2017 West Point Flooding Event: English Sole Tissue Monitoring Final Report

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King County

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2017 West Point Flooding Event: English Sole Tissue Monitoring Final Report

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Washington Department of FISH and WILDLIFE

Acknowledgements

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Citation

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EXECUTIVE SUMMARY

What happened at West Point?

The West Point Treatment Plant (West Point) experienced equipment failure in the winter of 2017. The resulting flooding on February 9, 2017 severely damaged the plant's mechanical and electrical systems. To prevent further damage, West Point discharged 244 million gallons of untreated stormwater and sewage to Puget Sound through a shallow-water emergency bypass outfall (EBO) during two separate events on February 9 and 15–16, 2017. The bypasses contained approximately 85–90% stormwater and 10–15% wastewater. West Point operated with reduced treatment until secondary treatment processes were restored on April 27, 2017. Thereafter, all effluent discharged from West Point received full secondary treatment and on May 10, 2017, West Point began meeting all National Pollutant Discharge Elimination System permit limits. During the facility restoration process, the Wastewater Treatment Division (WTD) carefully managed inflows to West Point during storm events to protect recovery of the biological treatment processes and prevent further damage to the plant. To reduce flows conveyed to West Point, WTD diverted inflows to three King County wet weather treatment facilities: Alki, Carkeek, and Elliott West. Following the initial overflow event, King County mobilized to evaluate the potential impacts of the emergency discharge and period of reduced treatment on the marine environment, leading to a series of monitoring events and reports, including this report.

What is this report about?

This document is one of seven technical reports that collectively evaluate the potential impacts of the West Point flooding event on Puget Sound water quality, subtidal and intertidal sediments, clams, zooplankton, crab, and English sole. Key findings will be synthesized in a final summary report. This report follows an earlier one (King County, 2021) which summarized 2017 English sole (*Parophrys vetulus*) muscle (i.e., fillet) chemistry data. This current report presents both 2017 and 2019 fillet tissue chemistry and biomarker[1] results for English sole collected as part of ongoing King County and Washington Department of Fish and Wildlife (WDFW) marine fish monitoring surveys. This report compares patterns in space and time using data from samples collected at King County stations in 2015 (i.e., pre-event data), 2017 (i.e., post-event data, year 0), and 2019 (i.e., year 2), and some other areas of the Puget Sound.

The key research questions driving the English sole tissue chemistry monitoring were:

1) Did proximity to the point of discharge from the West Point flooding event lead to higher chemical concentrations in English sole tissues? In other words, did fish sampled from West Point N and Myrtle Edwards in 2017 (post-event) have higher concentrations than fish sampled further away, or than fish from these locations before and 2 years after the event?

[1] 2019 PAH-metabolite and estrogen biomarker data not included in this report due to data processing delays.

2) Do biomarkers in English sole nearest the event discharge points indicate higher exposure to endocrine disrupting chemicals or PAHs that may be associated with the flooding event?
3) Did changes in fish tissue chemical concentrations following the flooding event result in any new exceedances of Washington Department of Health screening levels for seafood consumption advisories (i.e., would allowable levels of fish consumption decrease due to increased contaminant concentrations in fish tissue)?

To evaluate these questions, the available English sole tissue samples were analyzed for signs of exposure and chemical bioaccumulation and compared to data collected prior to the flooding event and data from areas not likely to have been directly affected by effluents from the West Point flooding event.

Did the West Point flooding event affect English sole tissue chemistry?
Puget Sound tissue monitoring programs are designed to track long-term trends in contamination from all the sources influencing water quality in an area (i.e., treated wastewater, wet weather treatment stations, combined sewer overflows, stormwater, rivers). Several confounding factors, including other contaminant sources, increased loadings from heavier than normal rainfall, and substantial dispersion of effluent in receiving water, make it difficult to definitively link changes in contaminant or biomarker levels related in part or exclusively to the West Point flooding event.

Within that frame, our analysis of English sole tissue data from this study did not show statistically higher metals, xenoestrogens, or PAH-metabolites after the flooding event (Year 0 or Year 2). The flooding event did not increase metals exposure in English sole beyond levels experienced by fish in other areas of King County, i.e., Shilshole, Alki or Quartermaster.

The West Point flooding event may have resulted in increased English sole exposure to some organic chemicals near the West Point outfall or the Elliott West wet weather treatment station in 2017, compared to exposures in 2015 and 2019. However, chemical concentrations from samples collected near the West Point and Elliott West outfalls were statistically similar to those in samples from one or more other urban areas in Puget Sound. Though vitellogenin (a biomarker for exposure to estrogenic compounds) data suggest environmental estrogens may have been higher at West Point N in 2017 than 2019, the lack of pre-event data and presence of a similar change from 2017 to 2019 across many urban and nonurban sites make it unclear whether this reflects influence from the flooding event, from the relatively high rainfall (stormwater) volumes in 2017, or some other factor.

Would Washington State Department of Health fish consumption advisories for people likely have changed because of the event?
We believe the English sole data collected following the West Point flooding event indicate there would be no changes to the existing seafood consumption advisory for flatfish in the areas near West Point. The Washington State Department of Health (WDOH) issues seafood consumption advisories to inform the public when concentrations of toxic chemicals in

seafood pose a health risk to people eating them. Several advisories that recommended people limit consumption of seafood from Puget Sound were in place before the West Point flooding event. Contaminants measured in English sole were compared to WDOH screening levels, which are used as guidelines to evaluate whether concentrations are high enough to warrant a change in seafood consumption advisories. Based on this comparison, we believe the West Point flooding event would result in no changes to existing Puget Sound seafood consumption advisories for English sole.

What were the overall findings?

Spatial and temporal analysis of the muscle chemistry and bile results suggest that concentrations of metals, xenoestrogens, and PAH-metabolites in English sole collected near West Point and the Elliott West wet weather treatment station were either not impacted by the West Point flooding event or were impacted to a small enough degree to be undetectable in our monitoring program. When considered in the context of spatial and temporal variation seen at other nearby stations, the data indicate the West Point flooding event by itself did not substantially change exposures of English sole to organic chemicals near West Point and the Elliott West wet weather treatment station. Though vitellogenin was detected in male fish near West Point, the limited amount of available data and high variability seen at other Puget Sound locations makes it unclear whether these differences reflect influence of the flooding event or pre-existing patterns at that location.

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1.0 INTRODUCTION

The West Point Treatment Plant (West Point) experienced equipment failure in the winter of 2017 (West Point flooding event). The resulting flooding on February 9, 2017 severely damaged the plant's mechanical and electrical systems. To prevent further damage, West Point discharged 244 million gallons of untreated stormwater and sewage through a shallow-water emergency bypass outfall (EBO). Following the initial overflow event, King County mobilized to evaluate the potential impacts of the emergency discharge and period of reduced treatment on the marine environment. This evaluation included collection of English sole (*Parophrys vetulus*) fillet, bile and blood plasma in 2017 and 2019 for chemistry and biomarker analyses. An earlier report (King County, 2021) presented the evaluation and findings from the 2017 English sole fillet chemistry analysis. This current report presents the 2017 and 2019 fillet tissue chemistry, vitellogenin in blood plasma for English sole, and the polycyclic aromatic hydrocarbon (PAH)-metabolites and xenoestrogens in bile results from 2017. The remaining biomarker results were delayed by COVID-19 and will ultimately be reported in an addendum.

The study questions were to determine:

1) whether proximity to effluent from the West Point flooding event led to increased chemical concentrations in English sole tissues.
2) whether biomarkers in fish nearest the event discharge points indicated increased exposure to endocrine disrupting chemicals or PAHs that may be associated with the event.
3) if any changes in fish tissue chemical concentrations following the flooding event resulted in greater exceedances of Washington Department of Health screening levels for seafood consumption advisories than currently exist.

In addition to evaluation of these questions, this report serves to meet the reporting requirements of King County and WDFW's 2017 and 2019 English sole monitoring program, by presenting summary information on the findings of those monitoring events.

1.1 Study Approach

King County and WDFW collaborate to conduct long-term monitoring of chemicals in English sole fillet tissue on a biannual basis. Chemicals monitored in English sole fillet for this program are:

- Metals
- Polychlorinated biphenyls (PCBs)
- Polybrominated diphenyl ethers (PBDEs)
- Dichlorodiphenyltrichloroethanes (DDT), its breakdown products, and other chlorinated pesticides.

WDFW's monitoring program also includes biomarkers of exposure to polycyclic aromatic hydrocarbons (PAHs) in fish bile and endocrine disrupters in fish blood.

Following the West Point flooding event, King County worked with WDFW to modify designs of their respective English sole monitoring programs; the team sought to evaluate the potential for changes in English sole exposures to these chemicals resulting from the West Point flooding event, to address the study questions listed above. The two agencies added sampling stations to represent conditions near the West Point outfall and sampled at these and at locations already included in their joint English sole monitoring program in 2017 (several months after the flooding event) and 2019 (2 years after event). Analyses of the chemistry results for English sole fillet tissue from 2015 (from pre-event monitoring), 2017 and 2019, including statistical comparisons between locations and over time, were conducted to address the study questions.

The data and sampling approaches for a long-term monitoring program, particularly for fish tissue, are not well-suited to evaluations of cause-effect relationships like those implicit in the study questions. A precipitation event like the one that triggered this evaluation increases the variability of chemical parameters in tissue and other environmental media throughout the region, not just near outfalls and CSOs affected by the West Point flooding event. Because of this variability, only a very strong response can be detected given the relatively few samples per location of the monitoring program. Our ability to identify and quantify any changes in fish tissue chemistry is limited by small sample sizes.

Additional detail on the scope of the sampling and data sharing between King County and WDFW, along with other methodological details, is presented in Section 3.

1.2 Report Organization

A summary of the flooding event and all the associated monitoring events is presented in Section 2. The remainder of the report summarizes the English sole sampling and analysis efforts (Section 3), a spatial and temporal comparison with other monitoring data to provide context (Section 4), a comparison to Washington Department of Health (WDOH) screening values (Section 5), and a summary of findings (Section 6).

2.0 PROJECT BACKGROUND

This section summarizes the West Point 2017 flooding event and associated monitoring response conducted by King County. A more detailed description of the monitoring response and water quality in Puget Sound's Central Basin waters is provided in the *West Point Flooding Event Water Quality Summary Report* (King County, 2018).

2.1 West Point Treatment Plant Characteristics

King County's West Point Wastewater Treatment Plant (West Point) is located near the west side of Magnolia Bluff, adjacent to Seattle's Discovery Park (Figure 1). This regional treatment plant serves a combined system that receives both municipal wastewater and stormwater. The plant began providing primary wastewater treatment in 1966 and was upgraded to provide secondary treatment in late 1995. The average annual secondary treatment volume of the plant is 95 million gallons per day (MGD). The average wet-weather flow is 133 MGD and the plant has a peak wet-weather capacity of 440 MGD. Secondary treatment at West Point consists of screening, grit removal, primary sedimentation, air-activated sludge, secondary sedimentation, disinfection by chlorination, and anaerobic digestion of solids. Secondary treated effluent is dechlorinated prior to discharge.

Treated effluent from West Point is discharged to the Central Basin of Puget Sound. Effluent is discharged through a marine outfall point approximately 3,600 feet (ft) (914 meters [m]) offshore to the west of West Point at a bottom depth of -240 ft (-73 m) referenced to mean lower low water (MLLW). Effluent exits the outfall through a multi-port diffuser that spans 610 ft (186 m) pipe. The diffuser produces rapid mixing of effluent with seawater. In addition to the main outfall, the plant has an emergency bypass outfall (EB0) located about 525 ft (160 m) offshore on the north side of West Point (see Figure 1). The discharge point is at the bottom at an approximate water depth of -40 ft (-12 m) MLLW.

2.2 West Point 2017 Flooding Event

Early in the morning on February 9, 2017, equipment failure and subsequent flooding of West Point during peak inflows resulted in an emergency bypass event. This bypass lasted approximately 18 hours and resulted in the release of 186 MG of untreated stormwater and wastewater into Puget Sound through the EBO. A smaller bypass event that occurred over the course of February 15 and 16 resulted in 58 MG of untreated discharge through the EBO. In total, about 244 MG of untreated flows were discharged via the EBO in February 2017.

The discharge of 244 MG of untreated flows from the EBO was higher than the total untreated discharges for any February for the previous 10 years from all combined sewer overflow (CSO) outfalls. The average untreated discharge for February (2007-2016) from all of the CSOs in the King County system was 52 MG. During this 10-year period, February

Figure 1. Location of the West Point Wastewater Treatment Plant and other outfalls.

CSO discharge volumes ranged from none for several years to a maximum of 214 MG in 2014.

The total untreated discharge from all King County CSOs in February 2017 increased substantially relative to the previous 10 years to 749 MG. February 2017 untreated CSO discharges increased due to the combination of record rainfall and the approach used to manage flows to West Point following the flooding event. The total annual untreated CSO discharge of 1.7 billion gallons in 2017 exceeded the average annual CSO volume of 918 MG for the last ten years.

Following the February 9 flooding event, West Point operated using reduced treatment while efforts to restore secondary treatment processes were underway. This reduced treatment included some solids settling, screening, disinfection, and dechlorination. The flooding event severely damaged the mechanical and electrical systems necessary to provide heat to the secondary system biological treatment, which essentially crippled West Point's ability to effectively remove the majority of incoming solids. During the restoration process, inflows to West Point during storm events were carefully managed to protect the recovery of the biological treatment processes and prevent further damage to the plant. King County relied on three wet weather treatment facilities (Alki, Carkeek, and Elliott West) to reduce flows conveyed to West Point. Some additional flows during storm events were also routed to King County's Brightwater and the City of Edmonds wastewater treatment plants. In addition, untreated overflows from the combined system were exacerbated due to reduced operations at West Point, both during the emergency bypass events and during other large storm events. It is not possible to estimate the volume of additional CSO discharges that resulted while West Point repairs were underway relative to the volume of discharges that would have occurred during heavy rainfall events had the plant been fully operational.

Restoration of West Point's primary and secondary treatment processes was completed by the end of April 2017 and all wastewater received full secondary treatment beginning April 27. However, recovery of the solids handling processes was still ongoing, which resulted in discharge of higher levels of suspended solids than normal. These higher levels of discharge affected West Point's ability to consistently meet its National Pollutant Discharge Elimination System (NPDES) permit limits for total suspended solids (TSS), carbonaceous biochemical oxygen demand (CBOD), and residual chlorine through May 9. In addition, from late March through mid-June, recovery of the solids handling processes was partially managed by using trucks to transport a portion of the solids produced at West Point to South Plant for additional treatment. The additional solids treatment at South Plant did not affect the ability of the plant to meet its NPDES permit requirements but did result in an increase (approximately 10%) in effluent ammonia levels at South Plant compared to typical concentration. West Point began meeting all NPDES permit limits on May 10, 2017. The current NPDES permit can be accessed at http://www.kingcounty.gov/depts/dnrp/wtd/system/npdes.aspx.

2.3 Response Monitoring

Less than eight hours after the emergency bypass began at West Point on February 9, King County posted warning signs and closed nearby beaches as a precautionary measure. Fecal indicator bacteria samples were collected from four beaches in the vicinity of West Point for 13 consecutive days, with the exception of February 14 at Carkeek Park and Golden Gardens. Sampling ended on February 21 when bacteria levels were safe for water contact and beaches re-opened.

Following an initial sampling response to monitor fecal indicator bacteria at nearby beaches, King County developed and implemented a monitoring plan to conduct additional sampling. Sampling beyond existing effluent NPDES permit requirements was included as well as receiving waters monitoring outside of the long-term Marine Water Quality Monitoring Program. The objectives of these sampling efforts were to:

- assess West Point effluent quality over time as repairs were made to the plant,
- evaluate any observed changes in West Point effluent quality in context of historical conditions,
- assess potential short-term changes to Puget Sound receiving waters following untreated discharges and during the period of reduced treatment,
- compare receiving water results to applicable Washington State Water Quality Standards for Marine Surface Waters and historical conditions, and
- assess potential for any effects on biological and sediment quality.

The first four objectives above were addressed in the West Point Flooding Event Water Quality Summary Report (King County, 2018). Sampling results presented in this report are related to the last objective.

Subtidal and intertidal sediments as well as butter clam, Dungeness crab, zooplankton, and English sole tissues were collected for analysis of chemical constituents, and benthic infauna abundance and community structure were assessed. Results from these sampling efforts were presented in separate reports and will be presented in context of each other and water quality data in a final West Point flooding event summary report. An earlier report summarized the 2017 English sole (*Parophrys vetulus*) fillet chemistry data (King County, 2021). This report includes a spatial and temporal analysis of English sole data from 2015 (when available), 2017, and 2019.

2.3.1 Effluent Monitoring

King County conducts routine effluent monitoring as required by the NPDES permit for each wastewater treatment facility. Samples analyzed for priority pollutant metals and organic chemicals are typically collected twice per year, once during the wet season and once during the dry season. However, 12 additional effluent samples were collected from February 9 to June 30, 2017 for the analysis of metals, while 7 samples were analyzed for organic chemicals. Acute and chronic toxicity tests were also conducted in March and April while the plant was being restored. Results from these sampling efforts can be found in *West Point Flooding Event Water Quality Summary Report* (King County, 2018) and are summarized in Section 2.4.

2.3.2 Puget Sound Central Basin Monitoring

King County collected and analyzed surface waters, sediments, and tissues from four types of marine organisms as part of the response monitoring to the West Point flooding event. A summary of the types of monitoring conducted is provided below and more detail is provided in *West Point Flooding Event Water Quality Summary Report* (King County, 2018).

2.3.2.1 Surface Water

During the West Point restoration period, water quality monitoring of Puget Sound (e.g., bacteria, physical parameters, and nutrients) was expanded beyond the existing routine monthly/bimonthly monitoring to assess potential changes in water quality. The routine long-term monitoring program helps provide an understanding of water quality within the Puget Sound Central Basin (see 2017 annual work plan [King County, 2016b]). Additional monitoring during the restoration period included:

- increased sampling frequency from bimonthly to weekly at a subset of four offshore long-term monitoring stations,
- the addition of a new (fifth) offshore monitoring station sampled weekly near the EBO,
- increased sampling and analysis frequency for bacteria from monthly to weekly at a subset of six beach stations,
- expanded nitrate monitoring in the water column at all offshore stations using a Submersible Ultraviolet Nitrate Analyzer (SUNA), and
- the measurement of metal concentrations in the water column at four stations.

2.3.2.2 Sediment

King County also collected and analyzed marine sediments and organisms near West Point's main outfall to identify potential adverse effects to sediment-dwelling organisms. These sediment monitoring efforts are detailed in the associated Sampling and Analysis Plans (SAPs) (King County, 2017a,b,c). A remotely operated vehicle[2] inspection of subtidal sediments near the EBO was also conducted shortly after the second release event to look for any indication of deposition from the release. Since no evidence was found, the County modeled the outfall discharges to determine the potential for the release to create any sediment quality concerns. Results of the intertidal and subtidal sediment monitoring efforts are presented in separate data reports and will be finalized along with other monitoring efforts in a final West Point flooding event summary report.

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[2] Remotely operated vehicle (ROV) is unoccupied, highly maneuverable underwater vehicle operated by someone at the water surface.

2.3.2.3 Tissue

King County collected marine organism tissues in the Central Basin of Puget Sound for chemical analysis to evaluate if discharges during the incident were associated with an increase in tissue contaminant concentrations. These sampling efforts include collection of butter clams, Dungeness crab, zooplankton, and English sole (described in King County, 2021 and in this report). The sampling efforts are detailed in project SAPs (King County, 2017a,d,e). The results of the other tissue monitoring efforts are presented in separate data reports and will be presented in context of each other and water quality data in a final West Point flooding event summary report.

2.4 Summary of Water Quality Results

The results of effluent and receiving water monitoring during the first half of 2017 were presented as part of the *West Point Flooding Event Water Quality Summary Report* (King County, 2018). The February 9 flooding of West Point resulted in changes to effluent characteristics from February 9 through May 9. The largest impact observed was an increase in bacteria levels at Seattle area beaches following the two untreated emergency discharge events in February. No other substantial water quality impacts were observed near the West Point outfall.

Increased loadings of effluent metals to Puget Sound during the period of reduced treatment did not appear to measurably affect water column concentrations. Given these monitoring results and observations, effluent discharged during the period of reduced treatment did not result in observable exceedances in marine water quality standards in receiving waters, which are intended to be protective of aquatic life.

2.5 Tidal Currents at the West Point Outfall

Tidal currents near the West Point outfall are a factor that influence how effluent is transported within, and out of, Puget Sound. Understanding tidal currents near the West Point outfall allows for an assessment of the potential for biological exposures at different locations. Tidal currents in the Central Basin average about 50 centimeters per second (cm/s). Typical tidal dispersion of the West Point effluent from the main outfall is depicted in Figure 2. Estuarine circulation is important for transporting water masses and is typically up to about 10 cm/s but can be higher during storms and bottom saltwater intrusion from Admiralty Inlet (King County, 2009). Mixing occurs near the outfall due to a combination of density differences, tidal currents, and the momentum of the discharge through diffusers. Currents may affect physical properties of sediment around the outfall, as well as effluent transport.

Currents were previously assessed in the vicinity of the West Point outfall for a five-week period beginning in February 2003. Current meters were deployed at multiple depths and locations were chosen to measure both nearshore and deep-water currents that may affect effluent transport. Results showed that tidal currents along the parallel transect aligned with the outfall flowed predominantly in the southwest/northeast directions, corresponding to the semi-diurnal tides. In addition, a clockwise eddy can form to the north of West Point during ebb tides, recirculating some water masses (Lincoln, 1976; King County, 2005). Tidal currents at an offshore station west of the outfall flowed in a more north/south direction. Currents at depths of 100-m and greater had a wider distribution of direction and aligned more towards the southwest/northeast than the currents at shallower depths. The 90th percentile current speeds ranged between 30 and 50 cm/s, including at depths greater than 100-m. Current direction is influenced by the topography of West Point as the shoreline is approached. A detailed description of the methods and results are provided in West Point Treatment Plant Marine Outfall Current Meter Analysis (King County, 2005).

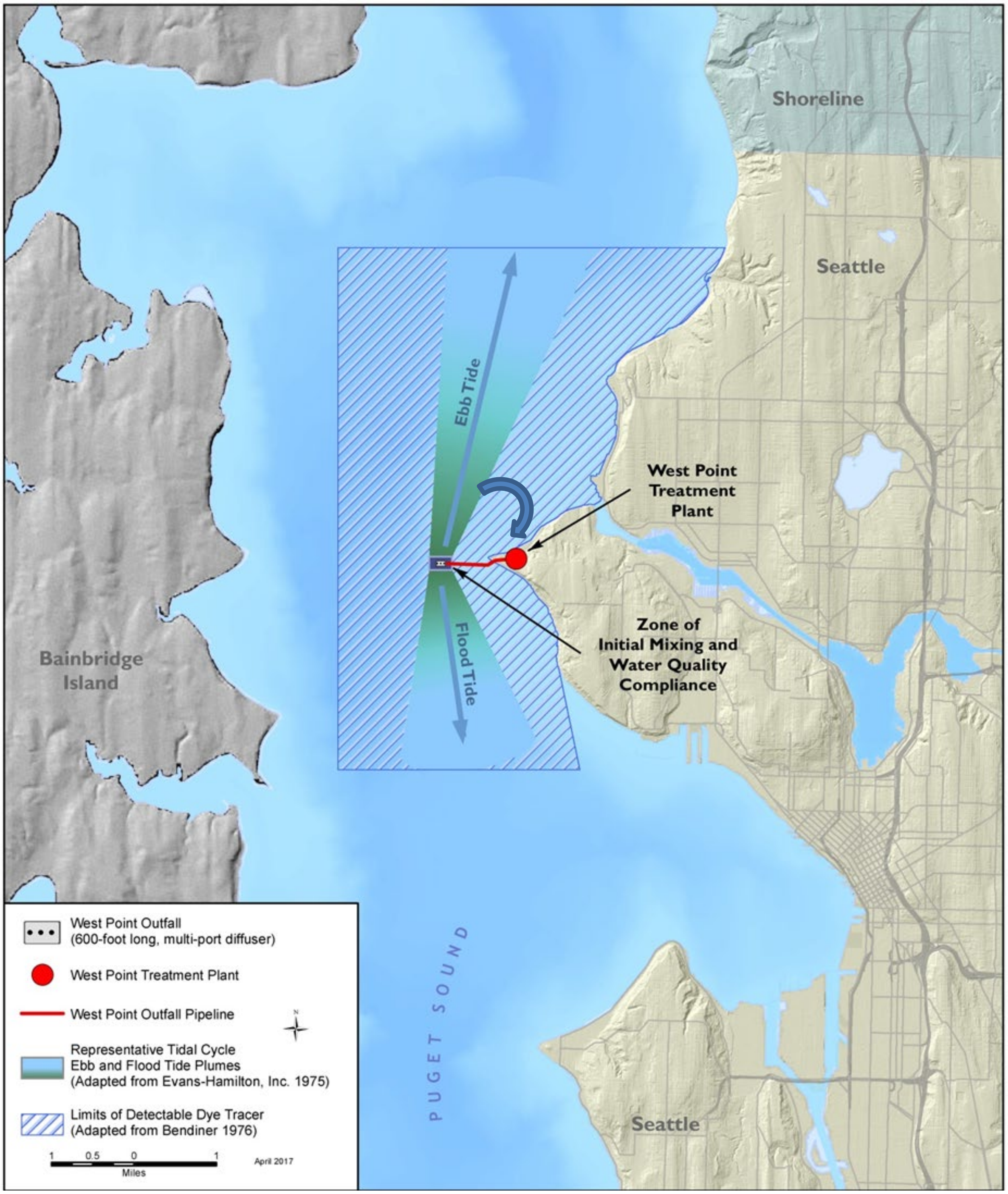

Figure 2. Typical dispersion in the winter and spring of the West Point treatment plant effluent over a tidal cycle. The blue box shows the extent of detectable dye tracer released from the main outfall in prior tracing studies (Bendliner, 1976). A clockwise eddy forming to the north of West Point has been observed in current data during ebb tides as well (Lincoln, 1976; King County, 2005). (Source: King County Dept. of Natural Resources, Wastewater Treatment Division)

3.0 METHODS

King County and WDFW adapted their existing English sole monitoring programs to conduct a joint program and address the study questions. The program was modified and executed as described below to evaluate the study questions identified in Section 1. The methods used in the modified monitoring program were consistent with existing monitoring programs but included additional sampling locations and analytes. This section presents the design used to collect the data analyzed for this report, including sampling locations, analytes, field sampling methods, sample processing, and the laboratory methods used to analyze the tissue samples. Results of quality assurance (QA) review of chemistry data and a brief description of deviations from the SAP are in Appendix A. Methods for data handling and data analysis are also summarized.

3.1 Monitoring Design

Sampling and analytical methods followed those described in King County's long-term marine tissue monitoring program SAP (King County, 2015) and the 2019 SAP addendum (King County, 2019), unless otherwise specified. Information on sampling performed for WDFW's TBiOS program is also presented in this section, because data from the TBiOS program is presented and analyzed to meet the reporting objective (Section 1). Within the collaborative program, individual samples of English sole fillet (as a composite of fillet tissue from up to 20 individuals) may be analyzed for the same constituent in two different laboratories or analyzed for different constituents (Table 1). Historically, WDFW's TBiOs program has composited multiple fish within individual samples as a cost-effective way of reducing variability in the data. King County's monitoring program follows the same protocols to ensure comparability between the two programs. The TBiOS program includes biomarker samples as well.

3.1.1 Sampling Locations

The King County and WDFW English sole monitoring locations, including modifications to address the study questions (Section 1), are as follows:

- King County monitors four stations in Elliott Bay, one station in Shilshole Bay, and one station in Quartermaster Harbor (Table 1).
- WDFW currently monitors 11 stations across Puget Sound as part of its Toxics in Biological Observation System (TBiOS) program (Table 1). WDFW's stations include one near Kellogg Island in the Duwamish River, one at Pier 62 in downtown Seattle (shared station with King County).
    - In 2017, WDFW added a new station in Elliott Bay, located off Myrtle Edwards Park. The Myrtle Edwards station had historically been included in both WDFW and King County programs (Figure 3).
    - Other WDFW stations are outside of Elliott Bay and do not overlap King County's monitoring stations.

- Following the West Point flooding event, King County and WDFW modified the monitoring design to include two new stations just north and south of the West Point outfall (West Point N and S). Samples at these locations were collected by WDFW (Figure 3).

All of the WDFW and King County monitoring stations (Figure 4) were sampled in May or early June of 2017, and all but the West Point S[3] monitoring station were resampled in June of 2019.

Table 1. Locations sampled, collectors, and laboratories used for 2017 and 2019 datasets

<table>
<tr><th rowspan="2">Station</th><th rowspan="2">Sampled By</th><th colspan="6">Analyzed By</th></tr>
<tr><th>Metals</th><th>PCBs, PBDEs, OC Pesticides</th><th>Stable Isotopes</th><th>Xeno-estrogens</th><th>PAHs[c]</th><th>Vtg</th></tr>
<tr><th></th><th></th><th>n=8</th><th>n=9</th><th>n=5</th><th>n=13</th><th>n=13</th><th>n=14</th></tr>
<tr><td>Shilshole</td><td rowspan="4">KC</td><td rowspan="8">KC</td><td rowspan="4">KCEL</td><td rowspan="4">NA</td><td rowspan="4">NA</td><td rowspan="4">NA</td><td rowspan="4">NA</td></tr>
<tr><td>Harbor Island[a]</td></tr>
<tr><td>Alki</td></tr>
<tr><td>Quartermaster Harbor</td></tr>
<tr><td>Myrtle Edwards</td><td>KC & WDFW</td><td rowspan="4">Both KCEL and NOAA</td><td rowspan="13">NOAA</td><td rowspan="13">NOAA</td><td rowspan="13">NOAA</td><td rowspan="14">NOAA</td></tr>
<tr><td>Pier 62</td><td rowspan="13">WDFW</td></tr>
<tr><td>West Point N</td></tr>
<tr><td>West Point S[a]</td></tr>
<tr><td>Duwamish</td><td rowspan="10">NA</td><td>NOAA</td></tr>
<tr><td>Sinclair Inlet</td><td rowspan="9">NU</td></tr>
<tr><td>Port Gardner</td></tr>
<tr><td>Vendovi</td></tr>
<tr><td>Strait of Georgia</td></tr>
<tr><td>Eagle Harbor</td></tr>
<tr><td>Commencement Bay</td></tr>
<tr><td>Hood Canal</td></tr>
<tr><td>Nisqually Bay</td></tr>
<tr><td>Port Madison[b]</td><td>NA</td><td>NA</td><td>NA</td></tr>
</table>

Vtg = vitellogenin; KC = King County; WDFW = Washington Department of Fish and Wildlife;
KCEL – King County Environmental Lab; NOAA – National Oceanic and Atmospheric Administration;
"Split" indicates both WDFW and KC analyzed the chemicals in the same sample by splitting homogenates into two fractions;
NA = not analyzed; NU = not used in the context of this study;
[a]Harbor Island and West Point S sampled only in 2017
[b]Port Madison sampled only in 2019
[c]PAH-metabolites

[3] The West Point S station was not resampled in 2019 due to budget constraints and because the 2017 data indicated the West Point flooding event likely did not affect contaminant levels in English sole from this location (King County, 2021).

Figure 3. Closest English sole sampling locations to the West Point Treatment Plant

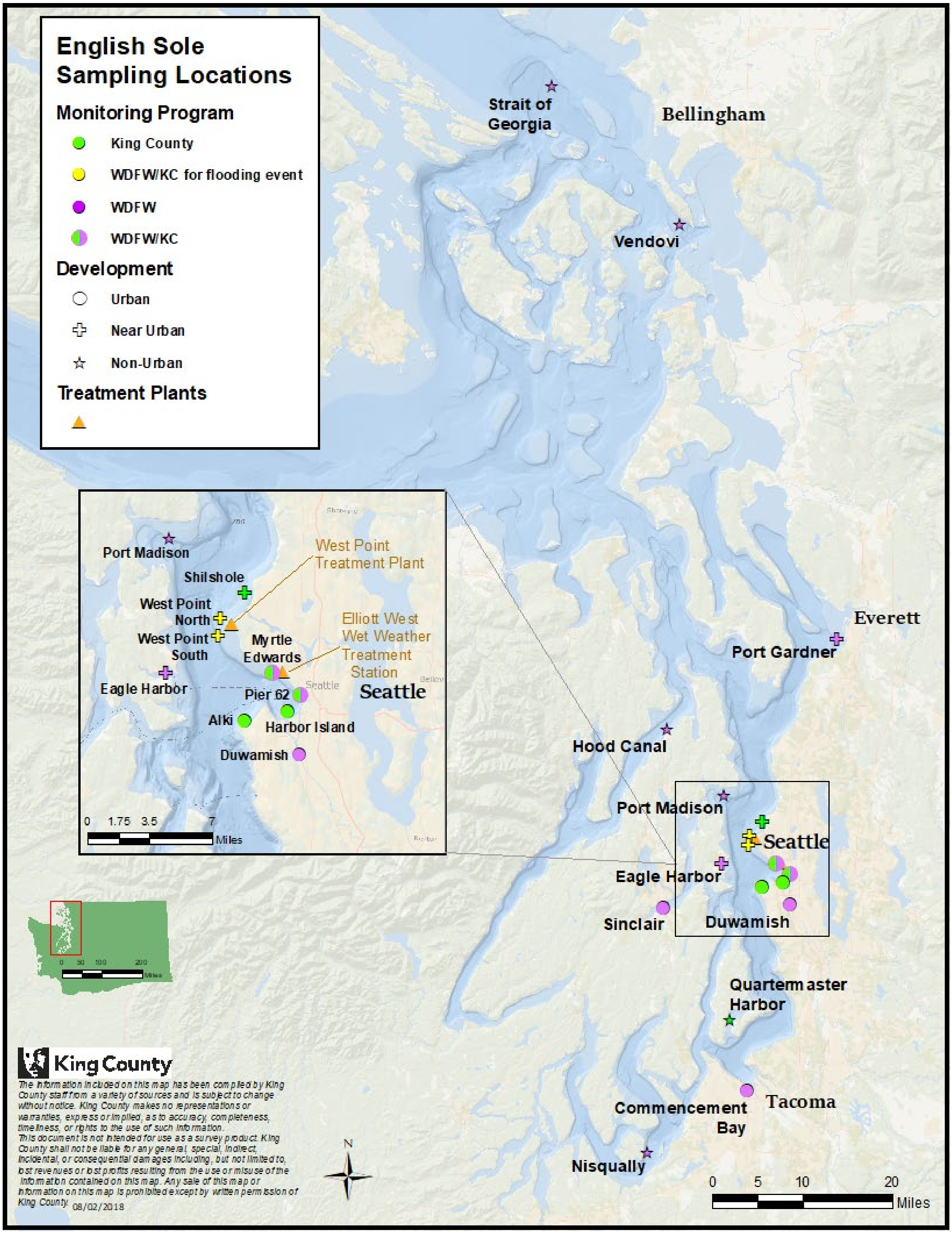
English Sole Sampling Locations
Monitoring Program
King County
WDFW/KC for flooding event
WDFW
WDFW/KC
Development
Urban
Near Urban
Non-Urban
Treatment Plants
Strait of Georgia
Bellingham
Vendovi
Everett
Port Gardner
Hood Canal
Port Madison
Seattle
Eagle Harbor
Sinclair
Duwamish
Quartermaster Harbor
Commencement Bay
Tacoma
Nisqually
0 5 10 20 Miles
Port Madison
Shilshole
West Point North
West Point South
West Point Treatment Plant
Elliott West Wet Weather Treatment Station
Myrtle Edwards
Eagle Harbor
Pier 62
Seattle
Alki
Harbor Island
Duwamish
0 1.75 3.5 7 Miles
King County
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Figure 4. All 2017 and 2019 English sole monitoring locations. Harbor Island and West Point S were only sampled in 2017, and Port Madison was only sampled in 2019. WDFW – Washington Departmet of Fish and Wildlife; KC – King County.

Selection of locations was opportunistic in part, based on upcoming King County and WDFW routine, long-term monitoring program sampling events that were scheduled for May and June 2017 and May and June 2019. Although King County's Shilshole station was the closest existing monitoring station to the West Point outfall, WDFW proposed to also collect English sole from two additional stations located north and south of the West Point outfall (West Point N and West Point S) in 2017 to characterize potential spatial differences in English sole exposures to effluents following the West Point flooding event. Additionally, WDFW and King County anticipated the King County monitoring station off Myrtle Edwards Park would provide information on possible effects of wastewater outfall discharges at the Elliott West wet weather treatment station outfall, one of three treatment facilities King County relied on during the flooding event to reduce flows conveyed to West Point (Figure 1). This site was historically monitored for English sole by WDFW prior to 2017, allowing comparisons to earlier data.

The final monitoring design for 2017 included English sole sampling from the two new stations near the West Point outfall (West Point N and S) and 16 stations previously monitored by the King County and/or WDFW marine tissue monitoring programs (Table 2, Figure 3 and Figure 4). In 2019, all of the same stations were sampled for comparison to 2017, except for West Point S and Harbor Island[4], and a new WDFW sampling location was added in Port Madison Bay (Figure 4).

Table 2. Locator ID and general coordinates for each sampling location.

Sampling Location	County Locator ID	Centroid Coordinates[a]		Mean Depth[b] (ft)	Monitoring Program
		Latitude	Longitude		
Shilshole	Shilshole_Trawl	47.68800	-122.41433	100 – 200	King County
Harbor Island[c]	HrbrIsl_Trawl	47.59186	-122.35951	150 – 200	
Alki	Alki_Trawl	47.584	-122.41075	~200	
Quartermaster Harbor	Qrtrmster_Trawl	47.36983	-122.47983	<100	
Myrtle Edwards	MyrtleEd_Trawl	47.62137	-122.37645	150 – 200	King County & WDFW
Pier 62[cd]	Pier62_Trawl	47.60535	-122.34782	~200	
West Point N	WPD01	47.66921	-122.43799	~100	None - New
West Point S[e]	WPD02	47.65135	-122.44008	0 – 100	
Duwamish	None	47.55790	-122.34408	~40	WDFW
Sinclair Inlet		47.55023	-122.63591	15 - 50	
Port Gardner		47.98465	-122.24266	40 - 200	

[4] The Harbor Island station was eliminated from King County's marine fish tissue contaminant monitoring program in 2019 due to budget constraints and because that station was not located near any current or historic King County outfalls. In addition, contaminant levels detected in English sole and rockfish at Harbor Island were generally similar to those in fish either from Myrtle Edwards or Pier 62 (Colton, 2019).

Vendovi		48.64503	-122.63859	100	
Strait of Georgia		48.85744	-122.95723	350	
Eagle Harbor		47.61924	-122.5135	~40	
Commencement Bay		47.26006	-122.43645	20 -30	
Hood Canal		47.83675	-122.64051	100 - 150	
Nisqually Bay		47.15549	-122.66839	240 - 450	
Port Madison[f]		47.73089	-122.50601	100 – 200	

[a] Coordinates represent the average location of trawl midpoints. For all but Shilshole, Harbor Island, and Quartermaster Harbor, station coordinates were weighted by the number of fish each trawl contributed to composites for each sampling locations. Coordinates are in State Plane North NAD83 US Survey Feet.
[b] Depths are approximated.
[c] These stations have historically been monitored periodically by WDFW and have been part of the King County monitoring program, sampled biannually, since program inception in 2015.
[d] Pier 62 station is the same location as the "Seattle Waterfront" station in the WDFW program (see West et al., 2017).
[e] West Point S and Harbor Island were not sampled in 2019.
[f] Port Madison was only sampled in 2019.

3.1.2 Laboratory Analytes

In a typical year, the monitoring stations, analytes monitored, and analytical methods used by King County differ from those of WDFW. In collaborating after the 2017 West Point flooding event, the two agencies sought to optimize the design and efficiency of the response program. A summary of the samples and analysis types for each organization is as follows:

- WDFW and King County shared composite fillet samples for analysis of total solids, lipids, and organic chemicals (PCBs, PBDEs, DDTs, chlordanes) from Myrtle Edwards, Pier 62, and the new West Point outfall station(s) in 2017 and 2019. Samples from these stations were split and analyzed by the King County Environmental Laboratory (KCEL) in Seattle and the National Oceanic and Atmospheric Administration (NOAA) Laboratory at Montlake, Seattle. The samples were split for two reasons: 1) the laboratories use different analytical methods for organic chemicals and 2) because some stations are regularly shared between the King County and the WDFW monitoring programs (i.e., Myrtle Edwards and Pier 62). Compatibility of datasets for splits and analysis decisions are discussed in Section 3.3.2.
- In addition to the analytical scheme for organics, King County's samples were analyzed for 15 metals (antimony, arsenic, barium, cadmium, chromium, copper, lead, mercury, molybdenum, nickel, selenium, silver, thallium, and zinc).
- According to the TBiOS monitoring program, WDFW also collected data on the following: stable isotopes of nitrogen and carbon in fillet tissue; polycyclic aromatic hydrocarbon (PAH) metabolites and estrogenic chemical levels in bile; and vitellogenin in blood plasma of male fish.

- Xenoestrogens, PAH metabolites in bile, and vitellogenin protein in blood of male English sole in 2017 and 2019 were collected at 14 stations. Five of these stations (i.e., Myrtle Edwards, Pier 62, West Point N, West Point S, and Duwamish) were in King County, four of these are locations where King County also collects data on fillet tissue chemistry (all but the Duwamish station; Table 1).

Table 1 summarizes sampling and analysis targets for each agency and station, and which samples were split and analyzed by both agencies. We do not report on WDFW's fillet tissue chemistry results for locations outside of King County (Table 1, stations marked as NU).

Results of the KCEL and WDFW muscle chemistry data, and the WDFW blood biomarker data for 2017 and 2019 samples are complete and included in this report. The WDFW bile results were incomplete[5] when this report was written; only data for the 2017 bile samples are addressed in this report. The 2019 bile sample results will be presented in an addendum.

3.1.3 Field Sampling

The sample handling, sample preparation and analysis procedures used in the 2017 and 2019 English sole sampling events are summarized below.

3.1.3.1 Sample Collection

WDFW and King County collected all English sole in 2017 and 2019 using a bottom otter trawl. The net was made of a modified commercial design, composed of polyethylene twine and 10 cm mesh size. The trawl has a 21.4 m head rope, a 28.7 m foot rope and a 3.2 cm mesh cod end liner. While fishing, the width of the net opening ranged between 9 m and 13 m, depending on speed, amount of trawl cable deployed and trawl depth.

During trawling, a vessel speed between 2 to 3 knots was maintained and the net width was maximized, by regulating the scope (fathoms of wire out per fathom of depth) of cable. Line out, water depth and positioning coordinates were recorded on the R/V Chasina navigation system. Station locations approximate the locations sampled from previous surveys (if available) to ensure comparable results between surveys. If the needed specimens could not be obtained with one tow, additional tows were made as time allowed. Detailed trawling methods can be found in Quinnell and Niewolny (2015).

Table 3 summarizes the number of English sole muscle composites made from fish at each station in 2017 and 2019, as well as the composites from 2015 which are used for comparison in this report. Table 4 summarizes the number of English sole from which bile and blood were sampled in all three years. In most cases these included individual male

[5] Laboratory analyses at the NOAA lab were delayed due to a shutdown related to the COVID-19 pandemic.

fish, except for analysis of PAH-metabolites in 2015 (composites of male fish) and 2017 (individual male and female fish).

Table 3. Number of English sole muscle composite samples (n), and mean number (#) and length of fish per composite in 2015, 2017, and 2019.

Sampling Location	Sampler	2015			2017			2019		
		n	Mean # fish	Mean Length (mm)	n	Mean # fish	Mean Length (mm)	n	Mean # fish	Mean Length (mm)
Shilshole	KC	2	19.5	255	4	19	270	2	14.5	256
Harbor Island	KC	4	20	263	6	18	255	NS		
Alki	KC	4	22.3	264	6	20	259	1	18	255
Quartermaster Harbor	KC	4	20	280	4	14.3	279	6	14.3	287
Myrtle Edwards	KC and WDFW	4	20	262	6	20	252	4	14[a]	244
Pier 62	WDFW	6	15.4	270	6	20	261	6	20	263
West Point N	WDFW	NS			6	18.2	275	4	14.3	269
West Point S	WDFW	NS			6	16.7	263	NS		
Duwamish	WDFW	6	16.7	274	6	19.5	293	5	14.2	274

[a] One of the four composites in this set had a substantially lower count than the others due to a processing error: counts were 6, 16, 17, 17.
NS = not sampled

Table 4. Number of English sole from which bile and blood were sampled for analysis of xenoestrogens, PAH-metabolites, and vitellogenin in 2015, 2017, and 2019. Samples are from individual male fish unless otherwise noted.

Sampling Location	2015 Bile (Xenos)	2015 Bile (PAHs)[a]	2017 Bile (Xenos)	2017 Bile (PAHs)[b]	2017 Blood (Vtg)	2019* Blood (Vtg)
Myrtle Edwards	NS	NS	7	14	14	14
Pier 62	7	3	6	8	21	31
West Point N	NS	NS	4	14	9	21
West Point S[c]	NS	NS	7	14	12	NS
Duwamish	14	3	8	9	16	30
Sinclair Inlet	8	3	6	7	16	26
Port Gardner	3	3	2	8	11	20
Vendovi	1	1	2	3	2	10
Strait of Georgia	2	3	4	6	6	8
Eagle Harbor	NS	2	6	8	20	30
Commencement Bay	4	3	6	8	18	42
Hood Canal	7	3	6	7	18	24
Nisqually Bay	NS	3	3	5	4	8
Port Madison[c]	NS	NS	NS	NS	NS	25

* Bile samples were collected in 2019, but laboratory sample results were not available at the time of publication of this report due to delays from COVID-19
Xenos – xenoestrogens; PAHs – PAH-metabolites; Vtg – vitellogenin; NS – not sampled
[a] Samples were composites of male fish (13-20 males/composite)
[b] Samples were individual male or female fish
[c] West Point S sampled only in 2017; Port Madison sampled only in 2019

3.1.3.2 Sample Processing and Homogenization

WDFW conducted initial onboard processing of English sole (as described in Niewolny and Langness, 2016) on the same day of collection to obtain blood and bile samples for analysis of vitellogenin in plasma and PAH metabolites and estrogenic chemicals in bile. After initial onboard processing was completed, the fish were placed in plastic bags labeled with location and date and stored in the onboard ship freezer until transport to WDFW office or KCEL in coolers with ice. At WDFW or KCEL, fish were stored frozen at -20^0 C until processed for muscle tissue analyses at a later date.

Following sorting of fish within each station to make composites with randomized fish size, all English sole composite fillet samples were prepared using the WDFW muscle resection technique described in the WDFW TBiOS Program standard operating procedures (SOP) (WDFW, unpublished reports) and the King County (2015) SAP. This technique results in collection of equal tissue mass from each fish for each composite. Tissue mass from each fish of the target total (20 fish per sample in 2017; 15 fish per sample in 2019) were included in each composite sample. When the target number of individuals per composite sample was not possible, equal numbers of fish from a sampling station were distributed into each sample (minimum of 15 individuals) to obtain the target number of composites.

Table 3 summarizes the mean number and length of English sole in each composite made in 2017 and 2019, as well as information on composites created in 2015 and used for comparison in this report. In 2017, the numbers of English sole collected at Shilshole and Quartermaster Harbor were only adequate to complete four of the targeted six composite samples per station. Following homogenization of samples from the locations listed in Table 3, the homogenate was split and shared with the partner agency's laboratory. According to the standard monitoring program protocol for King County and WDFW, English sole at or above 230 mm total length were targeted because this is the expected size at sexual maturity; however, sometimes slightly smaller fish were retained.

3.2 Laboratory Analytical Methods

The King County Environmental Laboratory (KCEL) and the NOAA laboratory at the Northwest Fisheries Science Center of the National Marine Fisheries Service (at the direction of WDFW) performed analyses separately, with some overlapping analytes. The analytes measured and the analytical methods followed by each laboratory are summarized in this section.

3.2.1 King County Environmental Laboratory

KCEL analyzed English sole samples collected by King County and split samples received from WDFW for the following parameters: conventional parameters (lipids and total solids), as well as total metals (arsenic, beryllium, cadmium, chromium, copper, lead, nickel, selenium, silver, thallium, and zinc), total mercury, 10 PCB homologs, 14 PBDE congeners, three DDTs, and 12 chlorinated pesticides. Table 5 summarizes the individual chemicals analyzed in each analyte group. Table 6 summarizes the KCEL laboratory methods, more detail is presented in the SAP and SAP Addenda (King County, 2015, 2017, and 2019).

Table 5. Chemicals analyzed by KCEL in English sole muscle tissue by analyte group..

PCB homologs*	PBDEs	DDTs	Other chlorinated pesticides
Monochlorobiphenyls	BDE-17*	4,4'-DDD (p,p'-DDD)	alpha-Chlordane
Dichlorobiphenyls	BDE-28/-33	4,4'-DDE (p,p'-DDE)	beta-Chlordane
Trichlorobiphenyls	BDE-47	4,4'-DDT (p',p'-DDT)	Heptachlor
Tetrachlorobiphenyls	BDE-66		Heptachlor Epoxide
Pentachlorobiphenyls	BDE-71*		alpha-Hexachlorohexane
Hexachlorobiphenyls	BDE-85		beta-Hexachlorohexane
Heptachlorobiphenyls	BDE-99		gamma-Hexachlorohexane
Octachlorobiphenyls	BDE-100		Hexachlorobenzene
Nonachlorobiphenyls	BDE-138*		Aldrin
Decachlorobiphenyl	BDE-153		Dieldrin
	BDE-154		Endosulfan
	BDE-183		Mirex
	BDE-190*		
	BDE-209*		

*Denotes analyte groups or individual analytes not analyzed in splits by the NOAA laboratory (see Table 7)

Table 6. KCEL methods by analyte group

Analytes	EPA or Standard Method	KCEL Standard Operating Procedure (SOP)
Lipids	Gravimetric	740v2
Total Solids	SM2540-G	307
Metals (except mercury)	PSEP1997 SW846 6020B	616
Total Mercury	PSEP 1997 SW846 7471B	604v5
PCB Homologs	SW846 3540C 680 SIM	782
PBDE Congeners	SW846 3540B and 3540C	781
Chlorinated Pesticides	SW846 3540C 8081B	733

N/A – Not applicable

3.2.2 NOAA Laboratory

English sole samples collected by WDFW were analyzed under contract by the NOAA laboratory at the Northwest Fisheries Science Center of the National Marine Fisheries Service. They analyzed the following chemistry parameters: conventional parameters (lipids and total solids), stable isotopes of carbon and nitrogen, 40 PCB congeners, 11 PBDE congeners, 6 DDTs, 16 chlorinated pesticides, 9 xenoestrogens, 33 PAH-metabolites (Table 7). Stable isotopes and biological covariates (fish age and lipid content) were measured to control for variation in fish condition, which may explain chemical patterns, but are independent of contaminant exposure. Table 8 summarizes the laboratory methods for analysis of chemicals in muscle tissue and bile; details of the analytical methods for PCBs, PBDEs and chlorinated pesticides are described in West et al. (2017), for xenoestrogen in da Silva (2013) and for PAH-metabolite in da Silva (in prep). Plasma vitellogenin measurements were conducted by WDFW according to the Multi Species Vitellogenin ELISA manufacturer's instructions (TECOmedical AG, Switzerland).

Table 7. Chemicals analyzed by NOAA in English sole muscle tissue and bile.

Analyzed in muscle tissue				Analyzed in bile	
Total PCB congeners*	**Total PBDEs**	**Total DDTs**	**Other chlorinated pesticides**	**Xenoestrogens***	**Total PAH-metabolites***
PCB 17	BDE-28	2,4'-DDD (o,p'-DDD)*	alpha-Chlordane	estrone	Me6OH2NPH
PCB 18	BDE-47	2,4'-DDE (o,p'-DDE)*	beta-Chlordane	17β-estradiol	MeOHSumNPH
PCB 28	BDE-49*	2,4'-DDT (o,p'-DDT)*	Oxychlordane*	17α-ethynylestradiol	OH1NPH
PCB 31	BDE-66	4,4'-DDD (p,p'-DDD)	cis-Nonachlor*	estriol	OH2NPH
PCB 33	BDE-85	4,4'-DDE (p,p'-DDE)	trans-Nonachlor*	Bisphenol-A	OH2FLU
PCB 44	BDE-99	4,4'-DDT (p',p'-DDT)	Nonachlor III*	Bisphenol-AF	OH3FLU
PCB 49	BDE-100		Heptachlor	Bisphenol-F	OH2DBT
PCB 52	BDE-153		Heptachlor Epoxide	Bisphenol-S	dihydroxy12dihydroPHN
PCB 66	BDE-154		alpha-Hexachlorohexane	Tetrabromo-bisphenol-A	dihydroxy910dihydroPHN
PCB 70	BDE-155*		beta-Hexachlorohexane		dihydroxydihydroPHN†
PCB 74	BDE-183		gamma-Hexachlorohexane		OH1PHN
PCB 82			Hexachlorobenzene		OH3PHN
PCB 87			Aldrin		OH4PHN
PCB 95			Dieldrin		OH9PHN
PCB 99			Endosulfan		PHN3carboxylic acid†
PCB 101			Mirex		PHN4carboxylic acid†
PCB 105					PHN9carboxylic acid†
PCB 110					bis18OHMeANT
PCB 118					dihydroxydihydroANT†
PCB 128					dihydroxy23dihydroFLA
PCB 138					dihydroxy34dihydro712dimethylBAA†
PCB 149					dihydroxy56dihydroBAA
PCB 151					dihydroxy89dihydroBAA
PCB 153					dihydroxy12dihydroCHR
PCB 156					dihydroxy34dihydroCHR
PCB 158					dihydroxy56dihydroCHR
PCB 170					transdihydroxy45dihydroBEP†
PCB 171					dihydroxy45dihydroBAP

Analyzed in muscle tissue				Analyzed in bile	
Total PCB congeners*	**Total PBDEs**	**Total DDTs**	**Other chlorinated pesticides**	**Xenoestrogens***	**Total PAH-metabolites***
PCB 177					dihydroxy78dihydroBAP
PCB 180					tetrahydroxytetrahydroBAP†
PCB 183					dihydroxy15ATQ
PCB 187					OH2ATQ
PCB 191					dihydroxyBPH
PCB 194					
PCB 195					
PCB 199					
PCB 205					
PCB 206					
PCB 208					
PCB 209					

*Denotes analyte groups or individual analytes not analyzed in splits by the KCEL laboratory (see Table 5).
PCB congeners in bold are those used to estimate total PCBs as the sum of detected concentrations multiplied by two (West et al., 2017; Lauenstein and Cantillo, 1993).
† Denotes PAH-metabolites added to the analyte list in 2017 but not included in 2015; see also Appendix C, Table 11.
ANT – anthracene
ATQ – anthraquinone
BAA – benzo(a)anthracene
BAP – benzo(a)pyrene
BEP – benzo(e)pyrene
BPH – biphenyl
CHR – chrysene
FLA – fluoranthene
FLU – fluorene
NPH – naphthalene
OH – hydroxy
PHN – phenanthrene

Table 8. Analytical Laboratory Methods by Analyte Group

Analytes	Standard Method
Lipids	Thin layer chromatography/flame ionization detector
Total Solids	Sloan et al., 2014
PCB Congeners (Limited list)	Sloan et al., 2014
PBDE Congeners	Sloan et al., 2014
Chlorinated Pesticides	Sloan et al., 2014
Stable Isotopes	IsoLab 2017
Xenoestrogens	LC-MS/MS (da Silva et al., 2013)
PAH-metabolites	LC-MS/MS[1]
Vitellogenin	TECO multi-species vitellogenin ELISA kit (TE1042) following manufacturer's instructions.

LC-MS/MS - liquid chromatography with tandem mass spectrometry
ELISA - enzyme-linked immunosorbent assay
[1] Method in development by NOAA.

3.3 Data Analysis

The sections below first present a description of the analytical approach to the English sole tissue chemistry data and then review issues of data comparability. Because data evaluated in this document were collected for long term monitoring, sampling was not designed to rigorously test hypotheses about cause-effect relationships between the West Point flooding event and English sole metrics of chemical exposure. However, the data collected for English sole is useful for ruling out whether a substantial change in chemical contaminant concentrations or biomarkers in English sole tissues occurred due to the West Point flooding event.

3.3.1 Approach

The statistical analysis approach to address the study questions emphasizes multiple comparisons across a range of locations for each chemical endpoint. English sole monitoring locations vary by their proximity to urban centers and to the West Point treatment plant. For each chemical analyte, we compare the English sole muscle tissue chemistry data from each location to results for West Point N and Myrtle Edwards stations. Data for biomarkers and stable isotopes, collected only by WDFW in 2017 and 2019 (Table 1), cover a larger geographic area and not all of the locations in King County.

English sole are abundant in Puget Sound. This species has been used by WDFW for monitoring since 1989. When King County initiated its marine fish monitoring program in 2015, it was in collaboration with WDFW, which had already selected English sole as one of the species of interest (rockfish are also used). English sole are an appropriate part of this joint monitoring effort because they are a benthic species closely associated with sediments and with a relatively long life span (King County 2015). They exhibit site fidelity to their spring and summer feeding areas (Day 1976; Moser et al. 2013), often in shallower bays. English sole travel significant distances, but exhibit homing behavior if displaced by scientists or by spawning migrations into deeper waters during winter (Day 1976; Moser et al. 2013). Their body burdens of persistent contaminants are considered representative of the local conditions in spring/summer habitats (West et al. 2017).

Because different locations were sampled for different endpoints, comparisons between stations vary by measured endpoint. To create a conceptual framework for the data analysis, sampling stations discussed in this report were assigned to one of four groups:

- Group 1. Higher Exposure: Locations where English sole were most likely to be exposed to untreated or primary treated discharges associated with the West Point flooding event due to proximity and/or predominant direction of flow related to the West Point Treatment Facility or to the Elliott West wet weather treatment station: West Point N and Myrtle Edwards.
- Group 2. Lower Exposure: Locations in King County where English sole were likely less or minimally exposed to untreated or primary treated discharges associated with the West Point flooding event due to proximity and/or predominant direction of flow and potentially exposed to storm-related inputs from surrounding urban areas in 2017: Shilshole, Alki, Pier 62, Harbor Island, Duwamish and West Point S.
- Group 3. Background Conditions: Locations that are not in King County, and where English sole were unlikely to be exposed to untreated or primary treated discharges associated with the West Point flooding event but were potentially exposed to storm-related inputs from surrounding urban areas in 2017: Eagle Harbor, Sinclair Inlet, Commencement Bay, Port Gardner.
- Group 4. Reference Conditions: Locations that were likely subject to the least amount of exposure to urban, storm-related chemical contaminant inputs in 2017 relative to the others in Groups 1 - 3: Nisqually Bay, Quartermaster Harbor, Port Madison, Hood Canal, Vendovi and Strait of Georgia.

These groups are presented as a general framework to guide interpretation. These designations were established after sample collection, not before, and sampling was not conducted evenly in each location or across locations (Table 1). Several of the analyses incorporate information from only a subset of all locations.

Although historical English sole tissue chemistry data directly adjacent to the West Point outfall were not available, data collected by both WDFW and King County prior to the flooding event (i.e., data from 2015), are used in this report to represent conditions prior to the West Point flooding event.

3.3.2 Interlaboratory Comparability

A subset of samples was split between the King County and WDFW labs to allow for an analysis of lab comparability. In an earlier report, King County analyzed differences in the 2017 organic chemical data between the KCEL and WDFW's NOAA laboratory due to differences in analytical methods (King County, 2021). PCB, PBDE, and DDT data from the

2017 and 2019 sampling events at the West Point N, West Point S (2017 only), Myrtle Edwards and Pier 62 stations were evaluated to determine comparability between the two labs to support data analysis decisions for this report.

3.3.2.1 PCBs

King County and WDFW estimate “total PCBs” using different summation methods: King County estimates total PCBs as the sum of detected homologs (Table 5) while WDFW estimates total PCBs by summing detected concentrations of 17 commonly detected congeners (Table 7) and then multiplies that sum by two (West et al., 2017; Lauenstein and Cantillo, 1993). Total PCB concentrations in split samples from 2017 and 2019 were consistently higher by NOAA’s method compared to KCEL, similar to findings from 2017 (King County, 2021). The relationship between total PCB results from the two labs is consistent (adjusted $R^2 = 0.86$; total PCBs $_{NOAA}$ = 9.3 + 1.39 * total PCBs $_{KCEL}$). This statistically significant relationship illustrates concordance between the two sets of results but with a high-bias in the WDFW results relative to the KCEL results (i.e., the slope coefficient >1, intercept of 9.3). Sample concentrations from KCEL methods ranged between 37.5 to 91.5% of concentrations from NOAA methods in split samples (mean = 62.4%) and variability was larger at higher concentrations. For these reasons, both datasets are reported herein but are analyzed separately in Section 4.

3.3.2.2 PBDEs

WDFW samples were analyzed for eleven PBDE congeners, nine of which are in common with the King County method (Tables 5 and 7). Only detected congeners are included in the total PBDE sums for both methods; however, congener detection limits by KCEL were generally two orders of magnitude lower than NOAA’s method; this differential was similar to earlier results (King County, 2021). Also similar to previous results, the relationship was weaker (adjusted $R^2 = 0.25$; total PBDEs $_{NOAA}$ = 2.58 + 0.353 * total PBDEs $_{KCEL}$) than that observed for total PCBs. Sample concentrations from KCEL methods ranged between 26.2 to 188.7% of sample concentrations from NOAA methods in paired samples (mean = 72.8%) and variability was larger at higher concentrations. Differences in analytical methods make direct comparison of total PBDE results from these two laboratories inappropriate.

3.3.2.3 DDTs

KCEL and NOAA use the same EPA method and instrumentation (GC/MS) but differ in the specific chemicals targeted for organochlorine pesticide analysis. The NOAA method includes three more isomers of DDTs and their metabolites than KCEL (Tables 5 and 7). Because of these differences, total DDT concentrations could potentially be higher in samples analyzed by NOAA. Only 4,4'-DDD and 4,4'-DDE isomers were detected by NOAA in the split samples, and 4,4'-DDE was the only detected organochlorine pesticide in samples analyzed by KCEL. Thus, higher total DDTs would be expected from the NOAA laboratory. A regression of the 2017 and 2019 data shows that total DDTs reported by KCEL ranged from 60 to 110% of total DDTs concentrations from NOAA methods in split samples (mean =

78%). Similar to PCBs, the relationship between the two labs is consistent ($R^2 = 0.84$; total $DDTs_{NOAA} = -0.0798 + 1.37 * total\ DDTs_{KCEL}$).

3.3.2.4 Interlaboratory Comparability Conclusions

On the basis of the above comparisons, the statistical analyses of the organic chemistry data (PCBs, PBDEs, and DDTs) were separated by laboratory. This approach avoids complications in data interpretation that would be introduced by the different analytical and summation methods used, which may have biased the data high or low relative to the results of each lab.

3.3.3 Data Evaluation Methods

Data evaluation included application of several data treatment and analytical steps.

3.3.3.1 Estimation of Non-Detects

For datasets with censored data, the method detection limit was used to estimate the concentration of chemicals reported as not detected by the laboratory (U-qualified). For calculation of total PCBs, total PBDEs, or total DDT and metabolites, non-detects were set to zero (ND = 0). Total concentrations for organic compounds were estimated by summing all detected values of congeners, homologs or isomers in each sample, except for total PCBs estimated by WDFW as described in Section 3.3.2.1 (see also Table 7). If all contributing values to organic compound totals were non-detect, the total concentration was estimated as the minimum detection limit of all the contributing congeners, homologs, or isomers.

3.3.3.2 Age- and Lipid-Adjustment of Tissue Concentrations

Stable isotopes, age, and lipid content data were used to help data interpretation. Incorporation of these parameters can help control for variations between individual tissue samples because each may explain chemical contaminant patterns but are independent of the contaminant exposure. For example, mercury concentration tends to increase with fish age because it bioaccumulates and is not readily excreted. Mercury concentrations would likely be higher in older fish, even if exposed to the same concentrations of mercury in their food and general environment as younger fish. Age as a covariate for mercury, and age and lipids as covariates for organic chemicals were tested using Analysis of Covariance (ANCOVA) in R. These covariates were not found to be significant ($p<0.05$). However, these covariates were found to be significantly correlated ($p<0.05$) with the parameters using Spearman's rank correlation. Because of this discrepancy, the data were also age- and lipid-adjusted and evaluated.

Age-adjusting the data is appropriate if the patterns in the data are influenced by the age of the fish sampled. However, if the patterns in the data are not influenced by age, then providing an age-based adjustment can cause erroneous age-based patterns to appear in the data. After age-adjusting and visually examining the English sole data, we observed a skewed pattern related to age that had not been present prior to age-adjusting. Thus, we concluded that age-adjusting the data introduced a pattern that was illogical and could have confounded the interpretation of the data.

Finally, we found that found that lipid-adjusting did not substantially change the outcomes of subsequent statistical tests for the English sole data. Therefore, we proceeded with the raw, unadjusted data and did not further evaluate fish age or lipids in this report.

3.3.3.3 Comparisons of Groups

All of the metal (except mercury), organic chemical, and isotope data were found to fail tests for normality and/or homoscedasticity. As a result, these parameters were tested for significant differences across stations using Kruskal-Wallis (nonparametric analysis of variance) followed by the post-hoc Wilcoxon rank-sum test. The Kruskal-Wallis test is a nonparametric test for comparing independent samples. The null hypothesis was there is no dominance among locations; a random drawing from location A has a 50% chance of being less than or equal to a random drawing from location B. To mitigate false discovery (i.e., Type I error), p-values were adjusted per Benjamini and Hochberg (1995). The "kruskal.test" function in the "stats" package and the "pairwise.wilcox.test" function in the "rcompanion" package for R v3.6.0 was used (R Core Team, 2019). After splitting the datasets by laboratory, each analyte was tested for significant difference across location and year using a Kruskal-Wallis test. The pairwise Wilcox post hoc test was used to determine whether the differences by location and year were significant at p-level 0.05.

3.3.3.4 Uncertainty Assessment

The risk of erroneously accepting the null hypothesis (i.e., Type II error) is a function of statistical power, which is controlled by sample size and shape of the data distribution. Given the high natural and likely storm-related variability in the English sole muscle tissue data and the uneven sample size at each site and year (*n* generally ranges from 3 to 6), we evaluated more closely the absence of significant differences between groups for evidence of Type II error.

Uncertainty Assessment - Approach
Type II error is a failure to reject the null hypothesis when a real effect exists. Our null hypothesis for each pairwise comparison was: there is no difference in the distribution of sample concentrations for 1) the same station between years, or 2) different stations in the same year (See section 3.3.3.3). A power analysis was not performed in advance of sampling, and retrospective power analysis is not appropriate; for further information about the misuse of retrospective power analysis and more appropriate alternatives, we refer the readers to the following literature (Lenth, 2001; Hoenig & Heisey 2001; Levine & Ensom 2001; Colegrave & Ruxton 2003; Heckman et al 2022).

Given the range of variation in English sole tissue parameters, we sought to characterize our confidence in conclusions from pairwise comparisons that resulted in rejecting the null hypothesis. To do this, we examined the 95% confidence interval for the estimated median difference between samples (Hoenig & Heisey 2001; Levine & Ensom 2001; Colegrave & Ruxton 2003; Heckman et al 2022). This analysis was conducted for all year-specific location comparisons *not* found to be significantly different. The median difference between *n* samples from Site A and *n* samples from Site B is the median value of A1 – B1, A1 – B2, A1 – B3, A2 – B1, etc. This 95% confidence interval provides a measure of certainty on the "true" differences between year-specific locations. Wider confidence intervals indicate less certainty about the "true" difference (i.e., higher chance of Type II error), while narrower intervals indicate more certainty (i.e., lower chance of Type II error).

The 95% confidence intervals for the estimated median difference between samples were calculated for the pairwise comparisons performed using the non-parametric Wilcoxon rank sum test (in the stats package of R) described above. Where sample sizes were inadequate to calculate the interval at the 95% confidence level, the statistical software automatically calculated the interval at the highest possible confidence level for the sample size provided. In cases where there was a sample size of 1, or if values were all identical (i.e., all samples were below detection limit and had MDL substituted), no confidence interval could be computed, and associated statistics are given as NA (not applicable). Associated p-values were adjusted for multiple comparisons using the Benjamini & Hochberg (1995) method of the p.adjust function.

While the use of confidence intervals is generally recommended to aid in the interpretation of non-significant results (Hoenig & Heisey 2001; Levine & Ensom 2001; Colegrave & Ruxton 2003; Di Stefano 2004; Heckman et al 2022), there are no hard rules to determine how wide of a confidence interval is too wide; it is open to interpretation by the investigators. Generally, given two confidence intervals that both contain zero, we have more certainty accepting the null hypothesis where the range is narrower. We more confidently reject the null hypothesis with confidence intervals not containing zero. Using this approach and our own review of the data, we have less confidence in the findings of "no significant difference" (i.e., failing to reject the null hypothesis) for comparisons where:

1. The confidence interval does not contain zero, but the adjusted p-value is not significant (if we had more samples, we would potentially see more significant adjusted p-values where the median difference is further away from zero. Closer examination of the confidence intervals is warranted.)
2. The confidence interval contains zero, but it is relatively wide (it takes up >25% of the possible confidence interval window)
3. Comparisons where one of the year-specific locations has an *n* of only 1 (i.e., Alki 2019).

We performed this uncertainty assessment only for pairwise comparisons found *not* to be significantly different from one another; comparisons that were found to be significantly different are not included because they were inherently powerful enough to discern differences. Figure 5 illustrates the output of this uncertainty analysis for the year-specific pairwise comparisons for PCBs. The dots in Figure 5 are the estimated median difference between samples, the colored lines (whiskers) are the 95% confidence interval of that estimate. Results shown in orange or red indicate cases in which comparisons met one of the three criteria shown above. For these comparisons, we have reduced confidence in our conclusion of no significant difference in the distribution of sample concentrations. Results shown in teal are accepted with higher certainty.

Results for PCBs indicate that there is a substantial number of cases in which we have less confidence in the finding of no difference for comparisons between years in the same site, especially for comparisons involving Elliott Bay sites (Harbor Island, Myrtle Edwards, Pier 62). This indicates a reduced confidence in conclusions that there is no difference in these pairwise comparisons. Likely an insufficient number of samples for the amount of variability in concentrations at each location is the cause of this lower confidence in our findings.

Outside of Elliott Bay, power appears to be adequate for PCBs (Figure 5, bottom panel). For example, we have higher certainty in the median difference between pairs (blue shading) for comparisons of PCB results at West Point North 2017 and 2019, and at West Point South 2017, as well as with those of Quartermaster Harbor or Alki locations. This result indicates higher confidence in results of the pairwise comparisons where they indicate no significant difference. The median difference between pairwise comparisons of these West Point locations and years with Elliott Bay sampling locations are less certain (orange and red shading). Where the pairwise comparisons indicate significant differences, this uncertainty does not affect our interpretation of the results, as there was sufficient power to detect the difference and so Type II error is not a relevant concern.

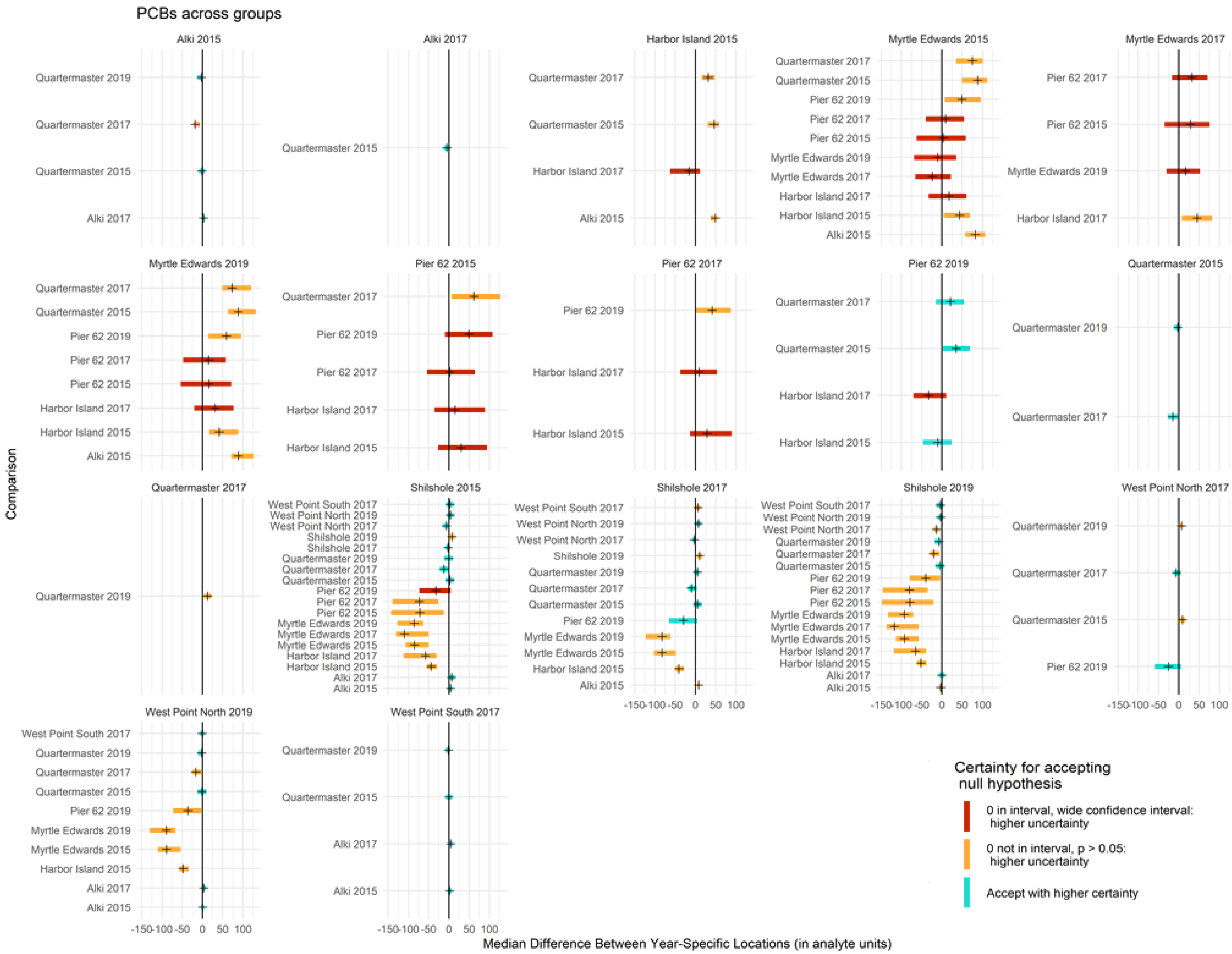

PCBs across groups
Comparison
Median Difference Between Year-Specific Locations (in analyte units)
Certainty for accepting null hypothesis
0 in interval, wide confidence interval: higher uncertainty
0 not in interval, p > 0.05: higher uncertainty
Accept with higher certainty

Figure 5. Median (plus sign) and 95% confidence intervals (colored line) for the estimated median difference between PCB concentrations where no significant difference was found (i.e., where the null hypothesis was accepted). The title of each sub-figure is the minuend[6] that the difference is calculated from, the group name on each y-axis is the subtrahends[6] (minuend – subtrahend = difference).

[6] A minuend is a number from which another is to be subtracted, a subtrahend is a number to be subtracted from another: minuend – subtrahend = difference.

The uncertainty assessment described above generates hundreds of results, which are shown in a series of figures for each of the metals and organic contaminants in Appendix D. To refine and summarize the evaluation, we isolated results involving within-site comparisons at West Point N (i.e., West Point N 2017 vs. West Point N 2019) and Myrtle Edwards (i.e., Myrtle Edwards 2015 vs. Myrtle Edwards 2017; Myrtle Edwards 2015 vs. Myrtle Edwards 2017; Myrtle Edwards 2017 vs. Myrtle Edwards 2019). These sampling locations make up the Group 1 dataset. Rejecting the null hypothesis in pairwise comparisons that include one of these locations could be interpreted as evidence that the West Point flooding event affected fish tissue concentrations. Failing to reject the null hypothesis when it should be rejected risks missing evidence that the event could have affected fish tissue concentrations. For this subset of results, we report the number of outcomes that indicate a comparison in which we have less confidence in the findings of "no difference" in Table 9.

Table 9. The number of within-site (temporal) comparisons at West Point N (2017, 2019) and Myrtle Edwards (2015, 2017, 2019) where we have less confidence in a finding of "no significant difference".

Chemical	West Point N	Myrtle Edwards
	(n/N)	(n/N)
Arsenic	0/1	3/3
Chromium	0/1	2/2
Copper	1/1	1/3
Lead	1/1	0/3
Mercury	0/1	3/3
Zinc	1/1	2/3
King County total PCBs	NA	3/3
WDFW total PCBs	NA	0/1
King County total PBDEs	1/1	1/3
WDFW total PBDEs	NA	NA
King County total DDTs	NA	3/3
WDFW total DDTs	NA	NA

n/N = number of comparisons (n) with unacceptably high uncertainty relative to the total number of comparisons with an adjusted p-value that was not significant (N).
NA = CI analysis not applicable because all comparisons had an adjusted p-value that was significant.

Our analysis indicated uncertainty for a number of comparisons with a finding of "no significant difference" for the English sole muscle tissue data (Appendix D). We have less confidence in the findings of "no difference" for most comparisons involving the Elliott Bay sites (Myrtle Edwards, Pier 62, and Harbor Island) for arsenic, chromium, copper, lead, mercury, and zinc. The same was true for many of the Elliott Bay site comparisons for the King County organic contaminant datasets (total PCBs, total PBDEs, and total DDTs), though these were limited to comparisons from Pier 62 in the WDFW organic contaminant dataset. In addition, we have uncertainty in the "no significant difference" findings for many of the between-year comparisons at the Myrtle Edwards location for metals (Table 9). We also have uncertainty in a number of "no difference" findings for comparisons involving West Point N (especially 2019), West Point S, and Shilshole for the metals and for the King County DDTs dataset (Appendix D). These findings confirm that the combination of noise in the data and the relatively low sample size at each location casts uncertainty on conclusions of "no difference" between specific pairs, particularly for locations in Elliott Bay and for the metals data overall. As a result, we limit our reporting of no significant differences in the results section to statistically robust comparisons that inform the questions of this report with low uncertainty in the finding.

3.3.3.5 Homogeneity of Variance

Given the complexity of the ecosystem where samples were collected, it is conceivable that elevated concentrations of wastewater-related contaminants in English sole tissue occurring in response to the West Point flooding event effluents could appear as greater variability of tissue contaminant concentrations within sites. English sole muscle tissue concentrations that are both higher and more variable in those proximal to the source of contaminant inputs have been observed previously. For example, West et al. (2017) report both higher concentrations and higher variability of PCB, PBDE, and DDT concentrations in English sole muscle collected from bays receiving waters from highly developed drainage basins than in those from moderately or less developed basins. For this reason, we examined the year-to-year homogeneity of PCB concentrations within sites to evaluate whether within-site sample variance may have been affected by the West Point flooding event.

To evaluate year-to-year sample variances within location, we compared King County data on total PCBs from 2015, 2017, and 2019 with historic WDFW total PCB data (from 2005-2019) taken from sites with a similar degree of anthropogenic influence. The WDFW historical dataset is used here because King County did not conduct this kind of sampling prior to 2015. Since the historical WDFW data gives us a baseline sense of the variance expected at each type of site, this comparison helps examine whether the West Point incident had any effects that would not be apparent from just a test of median sample concentrations at the King County study sites.

We examined only total PCB data for year-to-year (i.e., year-to-year homogeneity of variances) and did not include PBDEs or DDTs in this analysis because of the poor correlations between results generated by the KCEL and NOAA laboratories (Sections 3.3.2.2 and 3.3.2.3). We did not include metals in this analysis because of a lack of historical data from WDFW.

For each station at which 2017 and 2019 samples were analyzed by KCEL, we matched historical data using total PCBs from the appropriate WDFW sites. The WDFW sites we chose for comparison were expected to have similar levels of contaminant exposure, based on proximity to urbanized centers and excluding possible impacts of the West Point flooding event, to the King County sites against which they were compared. We then created distinct groupings based on site and year. WDFW and King County stations used in the comparison of total PCB year-to-year sample variances included:

- Nisqually station – compared to West Point N, West Point S, Shilshole, Alki, and Quartermaster Harbor stations
- Pier 62 and Myrtle Edwards stations – compared to Myrtle Edwards, Pier 62, and Harbor Island stations.

For each of these site-year groups, we conducted a Bartlett test for the homogeneity of variances using the bartlett.test function in the R stats package (R Core Team, 2020). We then adjusted the p-values for the pairwise homogeneity of variances tests using the "BH" method of the p.adjust function in the R stats package (R Core Team, 2020; Benjamini & Hochberg, 1995). Results are presented in Section 4.

4.0 RESULTS

The results of the data analyses performed on English sole tissue chemistry are presented in this section:

1. **Stable isotopes** of nitrogen and carbon establish whether fish represented by the samples were in substantially different food webs or subsist on fundamentally different food sources. If so, such differences impede comparison of bioaccumulative chemicals across locations.
2. **Metals and organic chemicals** in composite English sole fillet tissues, compared between sampling locations and over time.
3. **Biomarkers** (for WDFW sampling locations only) of exposure to PAHs, and endocrine disrupting compounds.

4.1 Stable Isotopes

Stable carbon and nitrogen isotopes exist naturally in lighter and slightly heavier forms in differing, but generally consistent, proportions. Normal biological processes, such as photosynthesis and carbohydrate formation, result in isotopic fractionation (i.e., selective release of lighter isotopes and retention of heavier isotopes; O'Leary, 1988, Schulze and Giese, 1993). This regular discrimination occurs during natural bio- and geo-chemical processes and results in an isotope "signature" in carbon and nitrogen sources as well as in organisms that take them in for biological use. Ratios[7] of carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) isotopes are expressed as measured differences from a standard reference in units of per mil (‰).

Results of analyses for stable isotopes are used in this report to evaluate whether trophic position or energy sources to English sole may confound comparisons among locations Analysis of $\delta^{15}N$ stable isotope ratios were used to investigate whether the English sole sampled were consuming diets at similar or different trophic levels (which can influence their tissue concentrations), while $\delta^{13}C$ was used to assist with characterization of different environmental sources of carbon. Values of $\delta^{15}N$ increase with trophic level in the food web. With each increase in trophic level, the heavier ^{15}N isotope is enriched in an organism's tissues resulting in greater proportions of the heavier isotope at the top of the food web. Generally, a difference of 2 to 4 ‰ in $\delta^{15}N$ is seen between trophic levels (McCutchan et al., 2003, Perkins et al., 2014).

Gradients of $\delta^{13}C$ have been used as an indicator of marine versus terrestrial carbon inputs in the diet (Hobson, 1999). $\delta^{13}C$ differs between freshwater and marine conditions, with freshwater (terrestrial-derived carbon sources) containing carbon more depleted (i.e., more negative) in ^{13}C (Hobson, 1999). Within marine environments, $\delta^{13}C$ can also be used to differentiate between two major sources of dietary carbon: nearshore (benthically-

[7]The ratio of heavier to lighter carbon or nitrogen isotope in a sample (R_{sample}) divided by that in a standard material ($R_{standard}$) is the basis for $\delta^{13}C$ or $\delta^{15}N$. That is: δX (‰) = $[(R_{sample}/R_{standard})-1] \times 1000$ (Peterson and Fry, 1987).

linked) vs. pelagic (open water) food webs (Hobson, 1999). More depleted $\delta^{13}C$ values are associated with pelagic production, while enriched (i.e., less negative) values are associated with nearshore production.

Stable isotopes were analyzed in 2017 (year 0) and 2019 (year 2) English sole muscle samples from the two West Point stations, Myrtle Edwards, and from Pier 62 and the WDFW Duwamish River station, both of which also included data from 2015 (pre-event) that was used for comparison. No other King County monitoring stations were evaluated for stable isotopes.

4.1.1 Nitrogen Isotopes

A change in trophic level generally occurs when differences in $\delta^{15}N$ reach between 2 to 4‰ (McCutchan et al., 2003, Perkins et al., 2014). There was some variability in nitrogen isotope ratios, but differences between locations were smaller than those between whole trophic levels (2 to 4 ‰). The $\delta^{15}N$ values within stations were generally within a tight range (0.5‰) across all years, although larger variability was observed between stations (Figure 6). Specifically, the Duwamish River $\delta^{15}N$ values for English sole muscle (mean 12.6‰) were statistically lower than all other sites measured (mean between 13.6 and 14.0‰), or a difference of 1 – 1.4‰. The overall narrow range of mean $\delta^{15}N$ ratios (≤1.5‰) indicates English sole were likely feeding at similar trophic levels at all these locations. Thus, dietary differences do not appear to substantially influence tissue chemical concentrations. However, the consistent difference between the $\delta^{15}N$ values in Duwamish River samples and those of the other locations indicates a potentially different diet (e.g., more benthic-driven) of English sole in this habitat.

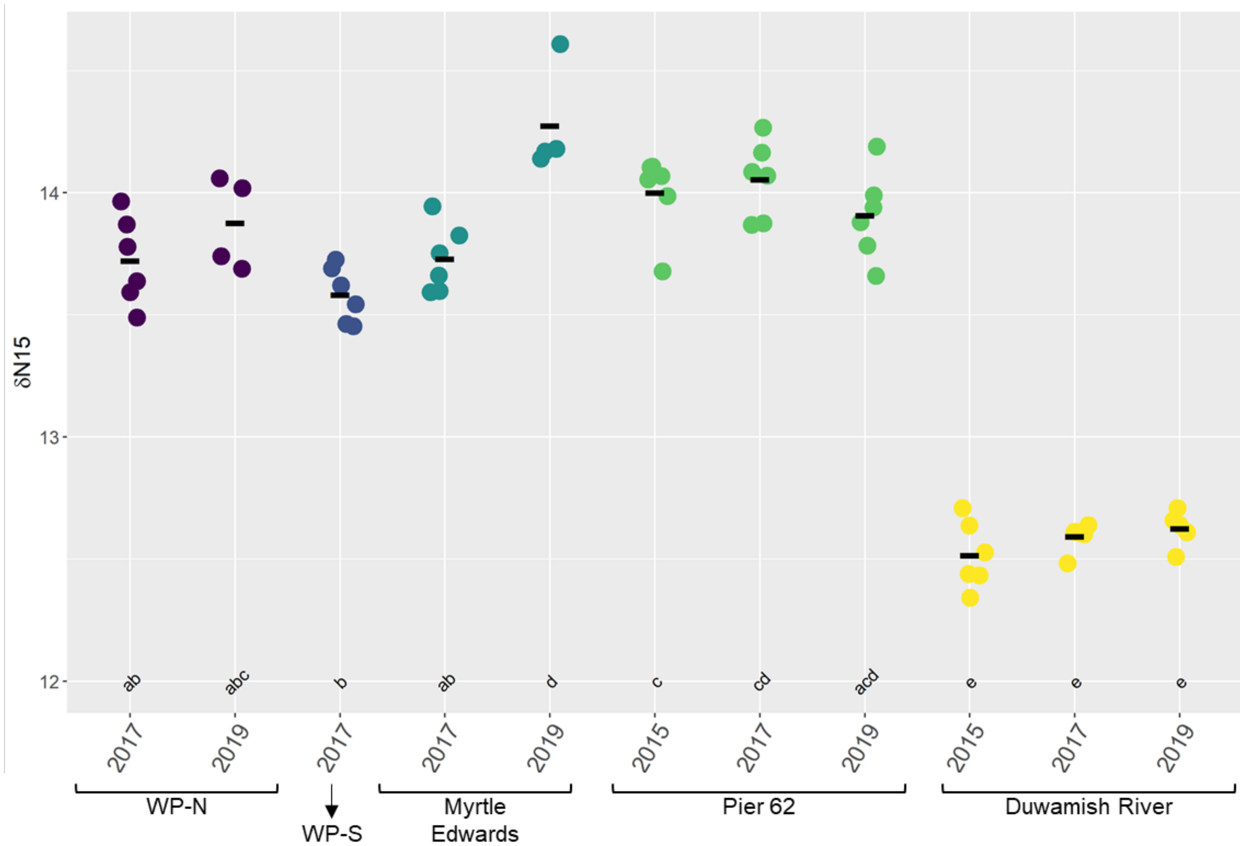


Figure 6. Nitrogen isotope ratios (‰) in English sole muscle from West Point, Elliott Bay, and Duwamish River stations. Stations are arranged from north to south and are denoted by color; horizontal lines repesent mean values; concentrations at locations that don't share letters at the bottom of the graph are statistically different ($p<0.05$).

4.1.2 Carbon isotopes

In 2017 (year 0), $\delta^{13}C$ at West Point N was statistically similar to Myrtle Edwards and Pier 62 (Figure 7), but all were significantly lower (i.e., more depleted) than West Point S. $\delta^{13}C$ from 2019 (year 2) West Point N was statistically lower than the 2017 value, but similar to values from the 2015 (pre-event), 2017, and 2019 Pier 62 station. Similar to West Point N, mean Myrtle Edwards results were lower in 2019 than 2017 although results were not significantly different. The 2019 West Point N samples were also similar to 2017 Myrtle Edwards, but lower than 2019 Myrtle Edwards; the similarity between years may be due to the higher variability in 2017 vs. 2019 Myrtle Edwards samples.

The elevated (enriched) $\delta^{13}C$ at West Point S in 2017 may indicate there was a different source of carbon in the food web (e.g., greater influence of pelagic production) in this area relative to West Point N and the Elliott Bay stations. This pattern supports the grouping of stations described above that considers West Point S (Group 2, lower exposure) distinct from West Point N (Group 1, higher exposure) in potential for English sole exposures. Similarities in $\delta^{13}C$ between Pier 62 (Group 2) and Myrtle Edwards (Group 1) do not support the idea that Pier 62 is distinct from Myrtle Edwards, at least not in the carbon sources in Elliott Bay. Finally, the significantly lower carbon isotope ratios measured at the Duwamish station (Group 2) are consistent with greater freshwater input at that location relative to the more marine-influenced environments of Elliott Bay and the West Point area.

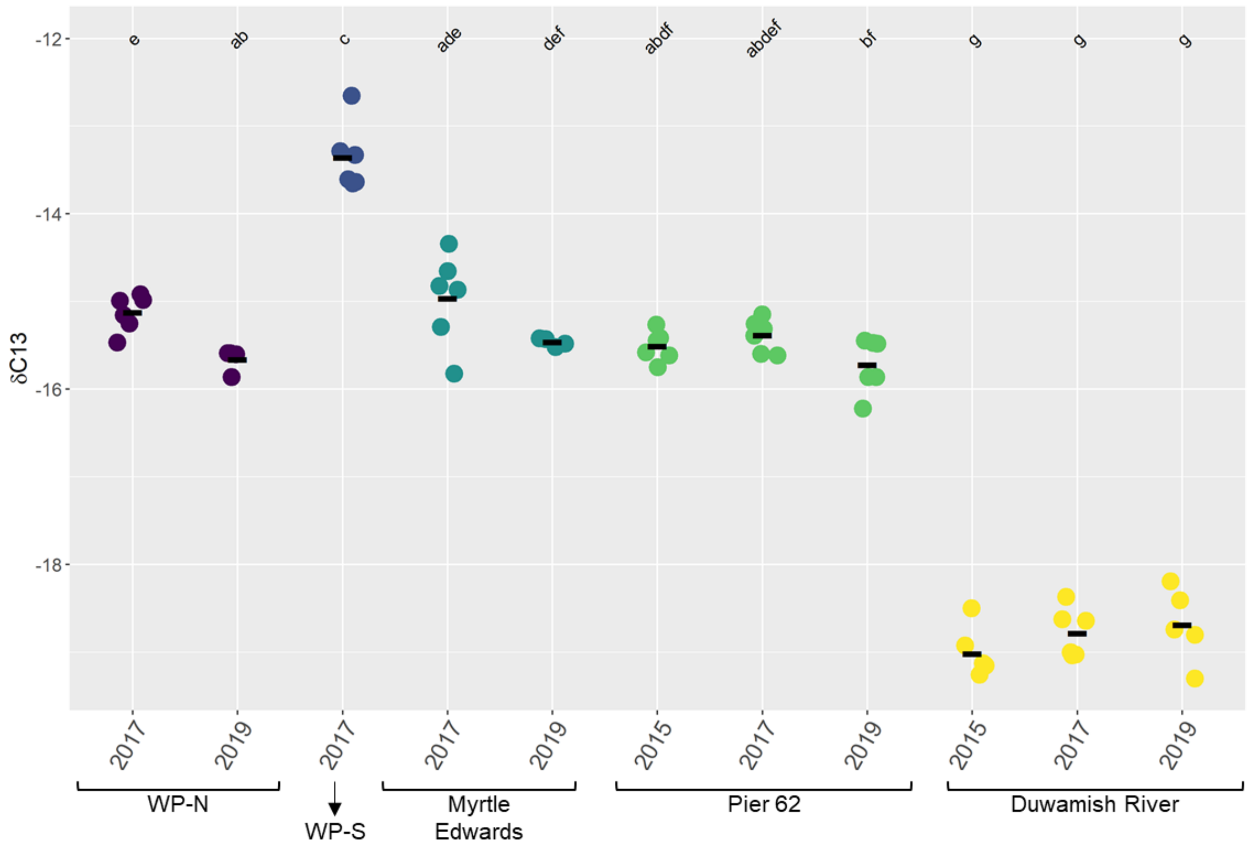


Figure 7. Carbon isotope ratios (‰) in English sole muscle from West Point, Elliott Bay, and Duwamish River stations. Stations are arranged from north to south and are denoted by color; horizontal lines are mean values; concentrations at locations that don't share letters at the top of the graph are statistically different ($p<0.05$). Less negative $\delta^{13}C$ are considered enriched; more negative $\delta^{13}C$ are considered depleted.

4.1.3 Stable Isotopes - Conclusions

Evidence from the stable isotope data suggests the trophic positions of English sole from all the sample stations were similar. Differences in mean $\delta^{15}N$ were not high enough to indicate changes in trophic position between sites. Carbon sources in the diets of English sole also appear to be similar between stations, with the distinct exception of sole from the Duwamish River and possibly West Point S. Differences in carbon in the Duwamish River are expected due to the relative influence of the marine and freshwater environments among the sampled habitats. Higher $\delta^{13}C$ and lower $\delta^{15}N$ in English sole from the West Point S location suggest those fish experience different conditions (e.g., more oceanic influence) and/or a slightly more pelagic-based food web than the other fish.

Overall, the similarity in trophic position and energy sources amongst stations suggests that English sole captured at the Myrtle Edwards (Group 1, higher exposure) and Pier 62 (Group 2, lower exposure) stations are not ecologically distinct groups. Further, $\delta^{13}C$ results for the West Point S and the Duwamish sole support the grouping of those stations into Group 2, which considers these locations as likely having been less or minimally affected by the West Point flooding event.

4.2 Metals and Organic Chemicals in Muscle Tissue

Results and analysis of muscle tissue samples analyzed for metals and organic compounds are presented in this section. To investigate whether the West Point flooding incident may have led to increases in exposures of English sole to metals or organic chemicals at the stations sampled in 2017 (flooding event, year 0), we compared muscle tissue (fillet) chemistry across sampling locations: from the Group 1, higher exposure (West Point N and Myrtle Edwards), Group 2, lower exposure (West Point S, Shilshole, Alki, Pier 62, Harbor Island, and Duwamish), Group 3, background conditions (Eagle Harbor, Sinclair Inlet, Commencement Bay, Port Gardner), and Group 4, reference conditions (Quartermaster Harbor, Nisqually Bay, Hood Canal, Vendovi and Strait of Georgia) stations in 2017.

If spatial differences were seen in 2017 (year 0), we evaluated whether differences were unique to that year. To do this, we compared concentrations within stations between years. This approach addressed questions such as: were concentrations in 2017 similar to those in 2015 (pre-flooding event), and were they different from values in 2019 (post flooding event year 2)? Finally, if we observed temporal differences at West Point N or Myrtle Edwards (Group 1) stations, we compared those differences to the degree of variation between years at the other stations. In this last step, we explore whether the interannual variance was unique to the Group 1 locations or occurred elsewhere as well.

All concentrations are presented on a wet weight basis unless otherwise noted, consistent with existing monitoring data and with fish consumption advisory screening levels. Mean values are available in Appendix B.

4.2.1 Metals

Metals were only analyzed in samples from West Point N and Myrtle Edwards (Group 1), West Point S (only sampled in 2017, year 0), Shilshole, Alki, Pier 62, Harbor Island (Group 2), and the Quartermaster Harbor (Group 4) stations (Table 1). Therefore, the spatial and temporal comparison for metals results was limited to those locations.

4.2.1.1 Infrequently Detected Metals

Of 15 metals analyzed, nine were detected in two or more English sole samples. Antimony and thallium were not detected in any samples. Barium, molybdenum, nickel, selenium, and vanadium were analyzed only in 2017 (year 0) and are discussed in an earlier report (King

County 2021). Silver was analyzed in all three years but was not detected in any samples. Cadmium was analyzed in all three years but was not detected from any West Point N or Myrtle Edwards (Group 1), or the West Point S (Group 2) samples. Cadmium was detected at Shilshole (Group 2) in 2/2 samples from 2015 at 0.0067 and 0.0072 mg/Kg. It was also detected at Quartermaster Harbor (Group 4) in 2015 (pre-event) and 2017 (4/4 samples each year); mean values were 0.00781 mg/Kg in 2015 and 0.00585 mg/Kg in 2017. Due to a lack of detects at all or most locations, the metals mentioned above are not included in the statistical analyses.

Of the remaining metals, eight were elevated in West Point effluent during the emergency bypass and the recovery period compared to historical conditions when the treatment plant was fully operational: arsenic, barium, chromium, copper, lead, mercury, nickel, and zinc (King County 2018). Therefore, although bioaccumulation of metals by fish is a complex process and does not follow simple and predictable patterns (USEPA, 2004; 2007), it is possible discharges from the West Point flooding event may have increased English sole exposure to these metals compared to previous years. Aside from barium and nickel, discussed in an earlier report (King County 2021), these metals are examined further in this section.

4.2.1.2 Metals

The following statistically robust findings suggest that the West Point flooding event had no effect on English sole muscle tissue chemistry:

- Arsenic:
    - There was no difference between the 2017 and 2019 (year 2) West Point N arsenic concentrations.
- Chromium:
    - Median chromium concentrations from the West Point N and S stations (Groups 1 and 2) were not significantly different from one another in 2017 and there was no difference in chromium between 2017 and 2019 at West Point N.
    - We might expect to see higher chromium concentrations in 2017 samples from West Point N and Myrtle Edwards if the West Point flooding event affected chromium exposure of English sole. However, chromium concentrations were higher at most locations in 2015 than 2017, suggesting the West Point flooding event did not elevate English sole exposure to chromium beyond concentrations that occurred prior to the event.
- Mercury:
    - There was no difference in mercury between 2017 and 2019 at West Point N.

There is some indication the West Point flooding event affected English sole muscle tissue chemistry by increasing the concentrations of mercury in fish tissues from West Point North in 2017: West Point N mean mercury concentrations were higher than West Point S in 2017. This difference is likely due to one of exposure rather than food web position since there was no difference in $\delta^{15}N$ values between English sole from the two locations, (Section 4.1.1). Whether it reflects relatively short exposure from the West Point flooding event discharge or a longer-term exposure is unknown.

There is also evidence of higher concentrations of chromium in fish tissues from Myrtle Edwards (Group 1) in 2017 compared to 2019. However, concentrations from Myrtle Edwards in 2017 were similar to those found in 2015 (pre-event). Additionally, chromium concentrations were higher at Harbor Island and Alki in 2015 than 2017, suggesting the West Point flooding event did not elevate exposure to chromium beyond concentrations that occurred prior to the event in Elliott Bay.

From this evaluation, the weight of evidence supports a conclusion that there was no effect on arsenic, chromium, and mercury concentrations in English sole muscle tissue as a direct result of the West Point flooding event.

For all other metals detected in English sole muscle tissue that were also elevated in West Point effluent during the emergency bypass and the recovery period (barium, copper, lead, nickel, and zinc), low statistical power prevents statistically robust conclusions. Though it does not appear the West Point flooding event had a significant influence on copper, lead or zinc concentrations in English sole muscle in the vicinity of West Point or Elliott Bay, given the higher risk of Type II error for multiple comparisons in these datasets we do not draw conclusions for those metals.

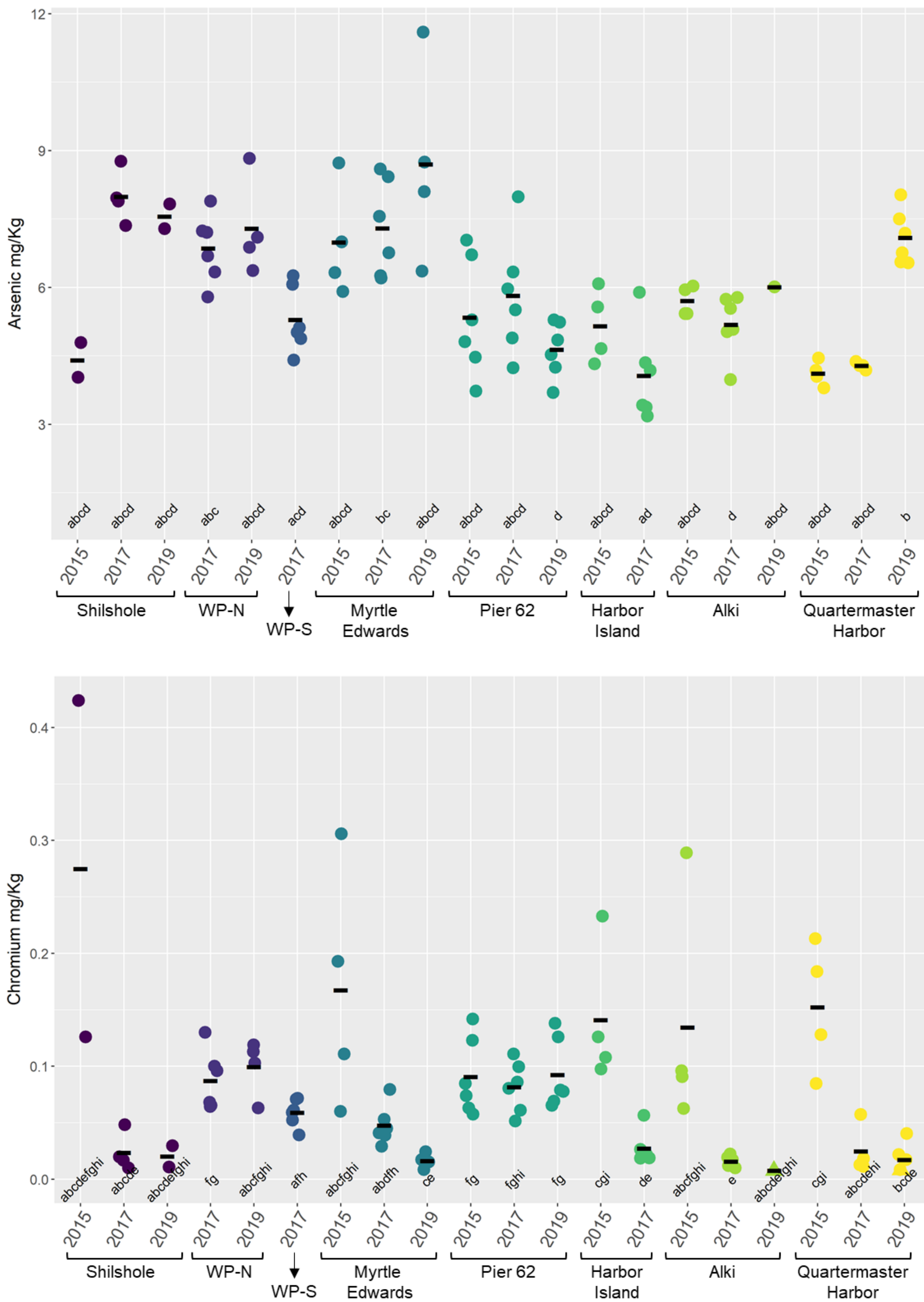


Figure 8. Arsenic (top) and chromium (bottom) concentrations in English Sole samples. Dots represent detected values, triangles represent detection limits of nondetects; stations are denoted by color; black lines represent arithmetic mean values; concentrations at locations that don't share letters at the bottom of each graph are statistically different (p<0.05).

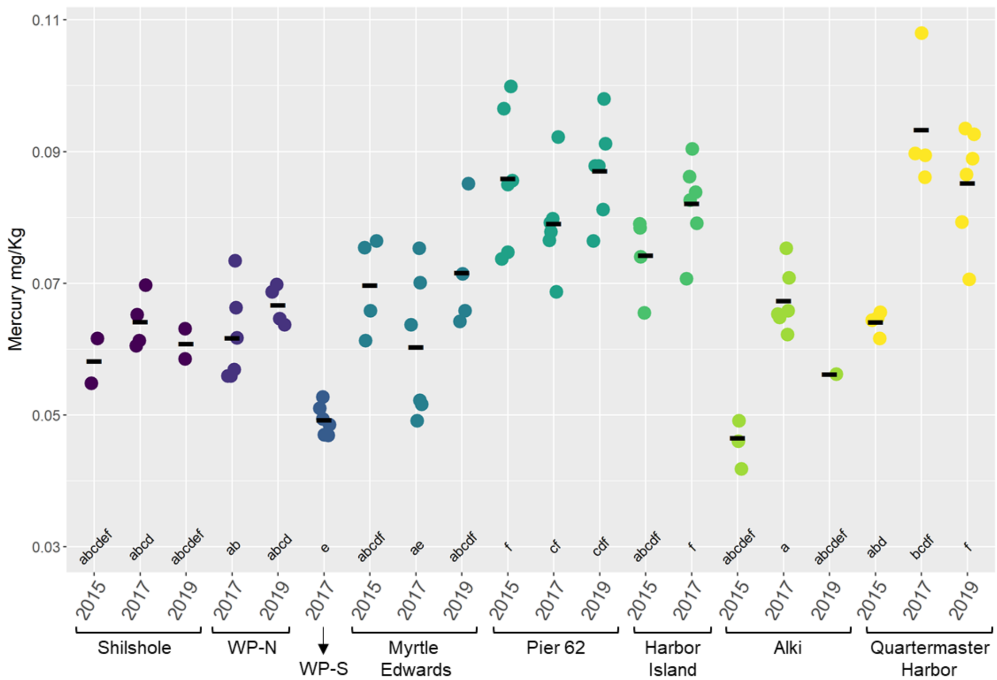


Figure 9. Mercury concentrations in English Sole samples. Stations are denoted by color; black lines represent arithmetic mean values; concentrations at locations that don't share letters at the bottom of each graph are statistically different (p<0.05).

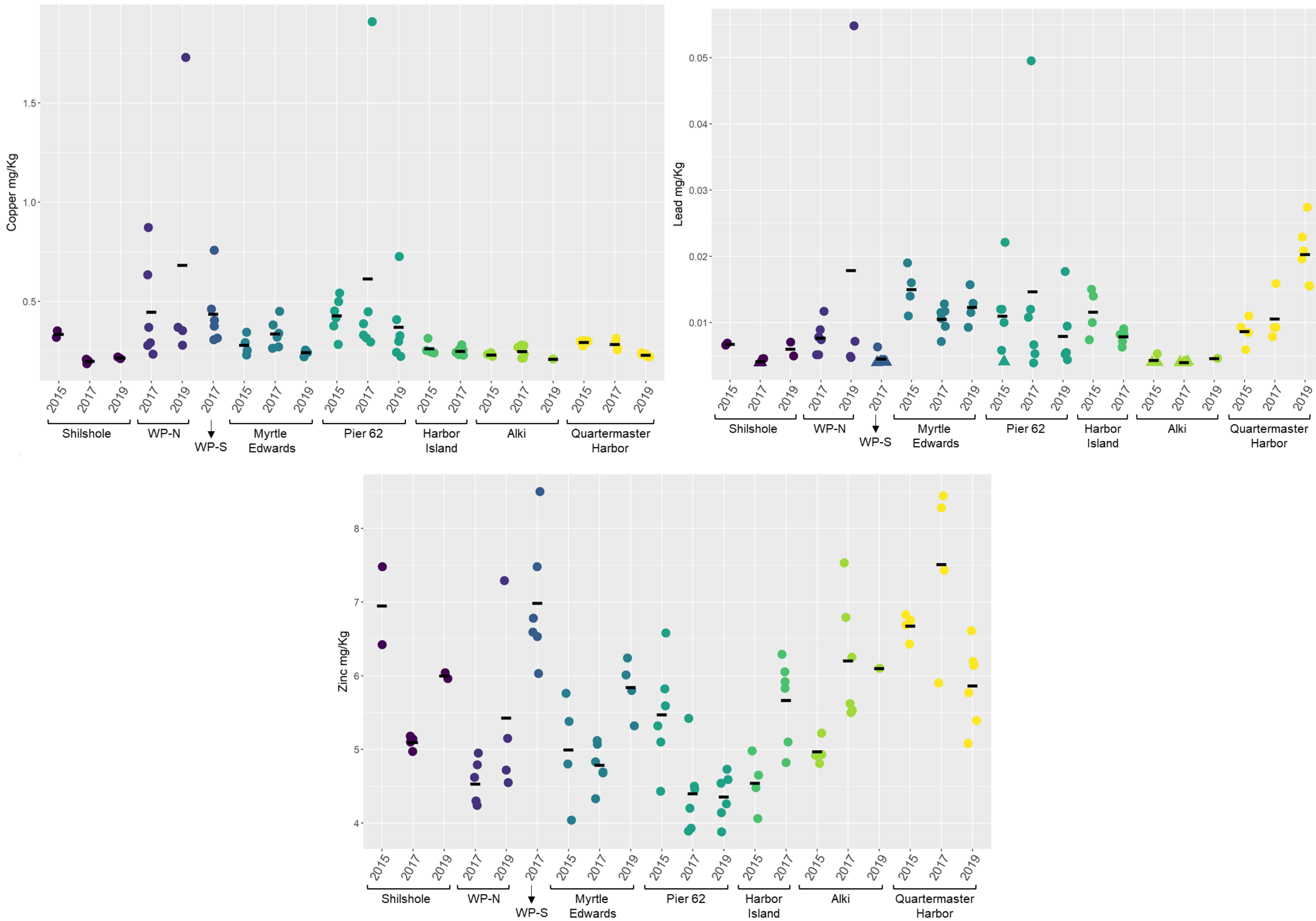


Figure 10. Copper, lead (top) and zinc (bottom) in English Sole samples, with no significant differences (p = 0.05) among any stations/years sampled. Stations are arranged from north to south; dots represent detected values, triangles represent detection limits of nondetects; stations are denoted by color; black lines represent arithmetic mean values.

4.2.2 Organic Chemicals

Organic chemicals were analyzed in all King County and WDFW English sole muscle tissue (i.e., fillet) composite samples. Organic chemicals evaluated were total PCBs, total PBDEs, and total DDTs. Because of low detection frequency, statistical analyses were not performed on results for chlordanes in English sole tissue. Due to differences in analytical methods for PCBs, PBDEs, and DDTs between King County and WDFW (Section 3.3.2), results from each of the two agencies for these chemicals are analyzed separately.

4.2.2.1 Total PCBs

PCB concentrations in fish tissues will increase in a matter of weeks in response to an increase in the exposure of the fish to PCBs in sediment, sediment porewater, water, and/or the foods of the fish (Rubenstein et al. 1984; Kobayashi et al. 2011; Fadaei et al. 2015). Relevant studies typically report concentrations in whole fish, not muscle tissue, but the evidence suggests PCBs are assimilated by fish on a time scale compatible with the sampling efforts in the current study (Section 3.1.1.). Specifically, Kobayashi et al. (2011) fed marbled sole (*Pseudopleuronectes yokohamae*) worms contaminated via exposure to contaminated sediments for 28 days. Following an additional 56 days eating clean worms, assimilation efficiencies (AEs) for 84 PCB congeners ranged from 0.21 to 0.78, with those having higher octanol-water partitioning coefficients (K_{OW}) also generally showing higher AEs. Fadaei et al. (2015) demonstrated that reducing dissolved PCBs (considered more bioavailable than those in food or sediment matrices) by 95% using a sediment amended with activated carbon, resulted in a corresponding 87% decrease in the PCB concentrations in whole zebrafish (*Danio rerio*) over 45 days. These studies show that uptake rates and assimilation efficiencies from either food or water (through ventilation) are sufficiently fast to result in measurable changes to PCB tissue concentrations over a period of weeks to months, consistent with our sampling design (Section 3.1.1).

To determine whether the West Point flooding event may have led to significant increases in total PCBs in English sole, concentrations in muscle tissue composites were compared across locations and years using the sum of homologs for King County data, and WDFW's summation of congeners method, as shown in two separate panels in Figure 11. If there had been a substantial impact from the West Point flooding event, we would have expected to see significantly higher concentrations of PCBs in English sole sampled from one or both of the Group 1 stations (West Point N and Myrtle Edwards) in 2017 (year 0) compared to the same location in 2019 (year 2) or 2015 (pre-event). We also would expect to see higher concentrations in the Group 1 (higher exposure) stations compared to the Group 2 (lower exposure), Group 3 (background conditions), and Group 4 (reference conditions) stations.

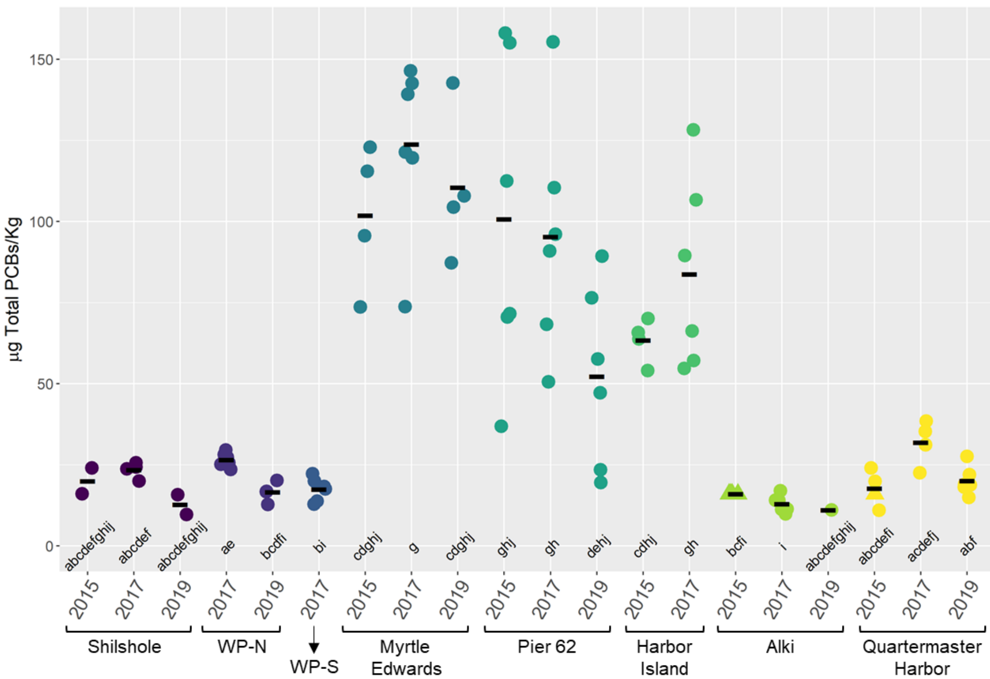
As an additional line of evidence, we evaluated whether sample variance may have been affected by the West Point flooding event by comparing year-to-year homogeneity of total PCB concentrations (Appendix E). We may have expected to see greater variance in PCB concentrations if there was a significant impact from the West Point flooding event, as some of the fish in composites would have been affected and some unaffected (instead of all unaffected).

Mean total PCB concentrations were significantly higher at West Point N compared to West Point S in 2017, and at West Point N in 2017 compared to 2019. By 2019 the mean total PCB concentration at West Point N dropped and was not significantly different from that at West Point S in 2017. This pattern is observed in both King County and WDFW PCB results. These results could be interpreted to indicate that the West Point flooding event caused a localized and temporary increase in total PCBs in English sole muscle tissue. However, concentrations similar to those from West Point N in 2017 were observed at Quartermaster Harbor (Group 4) in 2017 and at Shilshole (Group 2) in 2019. In addition, a similar (though non-significant) pattern of higher to lower concentrations from 2017 to 2019 occurred at Quartermaster Harbor and Shilshole. Though these differences were considered not significant, based on our uncertainty assessment we have less confidence in the findings for those comparisons (Appendix D).

Year-to-year homogeneity of total PCB concentrations (i.e., sample variance) was not significantly different between the King County stations measured in 2015 (pre-event), 2017 (year 0), and 2019 (year 2) and historical variance (data from 2005-2019) at comparable WDFW stations. The West Point incident did not significantly change the distribution of our sample concentrations relative to baseline variability: pairwise Bartlett test with p-level = 0.05, p-adjusted using Benjamini & Hochberg (1995). This suggests the West Point incident did not result in elevated PCB concentrations in English sole muscle tissue.

Within Elliott Bay, the Myrtle Edwards station (Group 1) showed no differences in total PCBs between years in either the WDFW or King County datasets, though based on our uncertainty assessment we have less confidence in the findings for the King County data comparisons (see Appendix D). Based on the WDFW dataset, mean total PCB concentrations at Myrtle Edwards in 2017 were not different from Pier 62. Though there were no significant differences in the King County dataset between Myrtle Edwards, Harbor Island, and Pier 62 in 2017, based on our uncertainty assessment we have less confidence in the findings for those comparisons (see Appendix D). There were also no significant differences between years in total PCBs at the Alki (King County), Pier 62 (KCEL and WDFW), Harbor Island (King County), or Shilshole (King County) stations (all Group 2, lower exposure stations). These results suggest the West Point flooding event did not significantly increase total PCBs in English sole in Elliott Bay.

Although the comparison of total PCBs between West Point N and West Point S in 2017 and the decrease at West Point N in 2019 suggest a possible effect of the flooding event on total PCB concentrations in fish around West Point, the effect was temporary and tissue concentrations were statistically similar to areas presumably unaffected by West Point. Considered in the context of the expected variation seen at other stations and years, the PCB tissue concentration data from the Myrtle Edwards stations does not provide evidence of an influence from the West Point flooding event at that location.



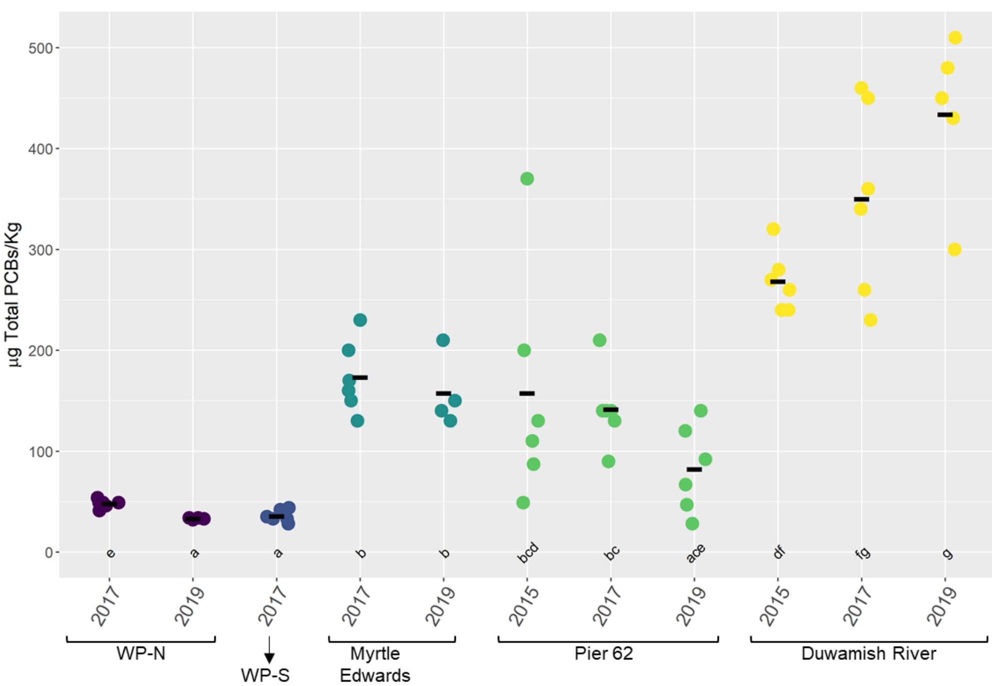


Figure 11. Total PCBs measured as homologues by KCEL for King County (top) and as congeners by NOAA for WDFW (bottom) in English sole from King County monitoring stations. Dots represent detected values, triangles represent detection limits of nondetects; stations are denoted by color; black lines represent mean values; concentrations at locations that don't share letters at the bottom of the graph are statistically different (p<0.05).

4.2.2.2 Total PBDEs

KCEL and WDFW analyze PBDEs separately, and both labs analyze for nine of the same congeners (Tables 5 and 7). In addition to these nine PBDE congeners, WDFW also reports concentrations of BDE-49 and -155; and King County reports five additional congeners: BDE-17, -71, -138, -190, and -209. Due to these differences, results for the sum of detected congeners (total PBDEs) are not directly comparable between the two agencies' datasets.

Fish can rapidly assimilate PBDEs when exposed in water (Mhabdi et al. 2014) or in food (Munschy et al. 2010), with measurable accumulations in various tissues, including muscle, occurring within weeks. However, PBDEs are metabolized in fish via debromination to form less brominated congeners and can be eliminated fairly rapidly. In common sole (*Solea solea*), Munschy et al. (2010) found rates of depuration following removal of exposure at about 1/10 to 1/3 of uptake rates for several common congeners (BDE-28, -47, -99, -100, 153, -209). Assimilation efficiencies of these congeners by the common sole from contaminated food ranged from 1 to 16.1 percent.

Although the bioaccumulation dynamics of PBDEs are more complex than for PCB, we would have expected to see significantly higher concentrations of PBDEs in English sole sampled from one or both of the Group 1 stations in 2017 (year 0) compared to 2019 (year 2) or 2015 (pre-event) if there had been a substantial impact from the West Point flooding event. We would also expect to see higher concentrations in 2017 in the Group 1 (higher exposure) stations compared to the Group 2 (lower exposure), Group 3 (background conditions), and Group 4 (reference conditions) stations if there had been a large impact.

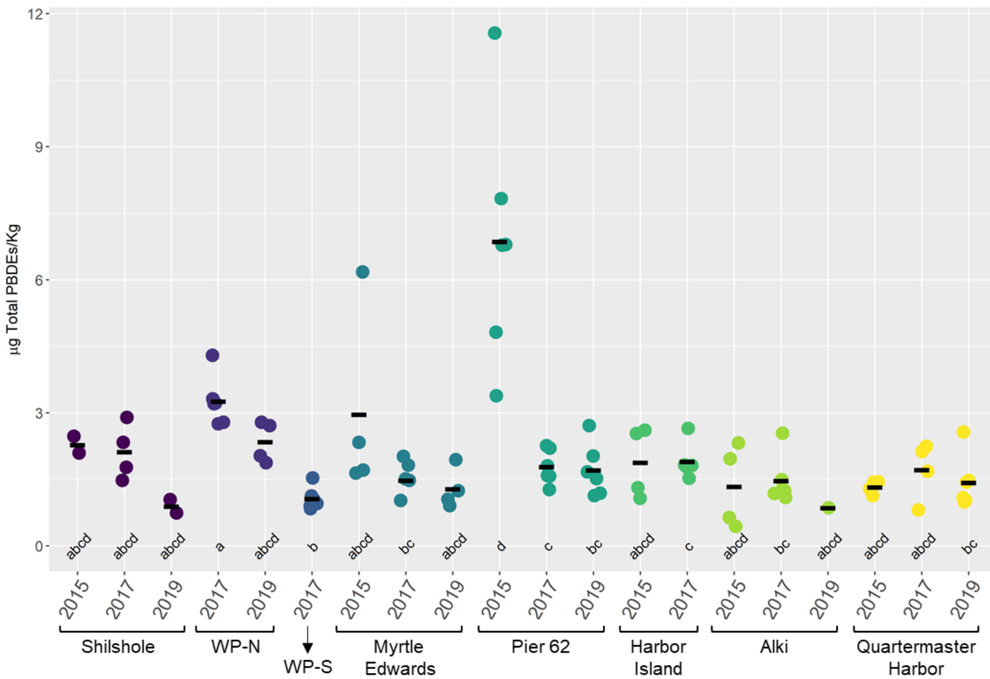
The mean total PBDEs was significantly higher at West Point N in 2017 compared to 2019 in the WDFW dataset. In both the King County and WDFW datasets, the mean total PBDE concentration at West Point N[8] (Group 1) in 2017 was significantly higher than in samples from West Point S (Group 2) in the same year. By 2019, the mean total PBDE concentration at West Point N decreased and was no longer significantly different from West Point S in 2017. However, a similar (though non-significant) pattern of higher to lower concentrations from 2017 to 2019 occurred at Quartermaster Harbor and Shilshole. Though the differences were considered not significant, based on our uncertainty assessment we have less confidence in the finding for Shilshole Bay than for Quartermaster Harbor (Appendix D).

Despite their temporal difference from one another, the King County samples from West Point N in 2017 and 2019 had concentrations that were statistically similar to values from two of three sampling years at Quartermaster Harbor (2015 and 2017). Though the concentrations at West Point N in 2017 were also statistically similar to other locations and years, based on our uncertainty assessment we have less confidence in the findings of "no significant difference" for many of those King County data comparisons (see Appendix D).

[8] In 2017, the congener BDE-99 was more commonly detected in West Point N samples than those from other locations but at concentrations below the quantitation limit (J-flagged). Therefore, the exact concentrations are uncertain.

For the WDFW samples, the total PBDE concentrations from 2017 West Point N were similar to all other stations that year, with the exception of West Point S. Though the comparison between West Point N and West Point S in 2017 and the decrease at West Point N in 2019 suggest a possible effect of the flooding event on PBDEs, any such effect was temporary and the resulting tissue concentrations were statistically similar to areas presumably unaffected by West Point.

Within Elliott Bay, the mean total PBDE concentration from Myrtle Edwards (Group 1) was higher in 2017 than 2019 in the WDFW dataset, but similar between years in the King County dataset (Figure 12). Samples from Myrtle Edwards from all years were statistically similar to values from Pier 62, Harbor Island, and Alki (all Group 2) in the King County dataset, though based on our uncertainty assessment we have less confidence in the findings for a few of these comparisons (see Appendix D). WDFW data comparisons suggest concentrations at Myrtle Edwards and Pier 62 were also similar (except Myrtle Edwards 2019 vs. Pier 62 2015); however, based on our uncertainty assessment we also have less confidence in the findings for these comparisons (Appendix D). Based on these results WDFW's data could indicate a localized increase in PBDEs in English sole muscle tissue at Myrtle Edwards. If this occurred, the change was temporary, and the result likely did not raise PBDE concentrations above those in surrounding areas.



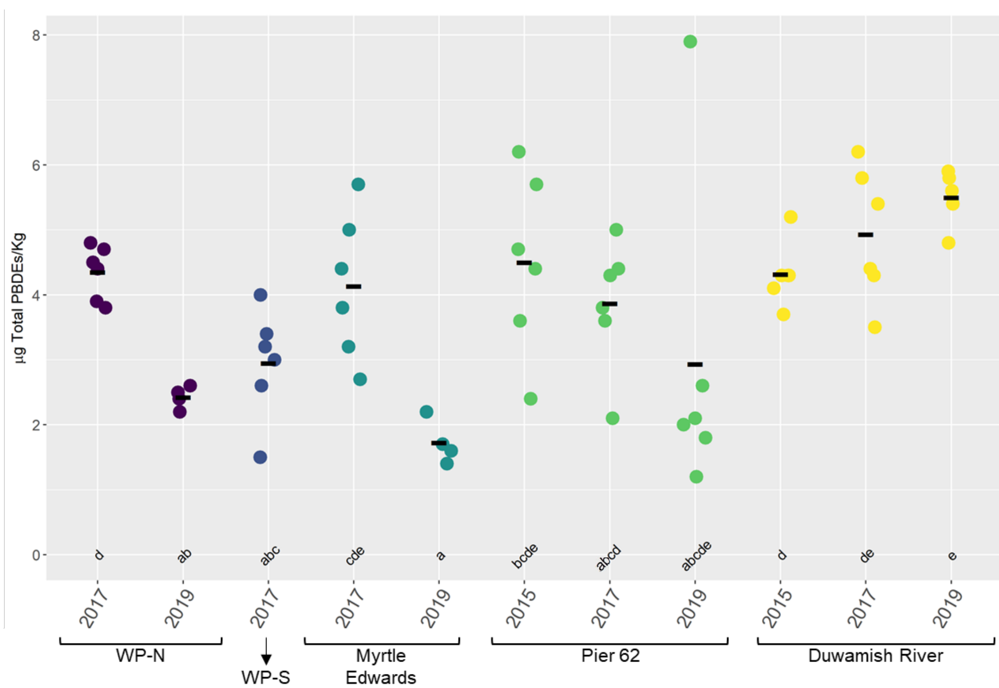


Figure 12. Total PBDEs by KCEL for King County (top) and by NOAA for WDFW (bottom) in English sole from King County monitoring stations. Stations are arranged from north to south; stations are denoted by color; black lines represent mean values; concentrations at locations that don't share letters at the bottom of the graph are statistically different (p<0.05).

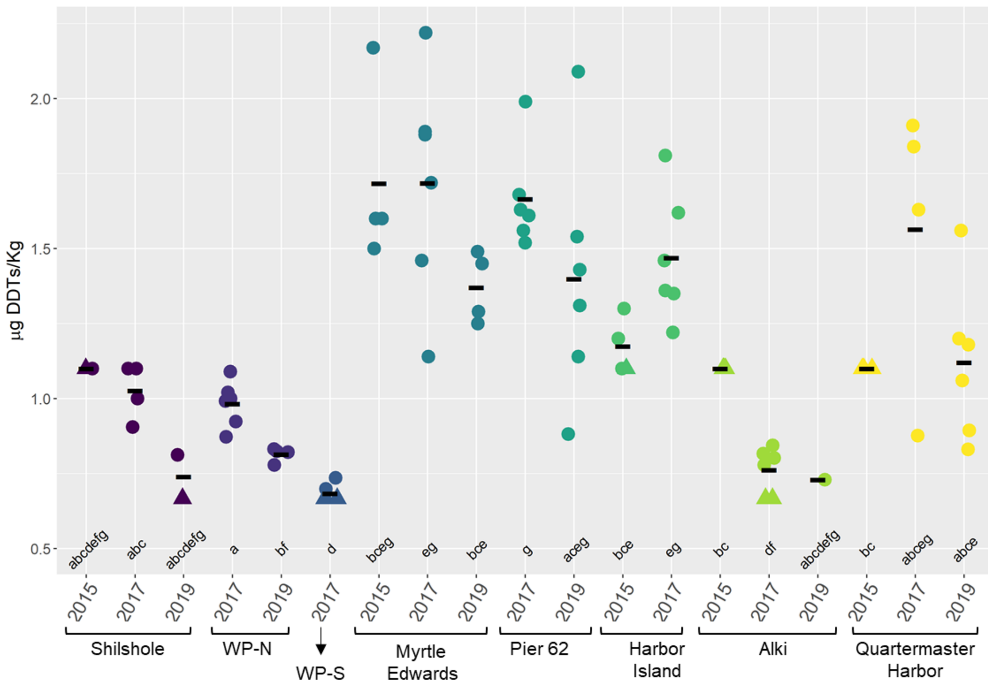
4.2.2.3 Total DDTs

In the discussion below, it is important to note that all 2017 samples from Myrtle Edwards (Group 1) and Quartermaster Harbor (Group 4), two-thirds of samples from 2017 Pier 62 (Group 2), and one of two samples from 2017 Shilshole (Group 2) had elevated QLs for 4,4'-DDT. This may have resulted in an underestimation of total DDTs for those samples, because in calculating sums, non-detects were not included (ND = 0).

Observations for Total DDTs include:

- Both the King County and WDFW data indicate total DDT concentrations in 2017 from the West Point N (Group 1) station were significantly higher than West Point S (Group 2), and values from West Point N were significantly higher in 2017 (year 0) than in 2019 (year 2) for both datasets (Figure 13). Like PBDEs, these results for total DDTs possibly indicate a temporary change in total DDT concentrations in English sole muscle tissue associated with the West Point flooding event. However, like the PCBs and PBDEs, a similar (though non-significant) pattern of higher to lower concentrations from 2017 to 2019 occurred at Quartermaster Harbor and Shilshole. Though these differences were considered not significant, based on our uncertainty assessment we have less confidence in the findings for those comparisons (Appendix D).
- Total DDTs from WDFW's Myrtle Edwards and Pier 62 samples were both statistically higher in 2017 compared to 2019. Though comparisons of total DDTs from the King County dataset for the Elliott Bay stations were not significant, however based on our uncertainty assessment we have less confidence in the findings for those comparisons (see Appendix D).
- In both King County and WDFW datasets, total DDTs at both West Point locations (Groups 1 and 2) were significantly lower than at Myrtle Edwards (Group 1) and Pier 62 (Group 2) in 2017; variability at these Group 1 and 2 Elliott Bay stations is high, but in general, those stations have higher total DDTs than outside of Elliott Bay (Groups 3 and 4, background and reference conditions).

Results for DDTs are similar to results for the other detected organic compounds, where within-year variability is high in a number of the stations, especially those from Elliott Bay in the King County dataset. Like the PCB and PBDE results, this study finds a significant difference between DDT concentrations in 2017 vs 2019 at West Point N. Because interannual variability was lower at the West Point stations, we have higher confidence in the findings for those comparisons. Though the WDFW dataset shows a significantly higher concentration at the Myrtle Edwards (Group 1) station in 2017 compared to 2019, this same pattern is seen at the Pier 62 (Group 2) station in that dataset as well. In fact, the total DDT concentration at Pier 62 was higher in both 2015 and 2017 compared to 2019. This suggests sources other than the West Point flooding event may be influencing DDT concentrations within Elliott Bay.



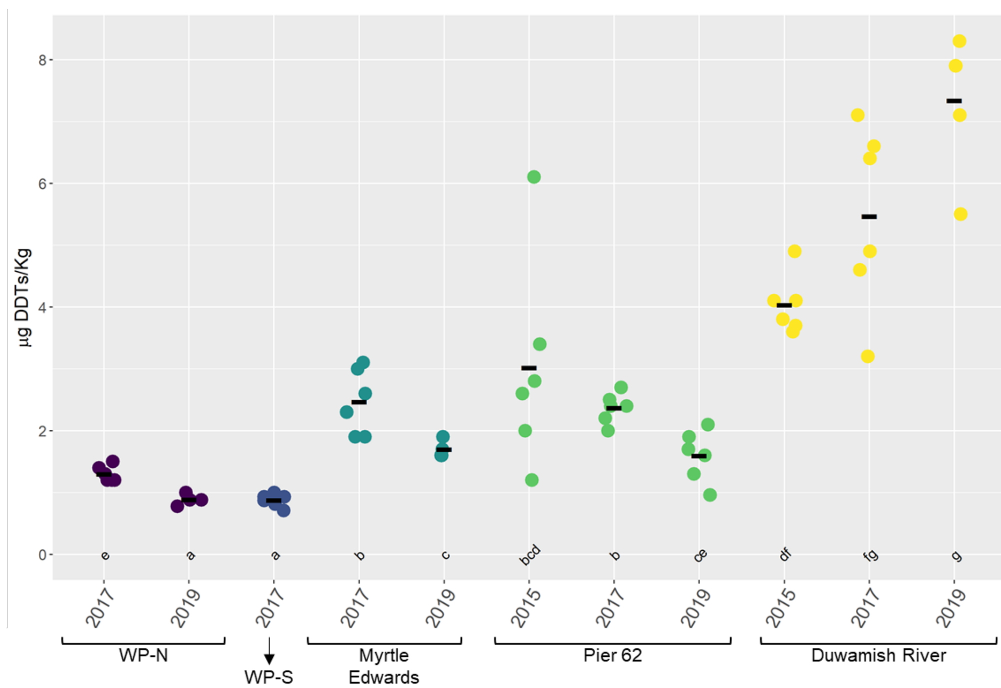


Figure 13. Total DDTs by KCEL for King County (top) and by NOAA for WDFW (bottom) in English sole from King County monitoring stations. Stations are arranged from north to south; stations are denoted by color; black lines represent mean values; dots represent detected values, triangles represent detection limits of nondetects; concentrations at locations that don't share letters at the bottom of the graph are statistically different (p<0.05).

4.2.3 Muscle Tissue Metals and Organics – Conclusions

If there had been a substantial impact from the West Point flooding event and no storm-related variation affecting samples elsewhere in the study area, we would have expected significantly higher concentrations of metals and organic chemicals in English sole sampled from one or both Group 1 (higher exposure) stations in 2017 (year 0) compared to Group 2 (lower exposure), Group 3 (background conditions), or Group 4 (reference conditions) stations. If concentrations were higher at Group 1 stations due solely to the West Point flooding event in 2017, we might have also expected to see those values go down in 2019 (year 2). If an effect of the West Point flooding event on fish tissue was more subtle, sparse data and low statistical power would prevent this study from detecting it.

In our analysis of metals data, we did not see any significant between-station differences in metals concentrations in English sole muscle tissue that indicated a response specifically attributable to the West Point flooding event. This may be because there was no effect on muscle tissue metals concentrations. However, based on our uncertainty assessment we have less confidence in the findings for most comparisons involving the Elliott Bay sites (Myrtle Edwards, Pier 62, and Harbor Island) for arsenic, chromium, copper, lead, mercury, and zinc (See Section 3.3.3.4 and confidence interval results for metals in Appendix D). Therefore, the lack of significant between-stations differences may be because this monitoring study had low statistical power to detect changes in metals concentrations in English sole muscle tissue.

For organic contaminants, there is some evidence of higher concentrations in fish from West Point N and Myrtle Edwards (the Group 1, higher exposure stations) in 2017. Although these comparisons suggest a possible effect of the flooding event on some organic contaminant concentrations in fish around West Point, the effect was temporary and tissue concentrations were often statistically similar to areas presumably unaffected by West Point. Due to a lack of long-term historical data at the West Point N location and the multiple sources of variation in the dataset noted earlier, we cannot conclude with certainty if the higher concentrations of organic contaminants in 2017 and 2019 are attributable solely to the West Point flooding event. In other words, the findings from this study are not definitive. If the West Point flooding event did impact the concentrations of PCBs, PBDEs, and DDTs in English sole, the effect was likely temporary and tissue concentrations were statistically similar to areas presumably unaffected by West Point.

High variability is common in environmental chemistry datasets, especially when sample sizes are low. The English sole tissue data used in these analyses likely reflect multiple sources of variation occurring simultaneously, which makes it difficult to demonstrate with confidence either the presence or the absence of an effect on English sole exposures due to a specific pollutant source. For example, the variability in bioaccumulative organic compounds in English sole at the Group 2 (lower exposure) locations may reflect the presence of multiple and diffuse sources of PCBs, PBDEs and DDTs to benthic habitats throughout Central Puget Sound, the mobility of the fish within those habitats, or some combination of these and other factors.

In addition, there is a larger context of pollutant loading that occurred in 2017. During February and March of 2017, the Puget Sound region experienced record-breaking high precipitation (PSEMP, 2018) that may have subjected English sole from some monitoring stations to relatively higher chemical contaminant exposures. The spring rainfall intensity combined with temporary flow diversions within the combined sewer system to reduce the stormwater volume received at West Point during the repair period, resulted in more discharges from wet weather treatment stations (WWTSs), and to some extent CSOs, than typical between February and April 2017 (King County, 2020). The Alki, Carkeek, Elliott West, and Henderson/Martin Luther King WWTSs experienced increased flows and events from flow management because of West Point's limited capacity.

Because of multiple potential chemical inputs, any effect specifically from the West Point flooding event would need to be substantial to be detected amid both normal variation in fish tissue chemistry and that induced by widespread mobilization of chemical contaminants simultaneous to the West Point flooding event. The English sole muscle chemistry data do not and could not have provided conclusive evidence of effects on benthic fish tissue quality from the West Point flooding event.

4.3 Biomarkers, Xenoestrogens, and PAH-metabolites

Data gathered by WDFW on the occurrence of vitellogenin (Vtg) in blood, and concentrations of xenoestrogens (exogenous substances that act as estrogen mimics) and PAH-metabolites in bile are presented in this section. All xenoestrogen and PAH-metabolite concentrations are presented on a wet weight basis unless otherwise noted, consistent with WDFW's prior reporting on data of this type.

4.3.1 Vitellogenin in Blood

Vitellogenin is an egg yolk protein produced by oviparous animals in response to steroid estrogens and estrogenic compounds. In English sole, this protein normally only occurs in sexually mature females with developing eggs. However, because males can synthesize Vtg when exposed to environmental estrogens, Vtg induction in male fish is a useful indicator of xenoestrogen exposure. In addition, the slow clearance of Vtg protein from blood plasma means fish have measurable levels of Vtg for months after initial exposure to an estrogenic compound, offering the possibility to detect influences that occurred months before measurement (Hemmer et al., 2002; Craft et al., 2004; Schmid et al., 2002).

We have summarized the occurrence (presence/absence) of Vtg in blood plasma samples from male English sole in 2017 (year 0) and 2019 (year 2) to investigate whether English sole were exposed to human estrogens and other estrogenic substances (i.e., xenoestrogens) due to the West Point flooding event. We compared results from stations sampled for this study (West Point N, West Point S, Myrtle Edwards, and Pier 62 - labeled in bold on Table 10) to each other, and to results from WDFW's long-term monitoring stations in Puget Sound for broader context.

Vtg was detected in blood of male English sole from West Point N and Myrtle Edwards (Group 1, higher exposure), and Pier 62 (Group 2, lower exposure) collected in both 2017 and 2019, and in male sole at West Point S (Group 2) in 2017 (Table 10). In 2017, a higher percentage of Vtg-positive males (44%) were detected from West Point N compared to West Point S (17%), Pier 62 (33%), and all the other WDFW stations in Puget Sound (Groups 3 and 4, background and reference conditions) except for Myrtle Edwards (Group 1; 50%) and the Duwamish River (Group 2; 69%). The percentage of Vtg-positive males was also higher at West Point N in 2017 (44%) than in 2019 (10%), suggesting a decline in exposure to environmental estrogens between years at that location. This is consistent with what we would expect to see if the West Point flooding increased the exposure of English sole to xenoestrogens.

Table 10. Locations where English sole were collected in 2017 and 2019, and percentages of male English sole from those sites with detectable vitellogenin (Vtg) in plasma.

Station	Group	2017	2019	Ratio 2017:2019
		% Vtg-positive males	% Vtg-positive males	% Vtg-positive males
West Point N	1	44	10	4.7
Myrtle Edwards	1	50	43	1.2
West Point S	2	17	NS	NC
Pier 62	2	33	52	0.6
Duwamish River	2	69	23	2.9
Port Gardner	3	27	5	5.5
Eagle Harbor	3	25	10	2.5
Sinclair Inlet	3	31	12	2.7
Comm. Bay	3	33	26	1.3
Strait of Georgia	4	0	0	--
Vendovi	4	0	0	--
Port Madison	4	NS	28	NC
Nisqually	4	25	0	--
Hood Canal	4	6	8	0.7

Note: Stations in bold are the focus of this study; stations in italics are part of WDFW's long-term contaminant monitoring program and are included for context.
NS, not sampled.
NC, missing data, ratio not calculated
Comm. Bay – Commencement Bay
'--' - Vtg not detected in males in at least one of the two years sampled.

The ratio of percent Vtg-positive males in 2017 to that in 2019 was 4.7 at West Point N, the highest for the dataset among locations in King County. The only higher value for this parameter was for Port Gardener, an area where water quality is likely also influenced by WWTP effluent. Notably, results for Group 4 locations indicate little to no Vtg in male English sole, in both 2017 and 2019. In the other results for Group 1, 2 and 3 locations, (Table 10) there is generally a higher percent of Vtg-positive males in 2017 than in 2019, except at Pier 62. It is unknown whether the relatively higher percent of Vtg-positive males at West Point N resulted from the flooding event, from the relatively high volumes of CSO discharged due to high rainfall (stormwater) volumes in 2017, or some other factor. Without data from pre-event years against which to compare, each alternative explanation is plausible.

Previous research has described Elliott Bay with the highest percentages of male English sole exhibiting Vtg synthesis relative to other locations in Puget Sound (Johnson et al., 2008). Therefore, we would expect substantial fractions of male English sole sampled from Myrtle Edwards and Pier 62 to have detectable blood levels of Vtg. In WDFW's data (Table 10), the Myrtle Edwards station had a higher percentage of Vtg-positive males in 2017 (50%) than in 2019 (43%), but the difference between years was not as large as at West Point N. Also, the percent of Vtg-positive males from Pier 62 increased from 33% in 2017 to 52% in 2019. Frequency of Vtg synthesis in male fish from Elliott Bay, was similar or lower than that seen in fish from Commencement Bay, another station within an urban embayment (Table 10). Considering the historical occurrence of Vtg-positive male English sole within Elliott Bay itself, it is likely multiple sources (O'Neill et al. 2016), in addition to the Elliott West WWTS, influenced Vtg production in male English sole in Elliott Bay in 2017 and 2019.

A previous study found significant levels of Vtg in male English sole sampled along the Seattle Waterfront and documented alteration of spawn timing in both male and female English sole from Elliott Bay (Johnson et al., 2008). In addition, a more recent study by WDFW identified high values of Vtg-induction in male sole from the Seattle Waterfront and continued altered reproductive timing in female fish from the Seattle Waterfront, likely from exposure to estrogenic chemicals (O'Neill et al., 2016). The authors of these studies speculate the sources and types of xenoestrogens present in Elliott Bay are likely associated with industrial effluent, surface runoff, and CSOs.

4.3.2 Xenoestrogens

Estrogenic compounds (ECs) include natural estrogens produced by vertebrates and xenoestrogens, which are exogenous (foreign) substances that are estrogenic (bind to estrogen receptors), and may act to feminize male fish. The presence of xenoestrogens or high levels of steroid hormones in fish can indicate exposure to industrial, pharmaceutical, human, or animal wastes. Municipal wastewater contains several human-derived hormones, such as the naturally produced female hormones (e.g., 17β-estradiol, estrone, and estriol), as well as synthetic estrogens (e.g., 17α-ethinylestradiol) used in birth control. While female fish produce steroid estrogens (e.g. 17β-estradiol), male fish would not be expected to have high levels of these hormones unless they were exposed in the environment. Municipal wastewater often contains nonsteroidal xenoestrogens that can act as endocrine disruptors. For example, a class of polycarbonate plasticizers called bisphenols are xenoestrogen compounds that mimic estrogen. Due to the ubiquitous use of plastics in homes and businesses, bisphenols are a common contaminant in wastewater and can be found in human waste. These endocrine-disrupting chemicals are commonly and widely detected in water and sediments and can disrupt hormonal and metabolic processes in fish even at relatively low concentrations (Pettersson et al., 2007; Scott et al., 2006a, Scott et al., 2007; Routledge et al., 2008). Because bisphenols do not occur naturally, high levels of these compounds in both male and female fish indicate exposure to xenoestrogens that could negatively impact reproduction.

To investigate whether the West Point flooding incident may have led to increased exposure of English sole to xenoestrogens at Group 1 stations, concentrations of estrogenic compounds in male English sole bile were evaluated from 2017 (year 0). We used two calculated variables in this evaluation:

- Sum of ECs
- 17β-estradiol (E2) equivalent (EEQ) concentrations

In calculating these sums, we assumed chemicals to be absent if reported as not detected (U-qualified) by the laboratory (ND = 0).

If there had been a substantial impact from the West Point flooding event, we would expect significantly higher concentrations in both parameters in English sole sampled in 2017 from West Point N (Group 1, higher exposure) or West Point S or Pier 62 (Group 2, lower exposure) compared to stations from Group 3 (background conditions) and Group 4 (reference conditions) samples that year. In addition, we might have seen higher concentrations in the Pier 62 station in 2017 compared to data from 2015 (pre-event).

A list of the analytes used to evaluate these possible outcomes is in Table 7 and descriptive statistics for each station are provided in Appendix C. We performed qualitative spatial comparisons between the sum of ECs detected in bile of male English sole from West Point N and Myrtle Edwards (Group 1), and West Point S and Pier 62 (Group 2). We compared these values to WDFW monitoring stations also sampled in 2017 (year 0), and to locations sampled by WDFW in 2015 (pre-event), mostly considered Groups 3 and 4 (background and reference conditions) stations in our study.

4.3.2.1 Uncertainty in the Dataset

We conducted our evaluation of the results for xenoestrogens in bile of male fish recognizing that there are sources of uncertainty within this dataset:

- Note that the 2015 (pre-event) analytical method included only bisphenol A (BPA), but four other BPA chemicals were added to the list of analytes in 2017 including bisphenol F (BPA-F), bisphenol AF (BPA-AF), bisphenol S (BPA-S), and tetrabromobisphenol (TB-BPA). The four bisphenol chemicals added in 2017 (year 0) are included in the Sum of Estrogenic Compounds (Section 4.3.2.2) analysis, but not in the Estrogen Equivalents (Section 4.3.2.3) calculations because 17β-estradiol equivalent factors (estrogenic potencies of various compounds relative to that of 17β-estradiol) were not available for those compounds.
- Sampling was conducted in May and June 2017, approximately two months after the start of the flooding event (February 2017). The timing of sampling likely reduced our ability to observe the full effect of the initial flooding release on this endpoint, because these chemicals in bile are short-lived; e.g., the half-life of nonylphenol residues in salmon (Arukwe et al., 2000) and of bisphenol A in male rats is on the order of several days (Kurebayashi et al., 2003). Nevertheless, measurement of this endpoint allows for a better overall characterization of the exposure environment in the West Point and Elliott Bay area and provides context for understanding the other measured endpoints.
- Results for this aspect of the study from 2019 (year 2) were not available for analysis at the writing of this report and will be reported in an addendum when they become available. The 2019 bile data will improve the understanding of between-year variation for xenoestrogens in English sole.

Although these uncertainties limit our ability to draw conclusions specifically about the West Point flooding event, the data has value in describing spatial patterns relevant to future study designs.

4.3.2.2 Sum of Estrogenic Compounds

Figure 14 shows the summed concentrations and relative proportions of ECs detected in male fish in 2015 (pre-event) and 2017 (year 0). Overall, the sum of detected ECs from West Point N and S (Groups 1 and 2) were among the lowest values in 2017 (27.7 and 37 ng/mL bile respectively); only sums from Hood Canal (Group 4; 24.6 ng/mL bile) were lower (Figure 14). The sum of ECs from the 2017 Myrtle Edwards station (Group 1; 119 ng/mL bile) was higher than at West Point stations. Pier 62 (Group 2) had the highest of total ECs in both 2015 (749 ng/mL bile) and 2017 (904 ng/mL bile). In both years, these elevated values were driven by 17β-estradiol, BPA, and estrone. Impacts of elevated ECs in English sole have been observed at Pier 62 for many years (Johnson et al., 2008; O'Neill et al., 2016): this area is near where female English sole have experienced altered reproductive status at least as far back as 1997 (Johnson et al., 2008).

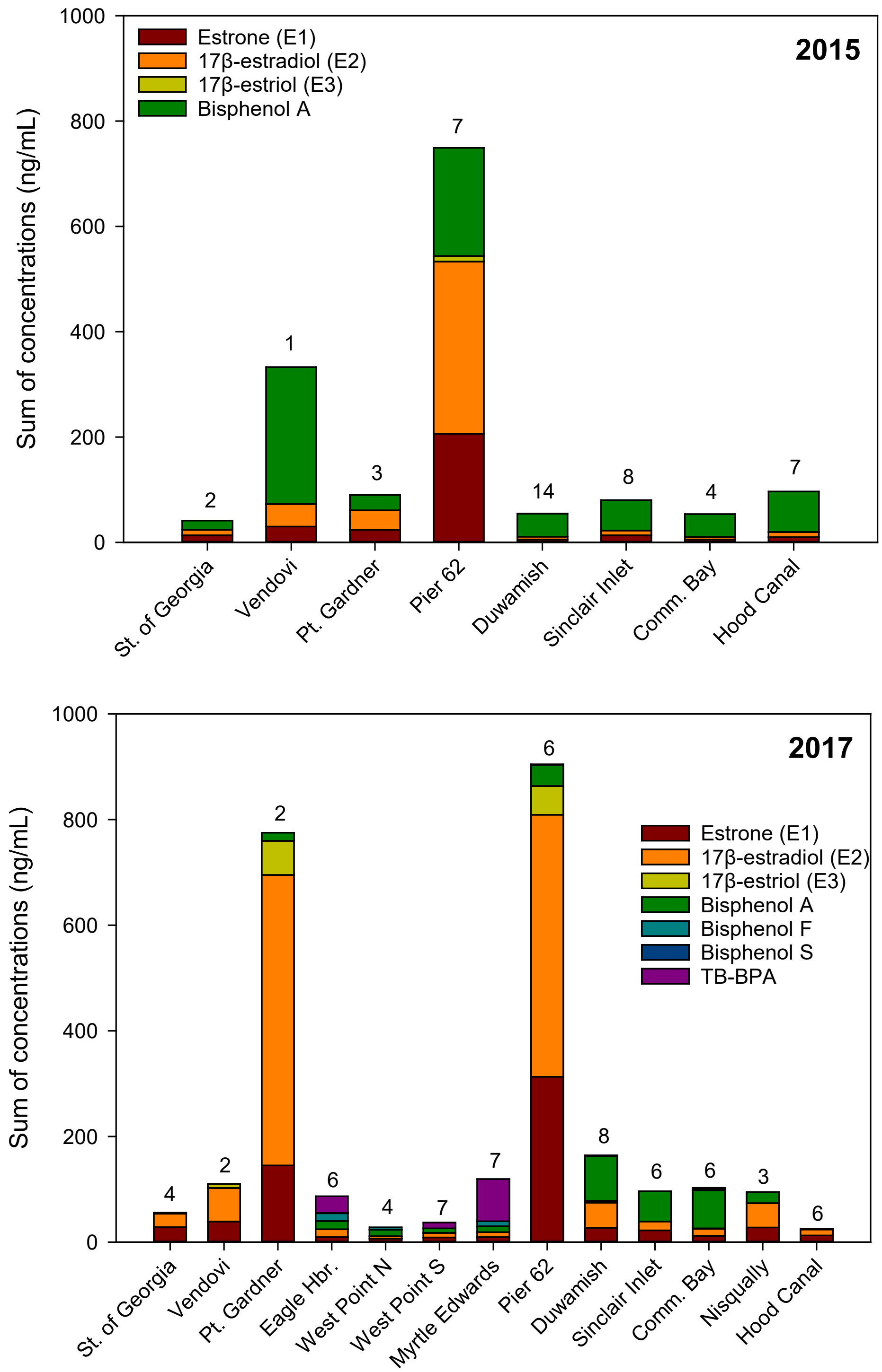


Figure 14. Detected estrogens and bisphenols in male English sole bile at 2015 and 2017 monitoring locations. Bars are summed averages of the concentration of detected compounds at each location, colors indicate the relative proportion of each compound in the sum. Values above each bar indicate the number of individual males. Note that three additional bisphenol chemicals were measured in 2017 compared to 2015. TB-BPA - tetrabromobisphenol. Comm. Bay = Commencement Bay.

4.3.2.3 Estrogen Equivalents

Total estrogenic activity was estimated from the combined concentrations of the ECs detected in each sample (not including BPA-F, BPA-S, or TB-BPA) as EEQ concentrations. EEQs were estimated for each fish by summing the E2 equivalent factors (EEFs) for each estrogenic compound measured in the bile. The EEFs for each EC were based on average values reported by Vega-Morales et al. (2013). The relative differences in estrogenicity among locations (Figure 15) are similar to those in the sums of detected compounds (Figure 14), particularly in 2017 (year 0).

Mean EEQ values in fish from West Point N (Group 1) were the lowest measured in the 2017 data (5.23 ng/mL bile), with West Point S (Group 2) values nearly as low (9.52 ng/mL bile; Figure 15). Values from the Myrtle Edwards (Group 1) station were also relatively low (10.4 ng/mL bile). In contrast, Pier 62 (Group 2) EEQ values were an order of magnitude higher in both 2015 (pre-event; 351 ng/mL bile) and 2017 (year 0; 536 ng/mL bile) than all other stations except Port Gardner (Group 3) in 2017 (573 ng/mL bile), which had the highest value measured that year. Across all samples, the majority of EEQ estrogenicity was accounted for by the natural hormone E2 (EEF = 1). Naturally produced hormones are often the main drivers of estrogenic activity in sewage effluent and receiving water (Desbrow et al., 1998; Rodgers et al., 2000; Aerni et al., 2004). Estrogenicity of estrone and estriol were reported as roughly one-tenth of E2; BPA contributed little to the overall EEQ.

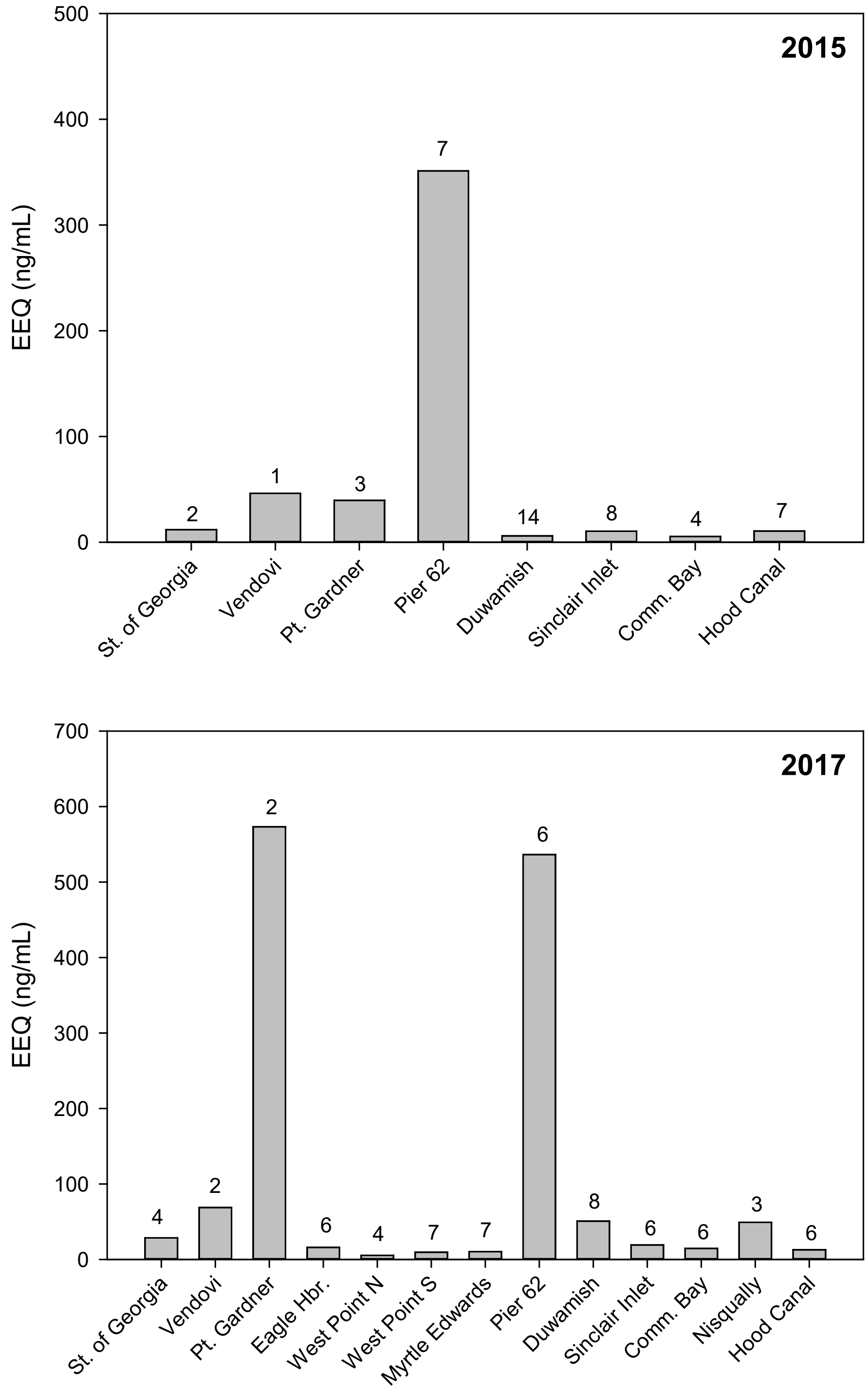


Figure 15. Estimated estrogenic potential of chemicals measured in bile of male English sole at each monitoring location. Bars are summed averages of the EEQs (17β-estradiol equivalents) for E1, E2, E3, and bisphenol A. Values above each bar indicate the number of individual males. Sums do not include EEQs for bisphenol F, bisphenol S, or tetrabromobisphenol A because no EEFs were available to calculate the EEQs for these compounds. EEF values used in calculations are from Table 3 in Vega-Morales et al. (2013). Comm. Bay = Commencement Bay.

4.3.2.4 Xenoestrogens Conclusions

Given the uncertainties of the xenoestrogen data, they cannot provide definitive evidence to address whether the West Point flooding event did or did not elevate ECs or EEQs in bile of male English sole collected in the vicinity of West Point or Elliott Bay in 2017. However, the results suggest insights about male English sole exposure to xenoestrogens that support development of future monitoring approaches, whether or not an event like the West Point flooding event occurs again.

The first observation is that Pier 62 is an estrogenic hot spot that requires further study. Multiple outfalls can deliver treated or untreated sewage to Elliott Bay via CSO discharges from the Duwamish River and from outfalls along the Seattle waterfront. However, if there were such multiple and diverse sources of xenoestrogens causing the higher EEQs in male English sole from the Pier 62 (Group 2) station, one would expect to also see higher values at the nearby Myrtle Edwards (Group 1) station (measured in 2017), which was not observed (Figures 14 and 15). The high values for both ECs and EEQs at the Port Gardner (Group 3) station indicate an association of increased EC and EEQ concentrations with sewage effluent - the Everett WWTP outfalls are situated relatively near that station. Together, the relatively elevated ECs and EEQs at Pier 62 and Port Gardner at a point in time after any effect of the West Point flood event in this endpoint had likely passed, and the absence of this effect at Myrtle Edwards, indicate an exposure condition for English sole and/or source of xenoestrogens at Pier 62 that is analogous to the Everett WWTP effluent. Further investigation is needed in the Elliott Bay area to determine the cause of the relatively high EEQ values in English sole at the Pier 62 station.

The second observation is that %Vtg and short-term expressions of exposure to xenoestrogens describe different phenomena, which may or may not relate to one another. Past studies have found high concentrations of biliary xenoestrogen in sampling locations surrounded by intensively developed areas, especially at the Pier 62 area of the Seattle Waterfront (da Silva et al., 2013; O'Neill et al., 2016). Though WDFW has also noted relatively high levels of Vtg induction in these locations, it has not reported a strong correlation between plasma Vtg and bile xenoestrogens (O'Neill et al., 2016). A lack of correlation between plasma Vtg and plasma E2 in male flounder has been reported previously (Scott et al., 2006b). This lack of correlation might be because the time period that xenoestrogens are measurable in bile may not overlap with the presence of measurable Vtg in plasma. Bile xenoestrogen concentrations reflect relatively short-term exposure (on the order of days); however, plasma Vtg can significantly increase within a week of exposure and can continue to increase up to one week after exposure has ended, depending on exposure magnitude (Hemmer et al., 2002; Craft et al., 2004; Schmid et al., 2002). For these reasons, we did not conduct an analysis of correlations between biliary xenoestrogen concentrations and the occurrence of Vtg in the blood plasma of male English sole. We know that elevated Vtg expression occurs in male English sole from Elliott Bay, and that it adversely affects English sole reproduction (Johnson et al., 2008). Future investigation to define the full range of xenoestrogens in English sole habitat, on their specific sources, and on dose-response relationships with population level response endpoints are necessary before the issue of xenoestrogens in English sole can be effectively addressed. Data allowing extrapolation to other fish species may also support targeted action.

4.3.3 PAH-metabolites

PAH-metabolites were measured in the bile of male and female English sole to investigate if the West Point flooding event may have led to increased English sole exposure to PAHs in the vicinity of West Point or the Elliott West WWTS. Spatial comparisons were made between West Point N, Myrtle Edwards (Group 1) and West Point S and Pier 62 (Group 2) in 2017 (year 0) and concentrations were compared to those from WDFW monitoring stations sampled before the flooding event (2015). Eight additional PAH-metabolites were measured in 2017 compared to 2015 (see Table 7 and Appendix C, Table 13), though four of these metabolites (PHN3carboxylic acid, PHN9carboxylic acid, dihydroxy34dihydro712dimethylBAA, and transdihydroxy45dihydroBEP) were not detected in 2017. Due to laboratory closure related to the COVID-19 pandemic, values from 2019 (year 2) were not available for analysis at the writing of this report and will be reported in an addendum when they become available.

Figure 16 shows the average concentrations of PAH-metabolites detected in 2015 (pre-event) and 2017 (year 0); descriptive statistics for each site are provided in Appendix C. PAH-metabolites in 2017 were highest at the Myrtle Edwards station (1,533 ng/mL bile total), while values at the West Point N and West Point S (Group 1 and 2) stations were some of the lowest that year (383 and 365 ng/mL bile total; Figure 16). The mean value of PAH-metabolites in the Pier 62 (Group 2) fish from 2017 were intermediate at 888 ng/mL bile, which was somewhat lower than values at that station in 2015 (1,250 ng/mL bile). In all cases, the phenanthrene metabolites were the highest measured values, with the fluoranthene metabolites also relatively high at the Myrtle Edwards station in 2017. Overall, it does not appear the West Point flooding event led to higher amounts of PAHs taken up by English sole at West Point N as compared to West Point S. Although the PAH-metabolite values in Myrtle Edwards fish in 2017 are higher than at Pier 62, this difference appears to be within the temporal variation seen at other sites; however, a lack of historical data limits further interpretation. The 2019 (year 2) bile data will improve the understanding of between-year variation for PAH metabolites in English sole.

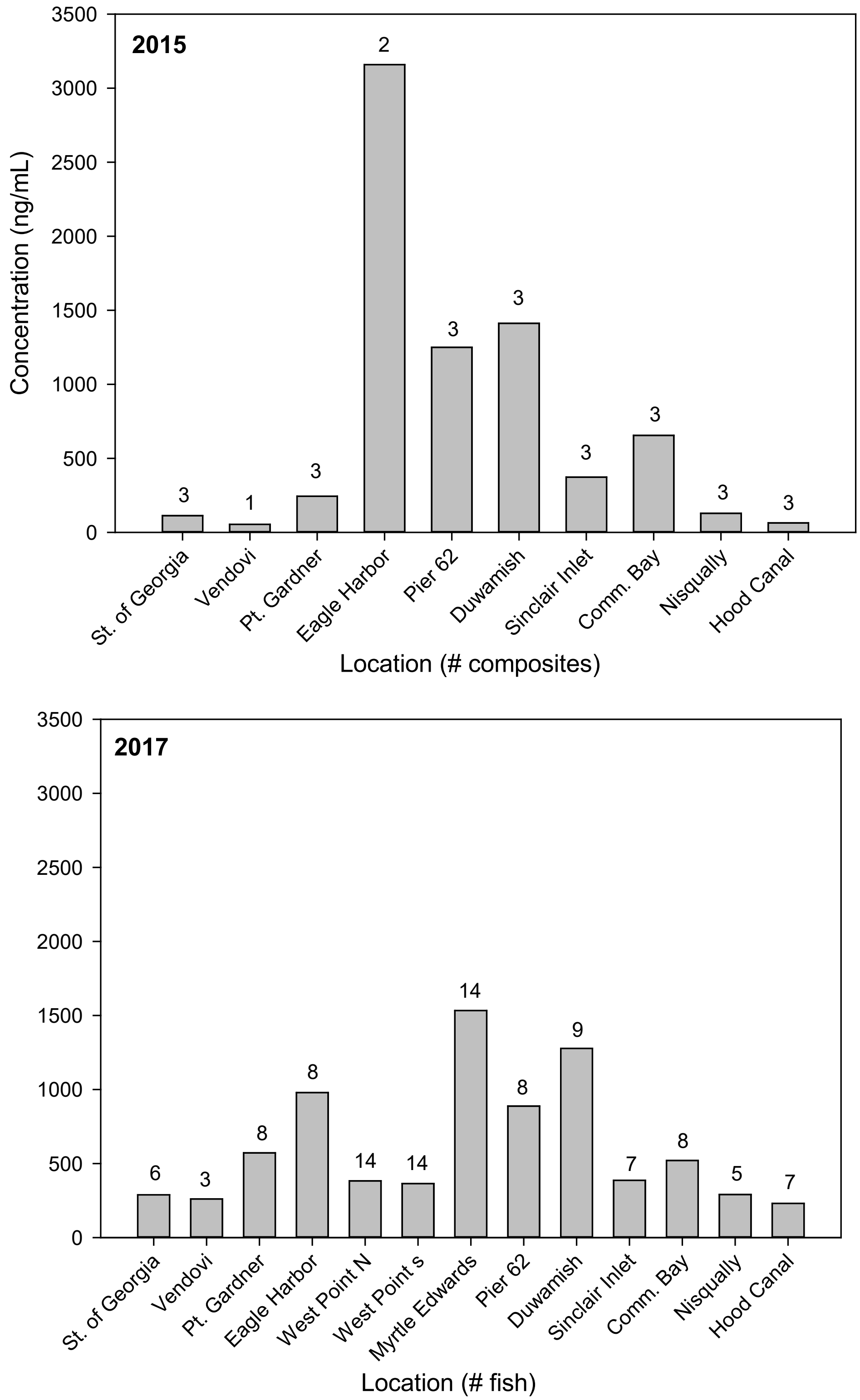


Figure 16. Average sum of detected PAH-metabolites in bile. 2015 - bile composites (13-20 males/composite); 2017 - individuals (male or female). Note that eight more PAH-metabolites were measured in 2017 compared to 2015 (see Table 7 and Appendix C, Table 13). Comm. Bay = Commencement Bay.Comparison of Metals and Organic Compound Results to Human Health Screening Levels

5.0 COMPARISON OF METALS AND ORGANIC COMPOUND RESULTS TO HUMAN HEALTH SCREENING LEVELS

Mean concentrations for the 2015 (pre-event), 2017 (year 0) and 2019 (year 2) English sole muscle tissue data from King County stations were compared to WDOH human health risk-based screening levels for seafood consumption advisories (McBride, 2018) (See Appendix B for mean values). Although other factors including data sufficiency would be evaluated before an advisory is issued by WDOH, comparison to these screening levels was used to evaluate consistency of the results presented in this study with existing fish consumption advisories regarding consumption of English sole muscle tissues. The screening levels are based on two exposure scenarios: (1) general population consumer eating 59.7 g per day of seafood and (2) high seafood consumer population eating 175 g per day of seafood (Table 11). Arsenic was not included in this comparison because only total arsenic results are available and the inorganic fraction of arsenic relevant for comparison to the screening value is unknown.

Currently, there is a flatfish (including English sole) consumption advisory for PCBs (not mercury or DDTs) in Elliott Bay limiting consumption to 2 meals per month for the general population, but English sole consumption is unlimited in other, non-urban areas of the WDOH Marine Area 10 (other than Eagle Harbor and Sinclair Inlet) (WDOH 2006). King County locations in Marine Area 10 without an English sole/flatfish advisory include West Point N and S, Alki, Quartermaster Harbor, and Shilshole. Data from Shilshole were included in the WDOH risk assessment that supports current advisories (WDOH 2006); data from other locations sampled for this study were not included in the prior WDOH (2006) analysis.

Table 11. WDOH screening levels (McBride, 2018) for seafood consumption advisories.

Compound (mg/kg unless noted)	General Population[1]	High Consumer[2]
Cadmium	1.17	0.400
Chromium (III)[3]	1759	600
Methylmercury[4]	0.101	**0.034**
Nickel	23.5	8.00
Selenium	5.86	2.00
Silver	5.86	2.00
Zinc	352	120
Total PCBs (μg/kg)	23	**8**
Total PBDEs (μg/kg)	117	40
Total DDTs (μg/kg)	**0.586**	**0.200**

All screening levels are based on non-cancer health effects.

Bolded levels were exceeded by mean concentrations at all King County stations in 2017 and 2019.
1 = Based on 59.7 grams per day seafood consumption
2 = Based on 175 grams per day seafood consumption
3 = Chromium was not speciated in tissue samples; chromium (III) is the most stable state of chromium in nature and the main form found in plants and animals (FDA 1993).
4 = Though methylmercury was not measured in this study, results for total mercury were assumed to reflect the methylmercury concentrations.
Note: WDOH does not use a standard screening level for lead but uses a predicted blood lead level in children from EPA's Integrated Exposure Uptake Biokinetic Model (IEUBK) model. Due to the complexity of model analysis, no screen for lead was performed with English sole data.

The 2017 and 2019 mean total PBDE concentrations in English sole at all King County stations were below both WDOH screening levels.

The mercury WDOH screening level is for methylmercury and we conservatively assumed the measured amount in English sole for this study was 100% methylated mercury. WDOH routinely compares total mercury values in seafood to this screening level on the assumption that nearly all mercury present in a fish muscle sample is the organic form (i.e., as methylmercury) (Bloom 1992). Aside from mercury, mean English sole muscle concentrations of all metals were also below the WDOH screening levels at all King County stations both years. As in 2015 (pre-event), mean total mercury concentrations in English sole muscle for all stations exceeded the high seafood consumer level for methylmercury, but not the general population screening level (Figure 17). Although we do not have pre-event data at West Point N or West Point S, the nearest station (Shilshole) appears to have similar or higher mean mercury concentrations than these two locations. In addition, mean mercury concentrations near West Point were similar or lower than at locations farthest away, i.e., Alki and Quartermaster.

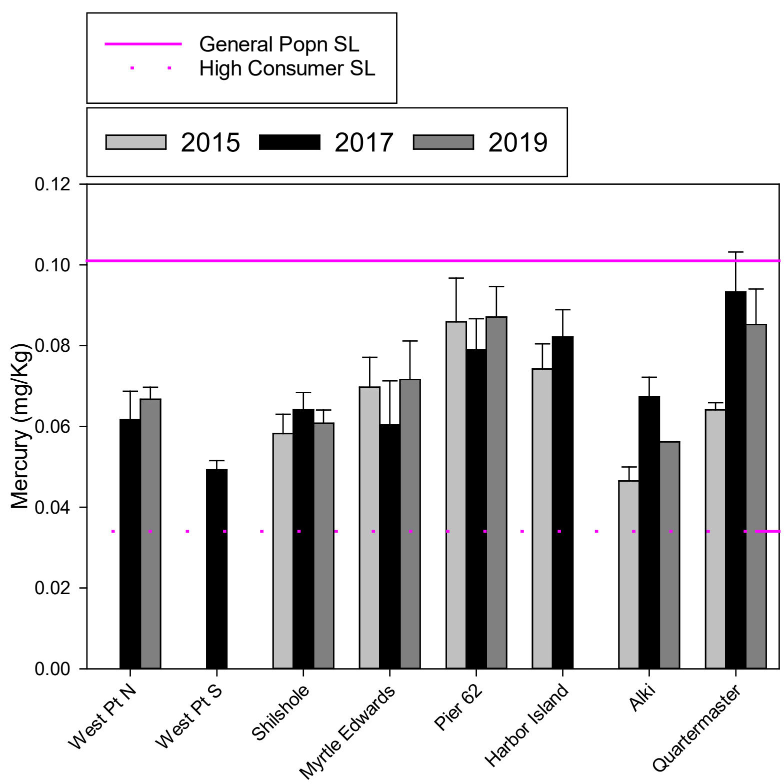


Figure 17. Mean (±SD) mercury concentrations from 2015, 2017, and 2019 compared to WDOH screening levels for seafood consumption.

As in 2015 (where available), mean total PCB concentrations measured by King County (total of homologs) and WDFW (total of congeners) at the West Point and other King County stations exceeded the high consumer screening level in 2017 and 2019 (Figure 18). Most samples also exceeded the general population screening level in all years, except for samples from the following sites analyzed by King County: West Point S (2017, Group 2), West Point N (2019, Group 1), Shilshole (2015 and 2019, Group 2), Alki (2017 and 2019, Group 2), and Quartermaster Harbor (2015 and 2019, Group 4) (Figure 18). Thus, PCB results were generally consistent with existing fish consumption advisories that limit consumption of English sole in Elliott Bay and do not limit English sole consumption within the main basin of Marine Area 10. Similarities in PCB concentrations at West Point N, West Point S (Group 1), Shilshole (Group 2) and Quartermaster (Group 4) are not consistent with a relationship to proximity to event discharges.

Mean total DDT concentrations in 2017 and 2019 also exceeded both WDOH screening levels at all stations, as measured both by King County and WDFW (Figure 19). Although mean values from 2015 (pre-event) in King County also exceeded both WDOH screening levels, a seafood consumption advisory has not been issued based on DDTs in the Central Basin[9] to date. In addition, in 2017 and 2019 mean total DDTs concentrations from all other Puget Sound stations also exceeded the WDOH high consumer screening level, and all but the following Group 4 stations exceeded the general consumer screening level: Hood Canal (2017 and 2019), Vendovi (2017 and 2019), Strait of Georgia (2017 and 2019), and Nisqually Bay (2019) and Port Madison (2019, only year measured) stations.

Overall, this evaluation indicates that the English sole muscle tissue PCB concentrations at Myrtle Edwards in 2017 or 2019 following the West Point flooding event were consistent with the existing fish consumption advisory status for Elliott Bay. In 2017, the PCB data for English sole muscle tissue near West Point and at Quartermaster Harbor were above fish advisory screening levels; however, with no pre-event data at West Point N or West Point S and the similarity to Quartermaster, this outcome is not necessarily linked to the 2017 flooding event.

[9] The WDOH (2006) health assessment for Puget Sound fish included DDTs and metabolites but their 90th percentile concentration was below the DDT screening value of 14 ug/kg and no further assessment was completed.

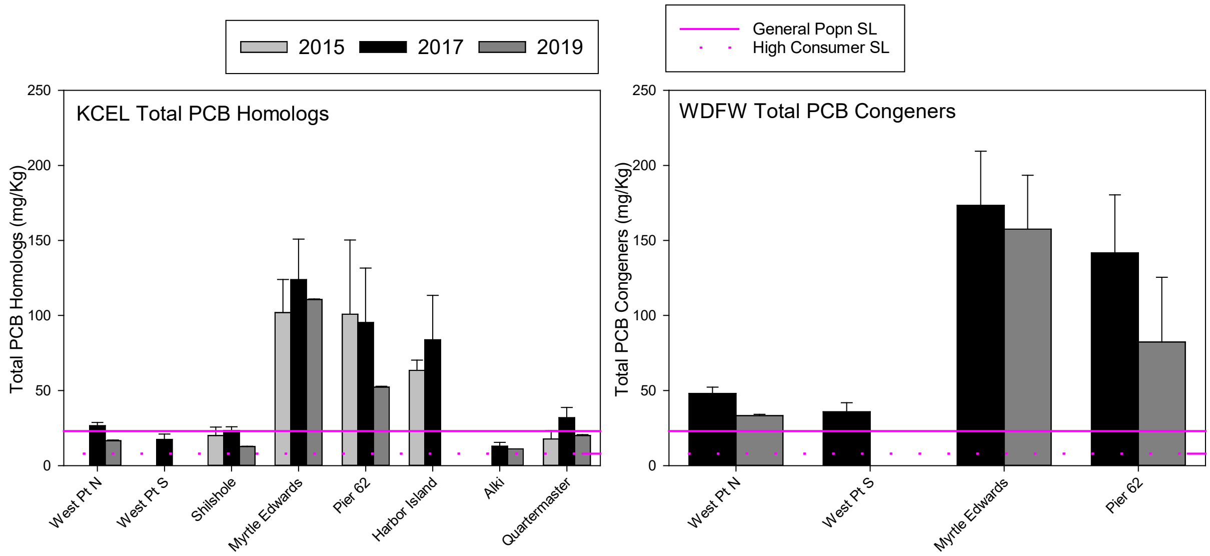


Figure 18. Mean Total PCB (±SD) concentrations for stations in King County as measured by sum of homologs at KCEL (left) from 2015 (pre-event), 2017 (year 0), and 2019 (year 2) and sum of congeners by WDFW (right) from 2017 and 2019, compared to WDOH screening levels for seafood consumption. Consumption advisory exists since 2006 in Elliott Bay including Myrtle Edwards, Pier 62 and Harbor Island.

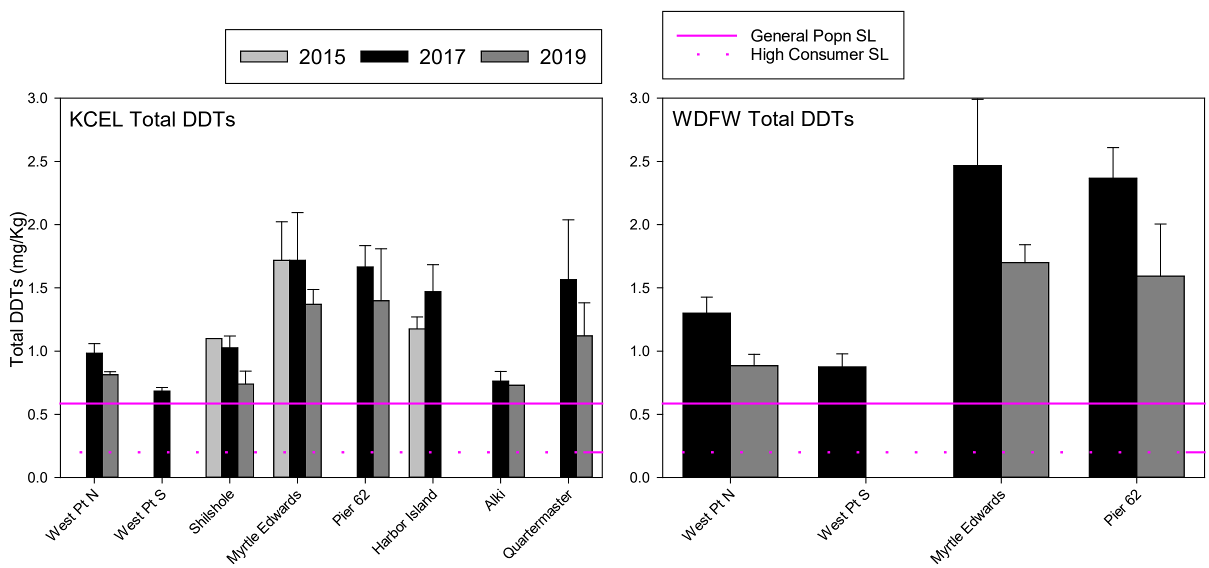


Figure 19. Mean Total DDT (±SD) concentrations for stations in King County as measured by KCEL (left, sum of 4 DDTs) from 2015 (pre-event), 2017 (year 0), and 2019 (year 2) and WDFW (right, sum of 6 DDTs) from 2017 and 2019, compared to WDOH screening levels for seafood consumption.

6.0 SUMMARY

After the 2017 West Point flooding event, King County and WDFW collaborated to collect and analyze English sole tissues in 2017 (year 0) and 2019 (year 2) to evaluate:

- whether proximity to effluent from the West Point flooding event led to increased chemical concentrations in English sole tissues
- whether biomarkers in fish nearest the event discharge points indicated increased exposure to endocrine disrupting chemicals or PAHs that may be associated with the event
- if any changes in fish tissue chemical concentrations following the flooding event resulted in greater exceedances of Washington Department of Health screening levels for seafood consumption advisories than currently exist.

The sampling design was opportunistic and was built from programs used to evaluate long-term trends. This underlying aspect, coupled with high natural variability in tissue chemistry data and the complexity of various chemical inputs to Puget Sound, impose limitations on our ability to detect site-specific outcomes tied directly to West Point. Elliott Bay site variability was higher than at other locations and in some years, low sample number further decreased the power to detect differences (e.g., Alki 2019, Shilshole all years, West Point 2019).

Did proximity to the point of discharge from the West Point flooding event lead to increased chemical concentrations in English sole tissues?

Metals

Seven metals (arsenic, barium, chromium, lead, mercury, nickel, and zinc) were the focus of the English sole tissue metals evaluation. These metals were elevated in West Point effluent during the emergency bypass and recovery period compared to when the treatment plant was fully operational (King County, 2018). Barium and nickel were analyzed only in English sole from 2017 and those results were included in an earlier interim report of that data; neither were found to be significantly different across stations (King County, 2021). Spatial and temporal comparisons of arsenic, copper, lead, and zinc concentrations measured in samples from West Point and Myrtle Edwards in 2017 and 2019 show no significant changes in these metals in English sole muscle.

Though chromium from Myrtle Edwards in 2017 was higher than 2019, the 2017 values were similar to those found at that location in 2015 (pre-event). Chromium concentrations were higher at most locations in 2015 than 2017 regardless of their proximity to discharges, suggesting the West Point flooding event did not elevate English sole exposure to chromium beyond levels that occurred prior to the event. Similarly, West Point N mean mercury concentrations were higher than West Point S in 2017. However, 2017 West Point N samples were lower than or similar to samples from other stations farther from the effluent discharges. We conclude the West Point flooding event did not increase chromium exposure in English sole beyond concentrations experienced by fish prior to the event or mercury exposure beyond concentrations in fish from other areas of King County, i.e., Shilshole, Alki or Quartermaster.

Organic Chemicals
We observed considerable variability in the three primary bioaccumulative organic compound groups (total PCBs, total PBDEs and total DDTs) detected in English sole across sampling locations, years, and over time. Simple, two-way comparisons between years (2017 vs. 2019) or locations (West Point N vs. West Point S) in some cases seem to suggest changes could be attributed to the West Point flooding event. For example, there were spatial differences in mean total PCB concentrations that suggested an impact from the West Point flooding event, such as significantly higher PCBs, PBDEs, and DDTs at West Point N compared to West Point S in 2017.

There was also evidence of a temporal effect; PCBs, PBDEs, and DDTs were higher in West Point N samples from 2017 compared to 2019, though for PBDEs this difference between years was only seen in the WDFW dataset, not the KCEL dataset. Although these comparisons suggest a possible effect of the flooding event on organic contaminant concentrations in fish around West Point, the increase was temporary and tissue concentrations were often statistically similar to areas presumably unaffected by West Point. Due to a lack of long-term historical data at the West Point N location and the multiple sources of variation in the dataset noted earlier, we cannot attribute the higher concentrations of organic contaminants in 2017 and 2019 to the West Point flooding event; nor can we rule it out.

Did biomarkers in fish nearest the West Point flooding event discharge points indicate increased exposure to endocrine disrupting chemicals or PAHs associated with the event?

Blood plasma data indicated a higher percent of vitellogenin-positive males occurred at West Point N relative to West Point S in 2017, and relative to West Point N in 2019, suggesting an impact from the West Point flooding event. However, there was limited data for this analysis, high variability at the other locations, and similar step reductions at a variety of other urban and nonurban sites between these years. Considering these factors, we are unable to determine whether the higher percent of vitellogenin-positive males seen at West Point in 2017 resulted from the flooding event or another phenomenon (e.g., high volume runoff into Puget Sound in 2017 or interannual variability).

Xenoestrogens and PAH-metabolites concentrations in bile were similar between stations and/or years. Results for xenoestrogens cannot conclusively address the study questions because of uncertainties inherent to the data; but tangential to the objectives of this study, they do identify a need for further investigation of endocrine disrupting chemicals and their effects on benthic fish health at Pier 62. PAH-metabolites in fish bile did not suggest an influence from the West Point flooding event in the vicinity of West Point or Elliott Bay.

Did any changes in fish tissue chemical concentrations following the flooding event result in any new exceedances of Washington Department of Health screening levels for seafood consumption advisories?

Several advisories that recommended people limit consumption of seafood from Puget Sound were in place before the West Point flooding event. Our evaluation indicates the West Point flooding event would not have changed the seafood consumption advisory status for English sole at Myrtle Edwards in 2017 or 2019, and fish consumption advisories for English sole near West Point (if issued) would have been the same as other stations in Elliott Bay and around Puget Sound in those years.

Conclusions

In summary, two of 14 tested metals and three organic contaminants (PCBs, PBDEs, and DDTs) were elevated in English Sole tissues near West Point in 2017, and the proportion of vitellogenin-positive males was higher in proximity to West Point in 2017 as compared to 2019. No other differences were detected. Similar (or higher) concentrations of the detected chemical contaminants described above occurred at other nearby sites and/or at different times near West Point, suggest the flooding event alone was not responsible for these elevated tissue concentrations. In addition, based on WDOH screening levels, the fish consumption advisories for English sole would not have changed as a result of the West Point flooding event.

The sampling for this study was opportunistic and (except for the addition of the West Point stations in 2017 and 2019) based on sampling locations and timing of already established tissue monitoring programs designed to track long-term trends in contamination from all the sources influencing water quality in the area. Coupled with several confounding factors (other contaminant sources, increased loadings from heavier than normal rainfall, substantial dispersion of effluent in receiving water), the resulting statistical design for this study was not well adapted to definitively link any increases in contaminant concentrations in part or exclusively to the West Point flooding event. If increases did occur, their magnitude was low enough to be obscured by the variability in contaminant concentrations exhibited by English sole in other areas in the year of the event and two years after.

Recommendations

Fish tissue chemistry is not an ideal tool for evaluating the effects of short-term untreated wastewater releases like the West Point flooding event. This is because individual fish experience variable contaminant exposures as they move across their home range resulting in highly variable tissue concentrations. In addition, muscle or whole-body tissues are typically monitored for bioaccumulative compounds that are not metabolized well by fish and collect in the body over a time scale of years. Organ and blood monitoring can provide better opportunities to examine effects of short-duration exposures (days/months).

We recommend King County's Water and Land Resources Division and Wastewater Treatment Division work together to expand King County's strategy to prepare for monitoring environmental impacts of future untreated or partially-treated large-scale wastewater discharges, like the West Point flooding event, on biota. The overall strategy for biota would include defined and targeted study questions with a sampling and analysis plan. The plan would address which biota to sample, sample timing, sample numbers, analytical parameters and other specifications. Wastewater Treatment Division and the marine monitoring program leads can prepare a study plan in advance to ensure that a technically robust program can be executed quickly in response to such an event. If biological tissues are part of the approach, we suggest the following technical considerations:

- Performa a power analysis using available historical data to support the sampling design.
- Focus sampling on contaminants, contaminants tracers, or metrics of fish exposure unique to wastewater effluent (e.g., bioaccumulative drugs like metformin, biomarkers of exposure to xenoestrogens).
- Fish may not be the most sensitive organism to monitor events like the West Point flooding event. Consider sampling other taxa instead for this purpose.
- If biomarkers are used, choose those appropriate to the time scale at which monitoring can occur.
- Explore additional short-term metrics (biomarkers) of fish health for use in events like these.

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Appendix A: Quality Assurance and Quality Control Analysis

Quality Assurance and Quality Control (QA/QC) Results

The QA/QC results for the 2017 stable isotopes, metals, PCBs, PBDEs, and OC pesticides analyses are detailed in the West Point Flooding Event 2017 English Sole Muscle Tissue Chemistry Report (King County, 2021). Below are the QA/QC results for the 2017 analyses of xenoestrogens, PAH-metabolites, and vitellogenin, and for the 2019 stable isotopes, metals, PCBs, PBDEs, OC pesticides, and vitellogenin analyses.

Stable Isotopes
The stable isotopes results met all acceptable lab QA/QC limits. Results for all standards and the reference material met the standard deviation criteria of ≤ 0.3‰ for $\delta^{15}N$ and ≤ 0.2‰ for $\delta^{13}C$.

Metals
The KCEL analytical results for metals met all acceptable lab QA/QC limits with several exceptions. One Myrtle Edwards and one Shilshole sample were J-flagged as estimated for chromium because the matrix spike for the batch was outside acceptance limits, and one Quartermaster Harbor sample was J-flagged for lead because the lab duplicate fell outside of the relative percent difference (RPD). In addition, the mercury results for all samples in 2019 were H-flagged, indicating that they did not meet the 28-day holding time. It is unlikely mercury results were significantly biased because all samples were held frozen until analysis. There were no split samples for metals analysis.

PCBs
The KCEL PCB homolog results and NOAA laboratory PCB congener results met all acceptable lab QA/QC limits in 2019.

PBDEs
The KCEL and NOAA laboratory PBDE results met all acceptable lab QA/QC limits in 2019.

OC pesticides
The KCEL and NOAA laboratory analytical results for organochlorine pesticides met all acceptable lab QA/QC objectives, except for a few KCEL samples in 2019. The quantitation limits (QLs) for DDT isomer, 4,4'-DDT, were elevated in all four KCEL samples from Myrtle Edwards, all six samples from Quartermaster Harbor, four of six samples from Pier 62, and one of the two samples from Shilshole due to interference from PCBs. These elevated QLs may have resulted in more non-detects of 4,4'-DDT in those samples, which may have led to an underestimation of total DDTs for those samples.

Xenoestrogens, PAH-metabolites, Vitellogenin

The NOAA laboratory 2017 analysis results for xenoestrogenic compounds (i.e., e-EDCs) and for PAH-metabolites in bile samples met all acceptable QA/QC objectives. The 2019 results for xenoestrogenic compounds and PAH-metabolites were not available at the time this report was written and will be included with QA/QC results in an addendum when available. The NOAA 2017 and 2019 analytical results for vitellogenin in plasma samples met all QA/QC objectives for this study.

SAP Deviations

Sampling procedures, processing and analysis followed the King County (2015) SAP and addenda (King County 2017 and 2019), and the WDFW TBiOS Program SOP (WDFW, unpublished reports). Sampling locations in 2017 and 2019 were modified as described in Section 3.1.1. Any additional deviations from sampling and analysis methods described in original monitoring plan SAPs are noted in this section.

King County SAP

There were no major deviations from the KCEL SAP and addenda in 2017. However, there was one deviation from the KCEL SAP in 2019: due to a lack of available fish one Myrtle Edwards sample was comprised of only five six instead of the target of 15. Having fewer fish in this composite may have led to higher within-sample variability, however this was not expected to impact the study results as there were three other composite samples for this station with 16 to 17 fish/sample.

WDFW SAP

There were minor changes to the TBiOS Program protocols in 2017 and 2019. In 2017, the number of English sole per sample ranged from 16 to 20 fish per sample, rather than the targeted 20 individuals per sample (Table 3). In 2019, the number of fish included in each sample ranged from 14 to 20 individuals (Table 3).

Appendix B: 2017 Muscle Chemistry Results for Elliott Bay, Shilshole, and West Point stations

The chemistry results for English sole collected by King County and WDFW from stations in the vicinity of the West Point outfall and Elliott West wet weather treatment station outfall are summarized in this section. They include the two West Point stations, Shilshole, the four Elliott Bay stations, and the Duwamish River (a WDFW long-term monitoring location included for comparison when available) for metals and organic chemicals. Data from several other long-term monitoring stations from WDFW are included for comparison for the xenoestrogens and PAH-metabolites.

Muscle Tissue

The chemical concentrations measured in muscle tissue samples of English sole are summarized in this section. All concentrations are presented on a wet weight (ww) basis, which is consistent with the majority of WDFW and King County tissue monitoring. The effect of tissue water content is thought to be insignificant because it is consistent and exhibits low variability in English sole samples.

Conventional Parameters

The analytical results for total solids and lipids met all acceptable lab QA/QC limits, except for West Point station sample and lab duplicates for lipids with relative percent differences (RPDs) outside control limits. The lab duplicate RPDs did not impact use of the results.

Percent total solids in English sole analyzed by KCEL ranged narrowly from 16.7 to 17.7% in 2015, from 15.4 to 17.3% in 2017, and from 15.5 to 18.3% in 2019 (Tables B1 and B2). Even between years there was little variation in mean percent total solids between locations, with lowest at Shilshole (15.4%) and the highest at Myrtle Edwards (17.3%). Mean percent total solids at West Point N and S were in the middle of this range (16.1 to 17.4%). The RPDs between mean percent total solids for split samples analyzed at KCEL and NOAA laboratories in 2017 and 2019 ranged from 0.6 to 12.2% indicating the solids results from these two analytical laboratories are relatively comparable.

Percent lipids in English sole analyzed by KCEL also ranged narrowly from 0.434 to 0.537% in 2015, from 0.488 to 0.727% in 2017, and from 0.567 to 0.766% in 2019 (Tables B3 and B4). Samples with the highest measured lipids were from Quartermaster Harbor in 2015, from Alki in 2017, and from Myrtle Edwards in 2019. The lowest mean percent lipid content was in muscle tissue from Alki in 2015, West Point S in 2017, and West Point N in 2019. The RPDs between mean percent total solids for split samples analyzed at KCEL and NOAA laboratories were moderate at 27 to 57% indicating lipid results from these two analytical laboratories are different and not comparable.

Table B1. Frequency of detection and percent of total solids by location and laboratory in 2015.

Sampling Location	KCEL 2015				WDFW 2015			
	FOD	Min	Max	Mean	FOD	Min	Max	Mean
Shilshole	2/2	17.1	17.9	17.5	NS	--	--	--
West Point N	NS	--	--		NS	--	--	--
West Point S	NS	--	--		NS	--	--	--
Myrtle Edwards	4/4	16.6	17.9	17.3	NS	--	--	--
Pier 62	6/6	16.5	17.7	16.8	NS	--	--	--
Harbor Island	4/4	16.0	17.3	16.7	NS	--	--	--
Duwamish River	NS	--	--		12/12	16.8	17.8	17.2
Alki	4/4	16.8	17.5	17.1	NS	--	--	--
Quartermaster	4/4	17.2	18.2	17.7	NS	--	--	--

FOD = Frequency of detection or # samples detected out of total
NS = Station not sampled

Table B2. Frequency of detection and percent of total solids by location, and relative percent difference between laboratories in 2017 and 2019.

Sampling Location	KCEL 2017				WDFW 2017				2017	KCEL 2019				WDFW 2019				2019
	FOD	Min	Max	Mean	FOD	Min	Max	Mean	RPD of Means	FOD	Min	Max	Mean	FOD	Min	Max	Mean	RPD of Means
Shilshole	6/6	15.0	16.7	16.1	NS	--	--	--	--	2/2	16.3	16.9	16.6	NS	--	--	--	--
West Point N	6/6	15.1	17.1	16.1	NA	--	--	--	--	4/4	16.7	17.9	17.4	4/4	14.6	16.1	15.4	12.2%
West Point S	4/4	14.9	16.0	15.4	NA	--	--	--	--	NS	--	--	--	NS	--	--	--	--
Myrtle Edwards	6/6	15.0	15.9	15.5	6/6	14.4	16.0	15.1	2.5%	4/4	16.9	19.4	18.3	4/4	16.5	18.7	17.2	6.4%
Pier 62	6/6	14.9	16.2	15.6	6/6	14.8	16.4	15.5	0.6%	6/6	16.1	17.3	16.8	6/6	15.3	16.8	16.0	4.9%
Harbor Island	6/6	16.1	17.8	17.0	NS	--	--	--	--	NS	--	--	--	NS	--	--	--	--
Duwamish River	NS	--	--	--	6/6	15.2	17.2	16.1		NS	--	--	--	5/5	14.2	16.1	15.3	
Alki	6/6	16.9	17.8	17.3	NS	--	--	--	--	1/1	17.9	17.9	17.9	NS	--	--	--	--
Quartermaster	4/4	15.3	16.5	15.9	NS	--	--	--	--	6/6	13.8	16.3	15.5	NS	--	--	--	--

FOD = Frequency of detection or # samples detected out of total
RPD = Relative percent difference
NS = Station not sampled

Table B3. Frequency of detection and percent of lipids by location in 2015.

Station	KCEL 2015			
	FOD	Min	Max	Mean
Shilshole	2/2	0.389	0.515	0.452
West Point N	NS	--	--	--
West Point S	NS	--	--	--
Myrtle Edwards	4/4	0.419	0.493	0.465
Pier 62	6/6	0.303	0.684	0.526
Harbor Island	4/4	0.364	0.553	0.446
Alki	4/4	0.386	0.500	0.434
Duwamish River	NS	--	--	--
Quartermaster	4/4	0.517	0.590	0.537

FOD = Frequency of detection or # samples detected out of total
NS = Station not sampled

Table B4. Frequency of detection and percent of lipids by location and relative percent difference between laboratories in 2017 and 2019.

Sampling Location	KCEL 2017				NOAA 2017				2017	KCEL 2019				NOAA 2019				2019
	FOD	Min	Max	Mean	FOD	Min	Max	Mean	RPD of Means	FOD	Min	Max	Mean	FOD	Min	Max	Mean	RPD of Means
Shilshole	4/4	0.529	0.552	0.540	NS	--	--	--	--	2/2	0.572	0.583	0.578	NS	--	--	--	--
West Point N	6/6	0.481	0.667	0.557	6/6	0.361	0.495	0.406	0.315	4/4	0.464	0.699	0.567	4/4	0.27	0.39	0.315	0.571
West Point S	6/6	0.412	0.633	0.488	6/6	0.296	0.448	0.372	0.269	NS	--	--	--	NS	--	--	--	--
Myrtle Edwards	6/6	0.499	0.692	0.596	6/6	0.253	0.406	0.344	0.535	4/4	0.697	0.885	0.766	4/4	0.39	0.52	0.478	0.464
Pier 62	6/6	0.580	0.643	0.603	6/6	0.346	0.523	0.421	0.356	6/6	0.609	0.839	0.729	6/6	0.31	0.52	0.447	0.481
Harbor Island	6/6	0.532	0.724	0.641	NS	--	--	--	--	NS	--	--	--	NS	--	--	--	--
Alki	6/6	0.624	0.819	0.727	NS	--	--	--	--	1/1	0.664	0.664	0.664	NS	--	--	--	--
Duwamish River	NS	--	--	--	6/6	0.338	0.615	0.500	--	NS	--	--	--	5/5	0.27	0.48	0.392	
Quartermaster	4/4	0.657	0.700	0.675	NS	--	--	--	--	6/6	0.592	0.659	0.620	NS	--	--	--	--

FOD = Frequency of detection or # samples detected out of total
RPD = Relative percent difference
NS = Station not sampled

Stable Isotopes

The carbon ratios ($\delta^{13}C$) in English sole ranged from -15.5 to -19.1 in 2015, from -15.1 to -18.8 in 2017, and from -15.4 to -18.7 in 2019 (Table B5). The lowest mean $\delta^{13}C$ was measured in samples from West Point S. The highest mean $\delta^{13}C$ was measured in samples from the Duwamish.

The nitrogen ratios ($\delta^{15}N$) in English sole ranged from 12.5 to 14.0 in 2015, from 12.6 to 14.1 in 2017, and from 12.6 to 14.3 in 2019 (Table B5). The lowest mean $\delta^{13}N$ was measured in samples from the Duwamish. The highest mean $\delta^{15}N$ was measured in samples from Myrtle Edwards. The West Point samples were similar between years at 13.6 and 13.7 (2017) to 13.9 (2019).

Table B5. Stable isotope ratios of carbon (C) and nitrogen (N) for English sole muscle composite samples.

Year	Sampling Location	n	$\delta^{13}C$			$\delta^{15}N$		
			Min	Max	Mean	Min	Max	Mean
2015	Pier 62	7	-15.8	-15.3	-15.5	13.7	14.1	14.0
	Duwamish	7	-19.6	-18.5	-19.1	12.3	12.7	12.5
2017	West Point N	7	-15.5	-14.9	-15.1	13.5	14.0	13.7
	West Point S	6	-13.7	-12.7	-13.4	13.5	13.7	13.6
	Myrtle Edwards	7	-15.8	-14.3	-15.0	13.6	14.0	13.7
	Pier 62	7	-15.6	-15.2	-15.4	13.9	14.3	14.1
	Duwamish	7	-19.0	-18.4	-18.8	12.5	12.7	12.6
2019	West Point N	4	-15.9	-15.6	-15.7	13.7	14.1	13.9
	Myrtle Edwards	4	-15.5	-15.4	-15.5	14.1	14.6	14.3
	Pier 62	8	-16.2	-15.4	-15.7	13.6	14.2	13.9
	Duwamish	5	-19.3	-18.2	-18.7	12.5	12.7	12.6

Metals

Of 15 metals analyzed, 12 were detected in two or more English sole samples. Antimony and thallium were not detected in any samples. Barium and silver were each detected in only one sample: barium in a Pier 62 sample in 2017 and silver in a Myrtle Edwards sample in 2015. In addition, cadmium was not detected in any samples from West Point or Myrtle Edwards in 2017 and 2019. Metals concentrations, and/or the mean detection limit (MDL) for samples with no detections, are presented below in Table B6.

Table B6. Frequency of detection and metals concentrations (mg/Kg ww) in English sole muscle composite samples from 2015, 2017, and 2019.

Metal	Station	2015				2017				2019			
		FOD	Min	Max	Mean	FOD	Min	Max	Mean	FOD	Min	Max	Mean
Antimony	Shilshole	NA	--	--	--	0/4	<0.0118	<0.0123	<0.0120	NA	--	--	--
	West Point N	NS	--	--	--	0/6	<0.0117	<0.0121	<0.0118	NA	--	--	--
	West Point S	NS	--	--	--	0/6	<0.0118	<0.0123	<0.0120	NS	--	--	--
	Myrtle Edwards	NA	--	--	--	0/6	<0.0117	<0.0121	<0.0119	NA	--	--	--
	Pier 62	NA	--	--	--	0/6	<0.0117	<0.0122	<0.0119	NA	--	--	--
	Harbor Island	NA	--	--	--	0/6	<0.0118	<0.0122	<0.0120	NS	--	--	--
	Alki	NA	--	--	--	0/6	<0.0118	<0.0124	<0.0119	NA	--	--	--
	Quartermaster	NA	--	--	--	0/4	<0.0116	<0.012	<0.0118	NA	--	--	--
Arsenic	Shilshole	2/2	4.03	4.79	4.41	4/4	7.36	8.77	8.00	2/2	7.29	7.83	7.56
	West Point N	NS	--	--	--	6/6	5.79	7.89	6.86	4/4	6.37	8.83	7.30
	West Point S	NS	--	--	--	6/6	4.41	6.26	5.29	NS	--	--	--
	Myrtle Edwards	4/4	5.91	8.73	6.99	6/6	6.21	8.60	7.30	4/4	6.36	11.6	8.70
	Pier 62	6/6	3.73	7.04	5.34	6/6	4.24	7.99	5.82	6/6	3.70	5.29	4.64
	Harbor Island	4/4	4.33	6.08	5.16	6/6	3.18	5.89	4.07	NS	--	--	--
	Alki	4/4	5.43	6.03	5.71	6/6	3.98	5.78	5.19	1/1	6.01	6.01	6.01
	Quartermaster	4/4	3.80	4.45	4.12	4/4	4.19	4.38	4.29	6/6	6.54	8.03	7.10
Barium	Shilshole	NA	--	--	--	0/4	<0.0197	<0.0205	<0.0201	NA	--	--	--
	West Point N	NS	--	--	--	0/6	<0.0194	<0.0201	<0.0197	NA	--	--	--
	West Point S	NS	--	--	--	0/6	<0.0197	<0.0205	<0.0200	NS	--	--	--
	Myrtle Edwards	NA	--	--	--	0/6	<0.0196	<0.0202	<0.0199	NA	--	--	--
	Pier 62	NA	--	--	--	1/6	0.0194	0.0254	0.0207	NA	--	--	--
	Harbor Island	NA	--	--	--	0/6	<0.0197	<0.0203	<0.0200	NS	--	--	--
	Alki	NA	--	--	--	0/6	<0.0197	<0.0207	<0.0199	NA	--	--	--
	Quartermaster	NA	--	--	--	0/4	<0.0193	<0.02	<0.0196	NA	--	--	--

Metal	Station	2015				2017				2019			
		FOD	Min	Max	Mean	FOD	Min	Max	Mean	FOD	Min	Max	Mean
Cadmium	Shilshole	2/2	0.0067	0.0072	0.0070	1/4	0.0020	0.0024	0.0021	0/2	<0.00199	<0.00206	<0.00202
	West Point N	NS	--	--	--	0/6	<0.00194	<0.00201	<0.00197	0/4	<0.00201	<0.00207	<0.00204
	West Point S	NS	--	--	--	0/6	<0.00197	<0.00205	<0.00200	NS	--	--	--
	Myrtle Edwards	2/4	0.0020	0.0048	0.0031	0/6	<0.00196	<0.00202	<0.00199	0/4	<0.00195	<0.00205	<0.00201
	Pier 62	1/6	0.0020	0.0029	0.0022	0/6	<0.00194	<0.00203	<0.00198	0/6	<0.00194	<0.00208	<0.00202
	Harbor Island	1/4	0.0019	0.0023	0.0021	5/6	0.0020	0.0047	0.0035	NS	--	--	--
	Alki	0/4	<0.002	<0.002	<0.002	3/6	0.0020	0.0069	0.0039	0/1	<0.00193	<0.00193	<0.00193
	Quartermaster	4/4	0.0047	0.0072	0.0059	4/4	0.0053	0.0106	0.0078	5/6	0.0020	0.0026	0.0024
Chromium	Shilshole	2/2	0.126	0.424	0.275	4/4	0.01	0.0483	0.02375	2/2	0.0109	0.0296	0.02025
	West Point N	NS	--	--	--	6/6	0.064	0.130	0.087	4/4	0.063	0.119	0.100
	West Point S	NS	--	--	--	6/6	0.039	0.072	0.059	NS	--	--	--
	Myrtle Edwards	4/4	0.060	0.306	0.168	6/6	0.029	0.079	0.048	4/4	0.009	0.024	0.016
	Pier 62	6/6	0.058	0.142	0.091	6/6	0.052	0.111	0.082	6/6	0.065	0.138	0.093
	Harbor Island	4/4	0.098	0.233	0.141	6/6	0.019	0.057	0.027	NS	--	--	--
	Alki	4/4	0.063	0.289	0.135	6/6	0.010	0.023	0.016	0/1	<0.00772	<0.00772	<0.00772
	Quartermaster	4/4	0.085	0.213	0.152	4/4	0.011	0.057	0.025	4/6	0.008	0.041	0.017
Copper	Shilshole	2/2	0.319	0.352	0.336	4/4	0.186	0.208	0.199	2/2	0.212	0.220	0.216
	West Point N	NS	--	--	--	6/6	0.234	0.872	0.446	NS	--	--	--
	West Point S	NS	--	--	--	6/6	0.307	0.758	0.437	4/4	0.279	1.730	0.683
	Myrtle Edwards	4/4	0.230	0.345	0.280	6/6	0.264	0.450	0.338	4/4	0.221	0.255	0.243
	Pier 62	6/6	0.283	0.542	0.428	6/6	0.295	1.910	0.614	6/6	0.223	0.726	0.371
	Harbor Island	4/4	0.240	0.314	0.263	6/6	0.229	0.281	0.250	NS	--	--	--
	Alki	4/4	0.223	0.241	0.231	6/6	0.215	0.280	0.249	1/1	0.210	0.210	0.210
	Quartermaster	4/4	0.275	0.304	0.295	4/4	0.256	0.314	0.284	6/6	0.220	0.241	0.230
Lead	Shilshole	2/2	0.0066	0.0069	0.0068	1/4	0.0039	0.0046	0.0042	2/2	0.0050	0.0071	0.0060
	West Point N	NS	--	--	--	6/6	0.0051	0.0117	0.0077	4/4	0.0047	0.0548	0.0179
	West Point S	NS	--	--	--	3/6	0.0040	0.0063	0.0045	NS	--	--	--
	Myrtle Edwards	4/4	0.0110	0.0190	0.0150	6/6	0.0071	0.0128	0.0105	4/4	0.0093	0.0157	0.0123
	Pier 62	5/6	0.0041	0.0221	0.0110	6/6	0.0039	0.0495	0.0147	6/6	0.0044	0.0177	0.0079
	Harbor Island	4/4	0.0074	0.0150	0.0116	6/6	0.0063	0.0091	0.0079	NS	--	--	--
	Alki	1/4	0.0040	0.0053	0.0043	0/6	<0.00394	<0.00413	<0.00399	1/1	0.0046	0.0046	0.0046
	Quartermaster	4/4	0.0059	0.0110	0.0087	4/4	0.0079	0.0159	0.0106	6/6	0.0155	0.0274	0.0203
Me rcu	Shilshole	2/2	0.0548	0.0616	0.0582	4/4	0.0605	0.0697	0.0642	2/2	0.0585	0.0631	0.0608
	West Point N	NS	--	--	--	6/6	0.0559	0.0734	0.0617	4/4	0.0637	0.0698	0.0667

Metal	Station	2015				2017				2019			
		FOD	Min	Max	Mean	FOD	Min	Max	Mean	FOD	Min	Max	Mean
	West Point S	NS	--	--	--	6/6	0.0469	0.0527	0.0493	NS	--	--	--
	Myrtle Edwards	4/4	0.0613	0.0764	0.0697	6/6	0.0491	0.0753	0.0603	4/4	0.0642	0.0851	0.0716
	Pier 62	6/6	0.0737	0.0999	0.0859	6/6	0.0687	0.0922	0.0790	6/6	0.0764	0.0980	0.0871
	Harbor Island	4/4	0.0655	0.0790	0.0742	6/6	0.0707	0.0904	0.0821	NS	--	--	--
	Alki	4/4	0.0418	0.0491	0.0465	6/6	0.0622	0.0753	0.0674	1/1	0.0562	0.0562	0.0562
	Quartermaster	4/4	0.0616	0.0656	0.0641	4/4	0.0861	0.1080	0.0933	6/6	0.0706	0.0935	0.0852
Molybdenum	Shilshole	NA	--	--	--	4/4	0.004	0.008	0.005	NA	--	--	--
	West Point N	NS	--	--	--	4/6	0.004	0.004	0.004	NA	--	--	--
	West Point S	NS	--	--	--	2/6	0.004	0.004	0.004	NS	--	--	--
	Myrtle Edwards	NA	--	--	--	5/6	0.004	0.005	0.004	NA	--	--	--
	Pier 62	NA	--	--	--	2/6	0.004	0.004	0.004	NA	--	--	--
	Harbor Island	NA	--	--	--	6/6	0.005	0.008	0.006	NS	--	--	--
	Alki	NA	--	--	--	6/6	0.004	0.009	0.006	NA	--	--	--
	Quartermaster	NA	--	--	--	3/4	0.004	0.009	0.006	NA	--	--	--
Nickel	Shilshole	NA	--	--	--	4/4	0.020	0.042	0.026	NA	--	--	--
	West Point N	NS	--	--	--	6/6	0.022	0.042	0.031	NA	--	--	--
	West Point S	NS	--	--	--	6/6	0.023	0.046	0.029	NS	--	--	--
	Myrtle Edwards	NA	--	--	--	6/6	0.018	0.039	0.027	NA	--	--	--
	Pier 62	NA	--	--	--	6/6	0.019	0.029	0.023	NA	--	--	--
	Harbor Island	NA	--	--	--	6/6	0.027	0.047	0.040	NS	--	--	--
	Alki	NA	--	--	--	6/6	0.021	0.062	0.038	NA	--	--	--
	Quartermaster	NA	--	--	--	4/4	0.046	0.078	0.056	NA	--	--	--
Selenium	Shilshole	NA	--	--	--	4/4	0.590	0.675	0.634	NA	--	--	--
	West Point N	NS	--	--	--	6/6	0.563	0.673	0.607	NA	--	--	--
	West Point S	NS	--	--	--	6/6	0.379	0.496	0.443	NS	--	--	--
	Myrtle Edwards	NA	--	--	--	6/6	0.562	0.768	0.653	NA	--	--	--
	Pier 62	NA	--	--	--	6/6	0.623	0.821	0.709	NA	--	--	--
	Harbor Island	NA	--	--	--	6/6	0.601	0.780	0.648	NS	--	--	--
	Alki	NA	--	--	--	6/6	0.648	0.763	0.723	NA	--	--	--
	Quartermaster	NA	--	--	--	4/4	0.529	0.689	0.598	NA	--	--	--
Silver	Shilshole	0/2	<0.0015	<0.0016	<0.0015	0/4	<0.00157	<0.00164	<0.00160	0/2	<0.00159	<0.00165	<0.00162
	West Point N	NS	--	--	--	0/6	<0.00156	<0.00161	<0.00158	0/4	<0.00161	<0.00166	<0.00163
	West Point S	NS	--	--	--	0/6	<0.00157	<0.00164	<0.00160	NS	--	--	--
	Myrtle Edwards	1/4	0.00160	0.00170	0.00163	0/6	<0.00156	<0.00161	<0.00159	0/4	<0.00156	<0.00164	<0.00160

Metal	Station	2015				2017				2019			
		FOD	Min	Max	Mean	FOD	Min	Max	Mean	FOD	Min	Max	Mean
	Pier 62	0/6	<0.0016	<0.0017	<0.00161	0/6	<0.00155	<0.00163	<0.00158	0/6	<0.00155	<0.00167	<0.00162
	Harbor Island	0/4	<0.0015	<0.0016	<0.00157	0/6	<0.00158	<0.00163	<0.00160	NS	--	--	--
	Alki	0/4	<0.0016	<0.0016	<0.0016	0/6	<0.00157	<0.00165	<0.00159	0/1	<0.00154	<0.00154	<0.00154
	Quartermaster	0/4	<0.0016	<0.0017	<0.00162	0/4	<0.00154	<0.0016	<0.00157	0/6	<0.00154	<0.00165	<0.00159
Thallium	Shilshole	NA	--	--	--	0/4	<0.00393	<0.0041	<0.00402	NA	--	--	--
	West Point N	NS	--	--	--	0/6	<0.00389	<0.00402	<0.00395	NA	--	--	--
	West Point S	NS	--	--	--	0/6	<0.00394	<0.0041	<0.00401	NS	--	--	--
	Myrtle Edwards	NA	--	--	--	0/6	<0.00391	<0.00404	<0.00398	NA	--	--	--
	Pier 62	NA	--	--	--	0/6	<0.00389	<0.00407	<0.00397	NA	--	--	--
	Harbor Island	NA	--	--	--	0/6	<0.00394	<0.00407	<0.00401	NS	--	--	--
	Alki	NA	--	--	--	0/6	<0.00394	<0.00413	<0.00399	NA	--	--	--
	Quartermaster	NA	--	--	--	0/4	<0.00385	<0.00401	<0.00392	NA	--	--	--
Vanadium	Shilshole	NA	--	--	--	4/4	0.0261	0.0326	0.0291	NA	--	--	--
	West Point N	NS	--	--	--	6/6	0.0139	0.0253	0.0196	NA	--	--	--
	West Point S	NS	--	--	--	5/6	0.0030	0.0102	0.0069	NS	--	--	--
	Myrtle Edwards	NA	--	--	--	6/6	0.0046	0.0100	0.0081	NA	--	--	--
	Pier 62	NA	--	--	--	6/6	0.0046	0.0060	0.0053	NA	--	--	--
	Harbor Island	NA	--	--	--	6/6	0.0070	0.0171	0.0129	NS	--	--	--
	Alki	NA	--	--	--	6/6	0.0082	0.0394	0.0212	NA	--	--	--
	Quartermaster	NA	--	--	--	4/4	0.0148	0.0265	0.0200	NA	--	--	--
Zinc	Shilshole	2/2	6.42	7.48	6.95	4/4	4.97	5.18	5.10	2/2	5.96	6.04	6.00
	West Point N	NS	--	--	--	6/6	4.24	4.95	4.53	4/4	4.55	7.29	5.43
	West Point S	NS	--	--	--	6/6	6.03	8.50	6.99	NS	--	--	--
	Myrtle Edwards	4/4	4.04	5.76	5.00	6/6	4.33	5.12	4.79	4/4	5.32	6.24	5.84
	Pier 62	6/6	4.43	6.58	5.47	6/6	3.89	5.42	4.40	6/6	3.88	4.73	4.36
	Harbor Island	4/4	4.06	4.98	4.54	6/6	4.82	6.29	5.67	NS	--	--	--
	Alki	4/4	4.81	5.22	4.97	6/6	5.50	7.53	6.20	1/1	6.10	6.10	6.10
	Quartermaster	4/4	6.43	6.83	6.68	4/4	5.90	8.44	7.51	6/6	5.08	6.61	5.86

Values with "<" were not detected at the MDL presented
FOD = Frequency of detection or # samples detected out of total
NA = Samples not analyzed for this metal; NS = Station not sampled

Total PCBs

Total PCB homologues in English sole muscle ranged from 11 to 158 µg/kg ww in 2015, from 9.87 to 155 µg/kg ww in 2017, and from 9.7 to 143 µg/kg ww in 2019 (Tables B7, B8, and B9, respectively for each year). Mean total PCB homologue concentrations in samples were lowest at Quartermaster Harbor in 2015, and at Alki in 2017 and 2019, and highest at Pier 62 in 2015 and at Myrtle Edwards in 2017 and 2019. Mean total PCB homologue concentrations in samples at West Point stations were approximately 10 times lower than at Myrtle Edwards and similar to those at Shilshole.

Split samples from the West Point, Myrtle Edwards and Pier 62 stations were analyzed for PCBs by KCEL and NOAA laboratories. These laboratories use different analytical and total PCB estimation methods. Therefore, the concentrations for split samples were not expected to be necessarily similar. However, the mean total PCB concentrations, although higher from NOAA, rank the four stations in the same order from high to low concentrations: the highest mean total PCB concentrations are in samples from Myrtle Edwards, then decreasing in samples from Pier 62, and the lowest concentrations were in samples from West Point N, then West Point S. Though samples from the Duwamish River (a long-term monitoring site for WDFW) were not included in the splits with King County, it was included in the analysis of the WDFW data for context.

Total PBDEs

Total PBDEs (King County data) in English sole muscle ranged from 0.44 to 11.6 µg/kg ww in 2015, 0.811 to 4.29 µg/kg ww in 2017, and 0.740 to 2.79 µg/kg ww in 2019 (Tables B7, B8, and B9, respectively for each year). Mean total PBDE concentrations in muscle were lowest at Quartermaster Harbor in 2015, West Point S in 2017, and Alki in 2019. Mean total PBDE concentrations at Myrtle Edwards were lower than at Pier 62 in all years.

Split samples were analyzed at the same four stations for PBDEs as PCBs by KCEL and NOAA laboratories. The PBDE analytical methods are different at these two laboratories. Therefore, the concentrations for split samples were not expected to necessarily be similar. NOAA mean total PBDE concentrations were higher than KCEL mean concentrations in 2017 and 2019. The West Point N station ranked highest and West Point S lowest for mean concentration in 2017, similar to KCEL results. Mean total PBDE concentrations for Myrtle Edwards and Pier 62 samples fell into different rank orders for KCEL and NOAA in 2017 but not in 2019. Though samples from the Duwamish River (a long-term monitoring site for WDFW) were not included in the splits with King County, it was included in the analysis of the WDFW data for context.

Organochlorine Pesticides

Total DDTs in English sole muscle ranged from <01.1 to 2.17 µg/kg ww in 2015, from 0.677 to 2.22 µg/kg ww in 2017, and from 0.677 to 2.09 µg/kg ww in 2019 (Tables B7, B8, and B9, respectively for each year). Mean total DDT concentrations were lowest in samples from Shilshole in 2015, West Point S in 2017, and Alki in 2019. Mean values were highest in samples from Myrtle Edwards in 2015 and 2017, and Pier 62 in 2019. Mean total DDT concentrations in samples from West Point N were higher than those from West Point S in 2017, and similar to those from Shilshole in 2017 and 2019.

Split samples from the two West Point stations, Myrtle Edwards and Pier 62 stations were analyzed by NOAA. Both 4,4'-DDT and 4,4'-DDE isomers were detected in NOAA split samples. Total DDTs were higher in split samples analyzed by NOAA than those analyzed by KCEL indicating some differences in analytical method. In addition to DDTs, chlordanes were detected by NOAA at Myrtle Edwards (mean 0.237 µg/kg ww), Pier 62 (mean 0.278 µg/kg ww), and the Duwamish River (mean 0.775 µg/kg ww) in 2017, and again at Pier 62 (mean 0.168 µg/kg ww) and the Duwamish (mean 1.14 µg/kg ww) in 2019. KCEL did not detect chlordanes at any site in 2017 or 2019. The NOAA organochlorine pesticide method includes oxychlor and nonachlor isomers which are absent from the KCEL analyte list. No chlordanes were detected in English sole from West Point stations.

Table B7. Total PCBs, total PBDEs, total DDTs, and total Chlordanes (µg/kg ww) in English sole muscle composite samples from 2015.

Parameter	Station	KCEL				NOAA			
		FOD	Min	Max	Mean	FOD	Min	Max	Mean
Total PCB	Shilshole	2/2	16.0	24.0	20.0	--	--	--	--
	West Point N	NS	--	--	--	--	--	--	--
	West Point S	NS	--	--	--	--	--	--	--
	Myrtle Edwards	4/4	73.7	123	102	--	--	--	--
	Pier 62	6/6	36.9	158	101	--	--	--	--
	Harbor Island	4/4	54.0	70.1	63.4	--	--	--	--
	Alki	0/4	<16	<16	<16	--	--	--	--
	Duwamish River	NS	--	--	--	6/6	240	320	268
	Quartermaster	3/4	11.0	24.0	17.7	--	--	--	--
Total PBDEs	Shilshole	2/2	2.09	2.47	2.28	--	--	--	--
	West Point N	NS	--	--	--	--	--	--	--
	West Point S	NS	--	--	--	--	--	--	--
	Myrtle Edwards	4/4	1.64	6.17	2.96	--	--	--	--
	Pier 62	6/6	3.39	11.6	6.86	--	--	--	--
	Harbor Island	4/4	1.07	2.61	1.88	--	--	--	--
	Alki	4/4	0.440	2.32	1.34	--	--	--	--
	Duwamish River	NS	--	--	--	6/6	3.70	5.2	4.32
	Quartermaster	4/4	1.13	1.45	1.33	--	--	--	--
Total DDTs	Shilshole	1/2	1.10	1.10	1.10	--	--	--	--
	West Point N	NS	--	--	--	--	--	--	--
	West Point S	NS	--	--	--	--	--	--	--
	Myrtle Edwards	4/4	1.50	2.17	1.72	--	--	--	--
	Pier 62	NS	--	--	--	--	--	--	--
	Harbor Island	3/4	1.10	1.30	1.18	--	--	--	--
	Alki	0/4	<1.1	<1.1	<1.1	--	--	--	--
	Duwamish River	NS	--	--	--	6/6	3.60	4.90	4.03
	Quartermaster	0/4	<1.1	<1.1	<1.1	--	--	--	--
Total Chlordanes	Shilshole	0/4	<1.1	<1.1	<1.1	--	--	--	--
	West Point N	NS	--	--	--	--	--	--	--
	West Point S	NS	--	--	--	--	--	--	--

	Myrtle Edwards	0/4	<1.1	<1.1	<1.1	--	--	--	--
	Pier 62	NS	--	--	--	--	--	--	--
	Harbor Island	0/4	<1.1	<1.1	<1.1	--	--	--	--
	Alki	0/4	<1.1	<1.1	<1.1	--	--	--	--
	Duwamish River	NS	--	--	--	6/6	0.560	0.750	0.643
	Quartermaster	0/4	--	--	--	--	--	--	--

Values with “<” were not detected above the limit of quantitation (LOQ), which is shown instead
FOD = Frequency of detection or # samples detected out of total
NS = Station not sampled

Table B8. Total PCBs, total PBDEs, total DDTs, and total Chlordanes (µg/kg ww) in English sole muscle composite samples from 2017, including split samples analyzed by KCEL and WDFW.

Parameter	Station	KCEL				NOAA			
		FOD	Min	Max	Mean	FOD	Min	Max	Mean
Total PCB	Shilshole	6/6	23.5	29.6	26.5	--	--	--	--
	West Point N	6/6	12.9	22.3	17.4	6/6	41.0	54.0	48.0
	West Point S	4/4	20.0	25.7	23.4	6/6	28.0	44.0	35.8
	Myrtle Edwards	6/6	73.7	146	124	6/6	130.0	230	173
	Pier 62	6/6	50.6	155	95	6/6	90.0	210	142
	Harbor Island	6/6	54.7	128.2	83.8	--	--	--	--
	Alki	6/6	9.874	17	12.92	--	--	--	--
	Duwamish River	NS	--	--	--	6/6	230	460	350
	Quartermaster	4/4	22.6	38.4	31.9	--	--	--	--
Total PBDEs	Shilshole	4/4	1.48	2.90	2.12	--	--	--	--
	West Point N	6/6	2.75	4.29	3.26	6/6	3.80	4.80	4.35
	West Point S	6/6	0.84	1.53	1.06	6/6	1.50	4.00	2.95
	Myrtle Edwards	6/6	1.02	2.02	1.48	6/6	2.70	5.70	4.13
	Pier 62	6/6	1.27	2.26	1.78	6/6	2.10	5.00	3.87
	Harbor Island	6/6	1.52	2.65	1.90	--	--	--	--
	Alki	6/6	1.09	2.54	1.47	--	--	--	--
	Duwamish River	NS	--	--	--	6/6	3.50	6.20	4.933
	Quartermaster	4/4	0.811	2.25	1.72	--	--	--	--
Total DDTs	Shilshole	4/4	0.905	1.10	1.03	--	--	--	--
	West Point N	6/6	0.872	1.09	0.983	6/6	1.20	1.50	1.30
	West Point S	2/6	0.667	0.736	0.684	6/6	0.71	1.00	0.9
	Myrtle Edwards	6/6	1.14	2.22	1.72	6/6	1.90	3.10	2.47
	Pier 62	6/6	1.52	1.99	1.67	6/6	2.00	2.70	2.37
	Harbor Island	6/6	1.22	1.81	1.47	--	--	--	--
	Alki	4/6	0.667	0.844	0.763	--	--	--	--
	Duwamish River	NS	--	--	--	6/6	3.20	7.1	5.467
	Quartermaster	4/4	0.877	1.91	1.564	--	--	--	--
Total Chlordanes	Shilshole	0/4	<0.667	<1.33	<0.833	--	--	--	--
	West Point N	0/6	<0.667	<1.33	<1.22	0/6	<0.16	<0.2	<0.182
	West Point S	0/6	<0.667	<0.667	<0.667	0/6	<0.11	<0.2	<0.153
	Myrtle Edwards	0/6	<0.667	<1.33	<1.109	5/6	0.190	0.290	0.237
	Pier 62	0/6	<1.33	<1.33	<1.33	6/6	0.210	0.420	0.278
	Harbor Island	0/6	<1.33	<2	<1.44	--	--	--	--
	Alki	0/6	<0.667	<1.33	<1.11	--	--	--	--
	Duwamish River	NS	--	--	--	6/6	0.270	1	0.775
	Quartermaster	0/4	<0.667	<1.33	<1.16	--	--	--	--

Values with “<” were not detected above the limit of quantitation (LOQ), which is shown instead.

FOD = Frequency of Detection or # samples detected out of total.

Table B9. Total PCBs, total PBDEs, total DDTs, and total Chlordanes (µg/kg ww) in English sole muscle composite samples from 2019.

Parameter	Station	KCEL				NOAA			
		FOD	Min	Max	Mean	FOD	Min	Max	Mean
Total PCB	Shilshole	2/2	9.68	15.8	12.7	--	--	--	--
	West Point N	4/4	12.8	20.2	16.6	4/4	32.0	34.0	33.3
	West Point S	NS	--	--	--	NS	--	--	--
	Myrtle Edwards	4/4	87.2	143	111	4/4	130	210	158
	Pier 62	6/6	19.5	89.3	52.3	6/6	28.0	140	82
	Harbor Island	NS	--	--	--	--	--	--	--
	Alki	1/1	11.1	11.1	11.1	--	--	--	--
	Duwamish River	NS	--	--	--	5/5	300	510	434
	Quartermaster	6/6	14.9	27.6	20.0	--	--	--	--
Total PBDEs	Shilshole	2/2	0.740	1.04	0.89	--	--	--	--
	West Point N	4/4	1.87	2.79	2.35	4/4	2.20	2.60	2.43
	West Point S	NS	--	--	--	NS	--	--	--
	Myrtle Edwards	4/4	0.906	1.94	1.28	4/4	1.40	2.20	1.73
	Pier 62	6/6	1.14	2.71	1.71	6/6	1.20	7.90	2.93
	Harbor Island	NS	--	--	--	--	--	--	--
	Alki	1/1	0.856	0.856	0.856	--	--	--	--
	Duwamish River	NS	--	--	--	5/5	4.80	5.90	5.50
	Quartermaster	6/6	0.987	2.57	1.43	--	--	--	--
Total DDTs	Shilshole	1/2	0.667	0.812	0.740	--	--	--	--
	West Point N	4/4	0.779	0.832	0.814	4/4	0.780	1.00	0.885
	West Point S	NS	--	--	--	NS	--	--	--
	Myrtle Edwards	4/4	1.25	1.49	1.37	4/4	1.60	1.90	1.70
	Pier 62	6/6	0.88	2.09	1.40	6/6	0.960	2.10	1.59
	Harbor Island	NS	--	--	--	--	--	--	--
	Alki	1/1	0.730	0.730	0.730	--	--	--	--
	Duwamish River	NS	--	--	--	5/5	5.50	8.30	7.34
	Quartermaster	6/6	0.831	1.56	1.12	--	--	--	--
Total Chlordanes	Shilshole	0/2	<0.667	<0.667	<0.667	--	--	--	--
	West Point N	0/4	<0.667	<0.667	<0.667	0/4	<0.16	<0.2	<0.183
	West Point S	NS	--	--	--	NS	--	--	--
	Myrtle Edwards	0/4	<0.667	<0.667	<0.667	0/4	<0.14	<0.18	<0.165
	Pier 62	0/6	<0.667	<0.667	<0.667	2/6	0.150	0.180	0.168
	Harbor Island	NS	--	--	--	--	--	--	--
	Alki	0/1	<0.667	<0.667	<0.667	--	--	--	--
	Duwamish River	NS	--	--	--	5/5	0.920	1.30	1.14
	Quartermaster	0/6	<0.667	<0.667	<0.667	--	--	--	--

Values with "<" were not detected above the limit of quantitation (LOQ), which is shown instead
FOD = Frequency of detection or # samples detected out of total
NS = Station not sampled

Appendix C: 2015 and 2017 Biomarker Results for Elliott Bay, Shilshole, and West Point stations

The chemical concentrations measured in bile samples of English sole collected by King County and WDFW from stations in the vicinity of the West Point outfall and Elliott West wet weather treatment station outfall are summarized in this section. These stations include the West Point N, West Point S, Myrtle Edwards and Pier 62.

Xenoestrogens

Five estrogenic chemicals (ECs) were measured in the bile of male English sole in 2015 and nine were measured in 2017 (Table C1). Total estrogenic chemicals (ECs) detected included three estrogens (estrone, 17β-estradiol, and estriol) and four bisphenols; Bisphenol-A (BPA), Bisphenol-AF (BPA-AF), Bisphenol-S (BPA-S), and Tetrabromobisphenol-A (TB-BPA) (Table C2). 17α-ethinylestradiol and Bisphenol-F (BPA-F) were not detected in the bile of male fish. Total ECs ranged from ranged from 41.5 to 749 ng/mL bile in 2015, and from 24.6 to 904 ng/mL bile in 2017 (Table C2). Total ECs in male bile samples were lowest at Commencement Bay in 2015, and at Hood Canal in 2017, with value at West Point N (27.7 ng/mL bile) and S (37.0 ng/mL bile) in 2017 similar to values at Hood Canal. Total ECs were highest at Pier 62, by an order of magnitude compared to most stations, in 2015 and 2017 at 749 and 904 ng/mL bile respectively.

Estriol was not detected at West Point N or S or at Myrtle Edwards in 2017, though it was detected at Pier 62 in 2017 and at Pier 62 in 2015. Estradiol and estrone were the only estrogens detected in fish from the West Point or Myrtle Edwards stations in 2017. These hormones are naturally occurring in both genders of English sole but can also be elevated in male or female fish as a result of environmental exposure to human-derived hormones from treated and untreated sewage. In 2017, BPA, BPA-F, and TB-BPA were detected at Myrtle Edwards, BPA and BPA-F were detected at West Point N, and BPA and TB-BPA were detected at West Point S and Pier 62. BPA was detected at Pier 62 in 2015, but the other analytes were not available for analysis that year.

The estrogenic potential (EP) in the bile of male English sole matched closely the total ECs (Table C3). Total EP values in fish from West Point N and S were the lowest measured in the 2017 data (5.23 and 9.52 ng/mL bile respectively). Values from the Myrtle Edwards station sampled in 2017 were also relatively low (10.4 ng/mL bile). Values at Pier 62 were an order of magnitude higher both in 2015 and 2017 (351 and 536 ng/mL bile, respectively).

Table C1. Details for the estrogenic chemicals measured in bile of male English sole in 2015 and 2017.

	Chemical name	Year Analyzed	Notes

Chemical Class		2015	2017	
Hormones	estrone (E1)	x	x	Natural estrogen
	17β-estradiol (E2)	x	x	Natural estrogen
	17α-ethinylestradiol (EE2)	x	x	Synthetic estrogen hormone; not detected in either year
	estriol (E3)	x	x	Natural estrogen
Bisphenols	Bisphenol-A (BPA)	x	x	
	Bisphenol-AF (BPA-AF)		x	Not detected in 2017
	Bisphenol-F (BPA-F)		x	
	Bisphenol-S (BPA-S)		x	
	Tetrabromobisphenol-A (TB-BPA)		x	

Table C2. Mean detected xenoestrogens, including estrogens, bisphenols, and total estrogenic chemicals (ng/mL), in bile of male English sole collected in 2015 and 2017.

Year	Sampling Location	n	Estrone	17β-Estradiol	Estriol	BPA	BPA-F	BPA-S	TB-BPA	Total ECs
2015	St of Georgia	2	13.5	10.5	<1.5	17.5	NA	NA	NA	41.5
	Vendovi	1	30.0	43.0	<1.5	260	NA	NA	NA	333
	Pt Gardner	3	24.0	37.0	<2.1	29.0	NA	NA	NA	90.0
	Pier 62	7	206	327	10.4	206	NA	NA	NA	749
	Duwamish	14	5.49	5.47	<1.9	43.8	NA	NA	NA	54.8
	Sinclair Inlet	8	13.6	9.00	<2.1	57.9	NA	NA	NA	80.5
	Comm Bay	4	5.68	4.85	<4.5	43.3	NA	NA	NA	53.8
	Hood Canal	7	10.2	9.56	<3	77	NA	NA	NA	96.7
2017	St of Georgia	4	28.1	25.5	<4.5	2	<0.75	<4.5	<0.75	56
	Vendovi	2	39.0	63.5	7.55	<4.5	<0.75	<4.5	<0.75	110
	Pt Gardner	2	145	550	64.5	15.5	<4.5	<0.75	<4.5	775
	Eagle Harbor	6	9.27	14.9	<4.5	15.7	14.7	0.633	31.7	86.8
	West Point N	4	6.55	4.50	<4.5	12.2	4.50	<0.75	<4.5	27.7
	West Point S	7	8.60	8.57	<5.6	8.70	<5.6	<0.94	11.1	37.0
	Myrtle Edwards	7	9.27	9.37	<4.5	11.2	9.3	0.366	80	120
	Pier 62	6	313	496	54.3	40.5	<4.5	<0.75	0.767	904
	Duwamish River	8	27.2	47.4	3.63	83.9	0.8	1.28	<4.5	164
	Sinclair Inlet	6	22.2	16.7	<4.5	57.3	<4.5	<0.75	<4.5	96.3
	Comm Bay	6	11.7	13.2	1.03	72.2	1.8	2.77	<4.5	103
	Nisqually	3	27.7	46	<4.5	21.0	<4.5	<0.75	<4.5	94.7
	Hood Canal	6	12.6	11.3	<4.5	0.767	<4.5	<0.75	<4.5	24.6

Values for n represent individual males in 2015 and 2017
BPA = Bisphenol A; BPA-F = Bisphenol AF; BPA-S = Bisphenol S; TB-BPA = tetrabromobisphenol
ECs = estrogenic chemicals
Values with "<" were not detected above the limit of quantitation (LOQ), which is shown instead; total was calculated using only the detected values, not including the LOQs
NA – Not analyzed

Table C3. Estradiol equivalent concentrations (EEQs, ng/mL) for estrone, estradiol, estriol, and Bisphenol A and total estrogenic potential in bile of male English sole collected in 2015 and 2017.

Year	Sampling Location	n	Estrone	17β-Estradiol	Estriol	BPA	Total EP
2015	St of Georgia	2	1.49	10.5	0	0.0068	12.0
	Vendovi	1	3.30	43.0	0	0.101	46.4

Year	Sampling Location	n	Estrone	17β-Estradiol	Estriol	BPA	Total EP
	Pt Gardner	3	2.64	37.0	0	0.0113	39.7
	Pier 62	7	22.7	327	1.15	0.0802	351
	Duwamish River	14	0.60	5.47	0	0.0171	6.1
	Sinclair Inlet	8	1.50	9.00	0	0.0226	10.5
	Comm Bay	4	0.62	4.85	0	0.0169	5.5
	Hood Canal	7	1.12	9.56	0	0.0300	10.7
2017	St of Georgia	4	3.09	25.5	0	0.0008	28.6
	Vendovi	2	4.29	63.5	0.831	0	68.6
	Pt Gardner	2	16.0	550	7.10	0.0060	573
	Eagle Harbor	6	1.0	14.9	0.00	0.0061	16
	West Point N	4	0.721	4.5	0	0.0047	5.23
	West Point S	7	0.946	8.57	0	0.0034	9.52
	Myrtle Edwards	7	1.02	9.37	0	0.0044	10.4
	Pier 62	6	34.4	496	5.97	0.0158	536
	Duwamish River	8	2.99	47.35	0.399	0.0327	50.8
	Sinclair Inlet	6	2.44	16.7	0	0.0224	19.2
	Comm Bay	6	1.29	13.2	0.114	0.0281	14.6
	Nisqually	3	3.04	46.0	0	0.0082	49.1
	Hood Canal	6	1.38	11.3	0	0.0003	12.7

Values for n represent individual males in 2015 and 2017
BPA = Bisphenol A
EP = Estrogenic potential

Total PAH-metabolites

A total of 25 PAH-metabolites were measured in the bile of English sole in 2015 and 33 were measured in 2017 (Table C4). The following eight PAH-metabolites were not detected in either year:

- OH2DBT
- PHN3carboxylic acid
- PHN9carboxylic acid
- dihydroxy34dihydro712dimethylBAA
- dihydroxy56dihydroBAA
- dihydroxy34dihydroCHR
- transdihydroxy45dihydroBEP
- dihydroxy78dihydroBAP

The total PAH-metabolites detected ranged from 54.2 to 3159 ng/mL bile in 2015 and from 231 to 1533 ng/mL bile in 2017 (Table C5). Total PAH-metabolites were lowest at Vendovi in 2015, and at Hood Canal in 2017, with value at West Point N (383 ng/mL bile) and S (365 ng/mL bile) in 2017 slightly higher than the lowest value and similar to values from fish in Sinclair Inlet (387 ng/mL bile. Total PAH-metabolites were highest at Eagle Harbor in 2015 and at Myrtle Edwards in 2017, which was similar to the value at the Duwamish River that year (1278 ng/mL bile).

Table C4. Details of PAH-metabolites measured in bile of English sole in 2015 and 2017.

Chemical Class	Chemical Name	Year Analyzed		Notes
		2015	2017	
PAH-metabolites	Me6OH2NPH	x	x	Naphthalene metabolites
	MeOHSumNPH	x	x	
	OH1NPH	x	x	
	OH2NPH	x	x	
	OH2FLU	x	x	Fluorene metabolites
	OH3FLU	x	x	
	OH2DBT	x	x	Dibenzothiophene metabolite; not detected either year
	dihydroxy12dihydroPHN	x	x	Phenanthrene metabolites
	dihydroxy910dihydroPHN	x	x	
	dihydroxydihydroPHN		x	
	OH1PHN	x	x	
	OH3PHN	x	x	
	OH4PHN	x	x	
	OH9PHN	x	x	
	PHN4carboxylic acid		x	
	PHN3carboxylic acid		x	Phenanthrene metabolites; not detected in 2017
	PHN9carboxylic acid		x	
	bis18OHMeANT	x	x	Anthracene metabolites
	dihydroxydihydroANT		x	
	dihydroxy23dihydroFLA	x	x	Fluoranthene metabolite
	dihydroxy89dihydroBAA	x	x	Benzo(a)anthracene metabolite
	dihydroxy34dihydro712dimethylBAA		x	Benzo(a)anthracene metabolites; not detected either year
	dihydroxy56dihydroBAA	x	x	
	dihydroxy12dihydroCHR	x	x	Chrysene metabolites
	dihydroxy34dihydroCHR	x	x	
	dihydroxy56dihydroCHR	x	x	
	transdihydroxy45dihydroBEP		x	Benzo(e)pyrene metabolite; not detected in 2017
	dihydroxy45dihydroBAP	x	x	Benzo(a)pyrene metabolites
	tetrahydroxytetrahydroBAP		x	
	dihydroxy78dihydroBAP	x	x	Benzo(a)pyrene metabolite; not detected either year
Aromatic Organic Compounds	dihydroxy15ATQ	x	x	Anthraquinone metabolites
	OH2ATQ	x	x	
	dihydroxyBPH	x	x	Biphenyl metabolite

Table C5. Mean detected PAH-metabolites (ng/mL, by analyte group) and total PAH-metabolites in bile of English sole collected in 2015 and 2017.

Year	Sampling Location	n	NPH	FLU	PHN	ANT	FLA	BAA	CHR	BEP	BAP	ATQ	BPH	Total PAH-metabolites
2015	St of Georgia	3	7.60	1.67	68.3	<4.5	<15	<1.5	<15	<4.5	<15	36.3	<15	114
	Vendovi	1	3.80	<0.45	20.8	<4.5	<15	<1.5	<15	<4.5	<15	14.6	15.00	54.2
	Pt Gardner	3	4.00	8.67	98.1	<4.5	113	5.27	<15	<4.5	<15	14.7	<15	244
	Eagle Harbor	2	27.0	328	1827	4.40	840	58.0	54.2	<4.5	<15	21.0	<15	3159
	Pier 62	3	18.7	82.0	651	<4.5	390	31.3	30.1	<4.5	<15	47.6	<15	1250
	Duwamish	3	119	111	739	<4.5	380	17.7	27.1	<4.5	<15	19.1	<15	1413
	Sinclair Inlet	3	13.9	22.9	124	<4.5	123	7.20	3.47	<4.5	<15	79.3	<15	374
	Comm Bay	3	26.4	34.9	256	<4.5	237	27.7	33.6	<4.5	<15	40.0	<15	656
	Nisqually	3	8.80	7.13	82.1	<4.5	<15	<1.5	<15	<4.5	9.67	21.7	<15	129
	Hood Canal	3	2.33	1.87	49.7	<4.5	<15	<1.5	<15	<4.5	4.33	5.87	<15	64.1
2017	St of Georgia	6	3.65	0.58	270	<1.5	5.17	<3	<4.5	<4.5	<15	9.23	<15	289
	Vendovi	3	5.60	2.43	236	<1.5	<15	6.03	<4.5	<4.5	<15	10.9	<15	261
	Pt Gardner	8	9.91	21.9	305	0.29	98.6	18.7	12.4	<4.5	12.75	91.2	<45	571
	Eagle Harbor	8	7.79	45.0	514	<1.7	349	38.9	20.5	<4.5	<15	4.40	<17	979
	West Point N	14	1.91	8.00	306	<4.5	46.3	5.92	0.70	<14	<45	14.3	<45	383
	West Point S	14	2.16	9.58	281	<1.5	52.1	5.40	0.49	<14	<45	14.0	<15	365
	Myrtle Edwards	14	10.2	77.4	845	5.59	512	32.3	18.9	1.16	<15	29.3	1.36	1533
	Pier 62	8	24.6	25.8	479	0.38	199	32.5	19.7	<4.5	<15	107	<15	888
	Duwamish	9	26.5	79.3	772	<1.9	301	33.5	23.4	1.04	<45	40.6	<19	1278
	Sinclair Inlet	7	13.9	8.75	243	<1.5	51.3	6.76	3.89	<4.5	<15	59.2	<30	387
	Comm Bay	8	11.0	32.6	291	<4.5	122	18.3	8.14	<14	<15	38.5	<45	521
	Nisqually	5	4.22	5.54	239	<2.1	28.6	<3	<4.5	<14	<15	13.2	<21	290
	Hood Canal	7	2.01	4.30	214	<1.5	7.14	<3	<4.5	<4.5	<15	2.97	<15	231

Values for n represent composites of males in 2015 vs. individual fish (males and females) in 2017
Acronyms at column headings denote metabolites for the following PAHs: NPH = Naphthalene; FLU = Fluorene; PHN = Phenanthrene; ANT = Anthracene; FLA = Fluoranthene; BAA = Benzo(a)anthracene; CHR = Chrysene; BEP = Benzo(e)pyrene; BAP = Benzo(a)pyrene; ATQ = Anthraquinone; BPH = Biphenyl
Values with "<" were not detected above the limit of quantitation (LOQ) for any analyte in the group and the highest LOQ for the group is shown instead; total was calculated using only the detected values, not including the LOQs

Appendix D: Confidence intervals for estimated median difference between samples used in pairwise comparisons

We conducted an analysis to help determine whether the pairwise comparisons of all year-specific locations in this study had sufficient statistical power to appropriately reject the null hypothesis of no difference in the distribution of sample concentrations (see Section 3.3.3.4 for method details). To do this we examined the 95% confidence interval for the estimated median difference between all pairwise comparisons found *not* to be significantly different from one another. Using this approach, we would have less confidence in the findings of "no significant difference" and higher concerns about failing to reject the null hypothesis (i.e., higher risks of Type II error) for comparisons where:

1. The confidence interval does not contain zero, but the adjusted p-value is not significant (if we had more samples, we would potentially see significant p-values)

2. The confidence interval contains zero, but it is a relatively wide confidence interval (>25% of the possible confidence interval window)

3. Comparisons where one of the year-specific locations has an n of only 1 (i.e., Alki 2019).

Figures D1 – D12 below show the output of the confidence interval analysis for the metals and organic chemicals analyzed in this study; these figures include only pairwise comparisons found *not* to be significantly different from one another. Comparisons that were found to be significantly different were clearly powerful enough to discern differences. In each figure, the plus signs are the estimated median differences between samples and colored lines are the 95% confidence intervals of that estimate. Results shown in orange or red indicate cases in which comparisons met one of the three criteria shown above. For these comparisons, we have reduced confidence in the conclusion that there is no difference in the distribution of sample concentrations. Results shown in teal are accepted with higher certainty.

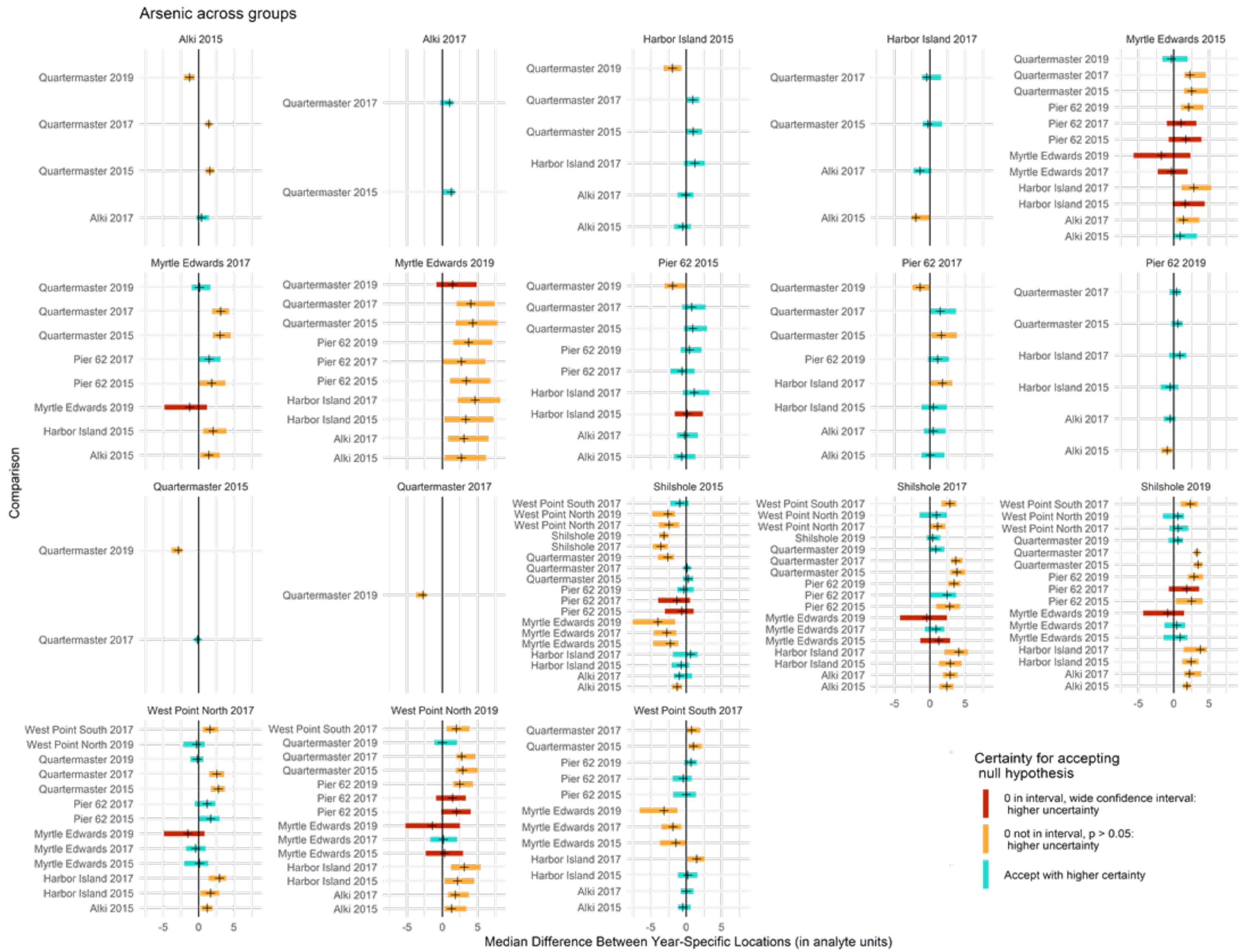


Figure D1. Median (plus sign) and 95% confidence intervals (colored line) for estimated median difference between arsenic concentrations where no significant difference was found (where null hypothesis was accepted). Title of each sub-figure is minuend that difference is calculated from, group name on each y-axis is subtrahends (minuend – subtrahend = difference).

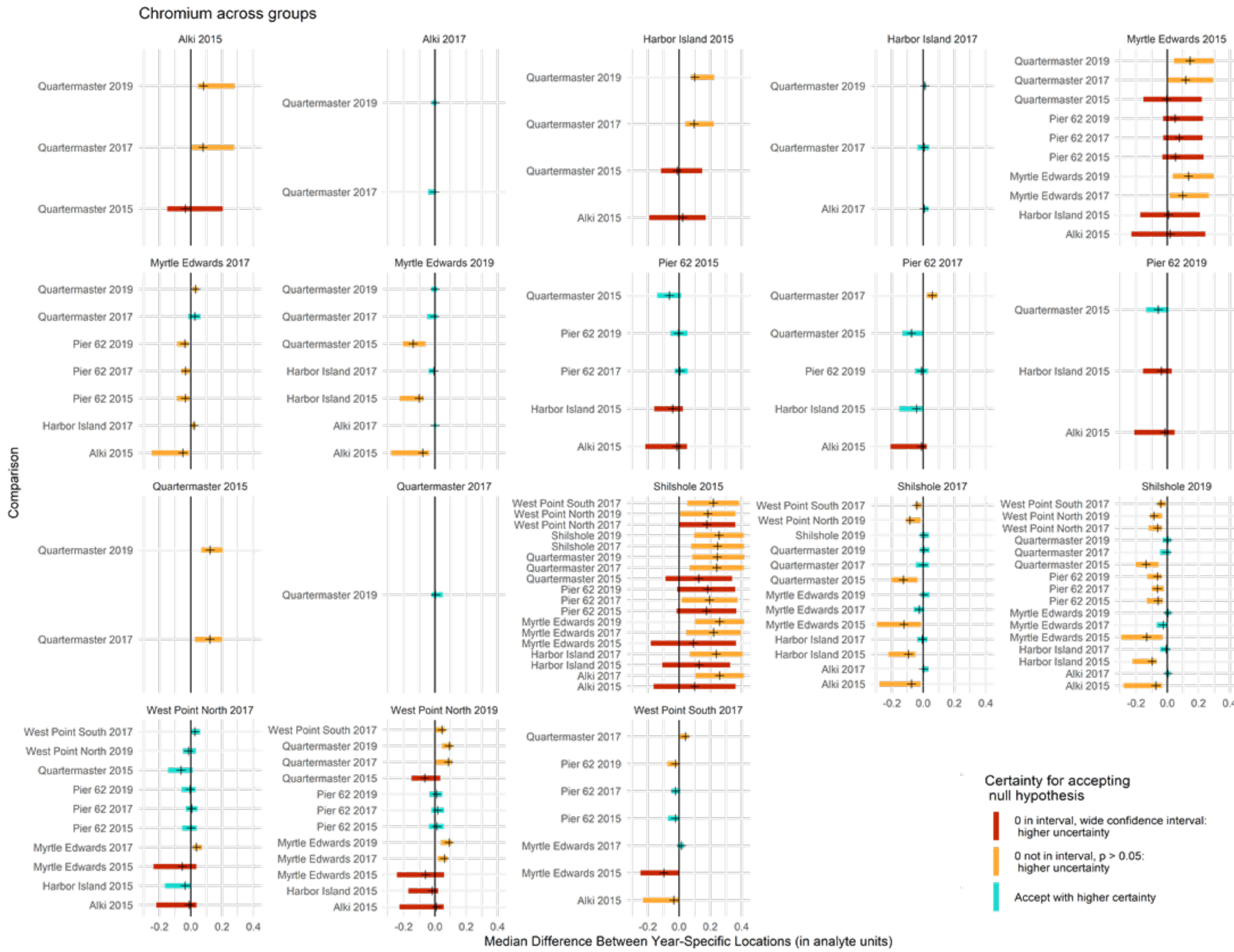


Figure D2. Median (plus sign) and 95% confidence intervals (colored line) for estimated median difference between chromium concentrations where no significant difference was found (where null hypothesis was accepted). Title of each sub-figure is minuend that difference is calculated from, group name on each y-axis is subtrahends (minuend – subtrahend = difference).

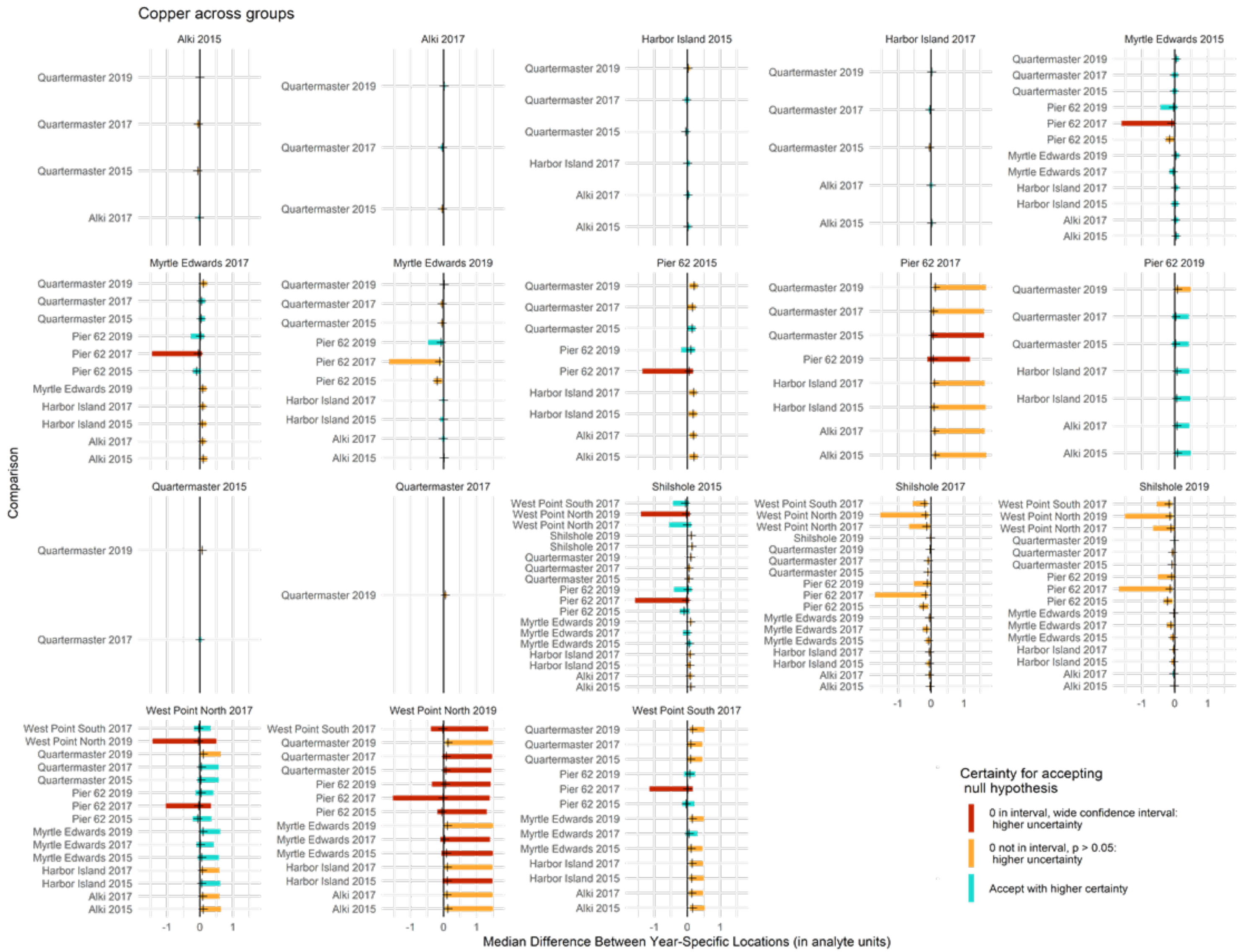


Figure D3. Median (plus sign) and 95% confidence intervals (colored line) for estimated median difference between copper concentrations where no significant difference was found (where null hypothesis was accepted). Title of each sub-figure is minuend that difference is calculated from, group name on each y-axis is the subtrahends (minuend – subtrahend = difference).

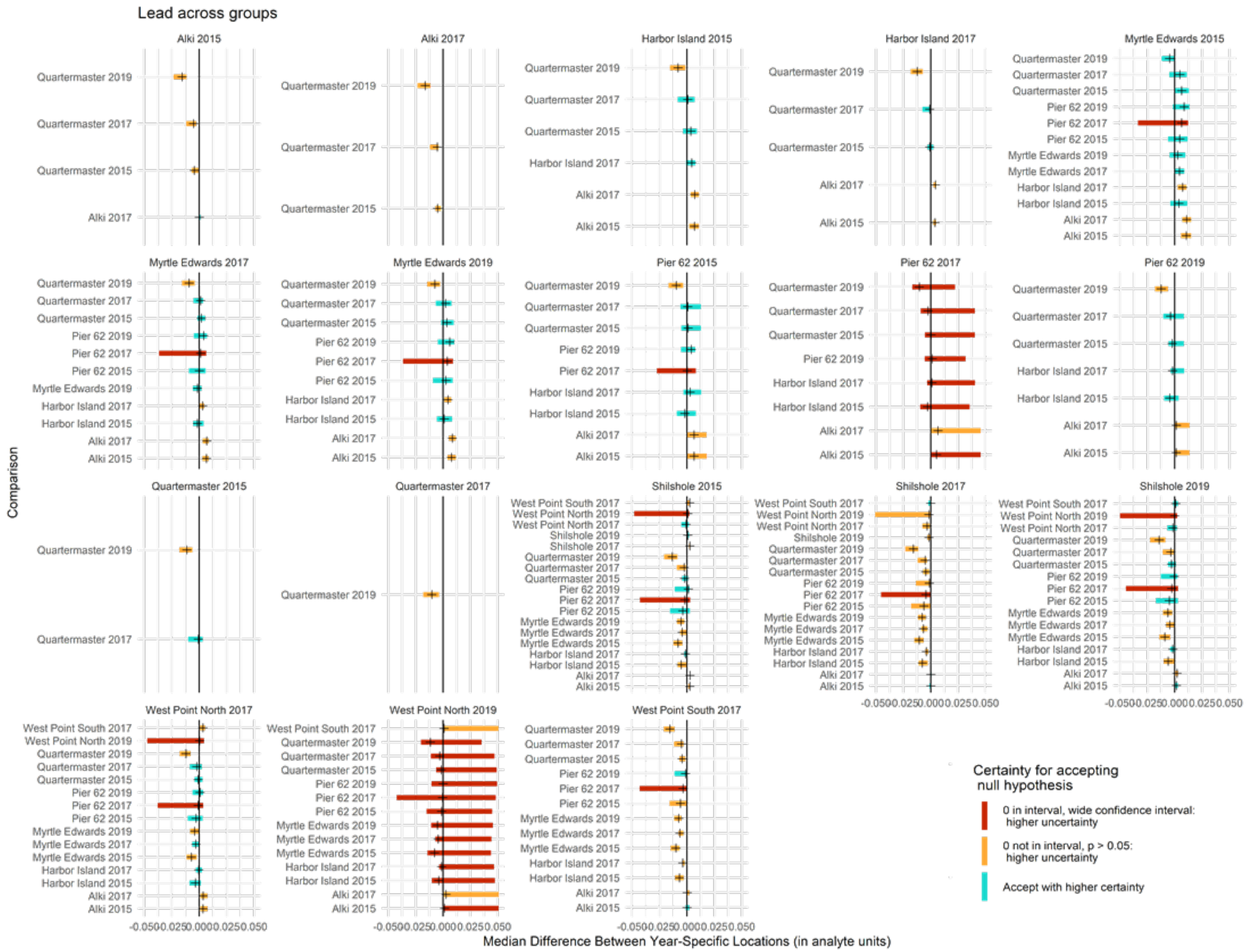


Figure D4. Median (plus sign) and 95% confidence intervals (colored line) for estimated median difference between lead concentrations where no significant difference was found (where null hypothesis was accepted). Title of each sub-figure is minuend that difference is calculated from, group name on each y-axis is subtrahends (minuend – subtrahend = difference).

Figure D5. Median (plus sign) and 95% confidence intervals (colored line) for estimated median difference between mercury concentrations where no significant difference was found (where null hypothesis was accepted). Title of each sub-figure is the minuend that difference is calculated from, group name on each y-axis is subtrahends (minuend – subtrahend = difference).

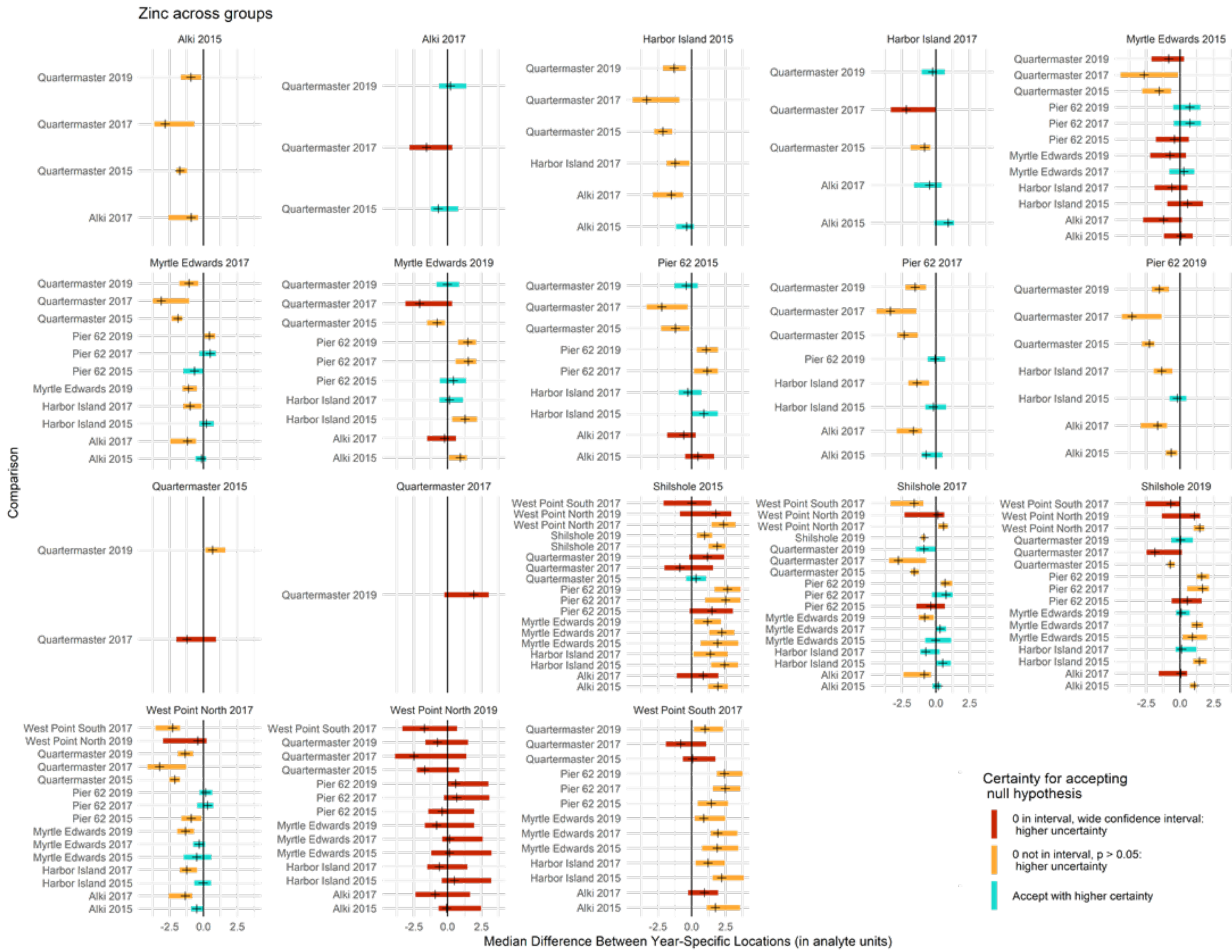


Figure D6. Median (plus sign) and 95% confidence intervals (colored line) for estimated median difference between zinc concentrations where no significant difference was found (where null hypothesis was accepted). Title of each sub-figure is minuend that difference is calculated from, group name on each y-axis is subtrahends (minuend – subtrahend = difference).

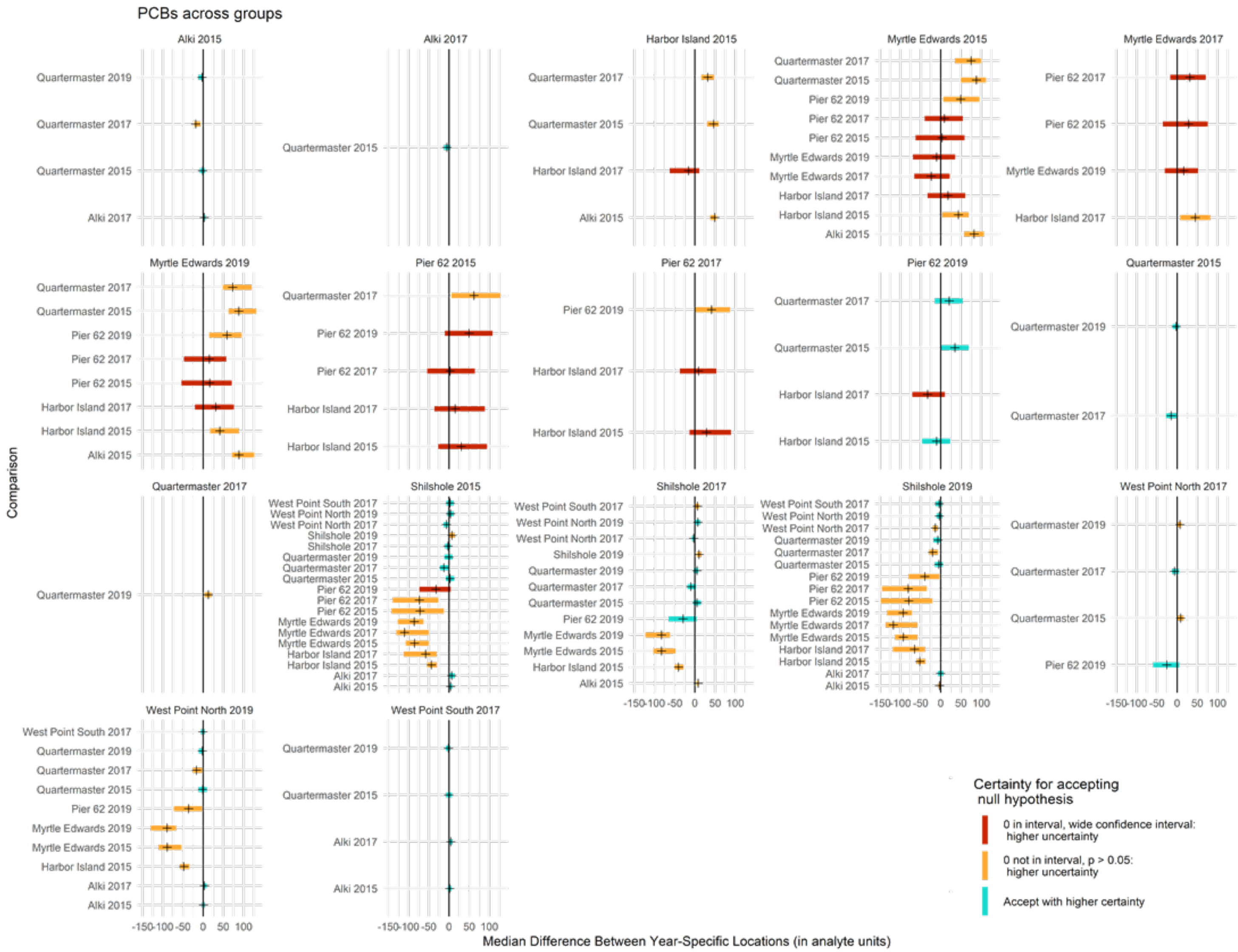


Figure D7. Median (plus sign) and 95% confidence intervals (colored line) for estimated median difference between King County PCBs concentrations where no significant difference was found (where null hypothesis was accepted). Title of each sub-figure is minuend that difference is calculated from, group name on each y-axis is subtrahends (minuend – subtrahend = difference).

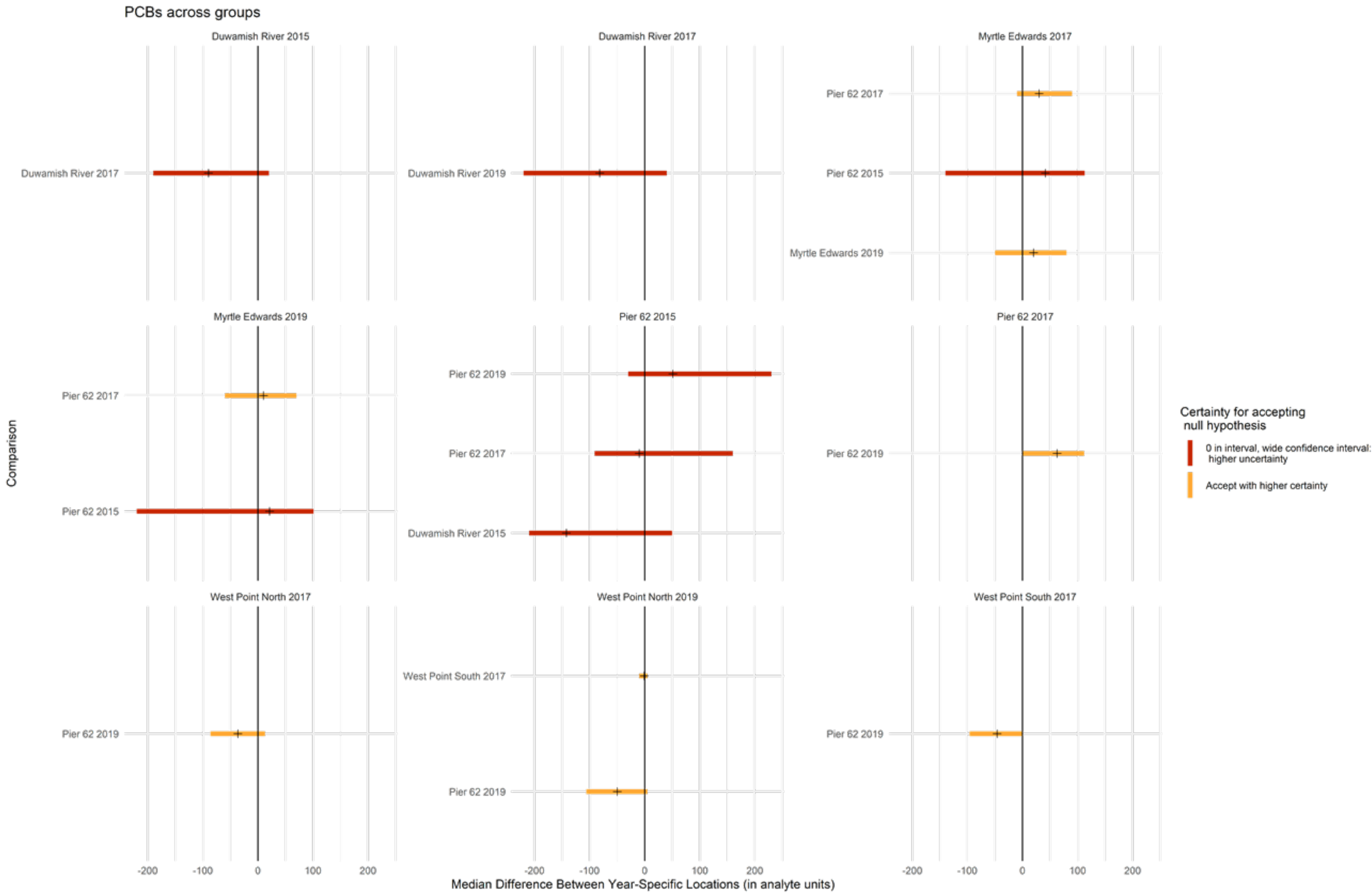


Figure D8. Median (plus sign) and 95% confidence intervals (colored line) for estimated median difference between WDFW PCBs concentrations where no significant difference was found (where null hypothesis was accepted). Title of each sub-figure is minuend that difference is calculated from, group name on each y-axis is subtrahends (minuend – subtrahend = difference).

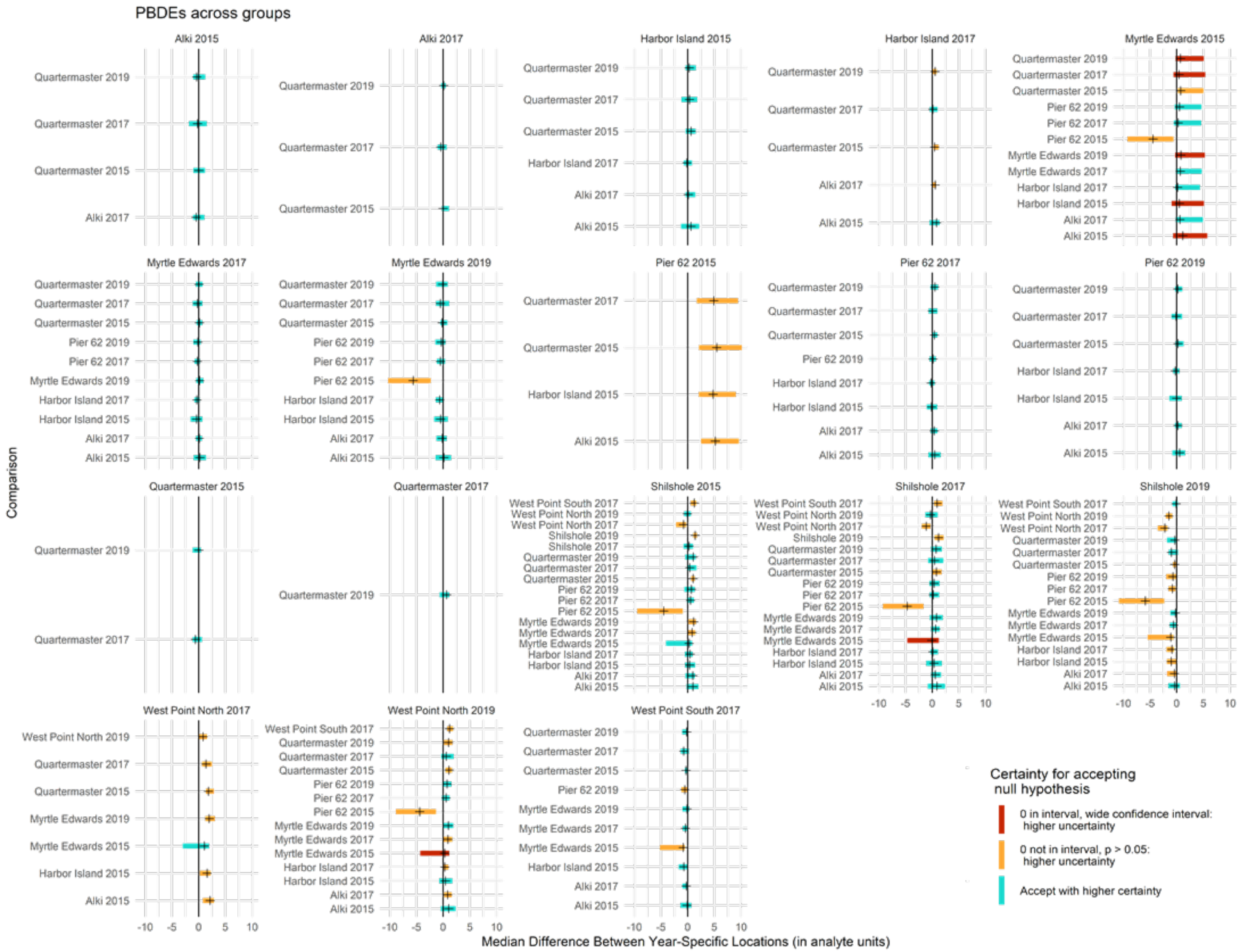


Figure D9. Median (plus sign) and 95% confidence intervals (colored line) for estimated median difference between King County PBDEs concentrations where no significant difference was found (where null hypothesis was accepted). Title of each sub-figure is minuend that difference is calculated from, group name on each y-axis is subtrahends (minuend – subtrahend = difference).

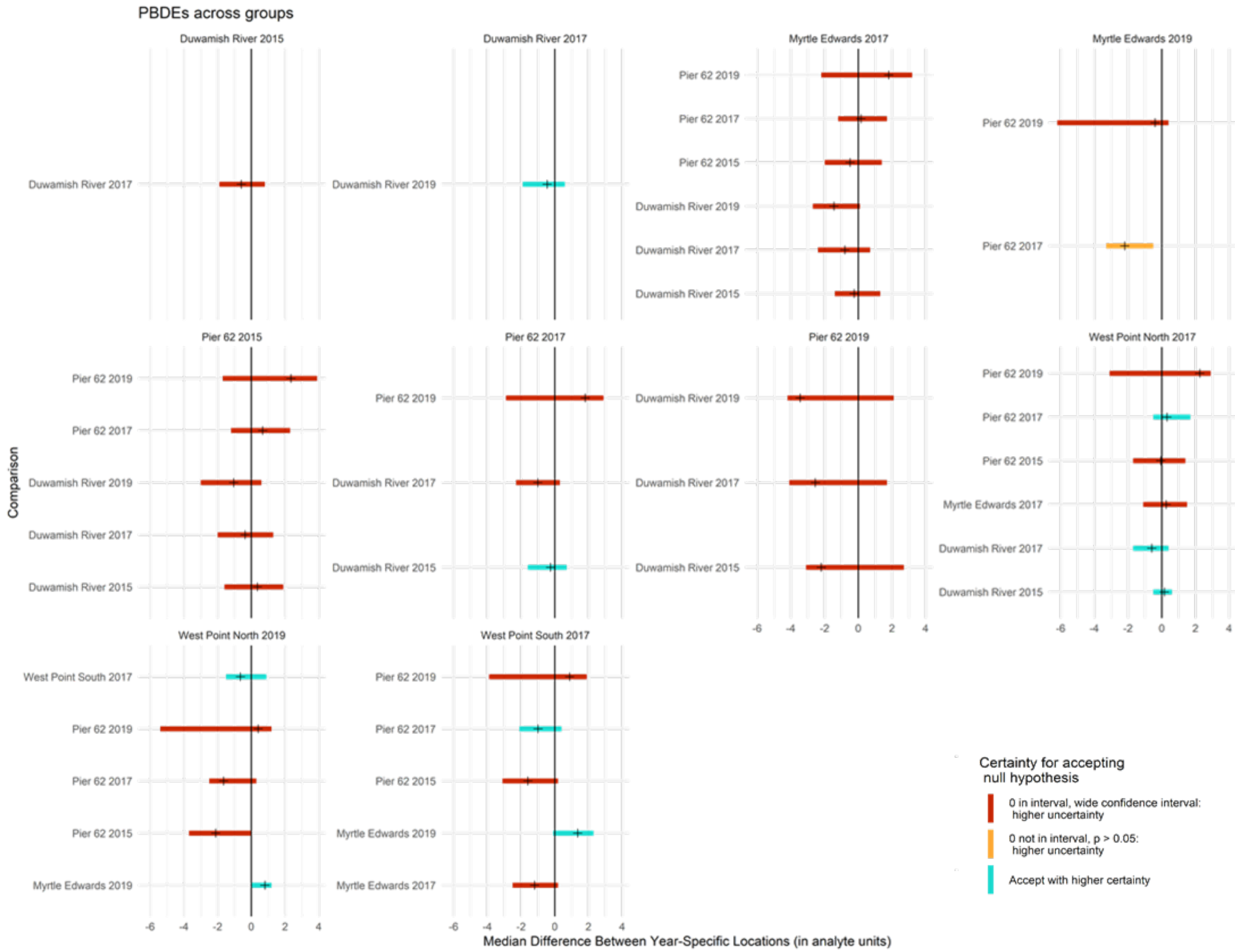


Figure D10. Median (plus sign) and 95% confidence intervals (colored line) for estimated median difference between WDFW PBDEs concentrations where no significant difference was found (where null hypothesis was accepted). Title of each sub-figure is minuend that difference is calculated from, group name on each y-axis is subtrahends (minuend – subtrahend = difference).

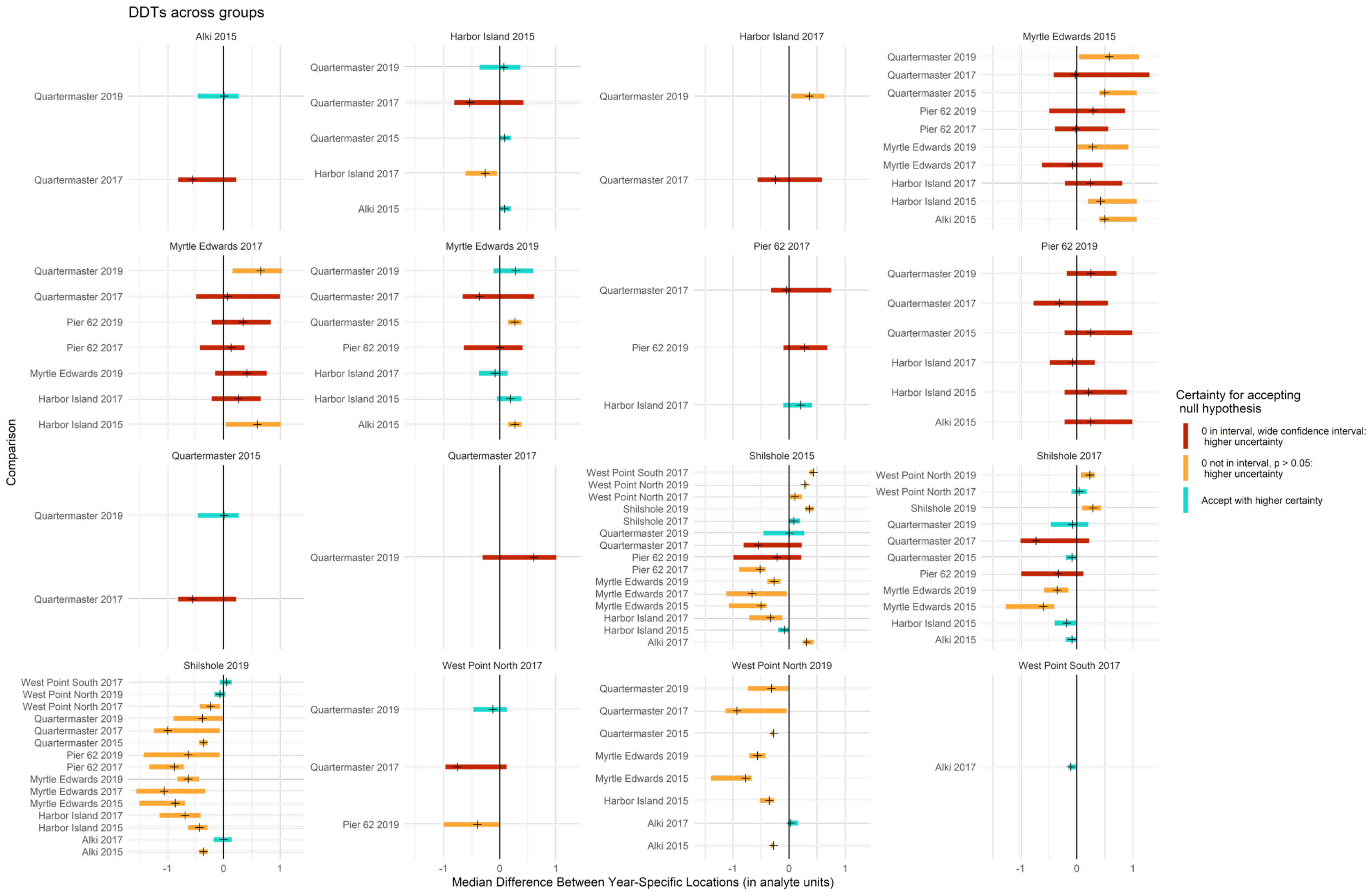


Figure D11. Median (plus sign) and 95% confidence intervals (colored line) for estimated median difference between King County DDTs concentrations where no significant difference was found (where null hypothesis was accepted). Title of each sub-figure is minuend that difference is calculated from, group name on each y-axis is subtrahends (minuend – subtrahend = difference).

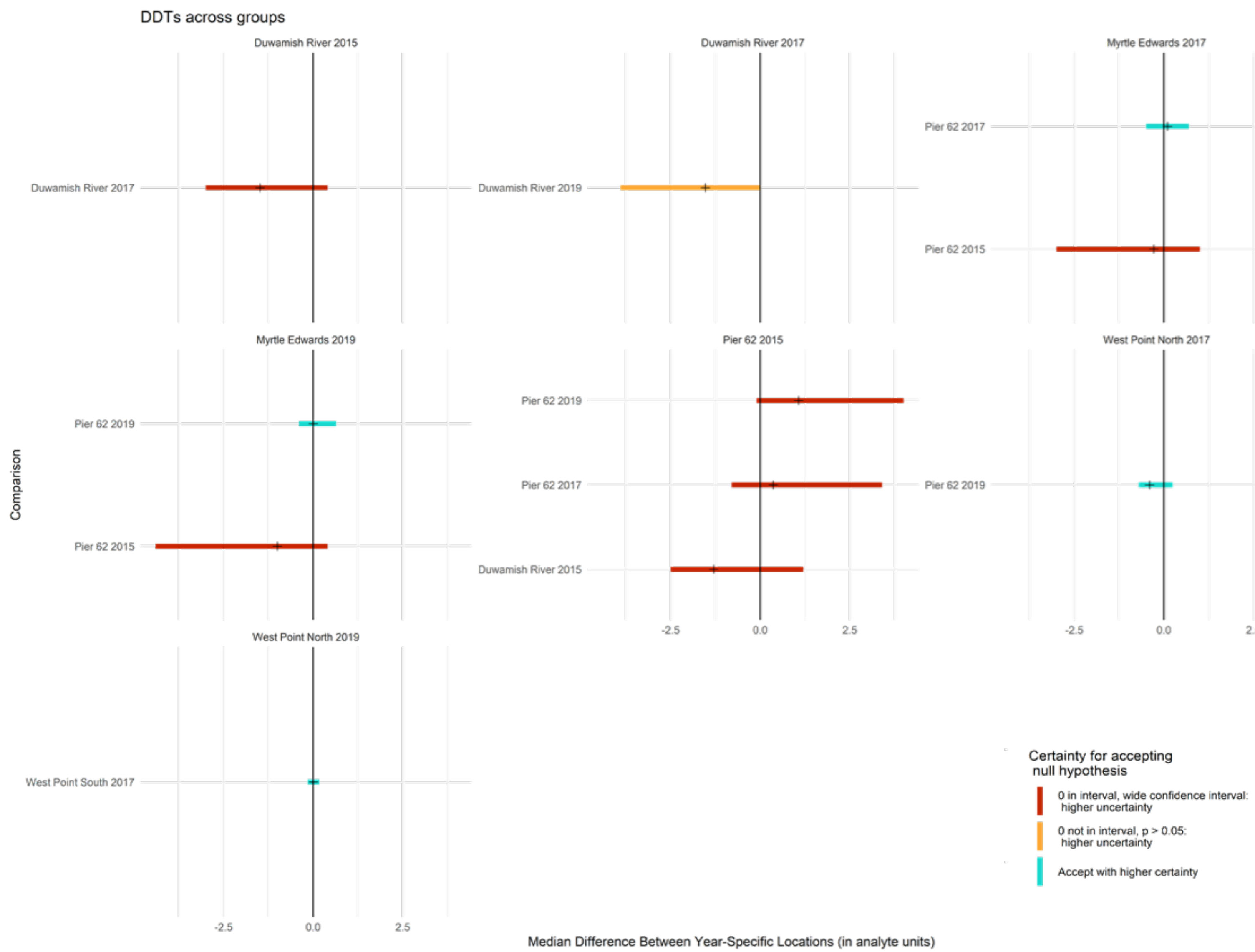


Figure D12. Median (plus sign) and 95% confidence intervals (colored line) for estimated median difference between WDFW DDTs concentrations where no significant difference was found (where null hypothesis was accepted). Title of each sub-figure is minuend that difference is calculated from, group name on each y-axis is subtrahends (minuend – subtrahend = difference).

Appendix E: Comparison of Variances in Total PCBs between King County Stations and Select Historic WDFW Stations

To evaluate year-to-year sample variances, King County data on total PCBs (2015, 2017 and 2019) was compared to historical WDFW total PCB data (2005-2019) taken from sites with a similar degree of anthropogenic influence. The following stations were compared.

- Nisqually Bay WDFW station compared to King County stations:
  - West Point N
  - West Point S
  - Shilshole
  - Alki
  - Quartermaster Harbor
- Elliott Bay (near Pier 62) and Elliott Bay 4 (near Myrtle Edwards) WDFW stations compared to King County stations:
  - Myrtle Edwards
  - Pier 62
  - Harbor Island

Comparison of year-to-year homogeneity of total PCB concentrations revealed that variance was statistically similar between King County and historical WDFW stations. The statistical comparisons are shown on Figure E1 (for West Point N and West Point S), Figure E2 (for Shilshole and Alki), Figure E3 (for Quartermaster Harbor and Myrtle Edwards) and Figure E4 (for Pier 62 and Harbor Island), where concentrations at stations and years that don't share letters are statistically different ($p<0.05$) and black lines represent mean values.

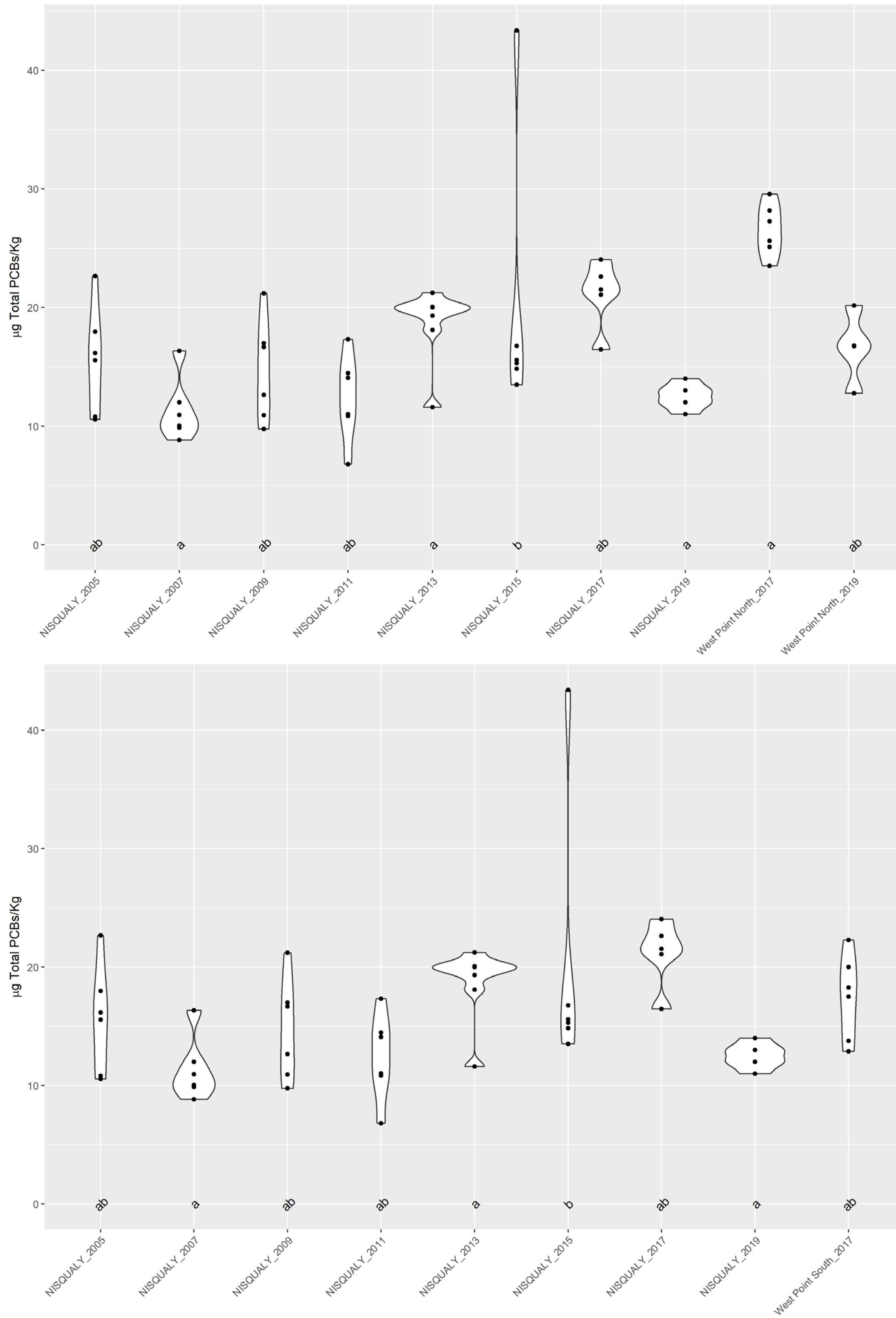


Figure E1. Comparison of year-to-year homogeneity of variances in total PCB concentrations at Nisqually Bay (WDFW historical sampling location) and the West Point N (top) and West Point S (bottom) stations. Concentrations at stations and years that don’t share letters are statistically different ($p<0.05$), black lines represent mean values.

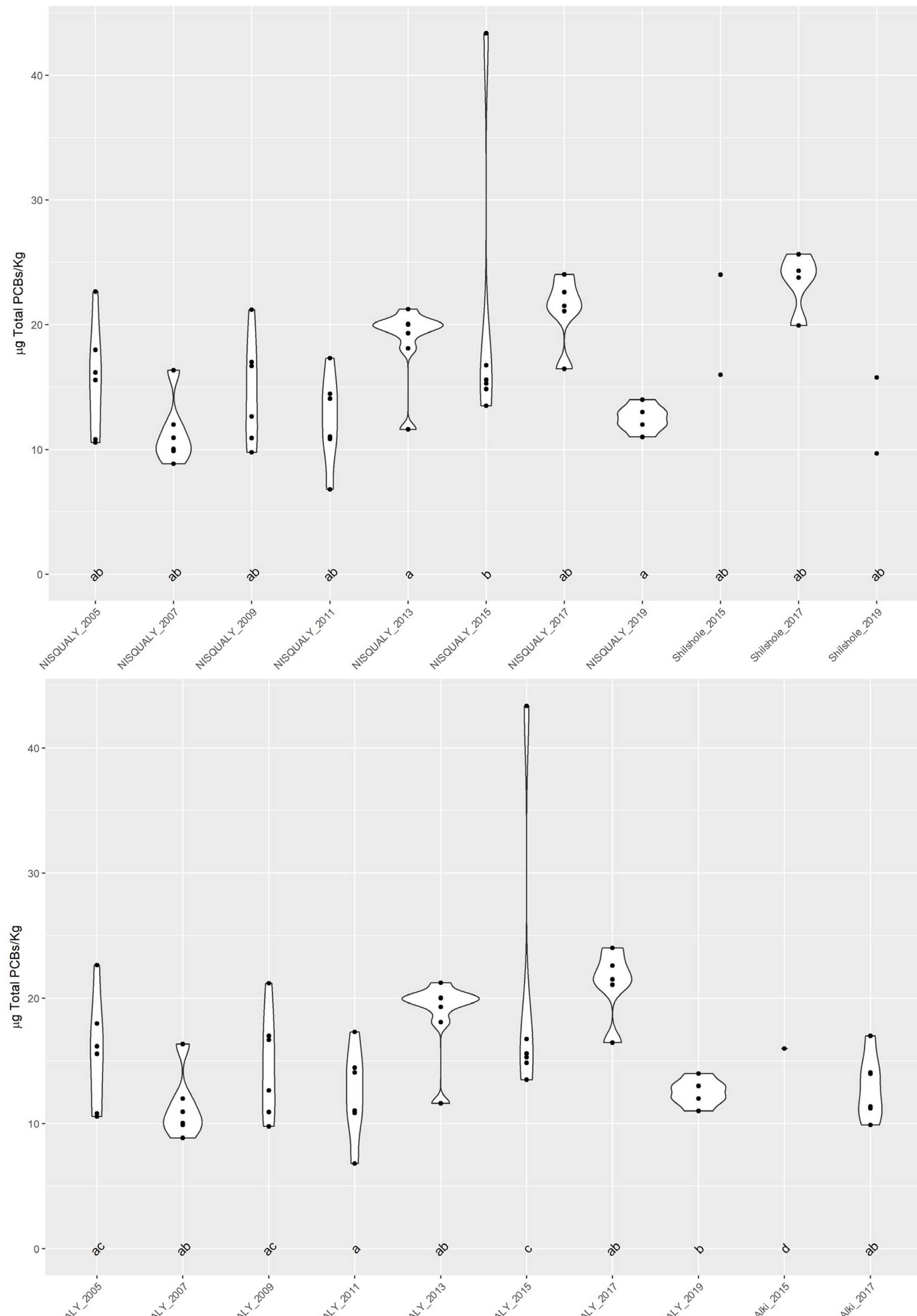
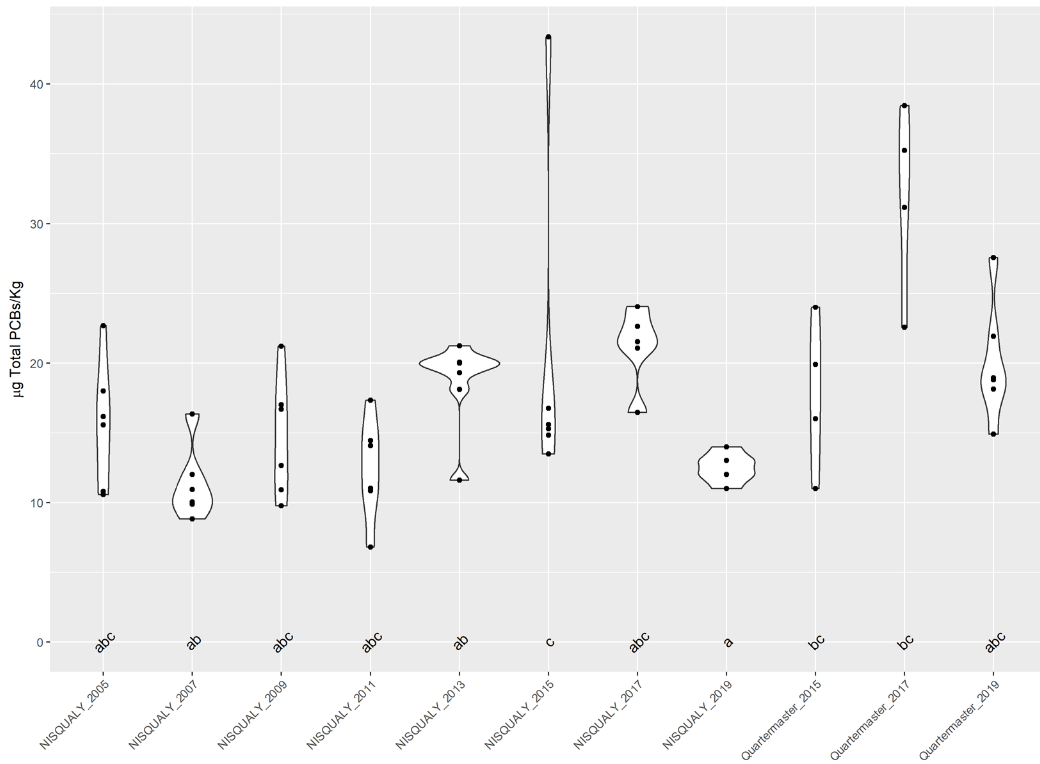


Figure E2. Comparison of year-to-year homogeneity of variances in total PCB concentrations at Nisqually Bay (WDFW historical sampling location) and the Shilshole (top) and Alki (bottom) stations. Concentrations at stations and years that don't share letters are statistically different ($p<0.05$), black lines represent mean values.



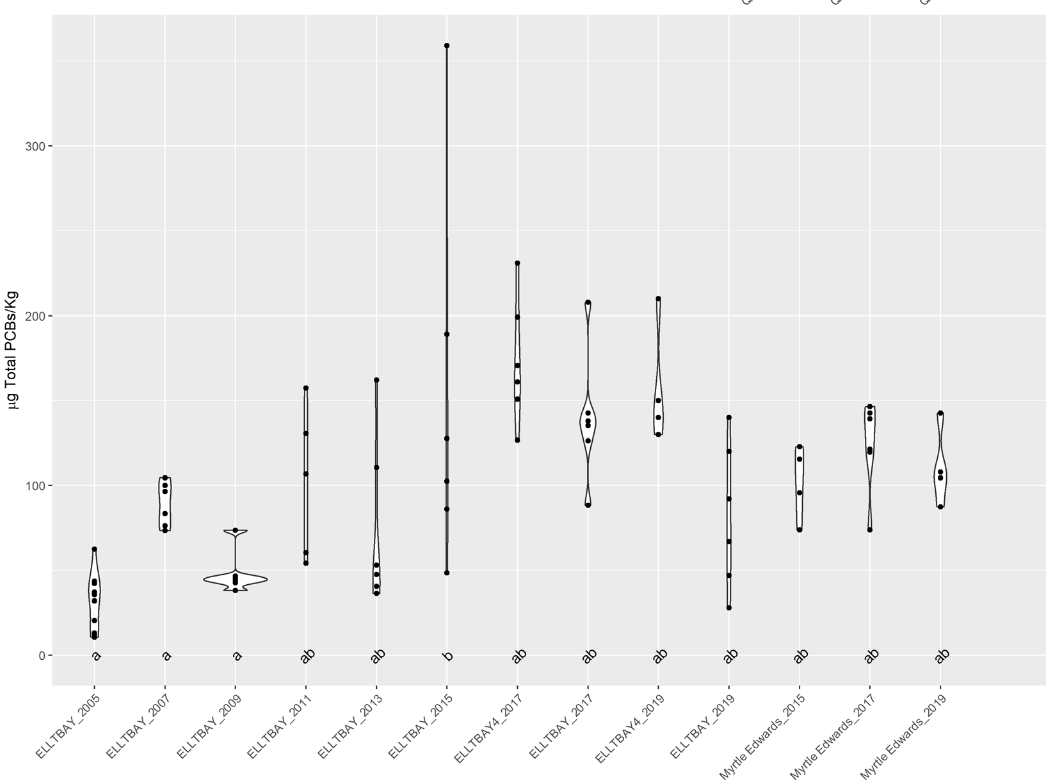


Figure E3. Comparison of year-to-year homogeneity of variances in total PCB concentrations at the Nisqually Bay (WDFW historical sampling location) and Quartermaster Harbor stations (top), and the Elliott Bay and Elliott Bay 4 (WDFW historical sampling locations) and Myrtle Edwards stations (bottom). Concentrations at stations and years that don't share letters are statistically different ($p<0.05$), black lines represent mean values.

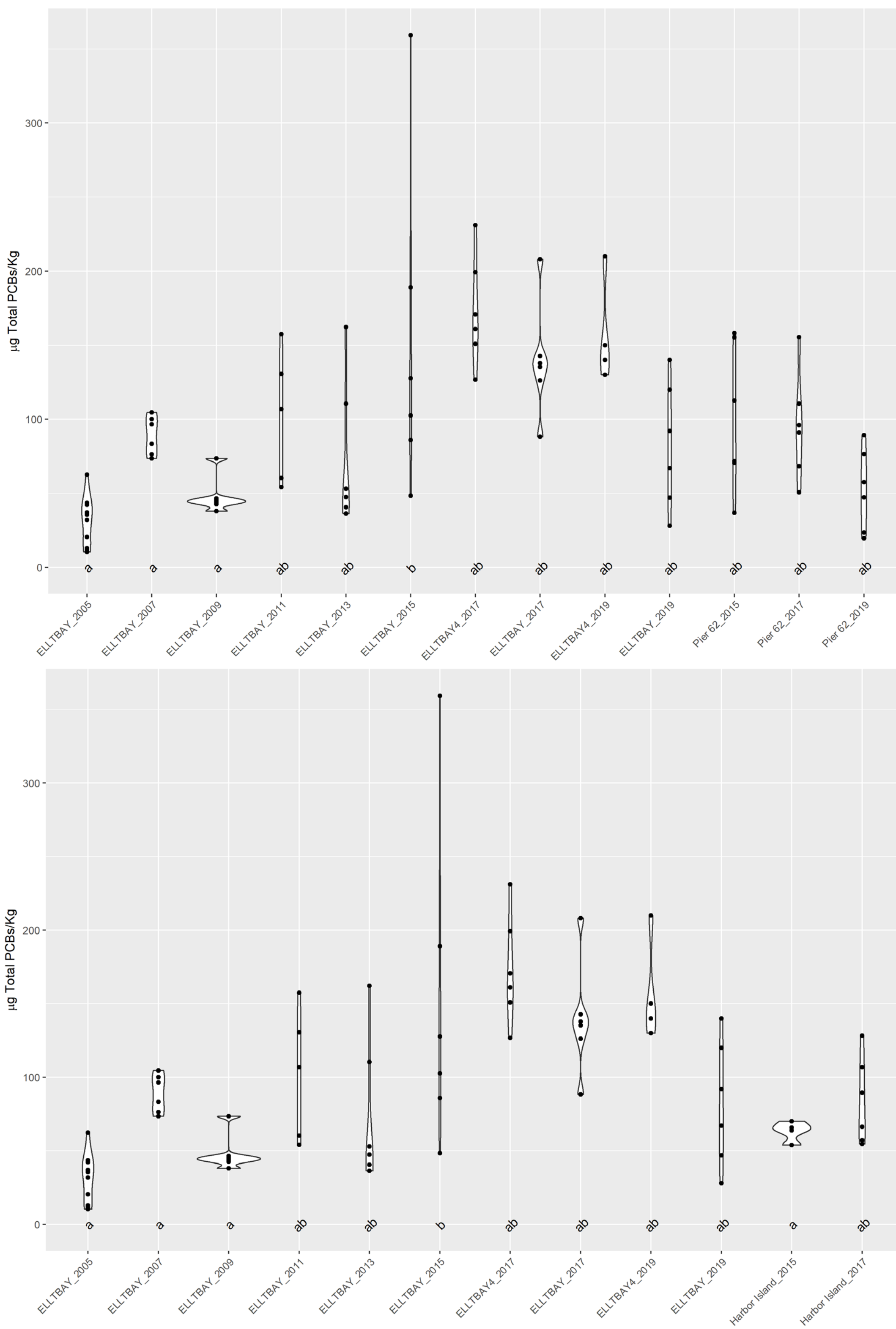


Figure E4. Comparison of year-to-year homogeneity of variances in total PCB concentrations at Elliott Bay and Elliott Bay 4 (WDFW historical sampling locations) and the Pier 62 (top) and Harbor Island (bottom) stations. Concentrations at stations and years that don't share letters are statistically different ($p<0.05$), black lines represent mean values.