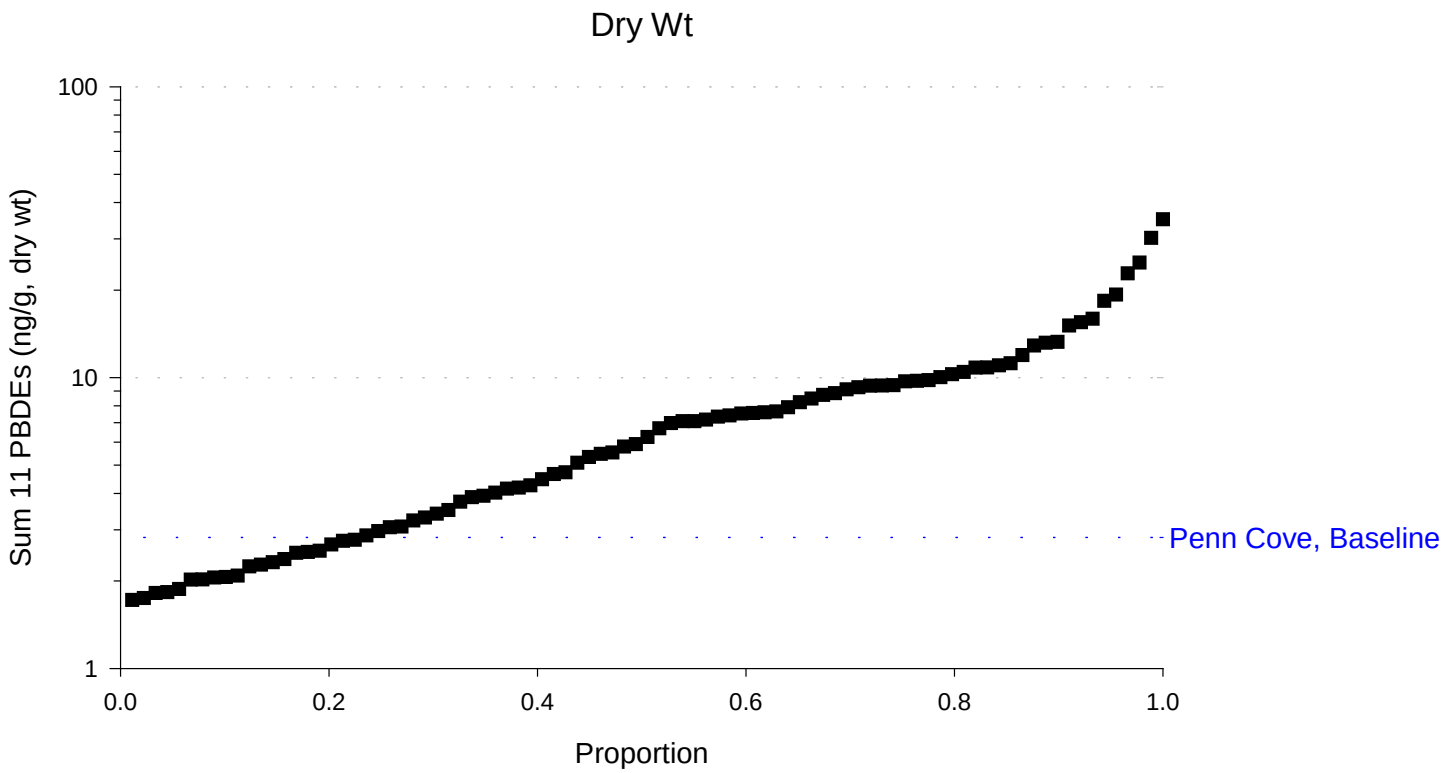
17 APPENDIX J: Details of PBDE Findings at Transplanted (i.e. Caged) Mussel Sites

17.1 Map of $\sum_{11}$PBDE Concentrations



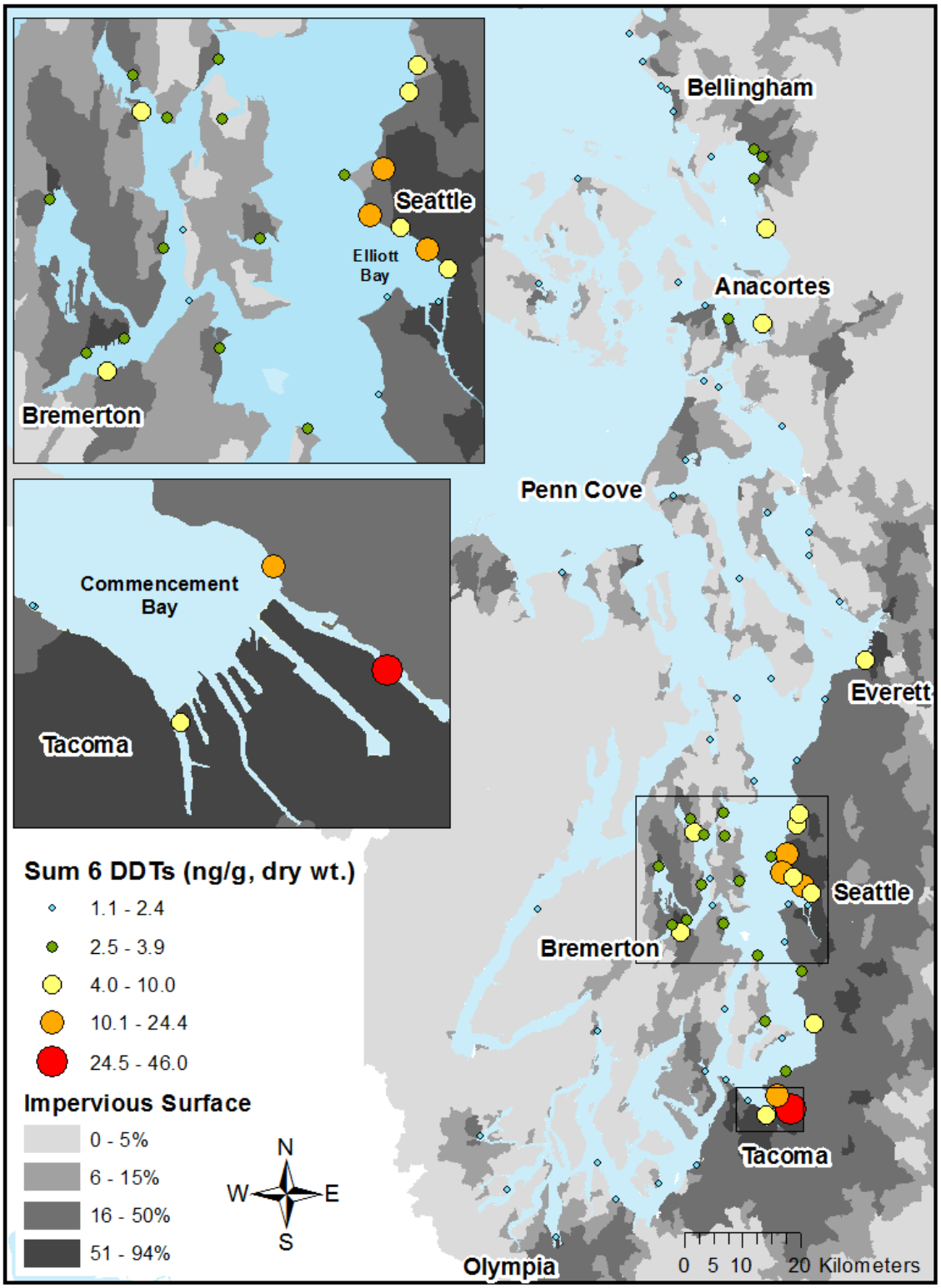
17.2 Cumulative Frequency Distribution of $\sum_{11}$PBDE Concentrations



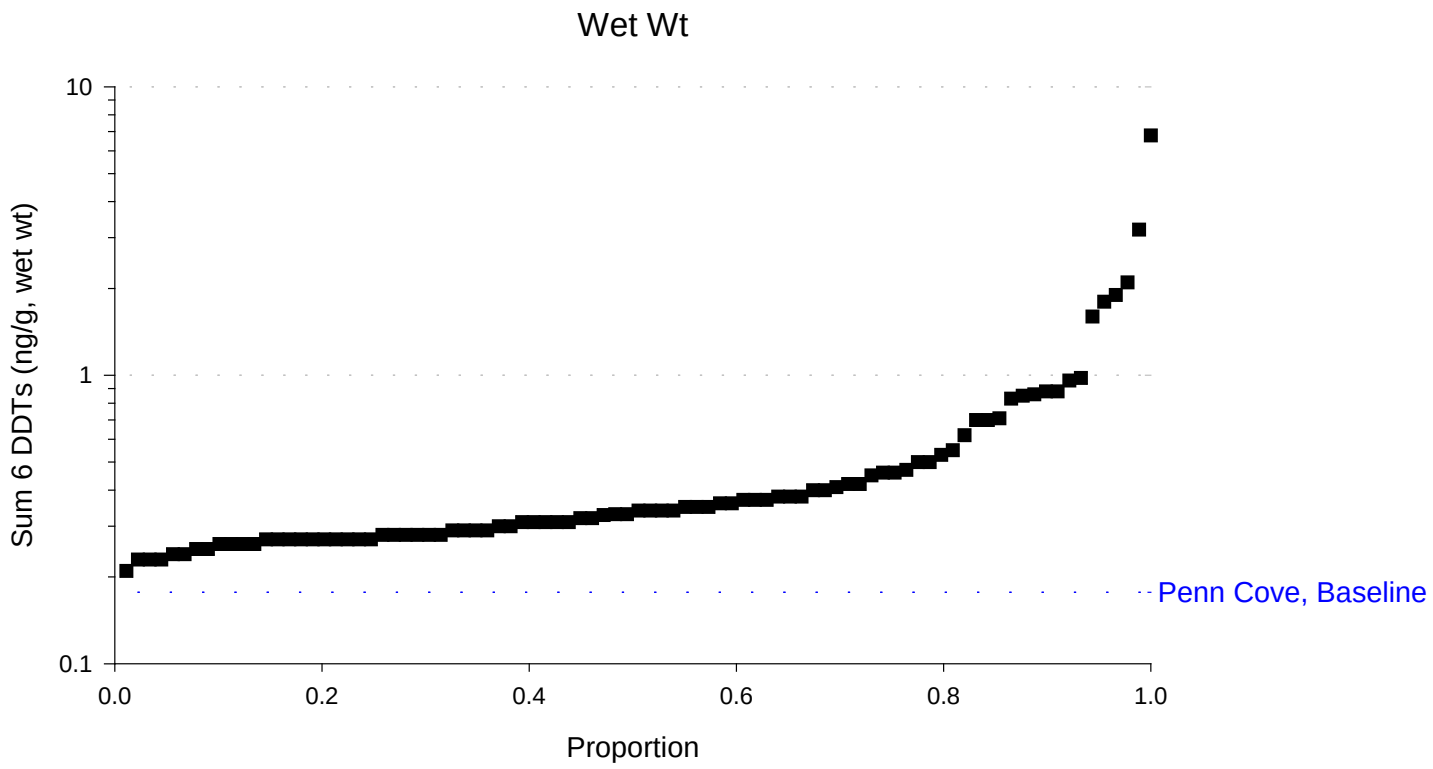


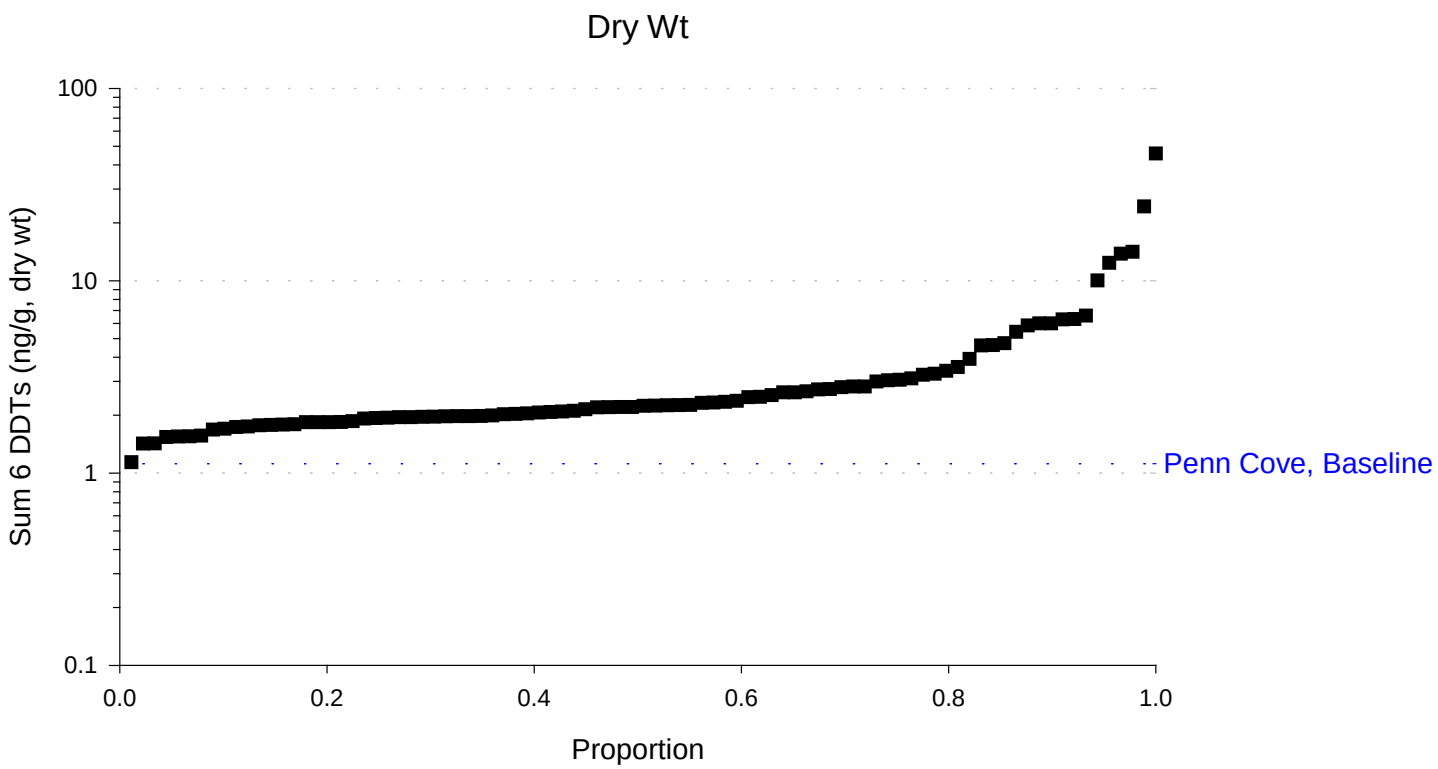
18 APPENDIX K: Detail of DDT Findings at Transplanted (i.e. Caged) Mussel Sites

18.1 Map of $\sum_6$DDT Concentrations



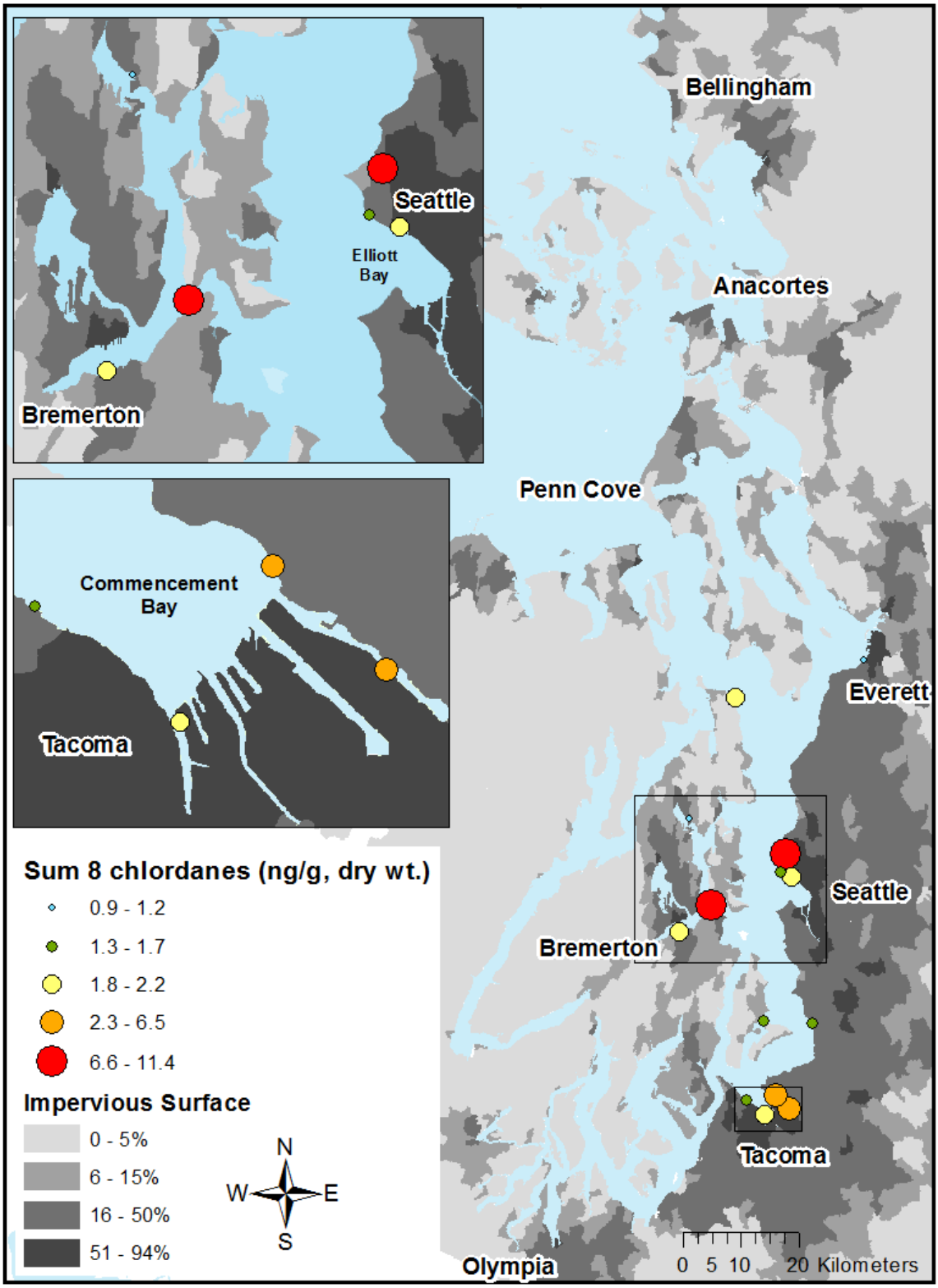
18.2 Cumulative Frequency Distribution of $\sum_6$DDT Concentrations





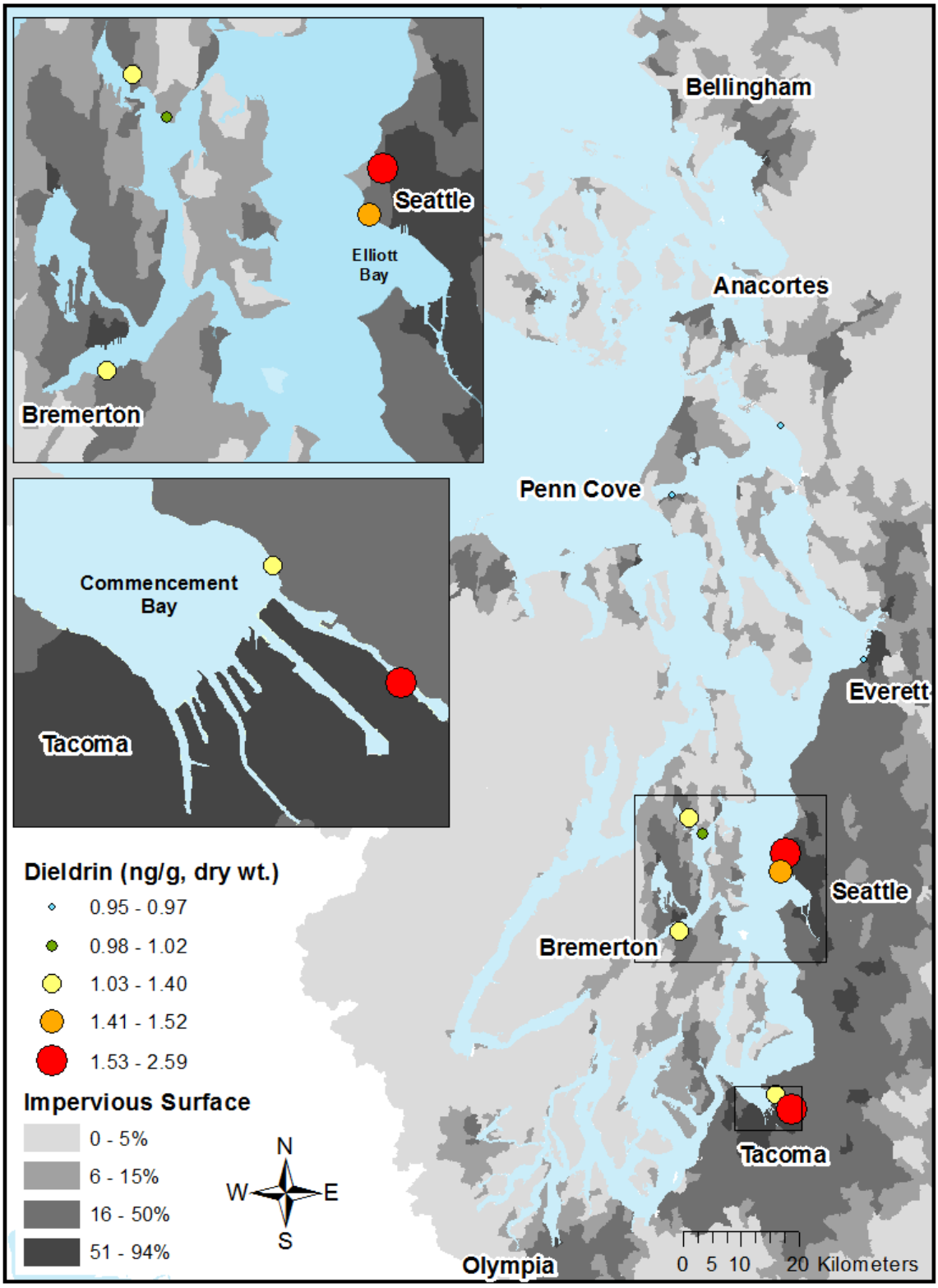
19 APPENDIX L: Chlordane Findings at Transplanted (i.e. Caged) Mussel Sites

19.1 Map of $\sum_8$Chlordane Concentrations



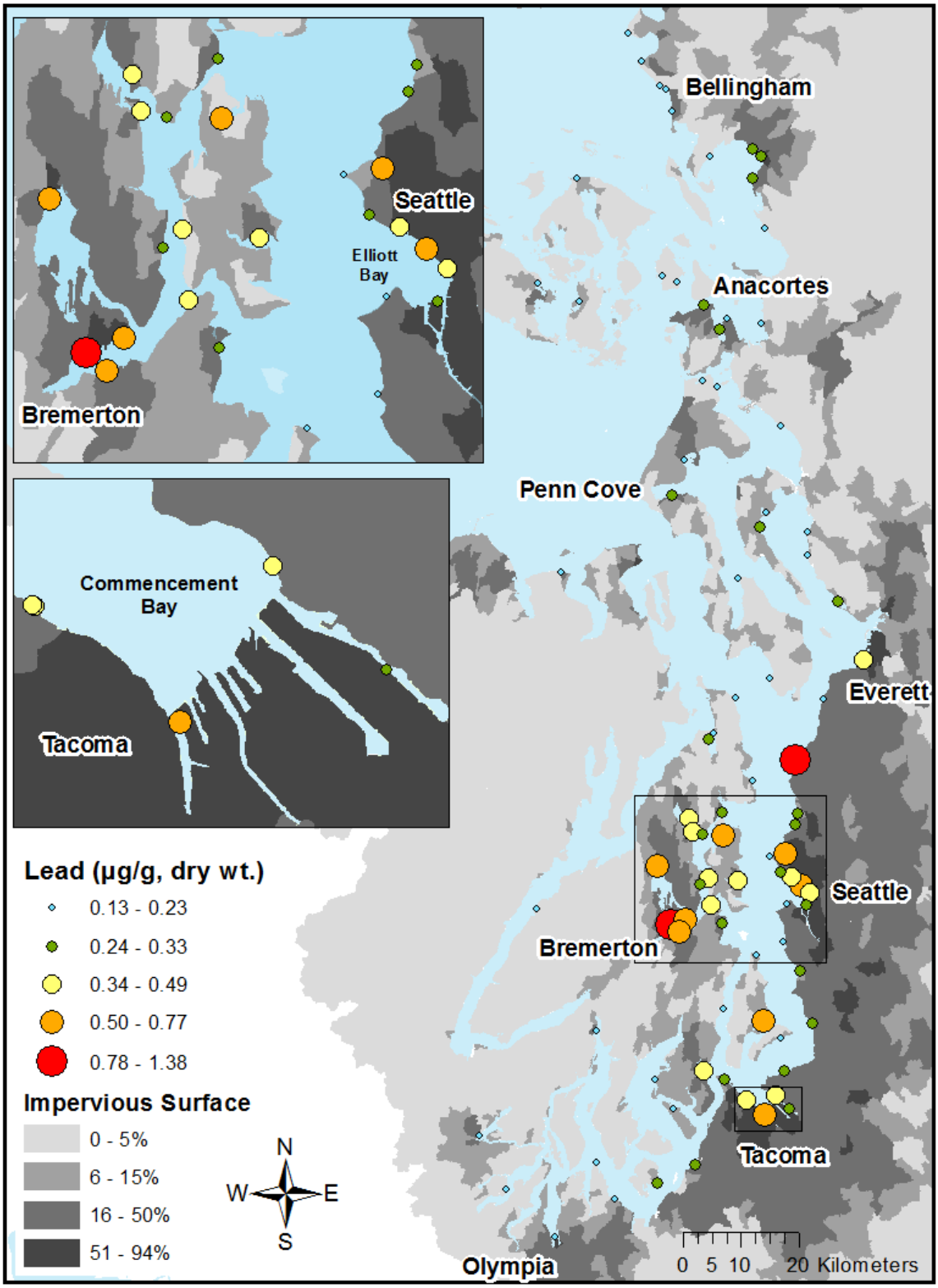
20 APPENDIX M: Dieldrin Findings at Transplanted (i.e. Caged) Mussel Sites

20.1 Map of Dieldrin Concentrations

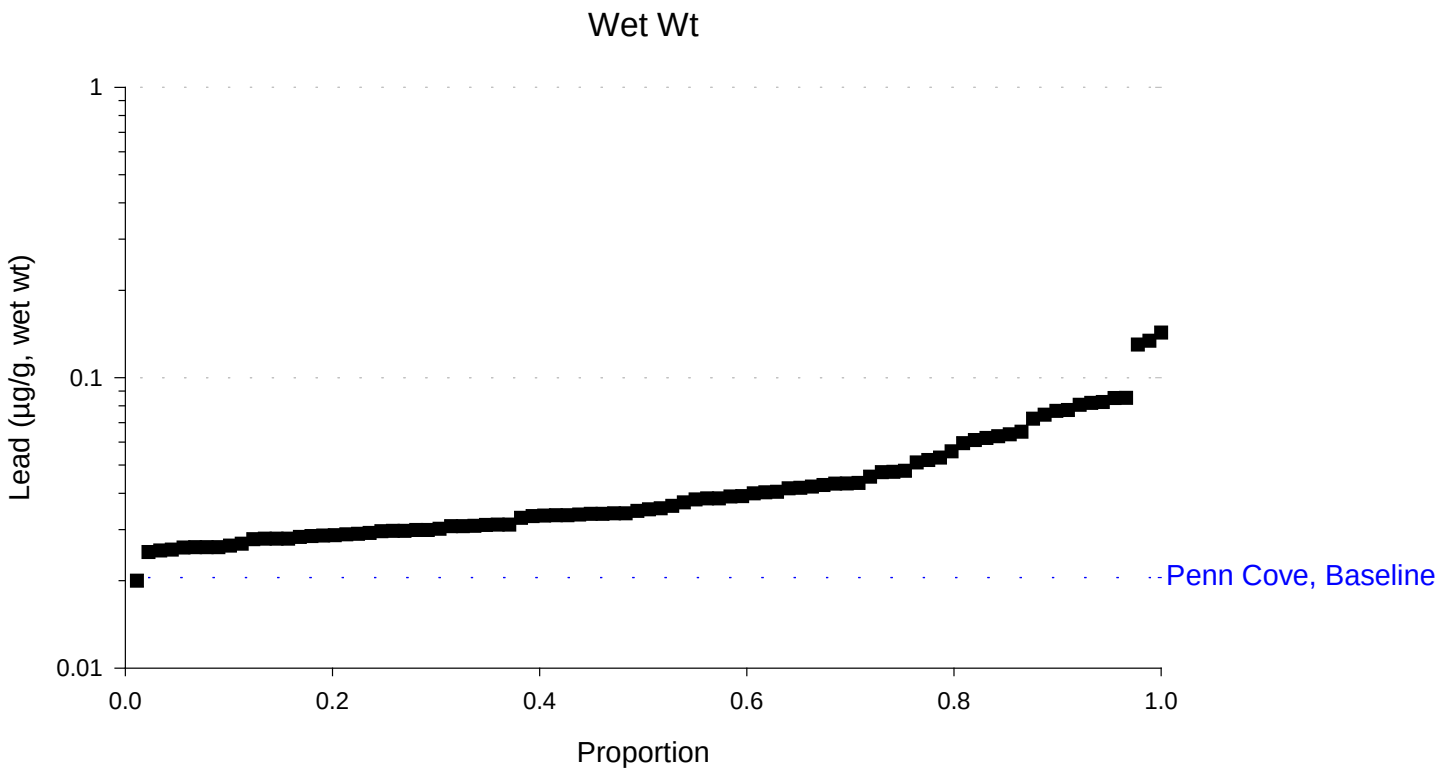


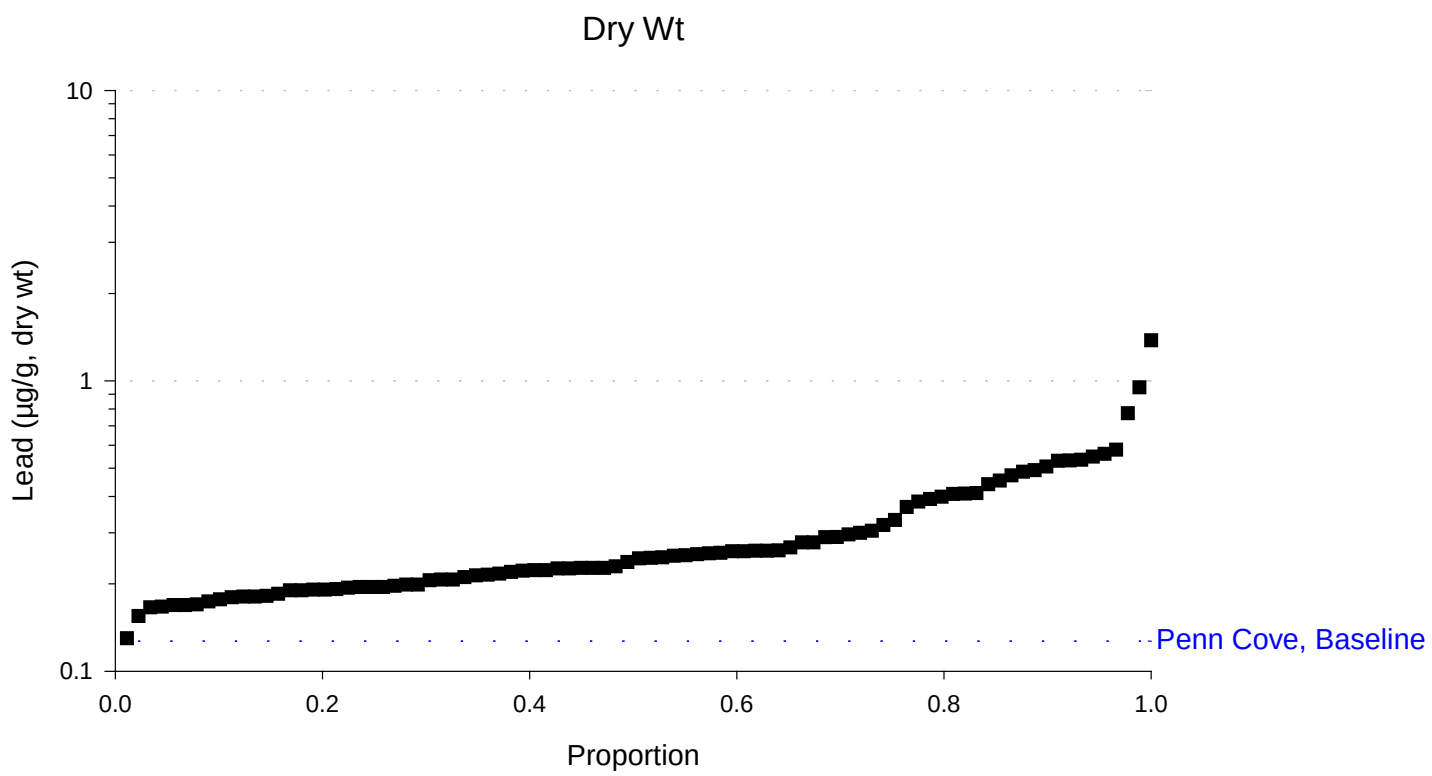
21 APPENDIX N: Details of Lead Findings at Transplanted (i.e. Caged) Mussel Sites

21.1 Map of Lead Concentrations



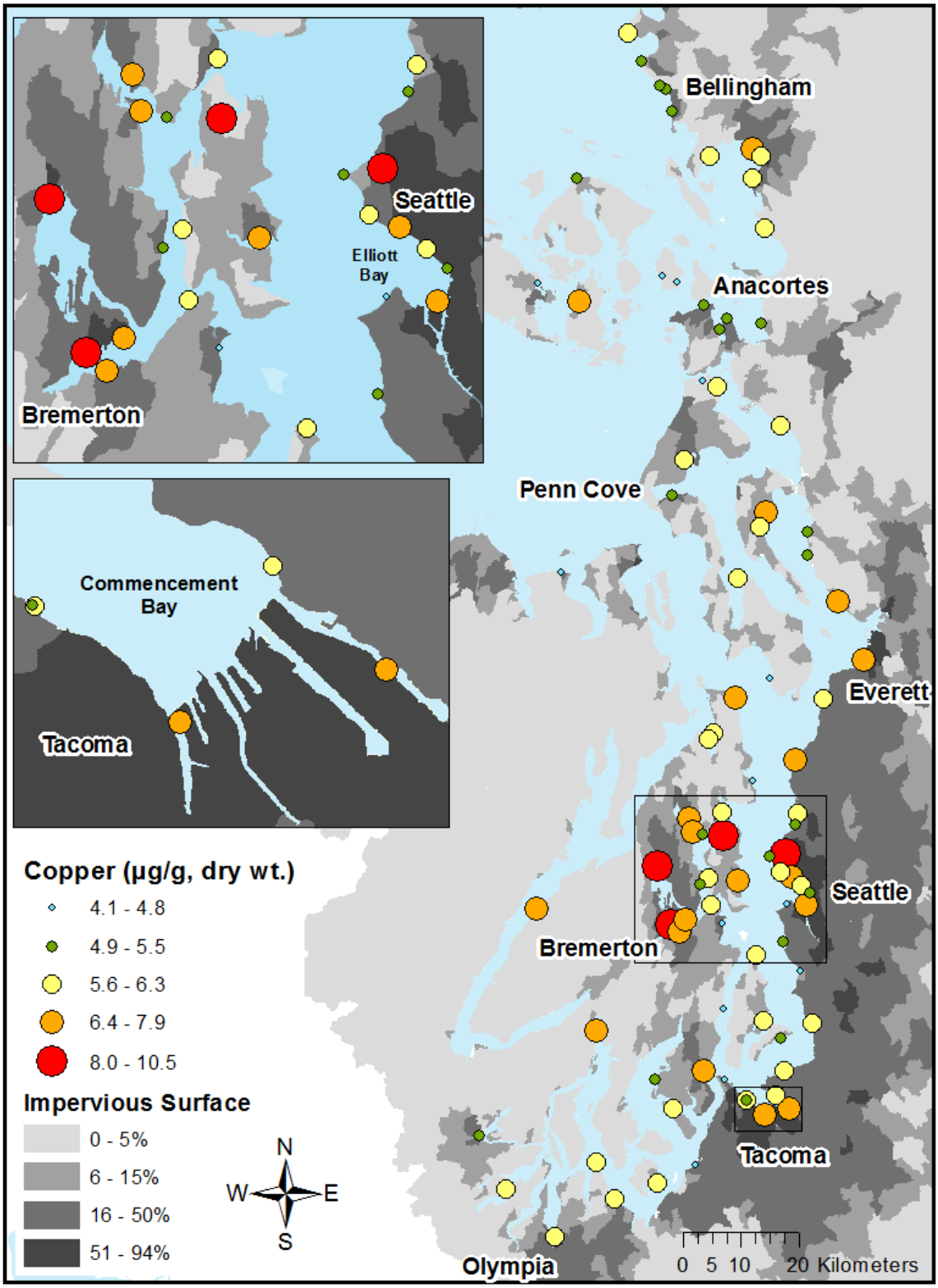
21.2 Cumulative Frequency Distribution of Lead Concentrations



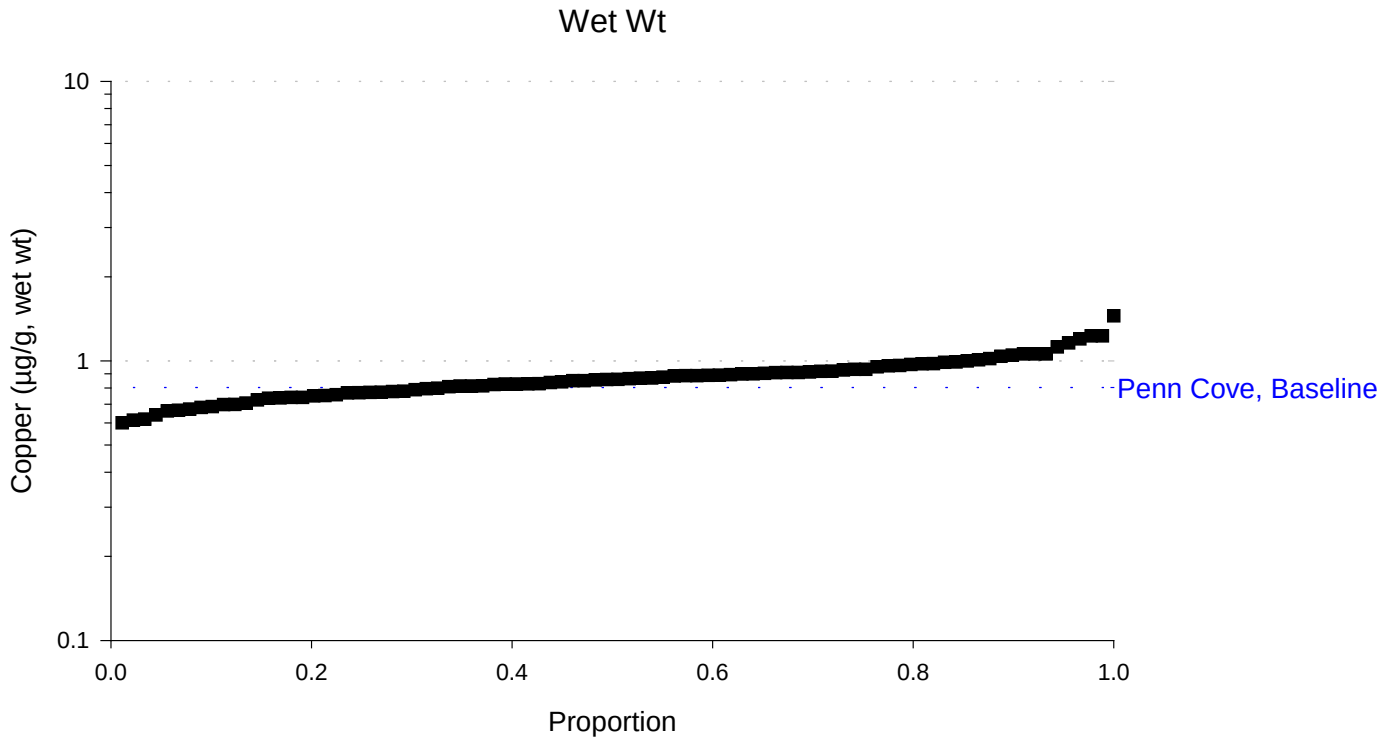


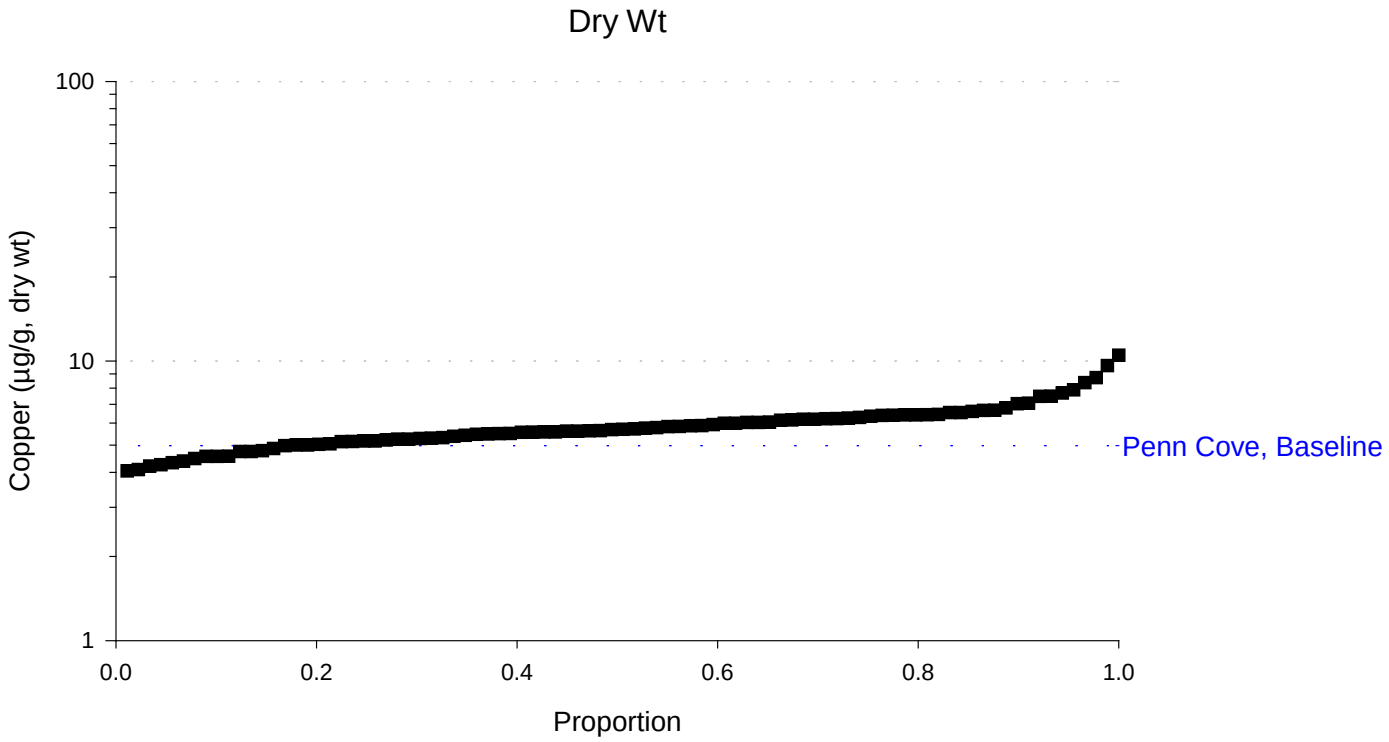
22 APPENDIX O: Details of Copper Findings at Transplanted (i.e. Caged) Mussel Sites

22.1 Map of Copper Concentrations



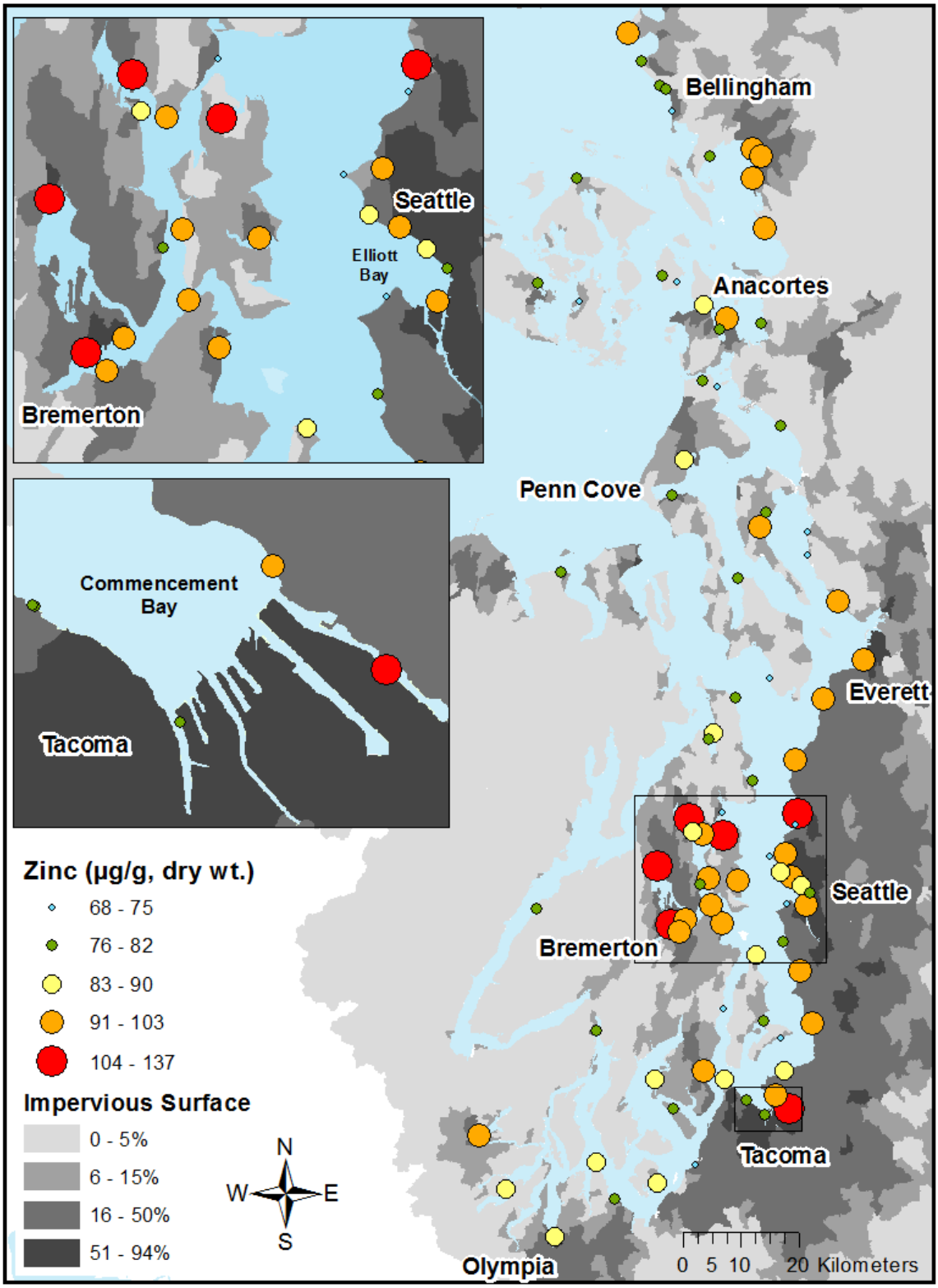
22.2 Cumulative Frequency Distribution of Copper Concentrations



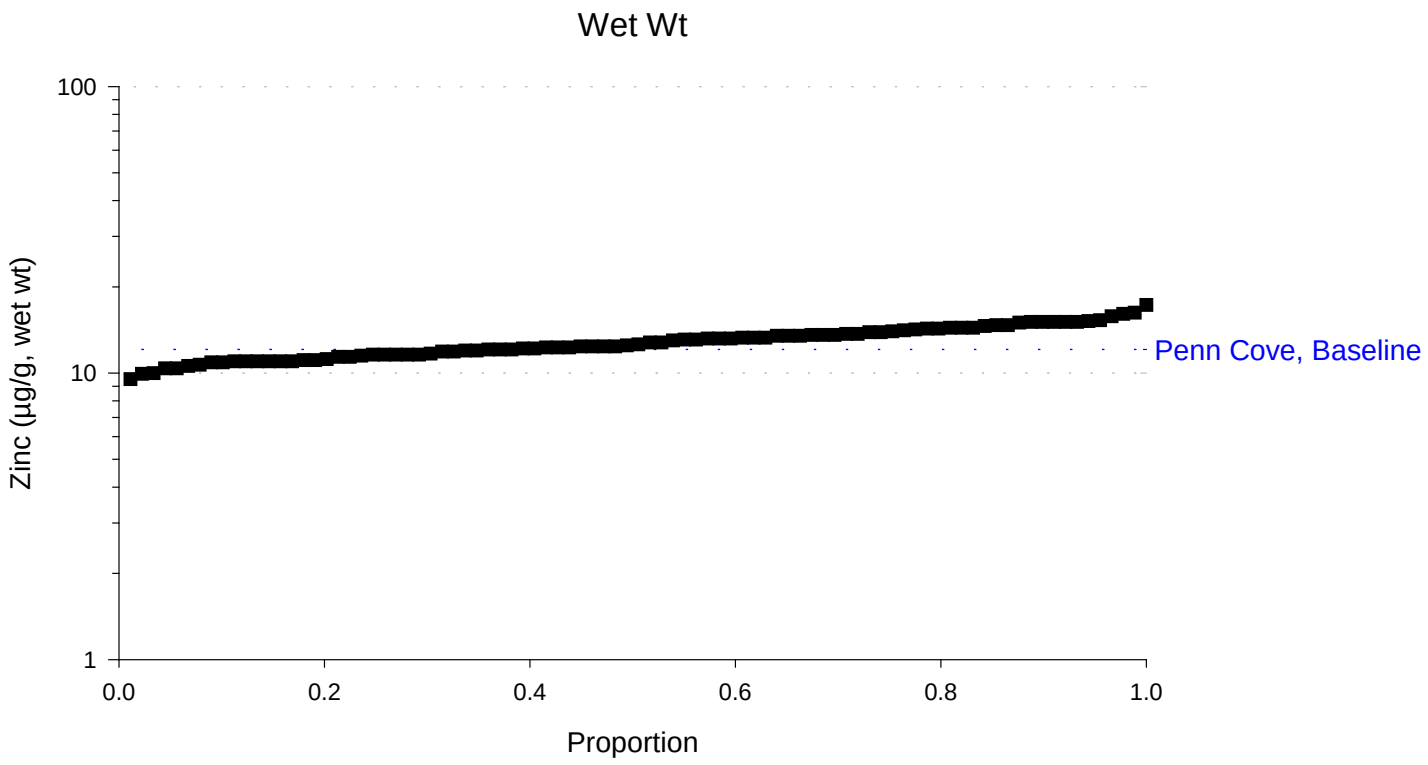


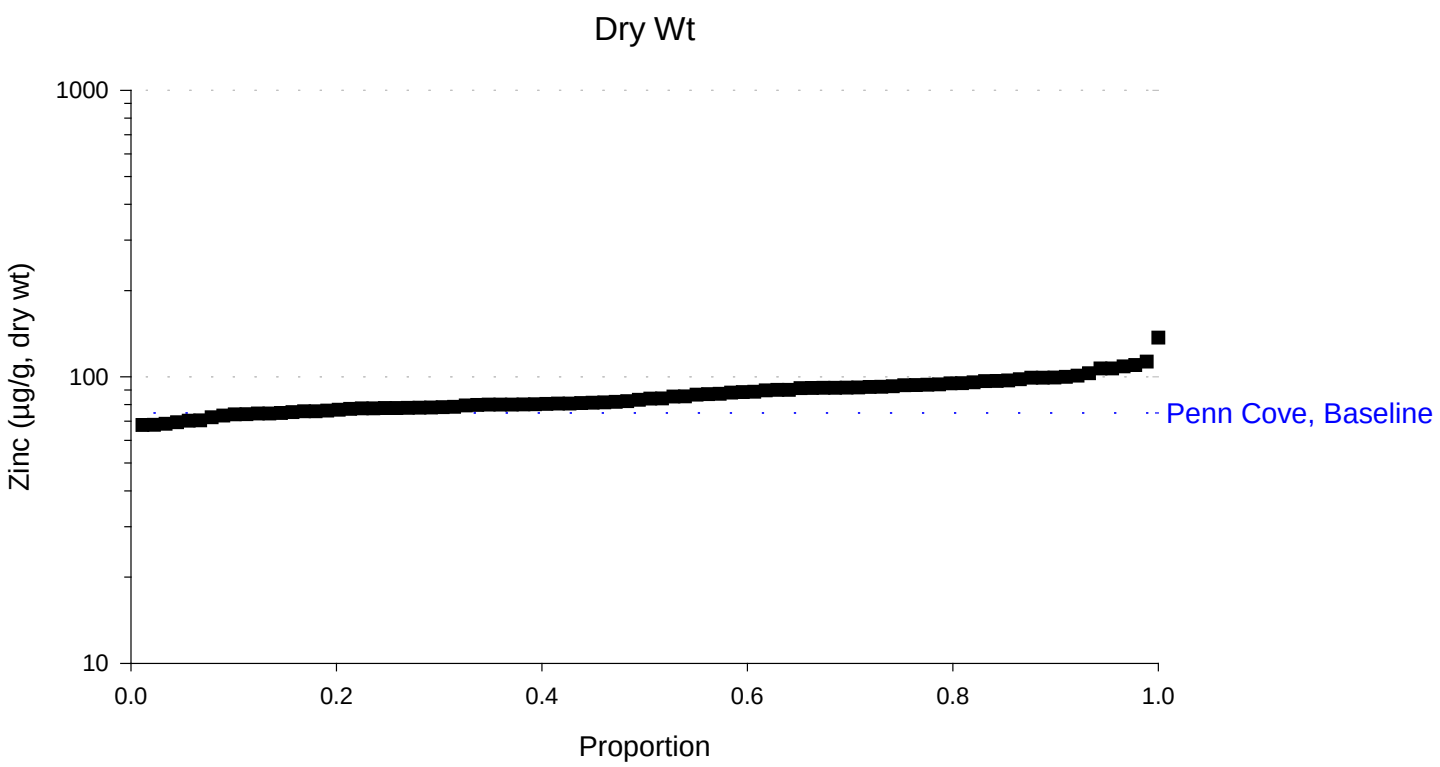
23 APPENDIX P: Details of Zinc Findings at Transplanted (i.e. Caged) Mussel Sites

23.1 Map of Zinc Concentrations



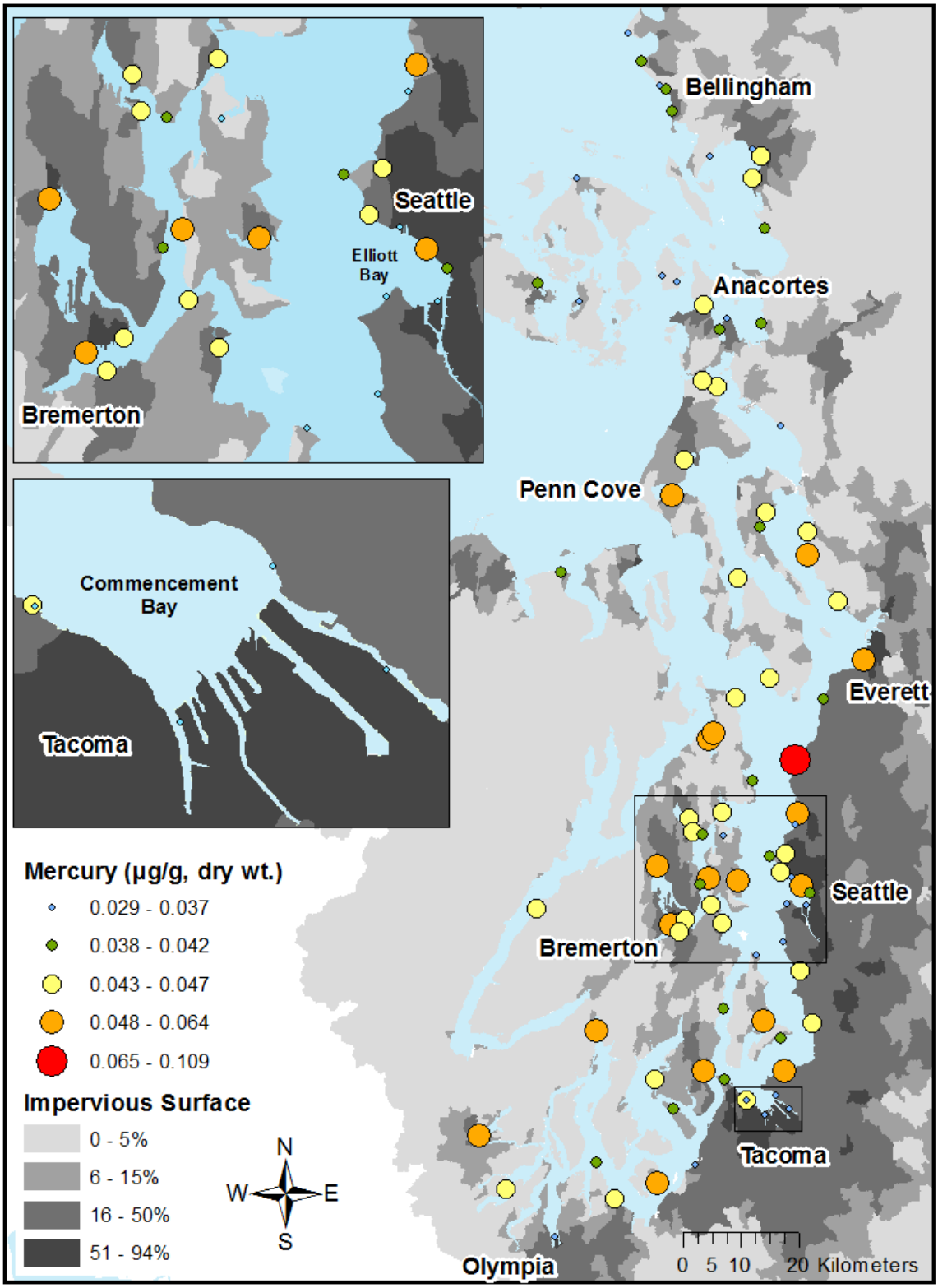
23.2 Cumulative Frequency Distribution of Zinc Concentrations



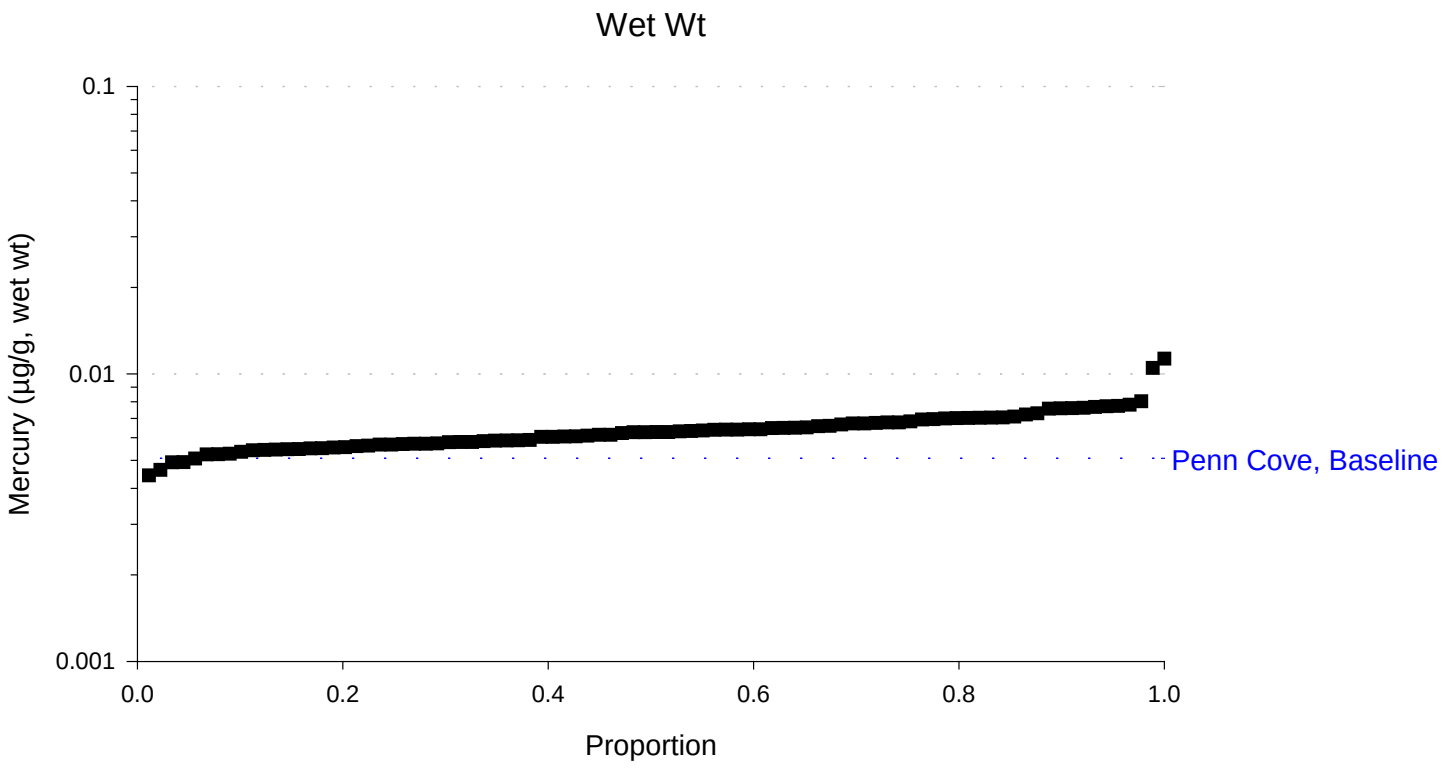


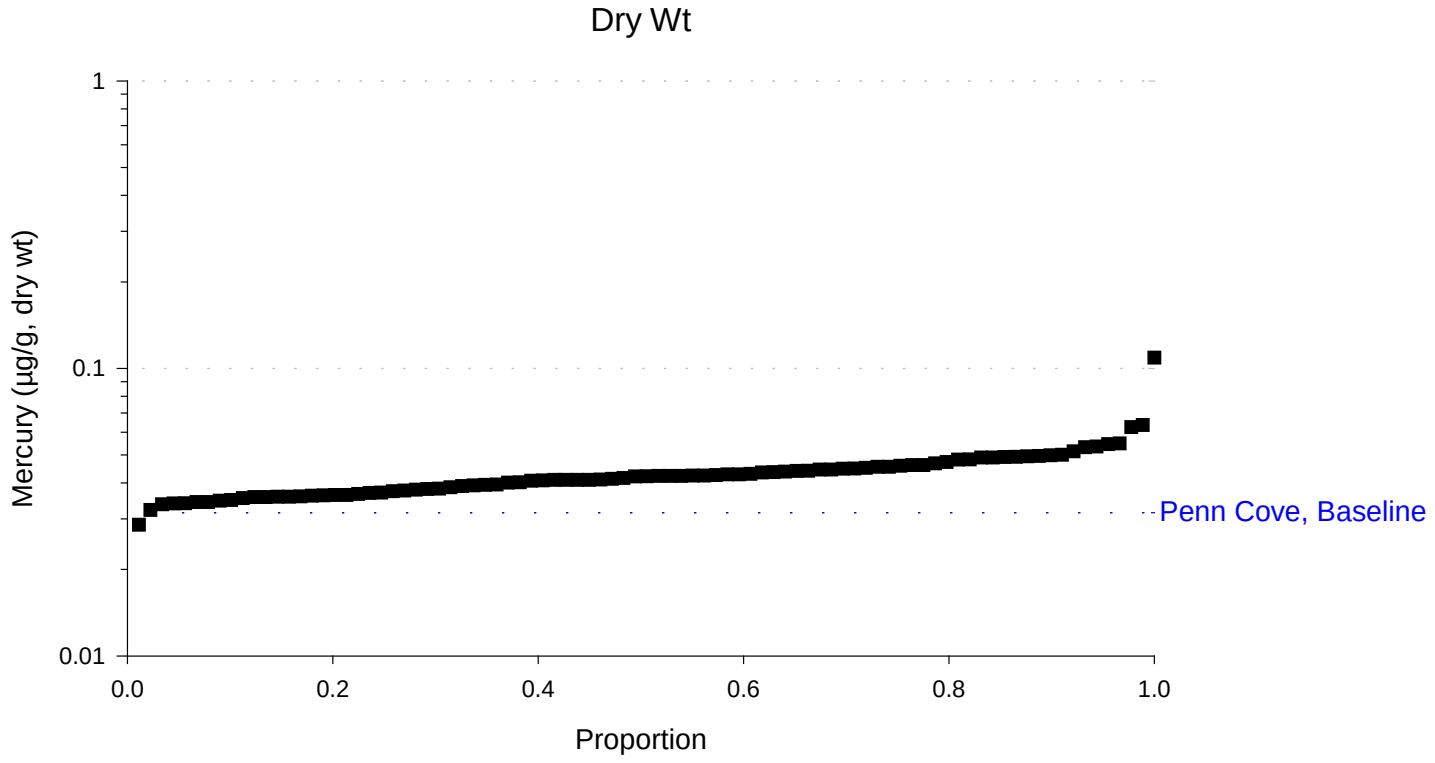
24 APPENDIX Q: Details of Mercury Findings at Transplanted (i.e. Caged) Mussel Sites

24.1 Map of Mercury Concentrations



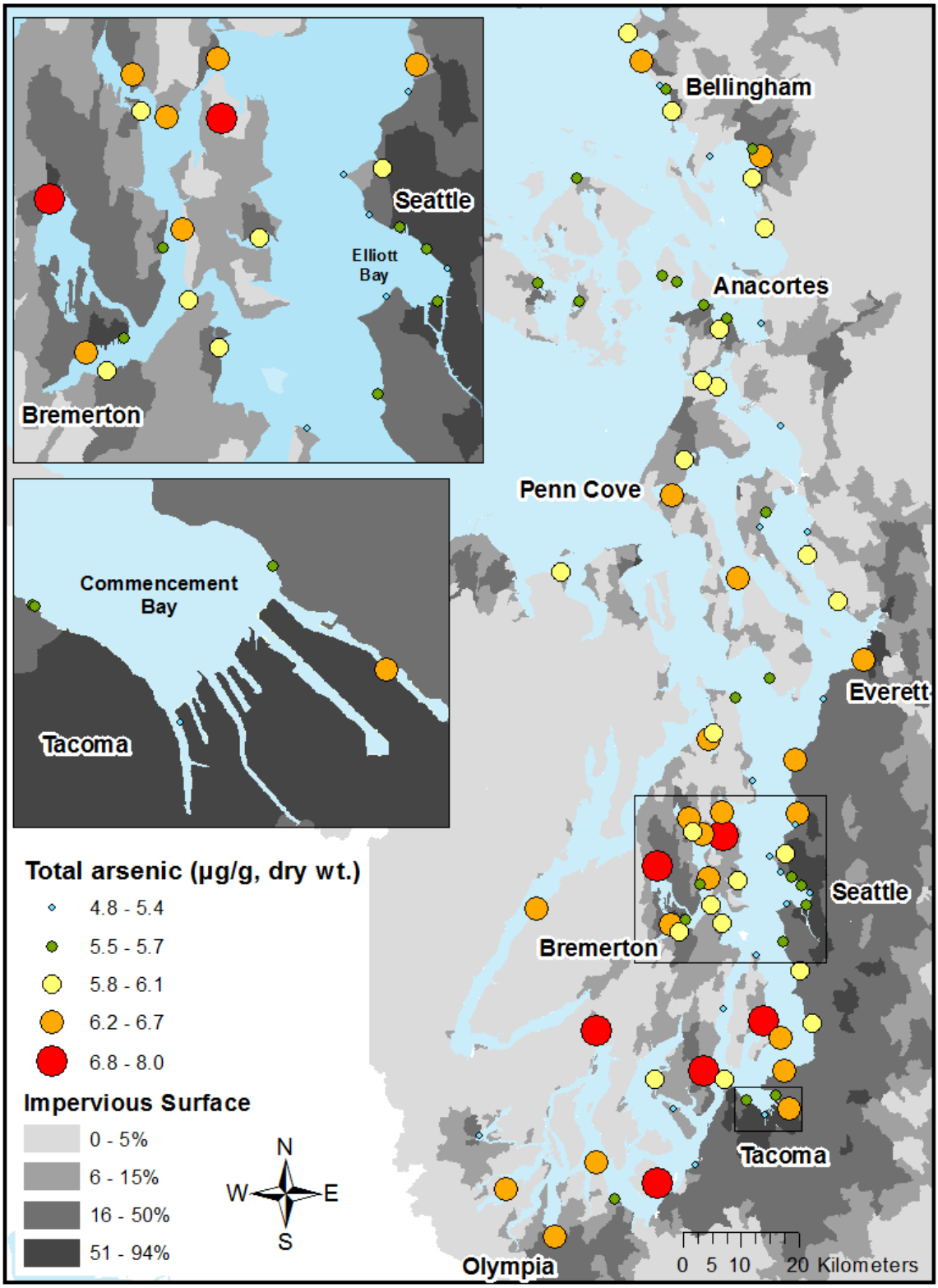
24.2 Cumulative Frequency Distribution of Mercury Concentrations



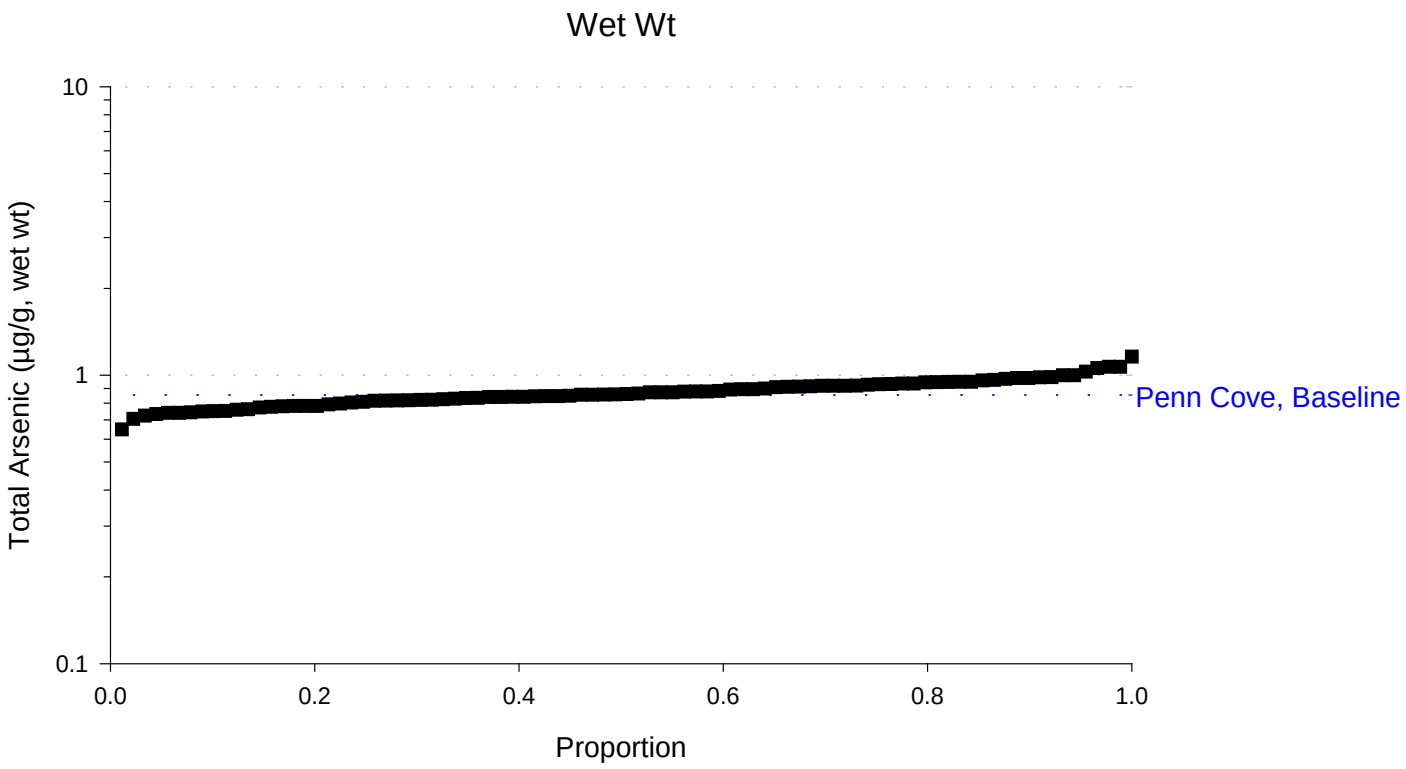


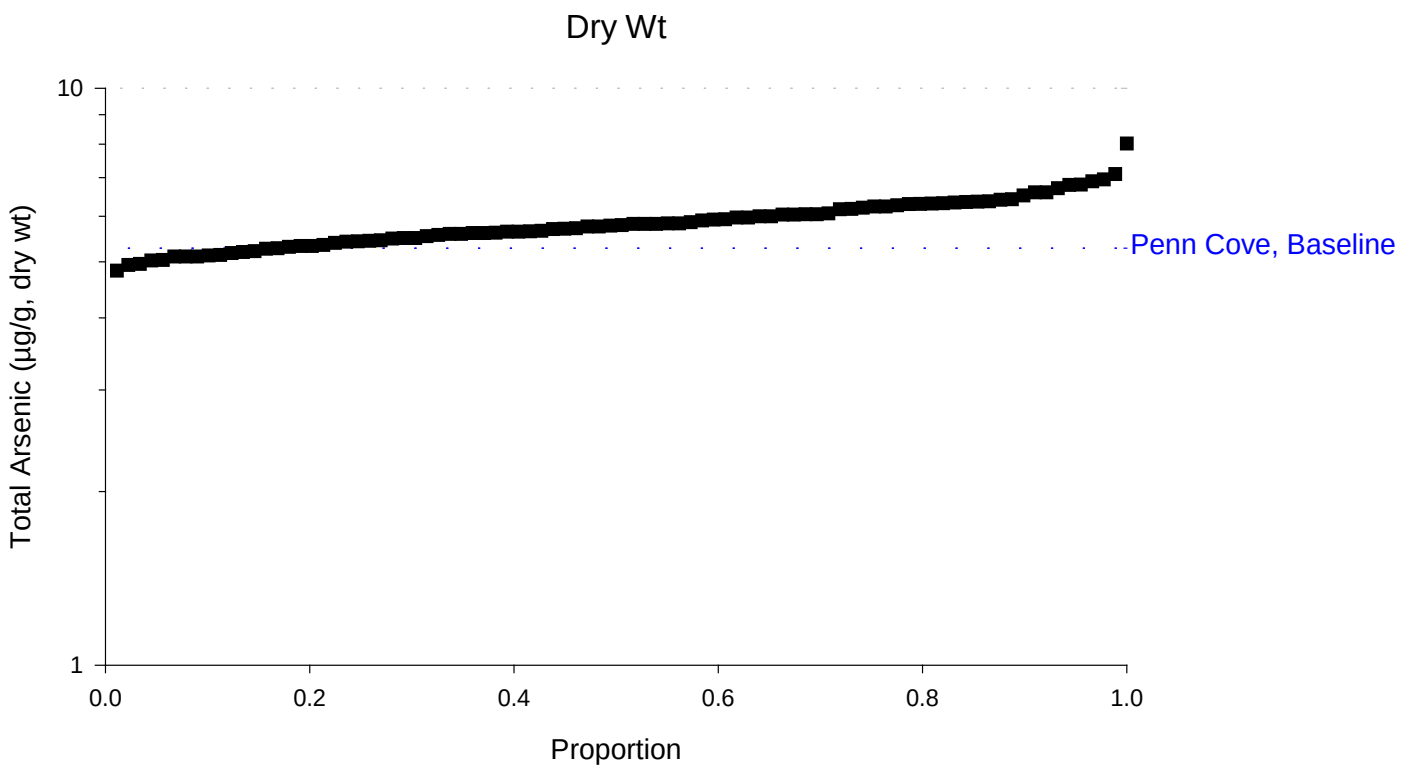
25 APPENDIX R: Details of Arsenic Findings at Transplanted (i.e. Caged) Mussel Sites

25.1 Map of Total Arsenic (Organic + Inorganic) Concentrations



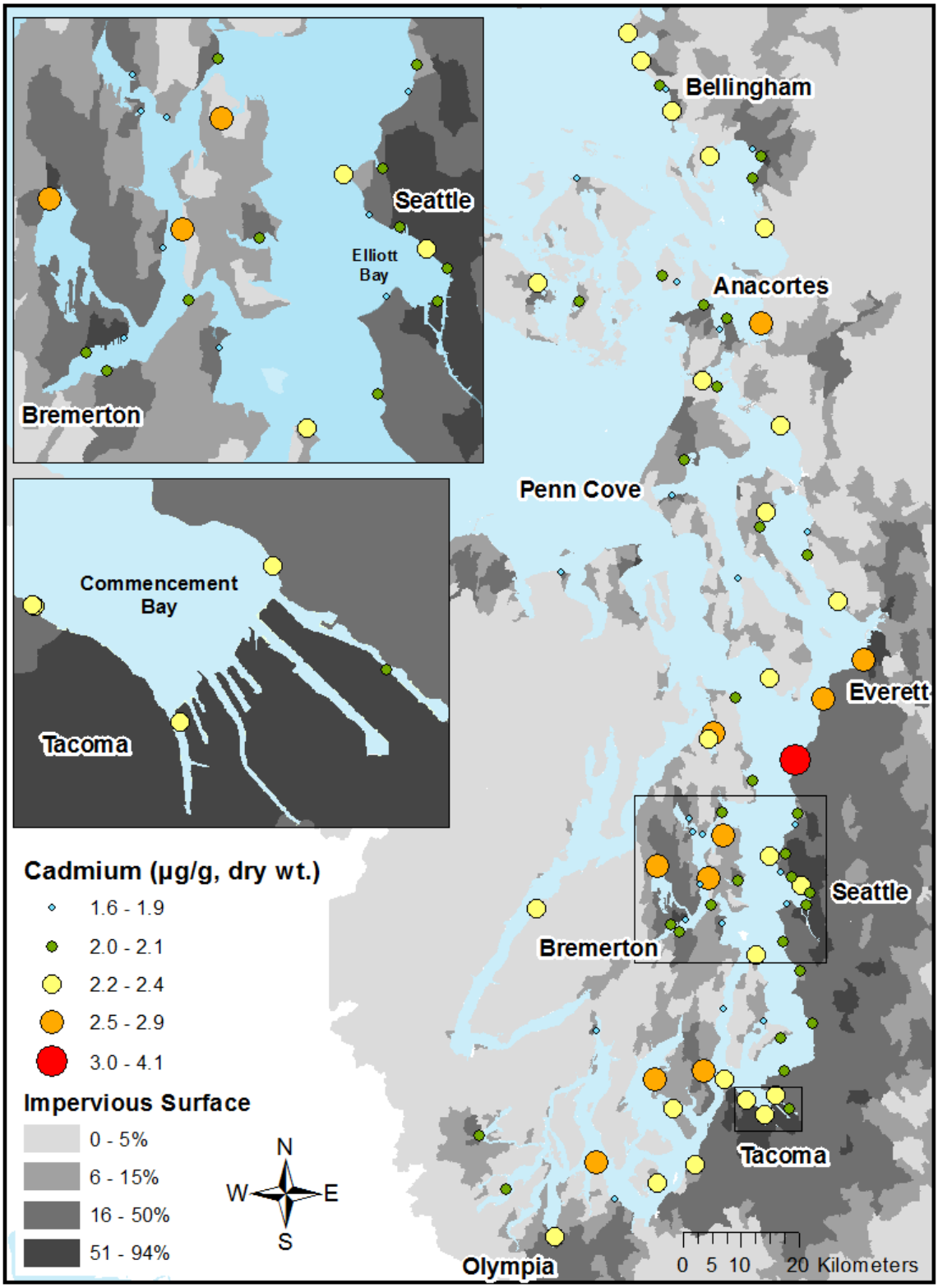
25.2 Cumulative Frequency Distribution of Total Arsenic (Organic + Inorganic) Concentrations



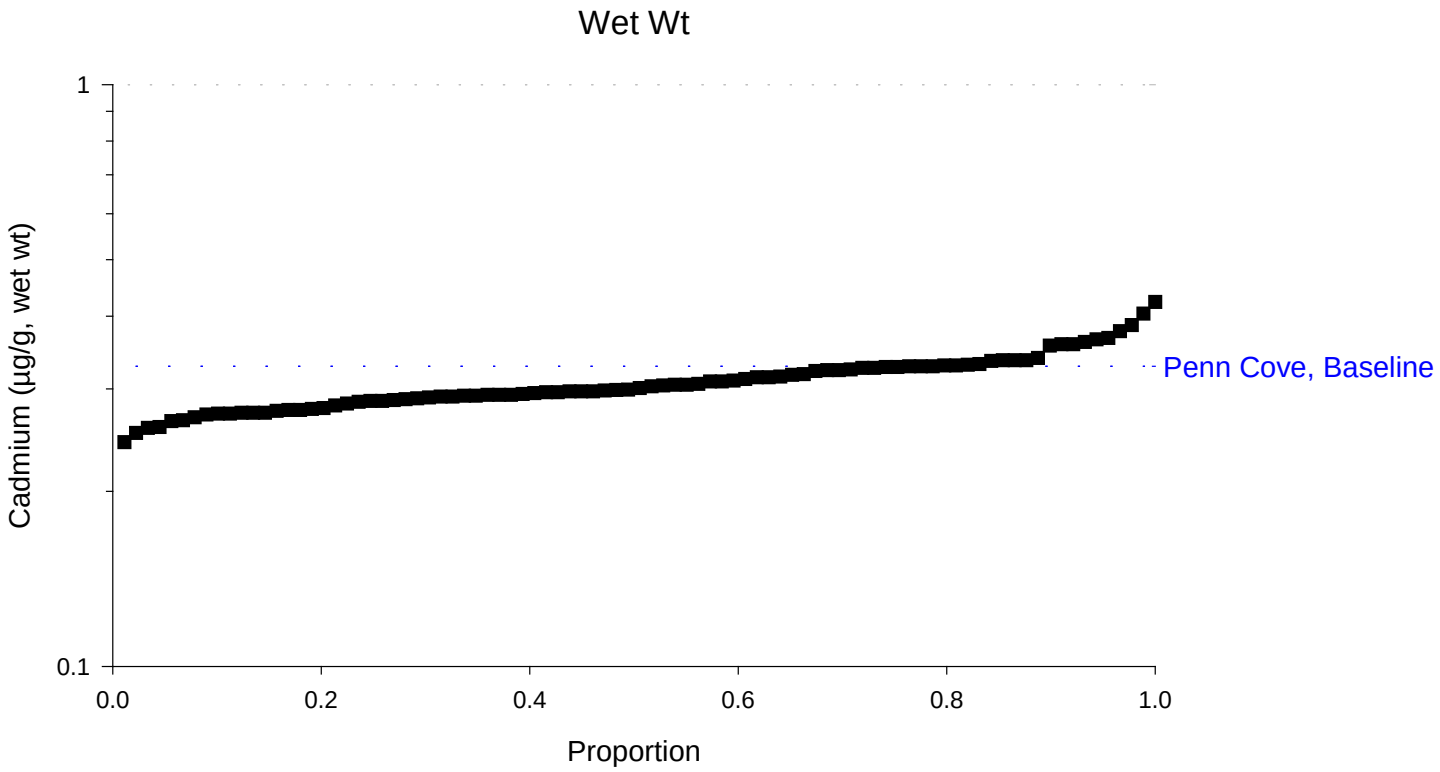


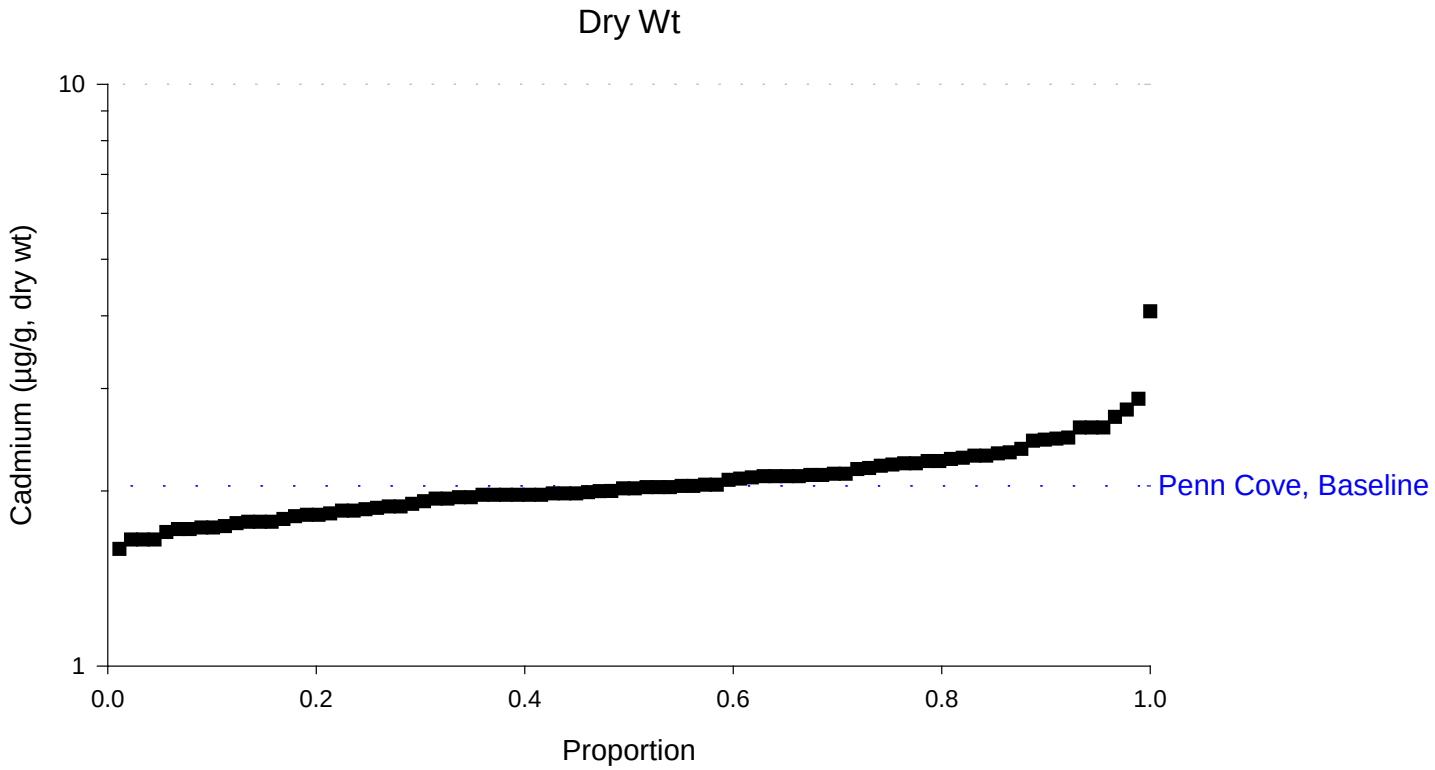
26 APPENDIX S: Detail of Cadmium Findings at Transplanted (i.e. Caged) Mussel Sites

26.1 Map of Cadmium Concentrations



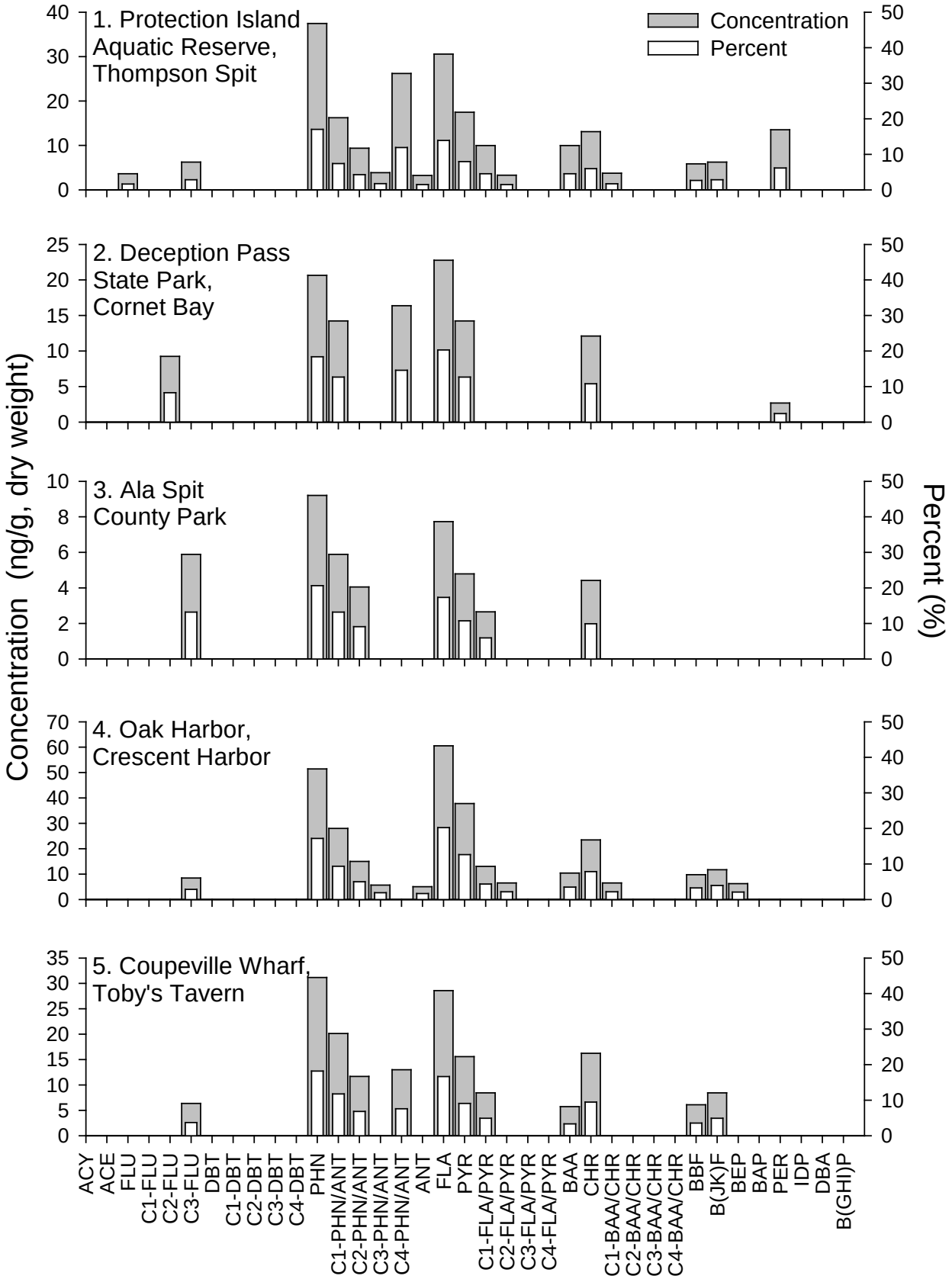
26.2 Cumulative Frequency Distribution of Cadmium Concentrations

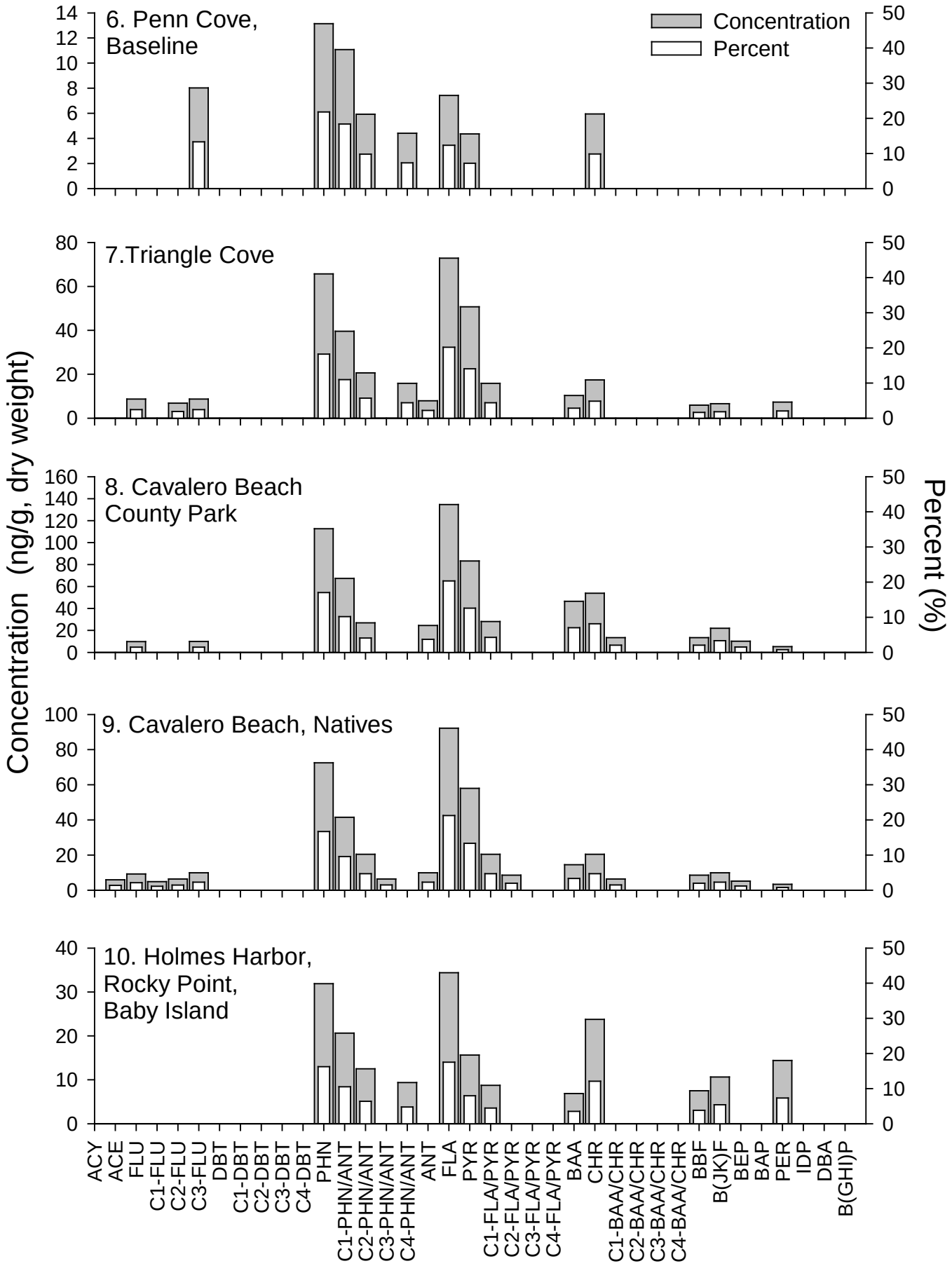


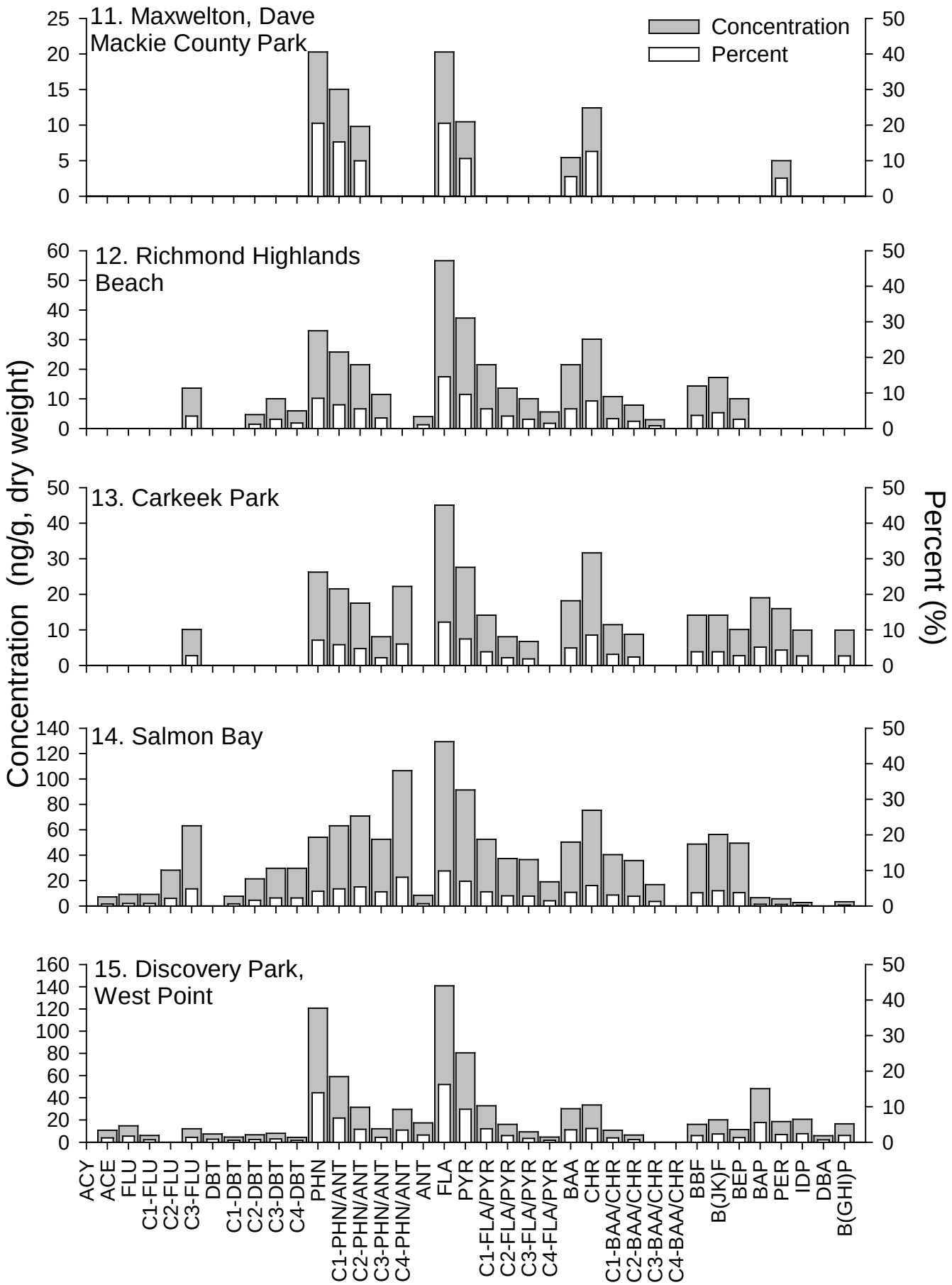


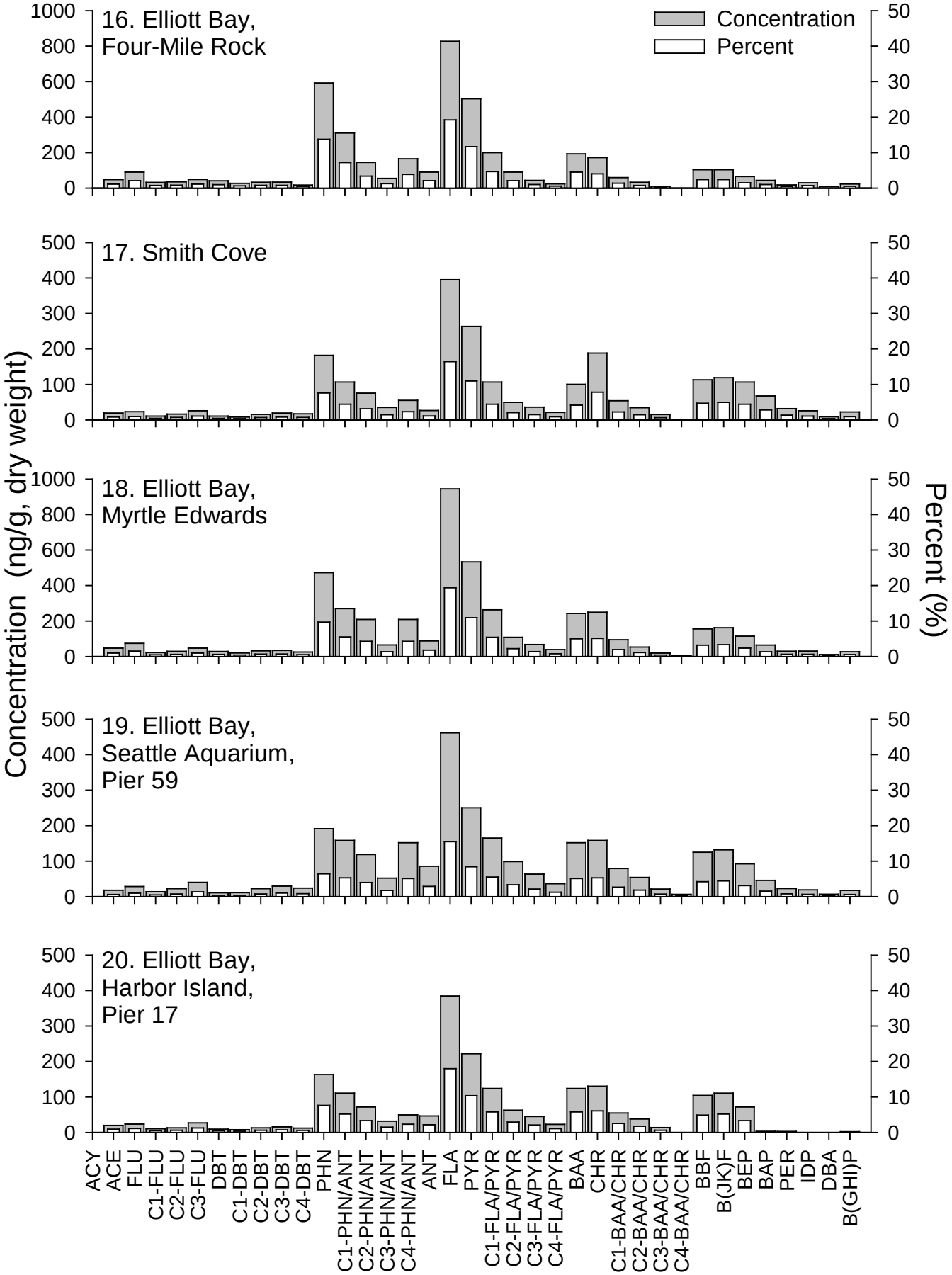
27 APPENDIX T: PAH FINGERPRINT FROM TRANSPLANTED (I.E. CAGED) MUSSEL SITES.

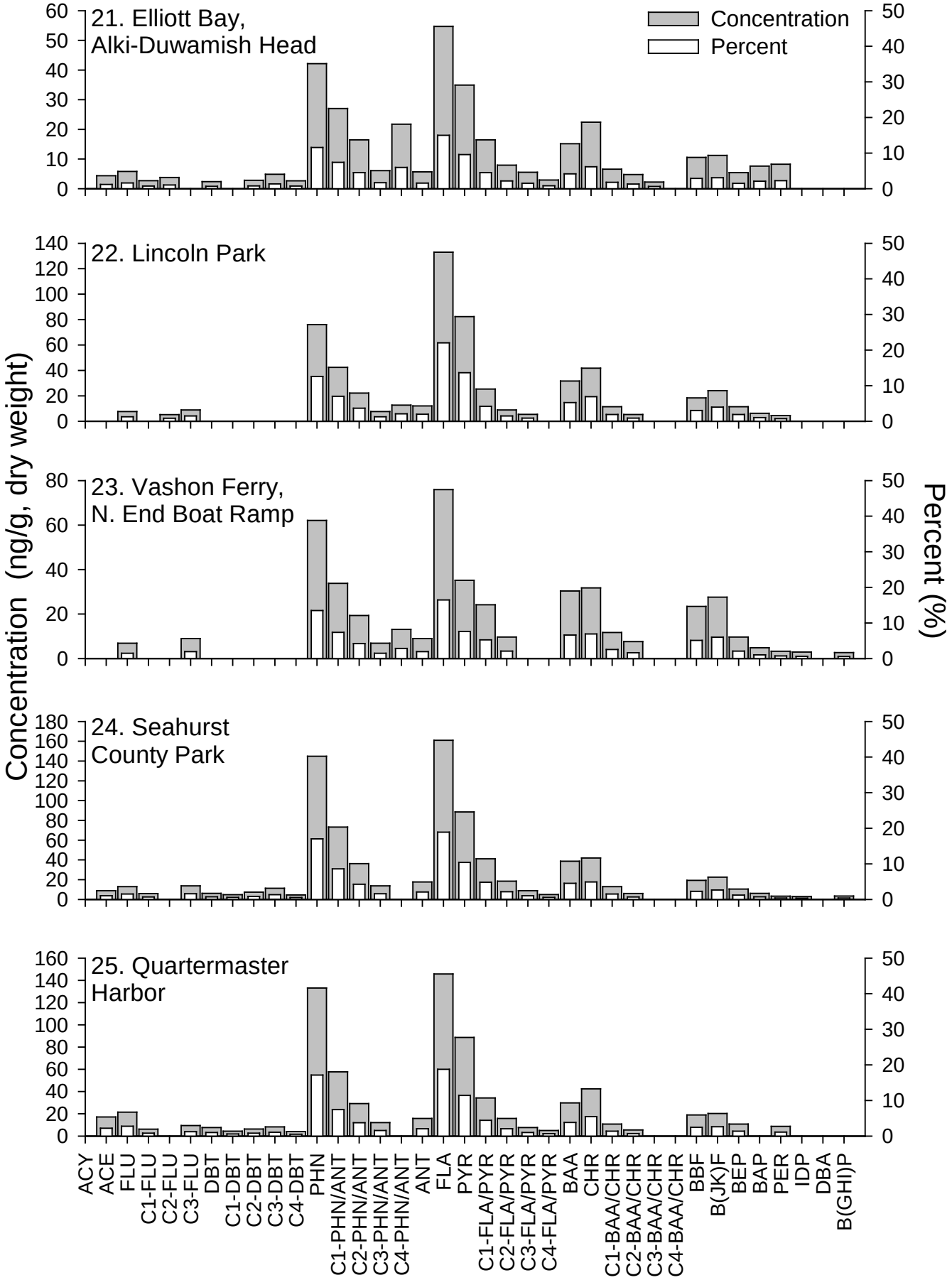
Site order is arranged alphabetically by county and then from north to south within each county. Scale on left side y-axis is adjusted in each plot so the visual focus is on proportion of PAHs, not concentration.

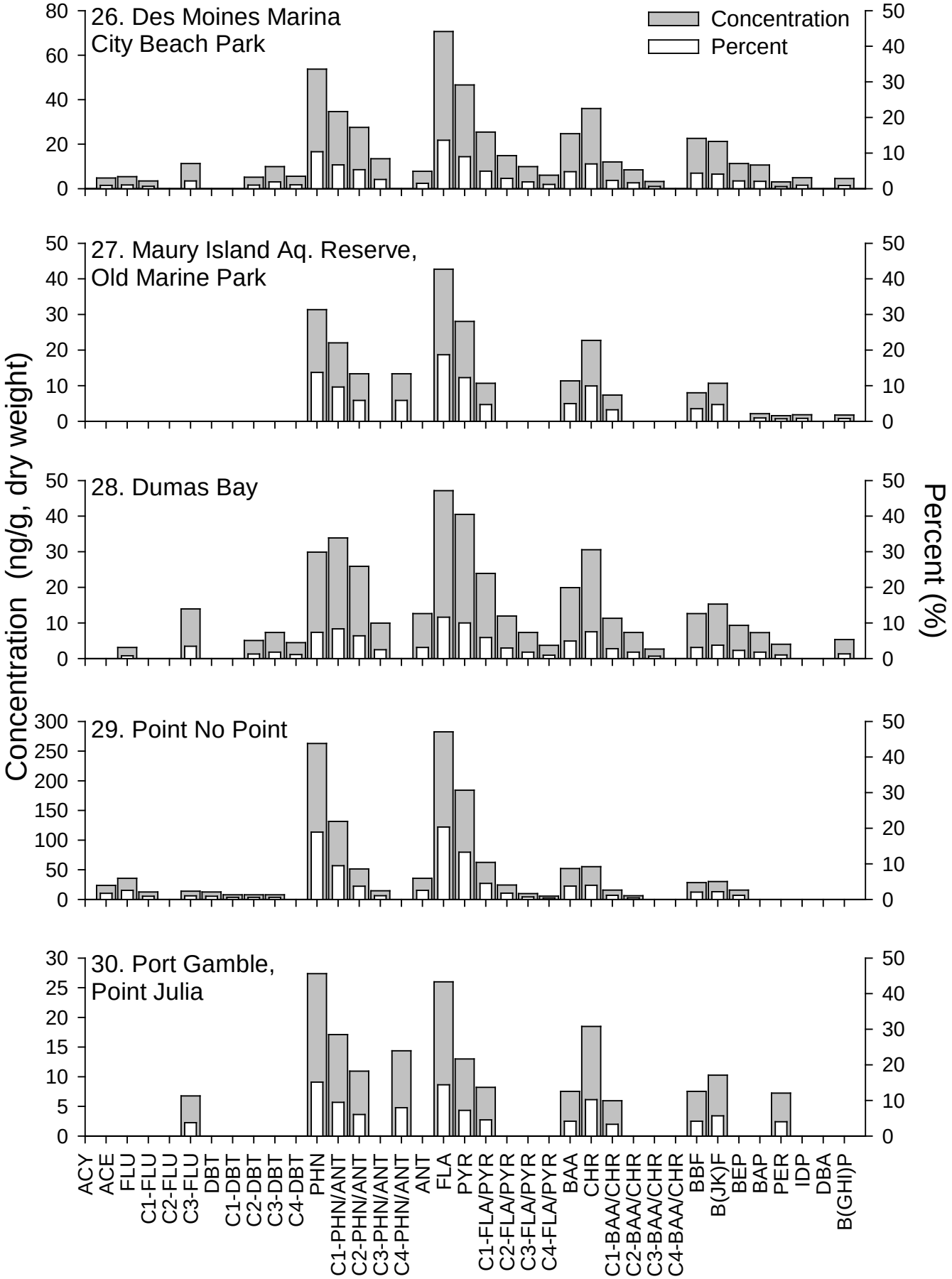


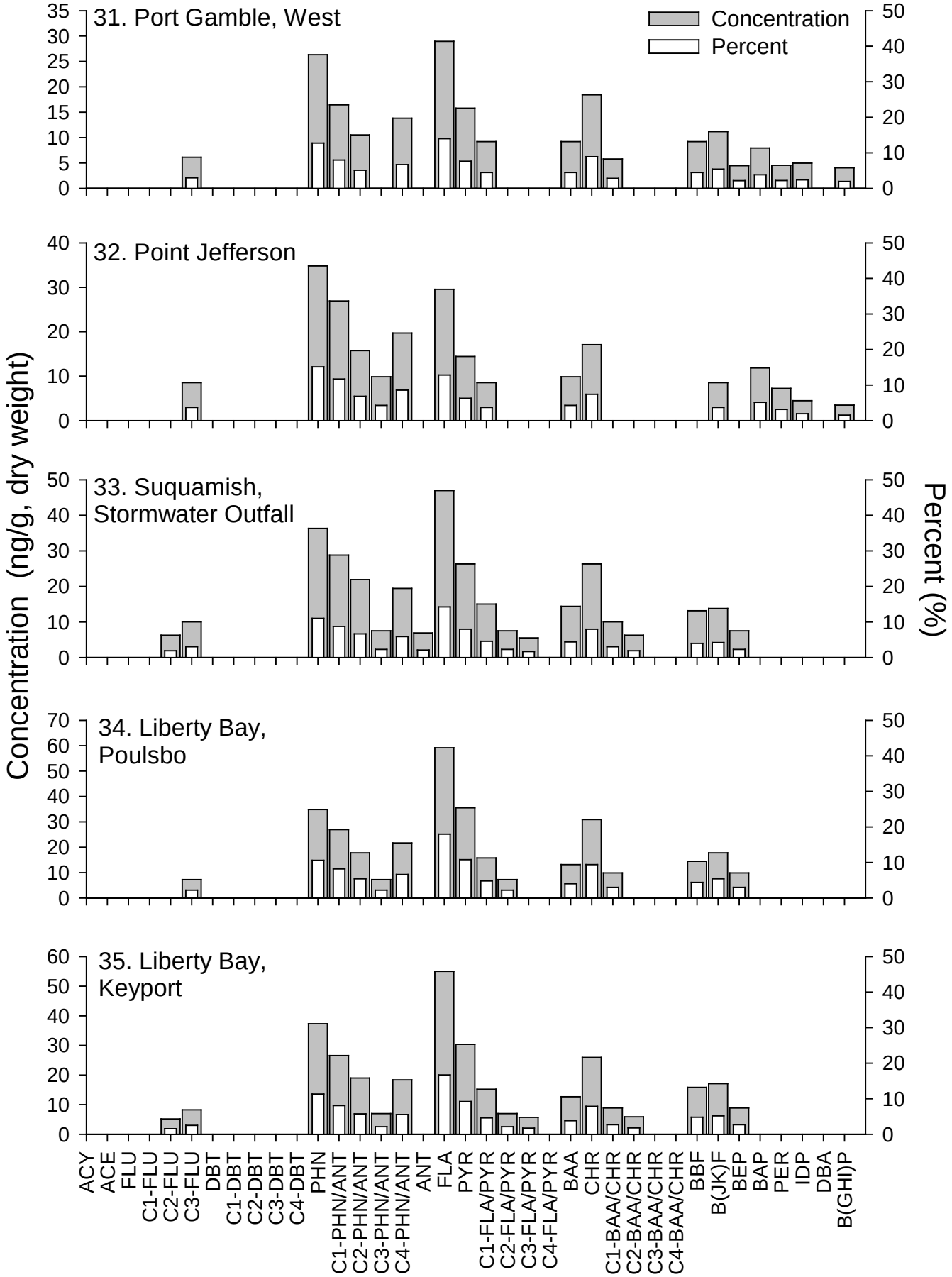

6. Penn Cove, Baseline
Concentration
Percent
7.Triangle Cove
8. Cavalero Beach County Park
9. Cavalero Beach, Natives
10. Holmes Harbor, Rocky Point, Baby Island
Concentration (ng/g, dry weight)
Percent (%)
ACY
ACE
FLU
C1-FLU
C2-FLU
C3-FLU
DBT
C1-DBT
C2-DBT
C3-DBT
C4-DBT
PHN
C1-PHN/ANT
C2-PHN/ANT
C3-PHN/ANT
C4-PHN/ANT
ANT
FLA
PYR
C1-FLA/PYR
C2-FLA/PYR
C3-FLA/PYR
C4-FLA/PYR
BAA
CHR
C1-BAA/CHR
C2-BAA/CHR
C3-BAA/CHR
C4-BAA/CHR
BBF
B(JK)F
BEP
BAP
PER
IDP
DBA
B(GHI)P

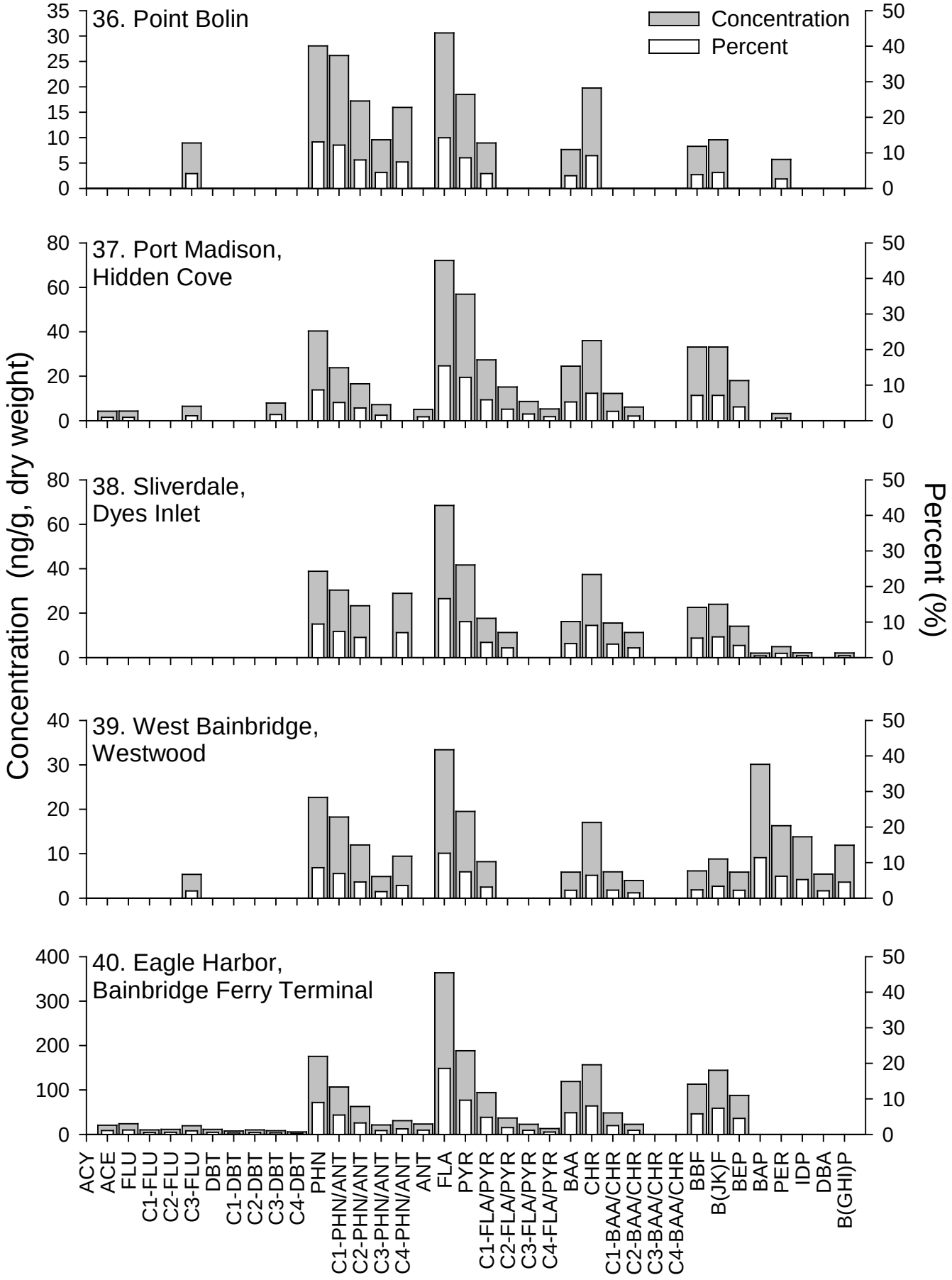

11. Maxwelton, Dave Mackie County Park
12. Richmond Highlands Beach
13. Carkeek Park
14. Salmon Bay
15. Discovery Park, West Point
Concentration
Percent
Concentration (ng/g, dry weight)
Percent (%)
ACY
ACE
FLU
C1-FLU
C2-FLU
C3-FLU
DBT
C1-DBT
C2-DBT
C3-DBT
C4-DBT
PHN
C1-PHN/ANT
C2-PHN/ANT
C3-PHN/ANT
C4-PHN/ANT
ANT
FLA
PYR
C1-FLA/PYR
C2-FLA/PYR
C3-FLA/PYR
C4-FLA/PYR
BAA
CHR
C1-BAA/CHR
C2-BAA/CHR
C3-BAA/CHR
C4-BAA/CHR
BBF
B(JK)F
BEP
BAP
PER
IDP
DBA
B(GHI)P

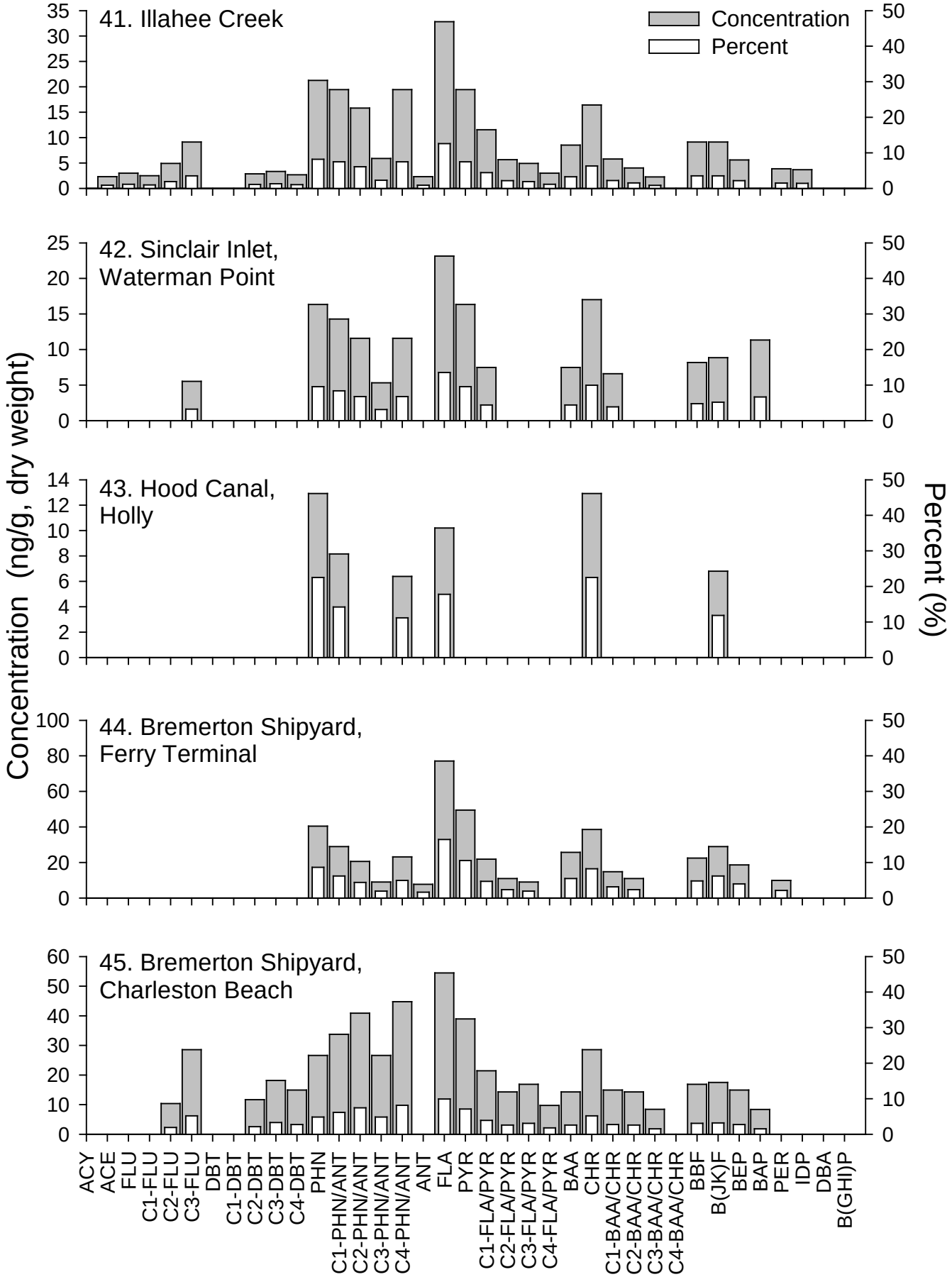

16. Elliott Bay,
Four-Mile Rock
Concentration
Percent
17. Smith Cove
18. Elliott Bay,
Myrtle Edwards
19. Elliott Bay,
Seattle Aquarium,
Pier 59
20. Elliott Bay,
Harbor Island,
Pier 17
Concentration (ng/g, dry weight)
Percent (%)
ACY
ACE
FLU
C1-FLU
C2-FLU
C3-FLU
DBT
C1-DBT
C2-DBT
C3-DBT
C4-DBT
PHN
C1-PHN/ANT
C2-PHN/ANT
C3-PHN/ANT
C4-PHN/ANT
ANT
FLA
PYR
C1-FLA/PYR
C2-FLA/PYR
C3-FLA/PYR
C4-FLA/PYR
BAA
CHR
C1-BAA/CHR
C2-BAA/CHR
C3-BAA/CHR
C4-BAA/CHR
BBF
B(JK)F
BEP
BAP
PER
IDP
DBA
B(GHI)P

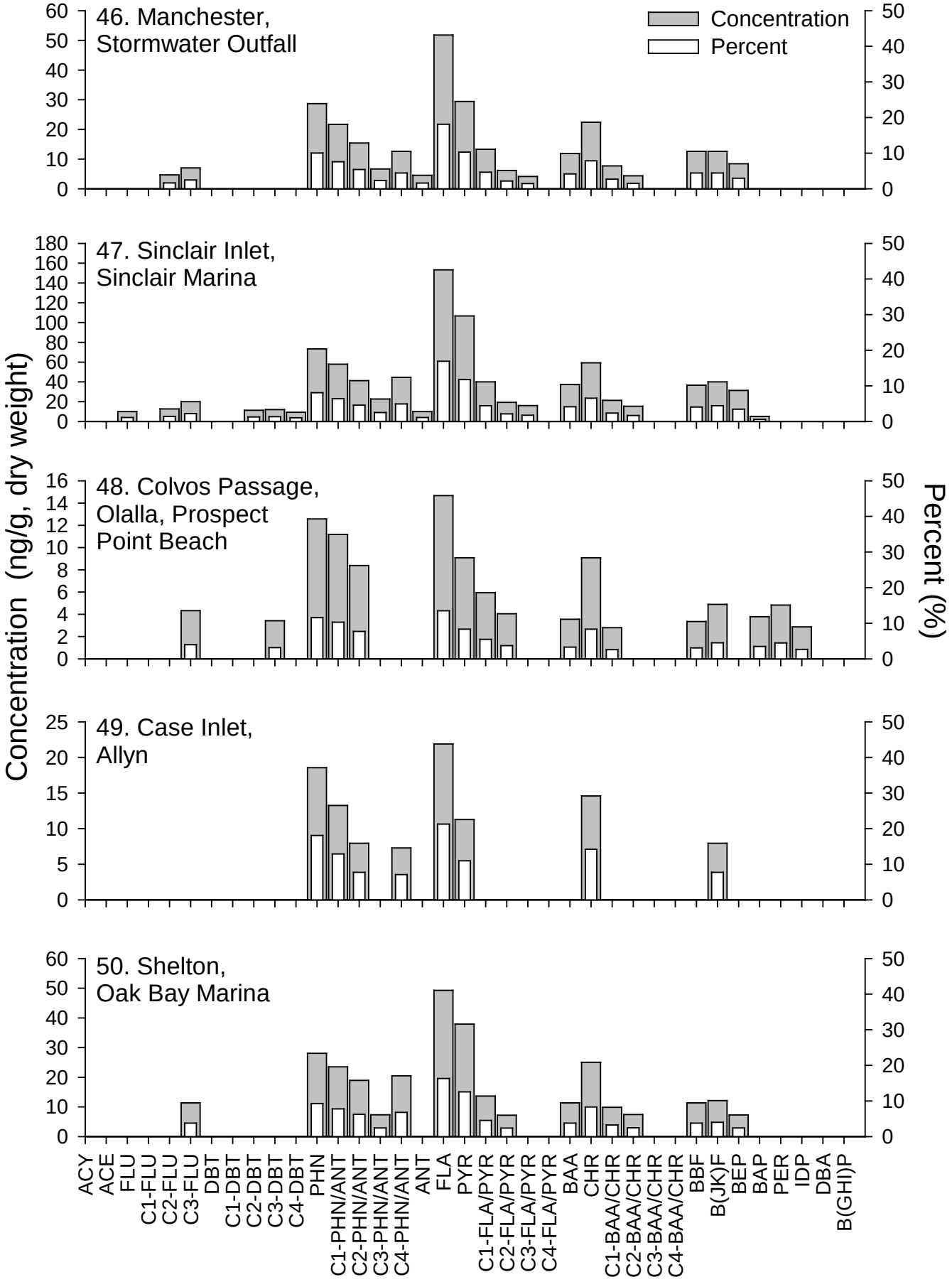

21. Elliott Bay,
Alki-Duwamish Head
Concentration
Percent
22. Lincoln Park
23. Vashon Ferry,
N. End Boat Ramp
24. Seahurst
County Park
25. Quartermaster
Harbor
Concentration (ng/g, dry weight)
Percent (%)
ACY
ACE
FLU
C1-FLU
C2-FLU
C3-FLU
DBT
C1-DBT
C2-DBT
C3-DBT
C4-DBT
PHN
C1-PHN/ANT
C2-PHN/ANT
C3-PHN/ANT
C4-PHN/ANT
ANT
FLA
PYR
C1-FLA/PYR
C2-FLA/PYR
C3-FLA/PYR
C4-FLA/PYR
BAA
CHR
C1-BAA/CHR
C2-BAA/CHR
C3-BAA/CHR
C4-BAA/CHR
BBF
B(JK)F
BEP
BAP
PER
IDP
DBA
B(GHI)P

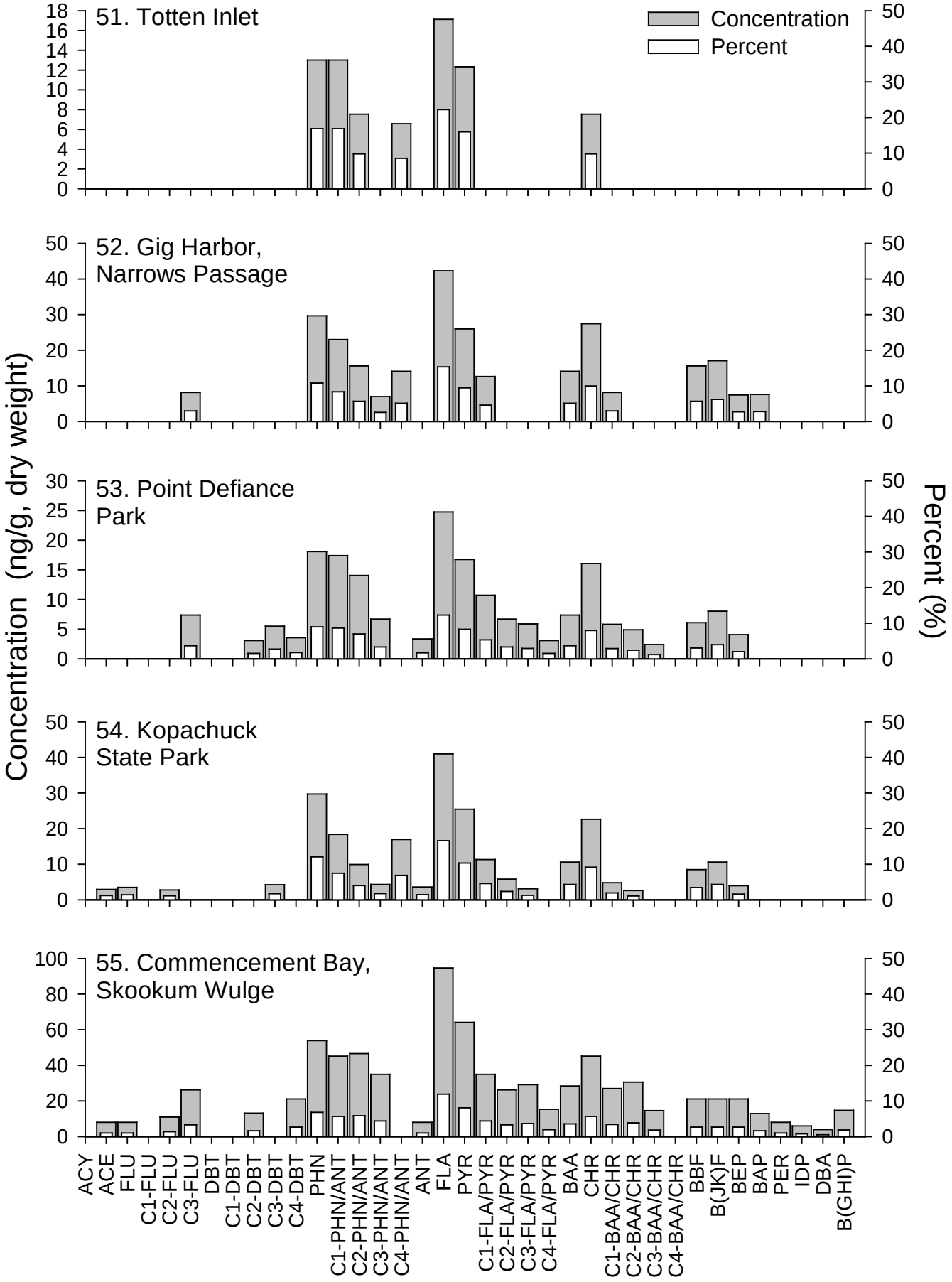

26. Des Moines Marina
City Beach Park
Concentration
Percent
27. Maury Island Aq. Reserve,
Old Marine Park
28. Dumas Bay
29. Point No Point
30. Port Gamble,
Point Julia
Concentration (ng/g, dry weight)
Percent (%)
ACY
ACE
FLU
C1-FLU
C2-FLU
C3-FLU
DBT
C1-DBT
C2-DBT
C3-DBT
C4-DBT
PHN
C1-PHN/ANT
C2-PHN/ANT
C3-PHN/ANT
C4-PHN/ANT
ANT
FLA
PYR
C1-FLA/PYR
C2-FLA/PYR
C3-FLA/PYR
C4-FLA/PYR
BAA
CHR
C1-BAA/CHR
C2-BAA/CHR
C3-BAA/CHR
C4-BAA/CHR
BBF
B(JK)F
BEP
BAP
PER
IDP
DBA
B(GHI)P

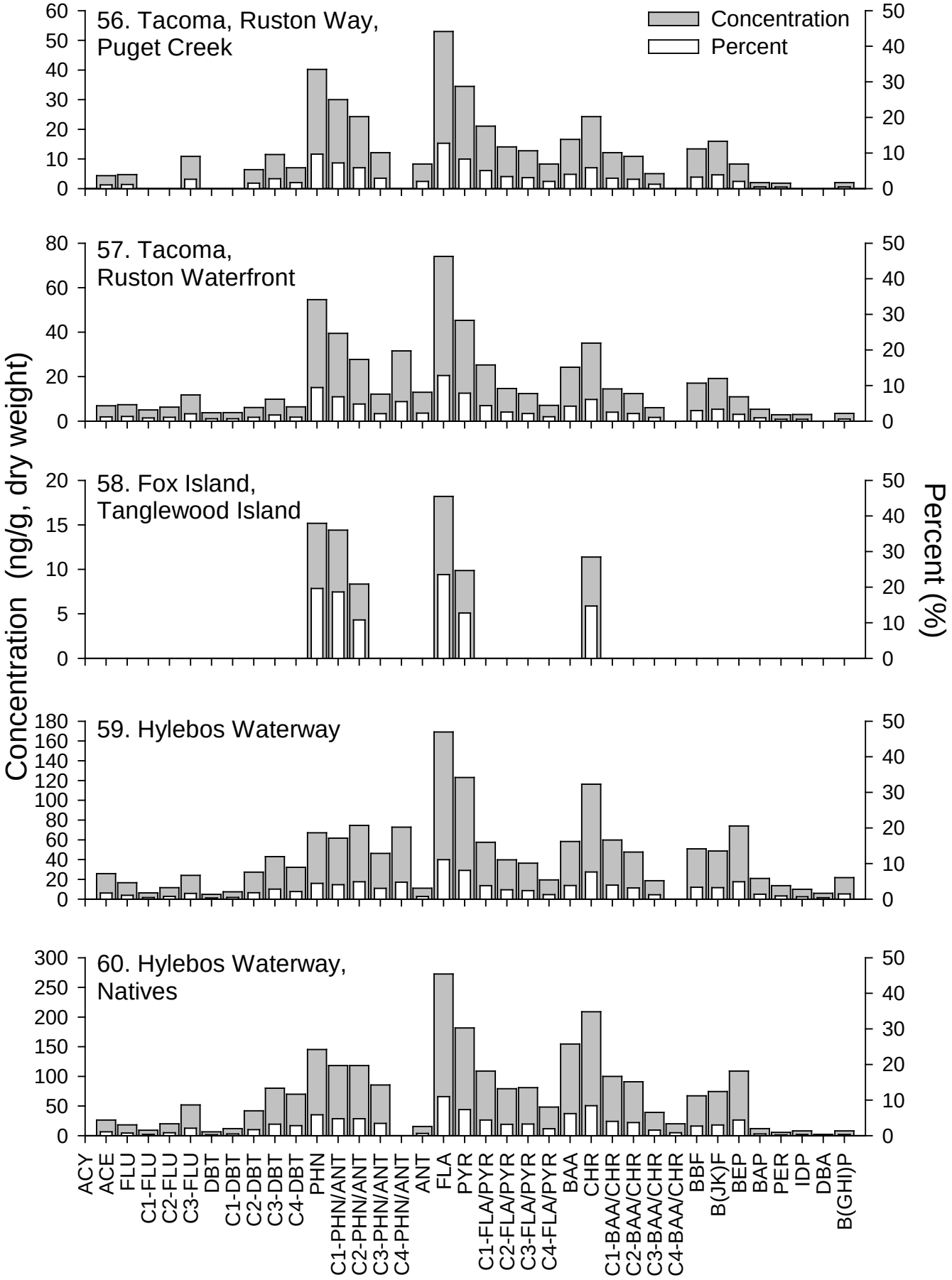

31. Port Gamble, West
Concentration
Percent
32. Point Jefferson
33. Suquamish, Stormwater Outfall
34. Liberty Bay, Poulsbo
35. Liberty Bay, Keyport
Concentration (ng/g, dry weight)
Percent (%)
ACY
ACE
FLU
C1-FLU
C2-FLU
C3-FLU
DBT
C1-DBT
C2-DBT
C3-DBT
C4-DBT
PHN
C1-PHN/ANT
C2-PHN/ANT
C3-PHN/ANT
C4-PHN/ANT
ANT
FLA
PYR
C1-FLA/PYR
C2-FLA/PYR
C3-FLA/PYR
C4-FLA/PYR
BAA
CHR
C1-BAA/CHR
C2-BAA/CHR
C3-BAA/CHR
C4-BAA/CHR
BBF
B(JK)F
BEP
BAP
PER
IDP
DBA
B(GHI)P

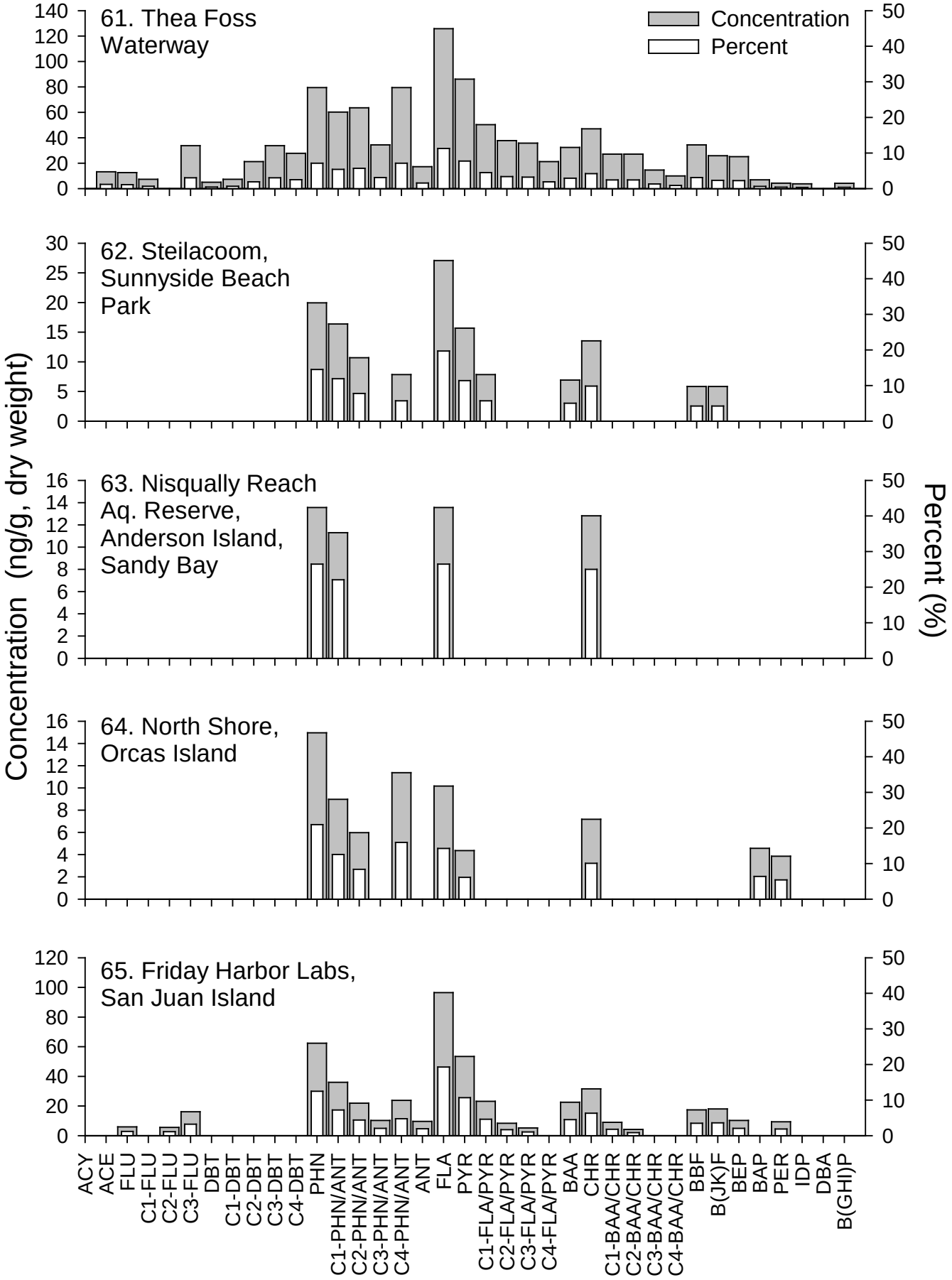

36. Point Bolin
Concentration
Percent
37. Port Madison, Hidden Cove
38. Sliverdale, Dyes Inlet
39. West Bainbridge, Westwood
40. Eagle Harbor, Bainbridge Ferry Terminal
Concentration (ng/g, dry weight)
Percent (%)
ACY
ACE
FLU
C1-FLU
C2-FLU
C3-FLU
DBT
C1-DBT
C2-DBT
C3-DBT
C4-DBT
PHN
C1-PHN/ANT
C2-PHN/ANT
C3-PHN/ANT
C4-PHN/ANT
ANT
FLA
PYR
C1-FLA/PYR
C2-FLA/PYR
C3-FLA/PYR
C4-FLA/PYR
BAA
CHR
C1-BAA/CHR
C2-BAA/CHR
C3-BAA/CHR
C4-BAA/CHR
BBF
B(JK)F
BEP
BAP
PER
IDP
DBA
B(GHI)P

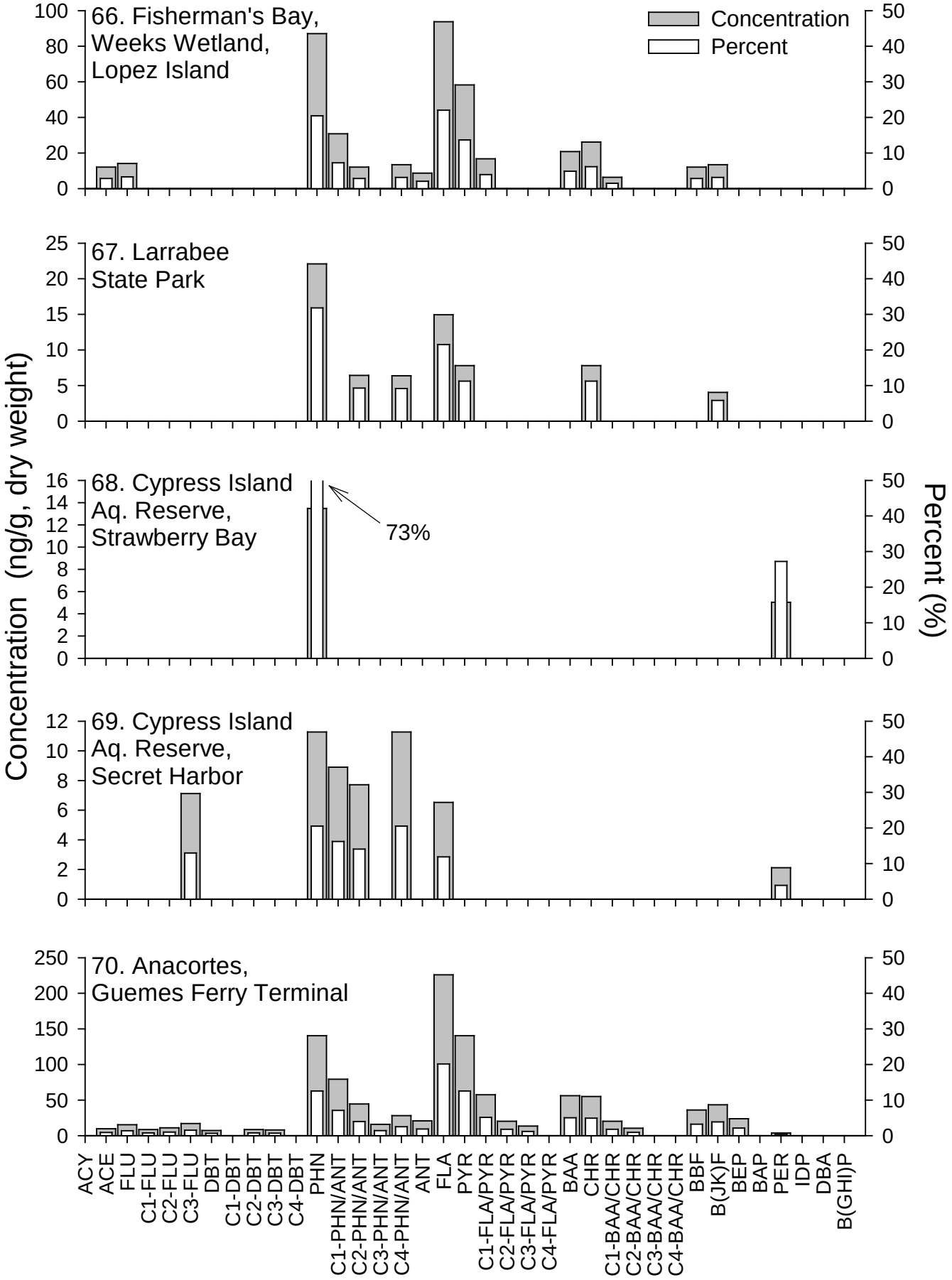

41. Illahee Creek
Concentration
Percent
42. Sinclair Inlet, Waterman Point
43. Hood Canal, Holly
44. Bremerton Shipyard, Ferry Terminal
45. Bremerton Shipyard, Charleston Beach
Concentration (ng/g, dry weight)
Percent (%)
ACY
ACE
FLU
C1-FLU
C2-FLU
C3-FLU
DBT
C1-DBT
C2-DBT
C3-DBT
C4-DBT
PHN
C1-PHN/ANT
C2-PHN/ANT
C3-PHN/ANT
C4-PHN/ANT
ANT
FLA
PYR
C1-FLA/PYR
C2-FLA/PYR
C3-FLA/PYR
C4-FLA/PYR
BAA
CHR
C1-BAA/CHR
C2-BAA/CHR
C3-BAA/CHR
C4-BAA/CHR
BBF
B(JK)F
BEP
BAP
PER
IDP
DBA
B(GHI)P

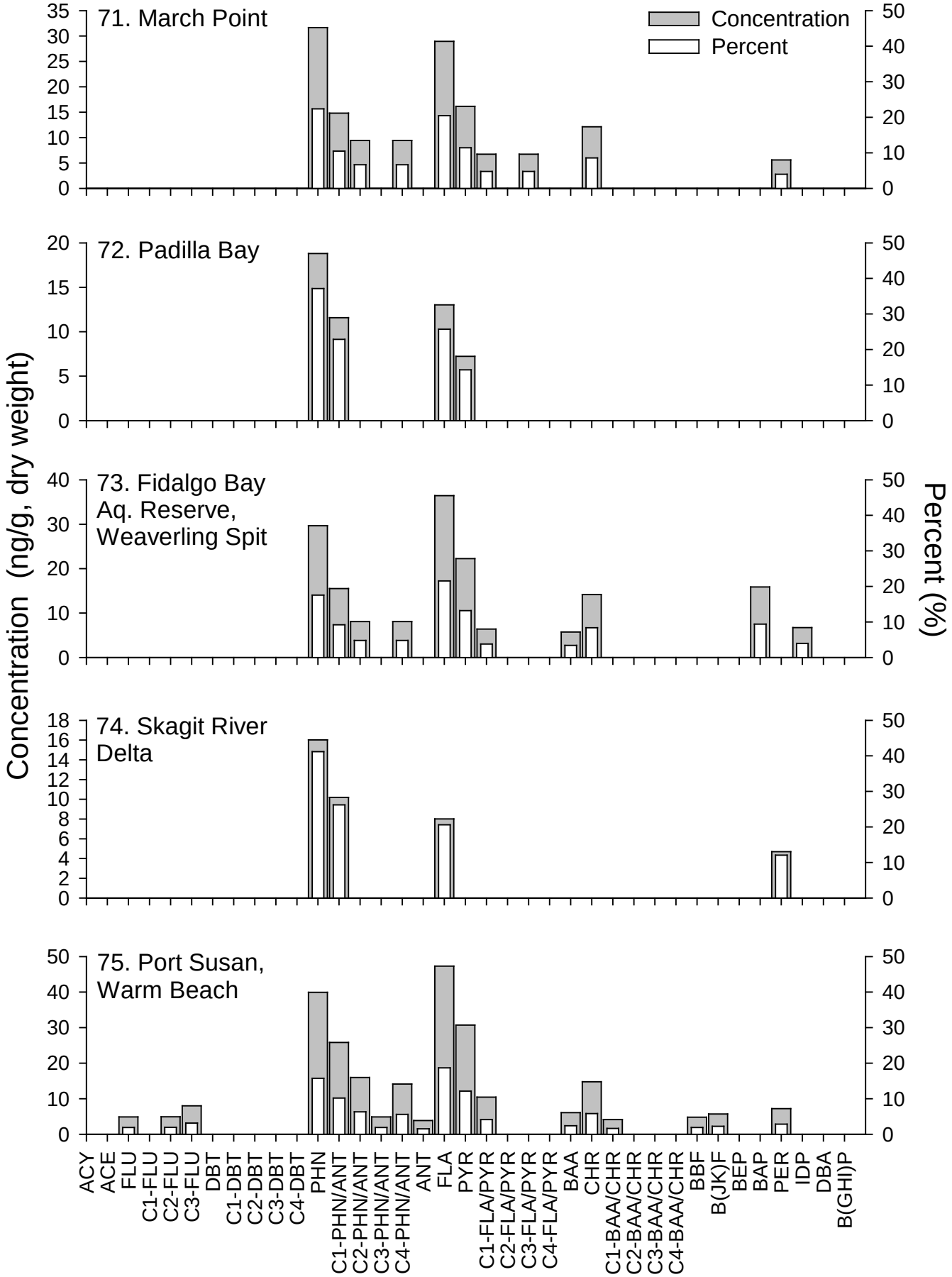

46. Manchester, Stormwater Outfall
Concentration
Percent
47. Sinclair Inlet, Sinclair Marina
48. Colvos Passage, Olalla, Prospect Point Beach
49. Case Inlet, Allyn
50. Shelton, Oak Bay Marina
Concentration (ng/g, dry weight)
Percent (%)
ACY
ACE
FLU
C1-FLU
C2-FLU
C3-FLU
DBT
C1-DBT
C2-DBT
C3-DBT
C4-DBT
PHN
C1-PHN/ANT
C2-PHN/ANT
C3-PHN/ANT
C4-PHN/ANT
ANT
FLA
PYR
C1-FLA/PYR
C2-FLA/PYR
C3-FLA/PYR
C4-FLA/PYR
BAA
CHR
C1-BAA/CHR
C2-BAA/CHR
C3-BAA/CHR
C4-BAA/CHR
BBF
B(JK)F
BEP
BAP
PER
IDP
DBA
B(GHI)P

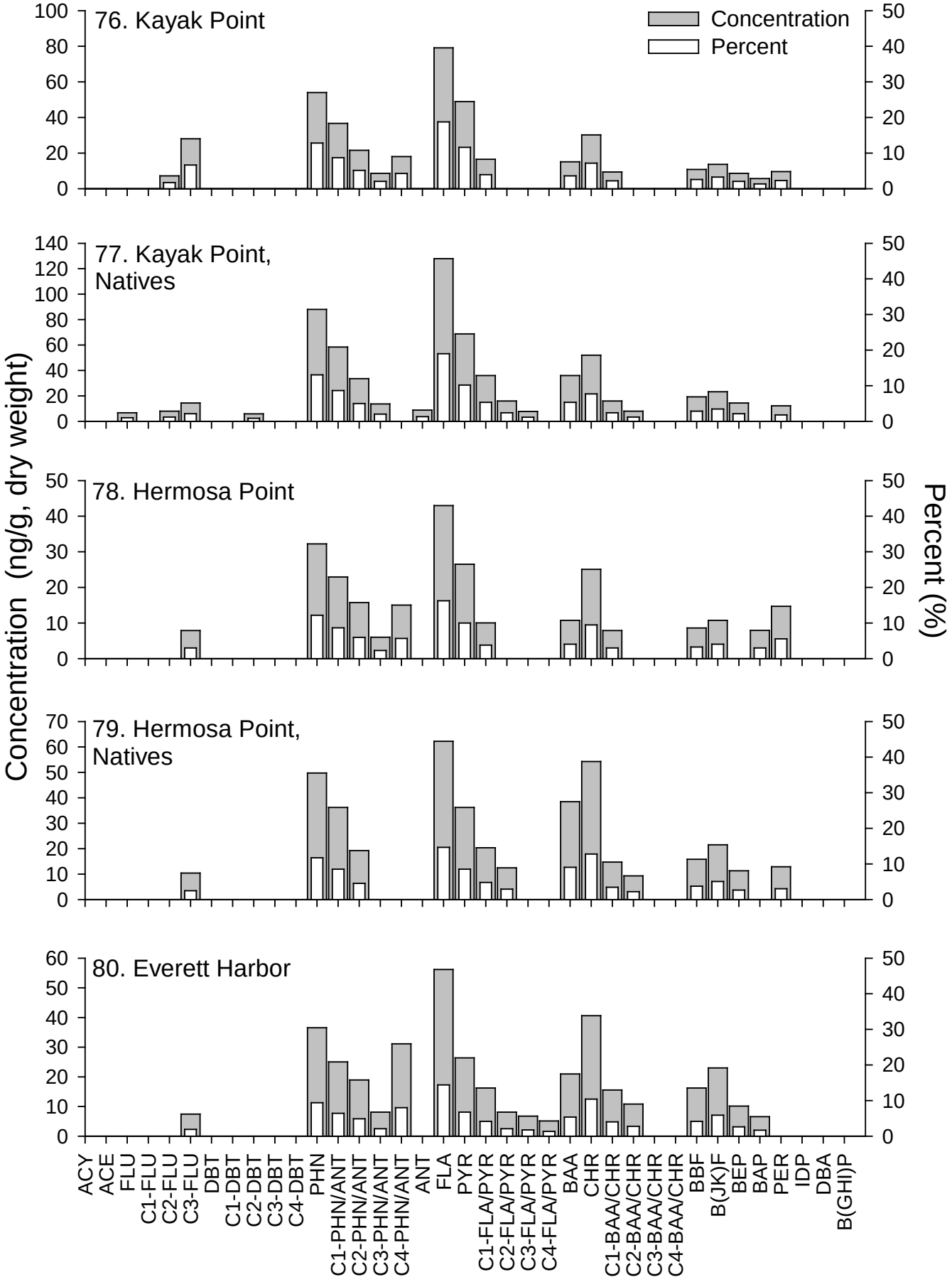

51. Totten Inlet
Concentration
Percent
52. Gig Harbor, Narrows Passage
53. Point Defiance Park
54. Kopachuck State Park
55. Commencement Bay, Skookum Wulge
Concentration (ng/g, dry weight)
Percent (%)
ACY
ACE
FLU
C1-FLU
C2-FLU
C3-FLU
DBT
C1-DBT
C2-DBT
C3-DBT
C4-DBT
PHN
C1-PHN/ANT
C2-PHN/ANT
C3-PHN/ANT
C4-PHN/ANT
ANT
FLA
PYR
C1-FLA/PYR
C2-FLA/PYR
C3-FLA/PYR
C4-FLA/PYR
BAA
CHR
C1-BAA/CHR
C2-BAA/CHR
C3-BAA/CHR
C4-BAA/CHR
BBF
B(JK)F
BEP
BAP
PER
IDP
DBA
B(GHI)P

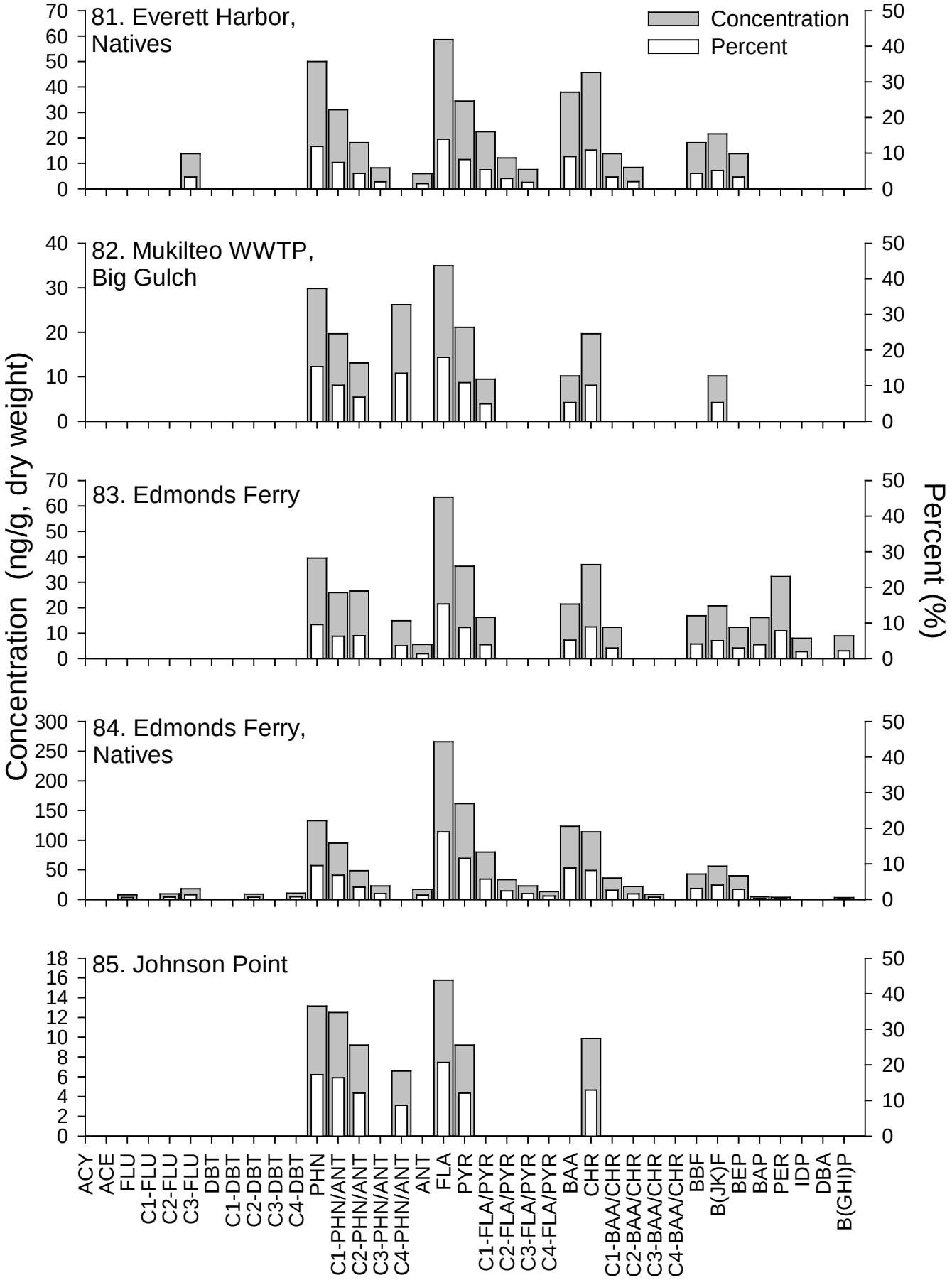

56. Tacoma, Ruston Way, Puget Creek
Concentration
Percent
57. Tacoma, Ruston Waterfront
58. Fox Island, Tanglewood Island
59. Hylebos Waterway
60. Hylebos Waterway, Natives
Concentration (ng/g, dry weight)
Percent (%)
ACY
ACE
FLU
C1-FLU
C2-FLU
C3-FLU
DBT
C1-DBT
C2-DBT
C3-DBT
C4-DBT
PHN
C1-PHN/ANT
C2-PHN/ANT
C3-PHN/ANT
C4-PHN/ANT
ANT
FLA
PYR
C1-FLA/PYR
C2-FLA/PYR
C3-FLA/PYR
C4-FLA/PYR
BAA
CHR
C1-BAA/CHR
C2-BAA/CHR
C3-BAA/CHR
C4-BAA/CHR
BBF
B(JK)F
BEP
BAP
PER
IDP
DBA
B(GHI)P

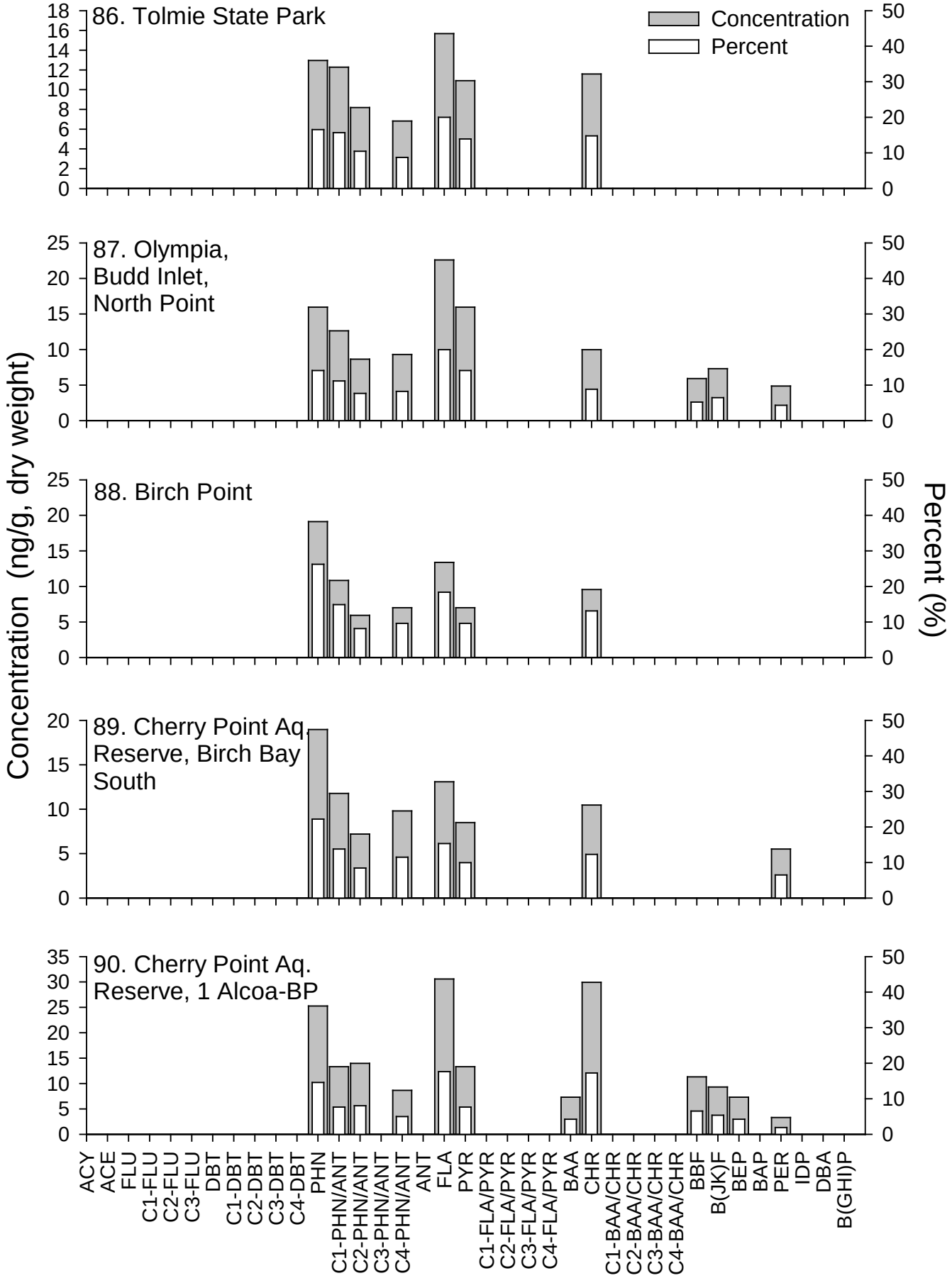

61. Thea Foss Waterway
Concentration
Percent
62. Steilacoom, Sunnyside Beach Park
63. Nisqually Reach Aq. Reserve, Anderson Island, Sandy Bay
64. North Shore, Orcas Island
65. Friday Harbor Labs, San Juan Island
Concentration (ng/g, dry weight)
Percent (%)
ACY
ACE
FLU
C1-FLU
C2-FLU
C3-FLU
DBT
C1-DBT
C2-DBT
C3-DBT
C4-DBT
PHN
C1-PHN/ANT
C2-PHN/ANT
C3-PHN/ANT
C4-PHN/ANT
ANT
FLA
PYR
C1-FLA/PYR
C2-FLA/PYR
C3-FLA/PYR
C4-FLA/PYR
BAA
CHR
C1-BAA/CHR
C2-BAA/CHR
C3-BAA/CHR
C4-BAA/CHR
BBF
B(JK)F
BEP
BAP
PER
IDP
DBA
B(GHI)P

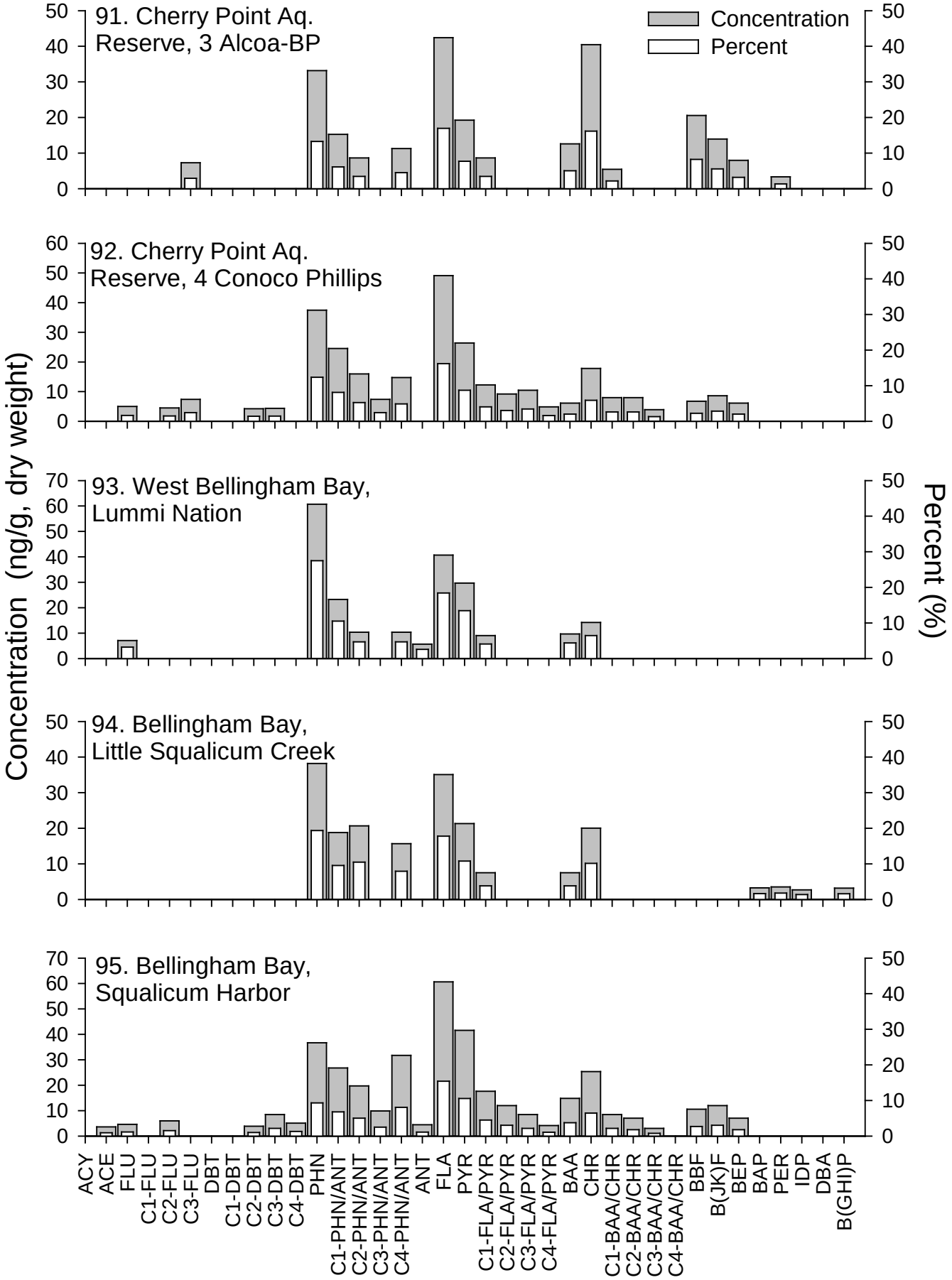

66. Fisherman's Bay, Weeks Wetland, Lopez Island
Concentration
Percent
67. Larrabee State Park
68. Cypress Island Aq. Reserve, Strawberry Bay
73%
69. Cypress Island Aq. Reserve, Secret Harbor
70. Anacortes, Guemes Ferry Terminal
Concentration (ng/g, dry weight)
Percent (%)
ACY
ACE
FLU
C1-FLU
C2-FLU
C3-FLU
DBT
C1-DBT
C2-DBT
C3-DBT
C4-DBT
PHN
C1-PHN/ANT
C2-PHN/ANT
C3-PHN/ANT
C4-PHN/ANT
ANT
FLA
PYR
C1-FLA/PYR
C2-FLA/PYR
C3-FLA/PYR
C4-FLA/PYR
BAA
CHR
C1-BAA/CHR
C2-BAA/CHR
C3-BAA/CHR
C4-BAA/CHR
BBF
B(JK)F
BEP
BAP
PER
IDP
DBA
B(GHI)P

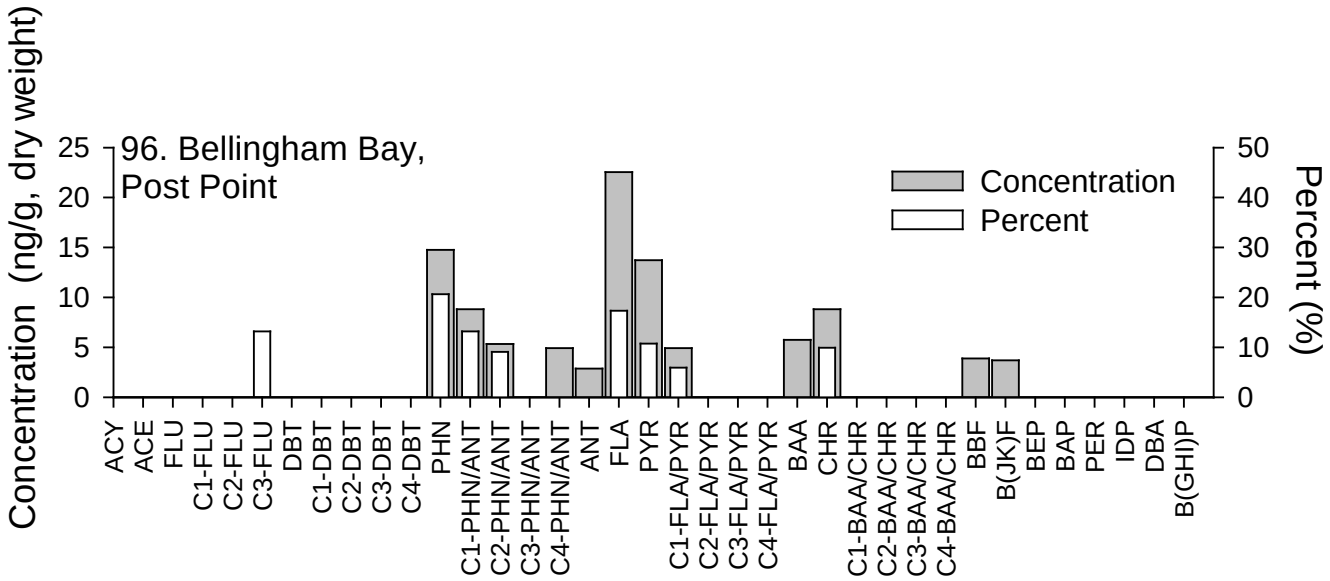

71. March Point
Concentration
Percent
72. Padilla Bay
73. Fidalgo Bay Aq. Reserve, Weaverling Spit
74. Skagit River Delta
75. Port Susan, Warm Beach
Concentration (ng/g, dry weight)
Percent (%)
ACY
ACE
FLU
C1-FLU
C2-FLU
C3-FLU
DBT
C1-DBT
C2-DBT
C3-DBT
C4-DBT
PHN
C1-PHN/ANT
C2-PHN/ANT
C3-PHN/ANT
C4-PHN/ANT
ANT
FLA
PYR
C1-FLA/PYR
C2-FLA/PYR
C3-FLA/PYR
C4-FLA/PYR
BAA
CHR
C1-BAA/CHR
C2-BAA/CHR
C3-BAA/CHR
C4-BAA/CHR
BBF
B(JK)F
BEP
BAP
PER
IDP
DBA
B(GHI)P


76. Kayak Point
Concentration
Percent
77. Kayak Point, Natives
78. Hermosa Point
79. Hermosa Point, Natives
80. Everett Harbor
Concentration (ng/g, dry weight)
Percent (%)
ACY
ACE
FLU
C1-FLU
C2-FLU
C3-FLU
DBT
C1-DBT
C2-DBT
C3-DBT
C4-DBT
PHN
C1-PHN/ANT
C2-PHN/ANT
C3-PHN/ANT
C4-PHN/ANT
ANT
FLA
PYR
C1-FLA/PYR
C2-FLA/PYR
C3-FLA/PYR
C4-FLA/PYR
BAA
CHR
C1-BAA/CHR
C2-BAA/CHR
C3-BAA/CHR
C4-BAA/CHR
BBF
B(JK)F
BEP
BAP
PER
IDP
DBA
B(GHI)P


81. Everett Harbor, Natives
Concentration
Percent
82. Mukilteo WWTP, Big Gulch
83. Edmonds Ferry
84. Edmonds Ferry, Natives
85. Johnson Point
Concentration (ng/g, dry weight)
Percent (%)
ACY
ACE
FLU
C1-FLU
C2-FLU
C3-FLU
DBT
C1-DBT
C2-DBT
C3-DBT
C4-DBT
PHN
C1-PHN/ANT
C2-PHN/ANT
C3-PHN/ANT
C4-PHN/ANT
ANT
FLA
PYR
C1-FLA/PYR
C2-FLA/PYR
C3-FLA/PYR
C4-FLA/PYR
BAA
CHR
C1-BAA/CHR
C2-BAA/CHR
C3-BAA/CHR
C4-BAA/CHR
BBF
B(JK)F
BEP
BAP
PER
IDP
DBA
B(GHI)P


86. Tolmie State Park
Concentration
Percent
87. Olympia, Budd Inlet, North Point
88. Birch Point
89. Cherry Point Aq. Reserve, Birch Bay South
90. Cherry Point Aq. Reserve, 1 Alcoa-BP
Concentration (ng/g, dry weight)
Percent (%)
ACY
ACE
FLU
C1-FLU
C2-FLU
C3-FLU
DBT
C1-DBT
C2-DBT
C3-DBT
C4-DBT
PHN
C1-PHN/ANT
C2-PHN/ANT
C3-PHN/ANT
C4-PHN/ANT
ANT
FLA
PYR
C1-FLA/PYR
C2-FLA/PYR
C3-FLA/PYR
C4-FLA/PYR
BAA
CHR
C1-BAA/CHR
C2-BAA/CHR
C3-BAA/CHR
C4-BAA/CHR
BBF
B(JK)F
BEP
BAP
PER
IDP
DBA
B(GHI)P


91. Cherry Point Aq. Reserve, 3 Alcoa-BP
Concentration
Percent
92. Cherry Point Aq. Reserve, 4 Conoco Phillips
93. West Bellingham Bay, Lummi Nation
94. Bellingham Bay, Little Squalicum Creek
95. Bellingham Bay, Squalicum Harbor
Concentration (ng/g, dry weight)
Percent (%)
ACY
ACE
FLU
C1-FLU
C2-FLU
C3-FLU
DBT
C1-DBT
C2-DBT
C3-DBT
C4-DBT
PHN
C1-PHN/ANT
C2-PHN/ANT
C3-PHN/ANT
C4-PHN/ANT
ANT
FLA
PYR
C1-FLA/PYR
C2-FLA/PYR
C3-FLA/PYR
C4-FLA/PYR
BAA
CHR
C1-BAA/CHR
C2-BAA/CHR
C3-BAA/CHR
C4-BAA/CHR
BBF
B(JK)F
BEP
BAP
PER
IDP
DBA
B(GHI)P


96. Bellingham Bay,
Post Point
Concentration
Percent
Concentration (ng/g, dry weight)
Percent (%)
0
5
10
15
20
25
0
10
20
30
40
50
ACY
ACE
FLU
C1-FLU
C2-FLU
C3-FLU
DBT
C1-DBT
C2-DBT
C3-DBT
C4-DBT
PHN
C1-PHN/ANT
C2-PHN/ANT
C3-PHN/ANT
C4-PHN/ANT
ANT
FLA
PYR
C1-FLA/PYR
C2-FLA/PYR
C3-FLA/PYR
C4-FLA/PYR
BAA
CHR
C1-BAA/CHR
C2-BAA/CHR
C3-BAA/CHR
C4-BAA/CHR
BBF
B(JK)F
BEP
BAP
PER
IDP
DBA
B(GHI)P