A risk-based screening of organic contaminants in juvenile Chinook salmon and their habitat in the Lower Fraser River and estuary, British Columbia

Bonnie P Lo[1,2], Vicki L Marlatt[1], KJ Sadler[3], Mitchell Hoyle[2], David C. Scott[4,5], Misty MacDuffee[4], Stephanie Holbert[1,2], Tanya M Brown[1,2*]

*Corresponding author Tanya_brown@sfu.ca

[1] Department of Biological Sciences, Simon Fraser University, Burnaby, British Columbia
[2] Pacific Science Enterprise Centre, Fisheries and Oceans Canada, West Vancouver, British Columbia, Canada
[3] Water quality monitoring and surveillance, Environment and Climate Change Canada, Vancouver, British Columbia, Canada
[4] Raincoast Conservation Foundation, Sidney, British Columbia, Canada
[5] Pacific Salmon Ecology and Conservation Laboratory, Department of Forest and Conservation Sciences, University of British Columbia, Vancouver, British Columbia, Canada

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Conflicts of interest

The authors declare no conflicts of interest.

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Tsawwassen First Nation, Musqueam Nation, and Sts'ailes Nation. Microsoft Copilot was used as a coding assistant alongside cited R packages.

Data availability

The authors declare that the data supporting the findings of this study are available within the article and its supplementary material.

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Abstract: Many populations of anadromous Chinook salmon (*Oncorhynchus tshawytscha*) in the Fraser River in British Columbia, Canada, have experienced decades of decline, with 85% (14/16) of assessed populations classified as 'Endangered' or 'Threatened'. During outmigration, these juvenile Chinook stocks transit through areas impacted by heavy agricultural use and urban and industrial development in the Fraser River watershed. The lack of biologically relevant assessments of environmental contamination in salmon habitats underscores a notable gap in conservation initiatives. Here, we screened organic contaminants measured in juvenile Chinook tissue and their habitat (i.e., water) to identify compounds that pose a potential risk to Fraser River Chinook populations. Over 595 organic contaminants (spanning eight contaminant classes) were measured in samples from five sites in the Lower Fraser basin. One hundred and thirty out of 288 (45%) of detected contaminants in water and 82 out of 368 (22%) detected in juvenile Chinook tissue were assessed against threshold values. These thresholds included published environmental quality guidelines (EQGs) and a variety of screening benchmark values, which were derived from in vivo, in vitro, or predictive modelling data across multiple databases. Sixteen contaminants were identified as "Priority" contaminants with the potential for adverse effects, and 23 were classified as "Watchlist" contaminants worthy of secondary monitoring. These included pharmaceuticals and personal care products, pesticides, polycyclic aromatic hydrocarbons, flame retardants and polychlorinated biphenyls. This preliminary risk-based screening of organic contaminants in the Lower Fraser River serves to inform monitoring and recovery efforts for culturally-valued Chinook populations, as well as the Endangered Southern Resident killer whales that depend on them.

Keywords: Chinook salmon; organic contaminants; emerging contaminants; Lower Fraser River; risk-based screening

1.0 Introduction

Chinook salmon (*Oncorhynchus tshawytscha*) the largest of the Pacific salmon, are an important recreational and commercial fisheries species and are the preferred prey for the

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Endangered Southern Resident killer whales (*Orcinus orca*; SRKW). In the summer up to 90% of SRKW prey is comprised of Chinook salmon from the Fraser River (Hanson et al., 2010). The Fraser River in British Columbia (B.C.), Canada, is one of the largest salmon rivers in the world, supporting all five Pacific salmon species and has served as the largest Canadian producer of Chinook salmon (Fisheries and Oceans Canada, 1999). Unfortunately, Chinook salmon numbers and average size along the west coast of North America have been steadily decreasing for decades in most populations (Atlas et al., 2023) and these declines are implicated in the lack of recovery for SRKW (Ward et al., 2009). More than 85% (14/16) of the assessed Chinook populations that inhabit the Fraser River have been classified as 'Endangered' or 'Threatened' (Committee on the Status of Endangered Wildlife in Canada, 2018, 2020). Identified threats to Fraser River Chinook salmon include unsustainable harvest, habitat loss, climate change, pathogens and changes in freshwater and marine habitat quality (Committee on the Status of Endangered Wildlife in Canada, 2018).

The Lower Fraser River has been heavily altered by anthropogenic activities including, but not limited to, forestry, mining, pulp and paper, wood preservation, chemical manufacturing, runoff (e.g. stormwater, agriculture) and wastewater (sewage) treatment (Shaw & Tuominen, 1999). These past and current activities have contributed to the presence of wide range of organic contaminants, including legacy contaminants and contaminants of emerging concern in the river. Such organic contaminants, include polycyclic aromatic hydrocarbons (PAHs), dioxins and furans, pesticides, petroleum hydrocarbons, fire retardants, per- and polyfluoroalkyl substances (PFAS), pharmaceuticals and personal care products (PPCPs), and vertebrate hormones/hormone-mimicking substances (MacDonald et al., 2011; Shaw & Tuominen, 1999). In the 1990s, the Fraser River Estuary Management Program sampled water in the estuary for metals and a variety of organic compounds (chlorophenolic compounds, nonylphenol, absorbable organic halides, industrial chemicals, resin and fatty acids, pesticides, and PCBs; Drinnan & Humphrey, 1997; Shaw & Tuominen, 1999). However, since the disbandment of this program, a large, coordinated effort to capture the breadth of possible organic contaminants and their risk to Chinook salmon and their habitat in the Lower Fraser has not been conducted.

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Studies conducted in Puget Sound, a large urban estuary, in the neighboring state of Washington, U.S.A. to the south of the Fraser River estuary, have demonstrated the impacts that contaminants can pose to juvenile Chinook salmon. For example, juvenile Chinook transiting through contaminated estuaries in the Greater Puget Sound area had a 45% lower survival rate compared to those migrating through uncontaminated estuaries (Meador, 2014). Meador et al. (2016) sampled water and fish tissue in three estuaries in the area and detected emerging contaminants (e.g., PPCPs, PFAS, and alkylphenols) that were linked to wastewater treatment plant (WWTP) effluent and at levels that may cause effects to fish. At two WWTP effluent-receiving estuaries in the area, juvenile Chinook were found to have evidence of metabolic disruption compared with reference site Chinook (Meador et al., 2020). Together, these studies focused on site-specific associations (juvenile Chinook in more contaminated or effluent-receiving estuaries exhibited reduced survival or metabolic disruption), rather than establishing chemical-specific concentration-effect correlations. In laboratory exposures using environmentally relevant (0.1–20%) WWTP effluent concentrations from the same region, juvenile Chinook displayed altered energy metabolism, suggesting potential adverse effects on growth, reproduction, behavior, and immune function (Meador et al., 2024). Sublethal effects were also reported in a study using WWTP effluent from Victoria, B.C., where juvenile Chinook exposed to concentrations between 0.05% and 10% effluent showed significant alterations in gene expression, further reinforcing the biological relevance of WWTP-derived contaminants in salmon-bearing waters (Osachoff et al., 2013). These findings highlight the need for conducting detailed contaminant profiling and an evaluation of the associated risks posed to juvenile Chinook in the Lower Fraser River.

Chinook, along with other Pacific salmon, have a varied life histories. While adult salmon spend considerable time at sea, spawning and the early life-stages take place in freshwater. Each spring, Chinook fry emerge and either migrate to sea within three months (ocean-type) or spend one or more years in freshwater before seaward migration (stream-type; Healey, 1998). Ocean-type Chinook fry utilize estuarine and nearshore habitats as key feeding areas, where they grow between 0.5 mm to 1 mm in length per day (Bottom et al., 2005). The Harrison (River) Chinook stock, which are ocean-type and numerically dominate the Lower Fraser River

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stocks, spend 30 to 50 days in the Fraser River estuary (Chalifour et al., 2021). Escapements of Harrison Chinook have been declining by approximately 6% per year for the last three decades and are currently considered *Threatened* (Atlas et al., 2023). Survival of ocean-type Chinook has been reported to be positively correlated with the percentage of estuary in natural condition across United States coastal specifically Washington, Oregon, and California; (Magnusson & Hilborn, 2003). Given that Harrison and other Fraser Chinook stocks utilize the heavily urbanized Lower Fraser River and estuary during an early, sensitive life stage, it is important to characterize and prioritize contaminants in both juvenile Chinook and their habitat.

The objective of this study was to measure organic contaminants in the Lower Fraser River and its estuary and juvenile Chinook tissues and prioritize these contaminants based on the potential risk of adverse effects to this Pacific salmonid. By comparing detected contaminant concentrations in tissue and water to not only available environmental quality guidelines (EQGs) but also multiple alternative screening benchmarks derived from either in vivo, in vitro, or modeled data, this study maximizes the currently available toxicological information to thoroughly assess the potential risk of adverse effects of organic contaminants on a threatened population of Fraser River Chinook (i.e., Harrison) and their habitat.

2.0 Methods

2.1. Water and tissue sample collection

Water and whole-body juvenile Harrison Chinook salmon samples were collected from four heavily urbanized sites in the Lower Fraser River and estuary and one upriver, semi-urban site (Table 1, see online supplementary material, S1; Figure 1). The Lower Fraser River leaves Hope, B.C., as a single stem and bifurcates into two arms (North and South) as it reaches the estuary. Before discharging into the Strait of Georgia, the Lower Fraser River flows through the populous regional districts of the Fraser Valley and Metro Vancouver, which together are home to over 3 million people (Statistics Canada, 2025). Impacted (heavily urbanized sites) sites included: one in the main arm of the lower Fraser upriver of bifurcation (FWM); two in the south arm (MAN, which is downriver of the largest wastewater treatment plant (secondary treatment) in Metro Vancouver and FSA which is near the mouth of the river); and, one in the north arm near the mouth of the river (FNA). In addition to wastewater effluent, these impacted sites are

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influenced by multiple anthropogenic activities, including industrial operations, agriculture, and major roadways. The up-river semi-urban site (i.e., HAR) was located near the mouth of the Harrison River, one of the largest salmon producing tributaries of the Fraser River, which is the origin of the Harrison Chinook stock examined in this study. The HAR site may have contaminant input from a wastewater treatment plant (tertiary treatment), albeit one that services a small population of approximately 1,500 (Statistics Canada, 2025). Sampling occurred in spring 2021 (March to May) at HAR, and over two outmigration seasons (April to June of 2020 and 2021) at the estuary sites. Up to six samples were collected per site, though the number varied due to logistical constraints (Table 1). Surface water grab samples totaled three at HAR (all collected in 2021) and five at each of the heavily urbanized sites (two in 2020 and three in 2021), while tissue samples ranged from three to six per site. Juvenile Chinook were collected using beach seines, following the sampling methods described in Scott et al. (2024). Tissue samples were not collected at FWM, as the site was not suitable for beach seining. All fish collections were performed under an approved Simon Fraser University Animal Care Committee protocol (#1313-B20). For tissue analysis, juvenile Chinook (n = 853 fish) from each outmigration season were collected to create 18 whole-body composite samples which were comprised of 23 to 87 individual fish (Table 1). Juvenile Chinook salmon mean weight across all sites was 0.883 g (± 0.530 g standard deviation), with individual weights ranging from 0.314 to 4.755 g. Tissue samples were frozen and stored at –20 °C prior to processing for analyses. To ensure only Harrison River Chinook samples were selected for analysis, genetic stock identification was conducted on a fin clip collected from each fish at the at the Pacific Biological Station Molecular Genetics Laboratory (Fisheries and Oceans Canada, Nanaimo, B.C.) using single nucleotide polymorphisms (SNPs) marker panel based on 547 SNPs (Beacham et al., 2021). Samples were analyzed for over 595 organic contaminants, from nine chemical classes including alkylphenols (n = 4, both matrices), polychlorinated dibenzo-p-dioxins (PCDDs, dioxins) and dibenzofurans (PCDFs, furans; n = 17, both matrices), polycyclic aromatic hydrocarbons (PAHs; n = 76, both matrices), flame retardants (polybrominated diphenyl ethers (PBDEs; n = 40 contaminants, both matrices) and non-PBDE flame retardants (n = 39 and 27 in water and tissue, respectively)), polychlorinated biphenyls (PCBs; n = 162, 159 in water and

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tissue, respectively), per- and polyfluoroalkyl substances (PFAS; *n* = 40 in both matrices), legacy and current use pesticides (*n* = 90 and 76 in water and tissue, respectively), pharmaceutical and personal care products (PPCPs; *n* = 146 and 141 in water and tissue, respectively) and short, medium and long-chain chlorinated paraffins (*n* = 66; tissue only). Samples were analyzed by SGS AXYS Analytical Ltd., Sidney, B.C. Note that hydrocortisone in juvenile Chinook tissue samples while measured, was excluded from data analyses as the compound cannot be differentiated from endogenous cortisol (M. Woudneh, SGS AXYS Analytical Ltd, Sidney, B.C., Canada, personal communication). To streamline data presentation, bisphenol A and bisphenol S (the only bisphenols detected), were grouped within the PPCP class because both have been associated with municipal WWTP effluent (Qian et al., 2021). The full list of analytes is listed in Table S2 and analytical methods are described in Table S3 (see online supplementary material). All analyte measurements below the detection limit were replaced with a zero and all measured concentrations were blank-corrected.

2.2. Screening benchmarks for biological response ratios

Chemicals detected in both water and tissue were prioritized using multiple approaches that involved evaluating the ratio of the measured contaminant concentration and available screening benchmarks (SBs) to calculate a Biological Response Ratio (BRR, adapted from James et al. 2023). Screening benchmarks used in BRR calculations were based on up to four data sources: environmental quality guidelines (EQGs); B.C. aquatic life benchmarks; hazard concentrations (at 5%; HC5); and activity concentration at cutoff (ACC). In this way, the SB was a concentration estimated to be protective of aquatic life from sublethal and lethal effects. All four SB types were used to prioritize contaminants detected in water, however only EQGs and ACCs were applied to contaminants detected in juvenile Chinook tissue samples because HC5 and B.C. aquatic life benchmarks are largely derived from in-vivo data across many taxa. When searching databases, contaminants were identified using Chemical Abstracts Service (CAS) numbers to minimize discrepancies related to contaminant name synonyms. Co-eluting PCB congeners were assigned a CAS number corresponding to the dominant co-eluter based on Frame et al. (1996). Contaminants that were measured but did not have a CAS number were

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omitted from the data analysis (55 contaminants, see online supplementary material, Table S7). Screening benchmarks are listed in online supplementary material, Table S4a-d.

2.2.1. Environmental quality guidelines

Derived by different jurisdictions, EQGs are science-based indicators of environmental quality. If a chemical is at or below an EQG, there is a low likelihood of direct adverse effects. For this study, EQG-based SBs for water and fish tissue were gathered from the B.C. Ministry of Water, Land, and Resource, Stewardship (Water Protection and Sustainability, B.C., 2024) and three national agencies/forum (Canadian Council of Ministers of the Environment (CCME), 2024; Environment and Climate Change Canada (ECCC), 2024; Pest Management Regulatory Agency (PMRA), 2024). Where applicable, the more conservative chronic EQGs were applied (instead of acute) and all B.C. water quality guidelines (i.e. approved, working, interim) were used.

2.2.2. Aquatic life benchmarks for contaminants of emerging concern

In the absence of published EQGs, some jurisdictions provide guidance on how to calculate aquatic life benchmarks to guide contaminant-related assessments. In B.C., the Ministry of Water, Land and Resource Stewardship developed the R package (wqbench) to derive aquatic life benchmarks of contaminants of emerging concern (Tillmanns & Pearson, 2024). wqbench extracts acute and chronic ecotoxicology data from the U. S. Environmental Protection Agency's (USEPA) ECOTOXicology Knowledgebase (ECOTOX; Olker et al., 2022), cleans, standardizes and applies assessment factors (when relevant) to derive chronic aquatic life benchmarks. ECOTOX, the world's largest compilation of curated in vivo ecotoxicity data, includes data regarding >12,000 chemicals from >50,000 references (Olker et al., 2022). The database is updated quarterly, and the September 2024 release was used in this study. Benchmark derivation encompassed a range of endpoints: effect concentrations (ECx), inhibition concentrations (ICx), lethal concentrations (LCx) with effects between 5% and 60%, as well as hypothesis-based concentrations (e.g. NOECs, LOECs). The program excluded time-based endpoints (e.g. LT50), feeding studies, studies without reported effect concentrations, and bioaccumulation/bioconcentration data. Toxicity data were grouped by species, life stage, and effect type, and the most sensitive available endpoint was used to represent each species. Benchmarks derived using wqbench employed one of two approaches: statistical, which is

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based on fitting a species sensitivity distribution to the data set and calculating the concentration at lower fifth percentile of the distribution; or deterministic, where the lowest effect concentration of most sensitive species is selected. To account for differences between acute and chronic data, a standardization factor (of 5 or 10) was applied where applicable. These benchmarks were not derived with the same level of scientific rigour as WQGs (e.g. toxicity data are not reviewed as extensively) and are therefore intended for screening purposes, where exceedances indicate that further investigation is warranted. For additional derivation details, refer to Tillmanns & Pearson (2024).

2.2.3. Hazard concentrations for 5 % of species

Species sensitivity distributions (SSDs) are derived using ecotoxicological data covering a range of species from multiple trophic levels. Hazard concentration (HC) estimates can be calculated from SSDs to estimate a concentration protective of a desired % of species (Aldenberg et al., 2002). Posthuma et al. (2019) modeled SSDs (based on chronic NOEC values) for >12,000 chemicals based primarily on in-vivo and modeled data from over 10 different sources including American and European government databases, reports and primary literature. These data, compiled prior to 2016, were obtained from resources such as ECOTOX (USEPA), the Registration, Evaluation, Authorization, and Restriction of Chemicals program (REACH, European Union), the European Food Safety Authority, the Swiss Federal Institute of Aquatic Science and Technology (EAWAG), and various peer-reviewed studies (see Posthuma et al., 2019 for full list of data sources). For this study, HC5 estimates (which are expected to be protective of 95% of exposed species), were calculated from the Posthuma et al. (2019) SSDs, based on methods used by Aldenberg et al. (2002) and James et al. (2023). Prior to HC5 calculations, SSDs were filtered for quality based on SSD quality scores, as assigned by Posthuma et al. (see online supplementary material, Table S5).

2.2.4. In vitro high throughput screening

The inclusion of in vitro high-throughput biological activity data broadened the scope of data available for BRR derivation and addressed some of the limitations of in vivo centric data sources (e.g. limited organic contaminant data). ToxCast is an USEPA program that provides publicly accessible medium and high throughput data from >20 assay sources (including cell

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lines, proteins, or receptor models) on the effects of chemical exposure on various biological targets for nearly 10,000 chemicals (Richard et al., 2016). The R package toxEval (DeCicco et al., 2025), uses ToxCast data to estimate ACC values and exposure activity ratios (EARs). Similar to a point of departure estimate, an ACC is the concentration at a bioactivity level that is significantly greater than baseline (above the cytotoxicity-associated response level indicative of non-specific activity rather than pathway-specific bioactivity; Blackwell et al., 2017). Exposure activity ratios are quotients of the environmental concentration divided by the ACC. Prior to EAR calculations, ACCs were filtered using data flag exclusions (Blackwell et al., 2017; Corsi et al., 2019; see online supplementary material, Table S6). As there can be multiple bioactivity endpoints (and thus multiple ACCs) for a contaminant, the toxEval derived BRR was calculated as a contaminant's sum EAR.

2.3. Contaminant prioritization framework

The contaminant prioritization framework was developed to assign detected contaminants into one of four groups: Priority, Watchlist, Low-risk and Undetermined (Table 2). Biological response ratio thresholds of >1 were used for the Environmental Quality Guideline (water and tissue), aquatic life benchmark, and HC5 screening approaches. For in vitro high throughput screening, a bioactivity BRR threshold of $>10^{-3}$ was used for water data to maintain comparability with water-quality benchmark evaluations (Corsi et al., 2019). For tissue data, a BRR threshold of >1 was used, and is consistent with previous EAR/toxEval chemical prioritization studies (Corsi et al., 2019). Contaminants that exceeded one or more thresholds were assigned to the Priority contaminants group. Tier classification was based on (i) the number of sites with threshold exceedances, and/or (ii) the frequency of exceedances at individual sites. The criteria for each tier were defined as follows: Priority Tier 1, which was associated with the highest potential risk level, included contaminants that either exceeded threshold values at four or five sites or recorded four or more exceedances within samples collected at a single site; Priority Tier 2 contaminants exceeded threshold(s) at three sites or recorded three exceedances at a single site and Priority Tier 3 contaminants exceeded threshold value(s) at one or two sites or recorded one or two exceedances at a single site.

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While the Priority group assessment focused on individual contaminant risk, the Watchlist assessment centered mixture-related risk. This approach acknowledged that despite not exceeding threshold(s), contaminants together may result in additive toxicity. First, total BRR (the sum of average contaminant BRR at a site [Blackwell et al., 2017]) were calculated to reflect potential mixture risk. Next, Watchlist contaminants were identified as those that did not exceed threshold(s) but were in the 95th percentile based on site BRR. As databases used for each BRR-type ranged in contaminant coverage (size of database), Watchlist assessments of each BRR data type were conducted independently. Watchlist group tiers included the following: Watchlist Tier 1 (primary) contaminants were in the 95th percentile of one or more BRR types at four or more sites and Watchlist Tier 2 (secondary) contaminants were in the 95th percentile of one or more BRR types at 2 or 3 sites. An assessment of total BRR based on tissue data were conducted using toxEval SBs only. There were only nine fish tissue guidelines, 6 of which were PBDE congeners/congener groups, with the remaining 3 for short-chain chlorinated alkanes, medium-chain chlorinated alkanes and perfluorooctane sulfonate (PFOS). As a results, the usefulness of assessing a tissue-centered BRR_{EQG} would be limited, as each guideline would carry disproportionate weight compared to an assessment with a significantly broader range and diversity of contaminants.

Detected contaminants with a compiled SB that were not included in either the Priority or Watchlist groups, were considered “low” priority as they did not exceed thresholds or contribute significantly to a site’s total BRR (see online supplementary material, Table S7). Lastly, detected contaminants without an SB were classified as “Insufficient information” (see online supplementary material, Table S7).

2.4. Statistical Analysis

Statistical analysis was conducted using R (Version 4.5.0; [R Core Team, 2025] via RStudio [Posit team, 2025]). Statistical significance was set at $\alpha = 0.05$. The Shapiro-Wilk test was used to test the normality of BRRs. As the data did not meet assumptions of normality, BRRs were evaluated using the Kruskal-Wallis test, followed by Dunn’s post hoc comparisons with Bonferroni correction to compare BRR among study sites. The analyses incorporated the following R packages: tidyverse (Wickham et al., 2024), dunn.test (Dinno, 2024), and rstatix (Kassambara,

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2023). For tissue analyses, samples from MAN were excluded as the sample size (available pooled homogenate mass) was insufficient for several classes (e.g. PAHs, flame retardants, PFAS, alkylphenols). Also, one sample each from HAR and FSA were excluded from tissue class-based statistical analyses due to insufficient homogenate (sample) weight for certain contaminant class analyses.

3.0 Results

3.1. Contaminant detection in water and juvenile Chinook samples

In water samples, of the >595 (595–612) contaminants measured, 157 were detected at least once at the up-river, semi-urban site (HAR; 26%), while contaminants detected at least once across the Lower Fraser River sites ranged from 204 to 219 (33–36%; Table 1). In terms of per-sample detection, the number of contaminants detected in water ranged from 71 to 103 (12–17%) at HAR, and 74 to 159 (12–26%) at estuary sites. In tissue samples (i.e., whole body juvenile Chinook), of the 646 contaminants measured, the number of contaminants detected in at least one sample at HAR was 259 (40% detection) and in the Lower Fraser River sites ranged from 239 to 339 (37–52%). On a per-sample basis, the number of analytes detected ranged from 205 to 246 (32–38%) at HAR, and 188 to 279 (29–43%) at estuary sites. Analytes were detected in juvenile Chinook from all assessed contaminant classes measured (*n* = 9, alkylphenols, dioxins and furans, PAHs, flame retardants, PCBs, PFAS, legacy and current pesticides, chlorinated paraffins (tissue only), and PPCPs).

3.2. Screening benchmarks

Using four sources was critical to obtaining the broad coverage of compiled SBs for the contaminants detected in both water and tissue. Environmental quality guidelines were limited to only 31 contaminants detected in water and nine in tissue, which included some guidelines that covered summed contaminants (e.g. total PCBs, total penta-PBDEs). Screening benchmarks for contaminants detected in water based on aquatic life benchmarks (derived from the ECOTOX database) totaled 100, while SBs based on HC5 values from Posthuma et al. were available for 49 contaminants. Additionally, 80 toxEval derived SBs using ACC values were identified for detected contaminants in both water and tissue. Some contaminants had SBs derived from more than one source. Excluding WQGs, 25 detected contaminants (in water) had

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values from all three of the other data sources, while 41 and 11, were unique to wqbench, and toxEval, respectively. Furthermore, 27 overlapped between wqbench and toxEval, 17 between toxEval and Posthuma and 7 between wqbench and Posthuma (Figure 2). Together, the four sources yielded SBs that covered 130 of 288 (45%) of individual detected contaminants in water and 82 of 368 detected (22%) tissue.

3.3. Priority and Watchlist contaminants

Fifteen contaminants detected in water (less than 6 % of the detected contaminants) exceeded BRR thresholds based on wqbench, toxEval, and EQGs derived screening benchmarks and therefore classified as Priority contaminants (Table 2). None of the contaminants exceeded HC5 based BRR thresholds. Of these 15 contaminants, over 50% were PPCPs, and the remaining were pesticides (one legacy, one current use), flame retardants, PCBs and PAHs (Table 2). In total five water contaminant compounds or classes (Bisphenol A, caffeine, metformin, endrin, and total PCBs) were identified as Tier 1 Priority (High). Tier 2 (medium high) and Tier 3 (medium) Priority contaminants in water were comprised of four (Bisphenol S, diltiazem, venlafaxine, PBDE-209) and six (cocaine, indeno[1,2,3-Cd]Pyrene, Methoprene, PCB-126, retene, theophylline) contaminants, respectively. For tissue, only PBDE-penta-total exceeded an EQG-based threshold and was classified as a Priority Tier 1 contaminant.

Watchlist contaminants were identified based on those that did not exceed threshold but ranked in the 95th percentile of site-level average contaminant BRR, highlighting compounds with elevated biological risk that fall outside the Priority list but warrant attention due to their potential contribution to mixture toxicity. Twenty-three contaminants were classified as Watchlist contaminants: 14 of those were assigned to Tier 1 (Primary; Table 3), while nine were assigned to Tier 2 (Secondary; Table 3). Furthermore, 13 of the Watchlist contaminants were identified based on water analyses, eight in tissue and two (N,N-diethyl-meta-toluamide (DEET) and Metolachlor) in both water and tissue analyses.

3.4. Biological response ratios totals and profiles differences among sites

Mixture effects at each site were evaluated using two approaches. First, total BRR values were calculated per sample to directly compare overall chemical risk across sites. Second, BRR values were averaged for individual contaminants across samples and then summed per site, providing

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a contaminant profile of cumulative exposure patterns relevant to juvenile Chinook salmon (Figure 3a, b). Contaminants ranking in the 95th percentile based on site-average $BRR_{wqbench}$ values were individually identified, while all remaining detected contaminants with SBs were grouped under 'Other'. Calculation of total BRR value and BRR contaminant risk profiles were conducted using the wqbench and toxEval based SBs only as they offered the most comprehensive toxicological data coverage.

The total water sample $BRR_{wqbench}$ revealed significant site-level differences (Kruskal-Wallis test $\chi^2 = 10.35$, df = 4, $p = 0.0376$), with FSA showing mean-rank scores 17-fold higher than the semi-urban site HAR (adjusted $p = 0.0134$). No other significant differences for total water sample $BRR_{wqbench}$ and no differences for contaminant class BRR profiles were found between or among the sites ($p>0.05$), respectively. The total water sample $BRR_{toxEVAL}$, provided an *in vitro*-based assessment of contaminant mixture risk. Total water sample $BRR_{toxEval}$ values did not vary across the study sites (Kruskal-Wallis $\chi^2 = 1.42$, df = 4, $p = 0.841$) nor did contaminant class BRRs ($p >0.05$).

Contaminant risk profiles generated using $BRR_{wqbench}$ showed that PPCPs contributed greatly to mixture-related risk at south arm sites (Figure 3a). At MAN, located downriver of the largest Metro Vancouver wastewater treatment plant, all contaminants in the 95th percentile were PPCPs (bisphenol S, caffeine, cocaine, diltiazem and metformin) and together, accounted for 90% of total average $BRR_{wqbench}$. Similarly, at the south arm estuary site FSA, caffeine, diltiazem, metformin, and venlafaxine together contributed to 85% of total average $BRR_{wqbench}$. In contrast, at the north arm estuary site FNA, the contaminant profile was more varied, with PPCPs (bisphenol S, caffeine, metformin) and pesticides (endrin and methoprene) accounting for 46% and 24% of total BRR, respectively.

Water $BRR_{toxEval}$ contaminant risk profiles showed high contribution from PPCPs across all sites (Figure 3b). Pharmaceutical and personal care products accounted for 86% of total contribution at HAR and ranged from 72% to 95% across the Lower Fraser sites (i.e., MAN, FSA, FNA). A large portion of the risk due to the PPCPs was driven by Bisphenol A across all sites, comprising of 85% at HAR, followed by 92%, 35%, 41% and 51% at FWM, MAN, FSA and FNA, respectively. Metformin was the second largest contributor to risk at Lower Fraser sites, particularly at MAN

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(48%), and FSA (36%). The remaining contaminants ranked in the 95th percentile were predominantly flame retardants and PAHs. Of the contaminants unique to sites, metolachlor (current use pesticide) was only present in the 95th percentile profiles of the Lower Fraser sites, 4-nonylphenol (4n-OP, an alkylphenol) was exclusive to the 95th percentile profiles at HAR and perfluorooctane sulfonamide (PFOSA, a PFAS) was unique to FNA.

For tissue BRR comparisons, the analyses focused on contaminant class only (not total contaminants BRR). $BRR_{toxEval}$ class-level comparisons for tissues revealed significant differences among the sites for total PCBs ($\chi^2 = 7.35$, df = 2, $p = 0.0253$), and total PFAS ($\chi^2 = 6.10$, df = 2, $p = 0.0474$). Average BRRs for total PCBs at HAR were 2- fold higher than FNA (adjusted $p = 0.0103$). Total PFAS BRRs at FNA were 1.4-fold higher than FSA (adjusted $p = 0.0308$). No other significant differences in tissue contaminant class $BRR_{toxEval}$ values were found among sites. Consistent with $BRR_{toxEval}$ water contaminant profiles, PPCPs also contributed greatly to the tissue contaminant total $BRR_{toxEval}$ profiles (Figure 3c). Pharmaceutical and personal care products accounted for 61% of the total average BRR at HAR, 64% at FSA and 47% at FNA. Bisphenol A was the dominant contributor to the PPCP BRRs at HAR (56%) and FNA (46%), however at FSA, norgestimate was the dominant contributor (50%). The phenolic compound (4-NP) was the second highest contributor to risk at FSA (24%) and FNA (27%), while the second highest contributor at HAR was PFOS (a PFAS; 11%).

4.0 Discussion

Water and juvenile Chinook tissue data were collected and analyzed for more than 595 (between 595 and 612 in water and 646 in tissue) analytes from eight contaminant classes from a semi-urban tributary (Harrison) that feeds into the Fraser River and then at four heavily urbanized sites located along the Lower Fraser River estuary. This comprehensive data set, which included both water and tissue samples enabled a screening and prioritization of contaminants that may pose a health risk to salmonids, particularly Chinook in the Lower Fraser. Overall, 288 and 368 different organic contaminants were detected at least once in water and juvenile Chinook (Harrison stock) tissue, respectively.

The prioritization framework identified PPCPs as the dominant Priority contaminants, alongside the pesticides endrin (legacy) and methoprene, total PCBs, PBDE-209 (BDE-209), several PAHs,

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and penta-BDEs. The Lower Fraser River estuary South and North Arm sites (FSA and FNA) showed higher contaminant risk relative to the semi-urban reference site (HAR). Pharmaceutical and personal care products linked to wastewater effluent dominated the risk profiles for both Fraser South Arm sites (MAN and FSA), while BRR profiles for tissue were dominated by Bisphenol A for HAR and FNA and progestin pharmaceutical, norgestimate, dominated the risk profile for FSA. Our study highlights the need to measure a wide range of chemicals as well as broaden toxicological screening benchmarks beyond published EQGs to assist in contaminant prioritization efforts.

4.1. Use of screening benchmarks derived from multiple sources

The use of screening values derived from multiple sources was key to increasing the number of detected contaminants to assess risk of adverse effects. Of the 288 individual contaminants detected in water, only 24 had either provincial (B.C.) or federal (CCME, ECCC, PMRA) EQGs and only 9 for fish tissue. Of the available EQGs for detected contaminants, most were for PAHs (33%) and pesticides (29%). By adding screening benchmarks from wqbench (based on ECOTOX data), toxEval, and Posthuma et al (2019), both contaminant classes and the number of contaminants within a class increased. For example, while 27 PPCPs were detected in the Lower Fraser only two EQGs were available for these analytes, but by including the additional databases/datasets, and additional 20 screening benchmarks were derived, resulting in 81% PPCP coverage. Also, of the 16 contaminants classified as 'Priority' due to threshold exceedances, only three were identified using EQGs. The remaining 13 were identified using alternative screening benchmarks: nine by wqbench, three by toxEval, and one by both wqbench and toxEval. It should be noted that BRRs generated from in vitro assays (i.e., $BRR_{toxEval}$) provide screening-level indicators of potential biological activity rather than organism-level effects; however, they remain valuable for identifying relative chemical drivers and prioritizing contaminants of concern. Support for the use of multiple databases/datasets (including ECOTOX and TOXCAST) to expand organic contaminant prioritization efforts have also been documented by Maloney et al. (2023) and James et al. (2023).

4.2. Priority and Watchlist contaminants

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The highest tier of Priority contaminants consisted of five contaminants: one pesticide (endrin), total PCBs and three PPCPs (bisphenol A, caffeine, metformin). Both PCBs and endrin are released primarily as a result of human activities and are designated Track 1 substances due their categorization as persistent and bioaccumulative, and thus are considered "CEPA-toxic" under the Canadian Environmental Protection Act (Canadian Council of Ministers of the Environment, 1999a, 2001). Since 1977, Canada has banned the import, manufacture, and sale (for re-use) of PCBs (Environment and Climate Change Canada [ECCC], 2017). However, closed use applications (e.g., electrical transformers) for PCBs continue with a deadline for their elimination in Canada of Dec. 31, 2029. This along with legacy sources of PCBs in the environment have resulted in PCBs being released to aquatic environments through leaks, spills, municipal and industrial effluents, runoff from contaminated soils, leachates from unsecured landfills, and long range atmospheric transport and deposition (CCME, 2001; ECCC, 2017). It is important to note that under this study's prioritization framework, the EQG for total PCBs was based on B.C.'s WQG of 0.1 ng/L, however federally, the CCME has a guideline value of 1 ng/L (published 1987) that is "no longer recommended because environmental exposure is predominantly via sediment, soil, and/or tissue"(Canadian Council of Ministers of the Environment (CCME), 2025b; Water Protection and Sustainability, 2025). While the CCME has not published a tissue guideline focused on the protection of aquatic biota, a tissue residue guideline for the protection of wildlife consumers of aquatic biota does exist and was published in 1998. Furthermore, the USEPA freshwater (chronic) WQG is 14 ng/L (U.S. Environmental Protection Agency, 2024). Given that the former 1998 CCME and current USEPA guidelines are approximately 10-fold and 140-fold higher, respectively, than the 1987 B.C. guideline of 0.1 ng/L, exceedances of the total PCB threshold in this study should be interpreted with caution, and further evaluation based on more current toxicological studies should be conducted. Endrin also has a complicated regulatory history with a previously published CCME water quality guideline that is no longer recommended for use for the same reason aforementioned for PCBs (Canadian Council of Ministers of the Environment [CCME], 2025a) therefore further evaluation into the applicability of a water-based prioritization of endrin may be warranted. Nonetheless, in this study, endrin was classified as a Priority contaminant due to exceedances of a wqbench-

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based threshold at four out of five sites (all heavily urbanized). Endrin, is a synthetic organochlorine insecticide used in Canada between the 1950s to 1980s and current environmental inputs include runoff from treated areas, spray drift, and aerial transport (Canadian Council of Ministers of the Environment, 1999a). Furthermore, neither endrin nor PCBs have biota or fish tissue quality guidelines in the EQG databases reviewed, suggesting that the potential adverse effects due to both water and dietary routes of these bioaccumulative and persistent contaminants (Canadian Council of Ministers of the Environment, 1999b, 2001) may not be adequately assessed.

All three of the water Priority Tier 1 PPCPs have been associated with wastewater treatment plant (WWTP) effluent (Meador et al., 2016). Bisphenol A is an industrial chemical with a wide range of uses including as an intermediate in polycarbonate plastic production and a heat-activated colour developer in thermal paper that is released into the environment during manufacturing, use and disposal of consumer products (Gewurtz et al., 2021). Bisphenol A has been found to affect fish physiology and reproduction through both genomic and non-genomic pathways, interacting with multiple hormone receptors including estrogen, androgen, and thyroid receptors (Faheem & Bhandari, 2021). Metformin, another Tier 1 Priority compound, is a widely used prescription drug used to treat type 2 diabetes mellitus. Metformin has been measured in many aquatic environments (e.g., surface water, ground water, drinking water) and has been associated with oxidative stress, genotoxicity, and growth and morphological alterations in fish (Zheng et al., 2024). The remaining Tier 1 Priority PPCP, caffeine, is commonly associated with food/natural products, and has been shown to cause changes in fish heart rate, ventilation, biochemistry, behaviour and physiology in fish (Bikker et al., 2024; Harada et al., 2022). Although bisphenol A is commonly associated with WWTP effluent, the consistent exceedances of its threshold across all sites (Table 2) suggest that in the Lower Fraser River, additional pathways or sources beyond WWTP discharge may be contributing to its presence and risk to juvenile Chinook. In contrast, the higher frequency of threshold exceedances of metformin (three at MAN and four at FSA) and caffeine (three at FSA) points to an increased localized pathway of contaminant risk, potentially linked to effluent discharge from the region's largest WWTP (secondary treatment) located in the south arm, upriver of both MAN and FSA.

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The water Tier 2 Priority contaminants were Bisphenol S, diltiazem, venlafaxine (all PPCPs) and PBDE-209 (a flame retardant). Bisphenol S, a common substitute for Bisphenol A, may cause a variety of adverse effects in fish including altered sex ratio in offspring, impaired reproductive capacity of male fish (Hao et al., 2022), oxidative stress (Shehna Mahim et al., 2021), and recognition memory (Naderi et al., 2020). Both diltiazem and venlafaxine are commonly prescribed pharmaceuticals, with the former used to treat angina pectoris and hypertension and the latter serving as an antidepressant. Diltiazem has altered the activity of antioxidant enzymes in liver and gill of rainbow trout (Steinbach et al., 2016), while venlafaxine has been associated with altered heart rates in zebrafish (Thompson et al., 2022). The brominated flame retardant, PBDE-209, has been shown to cause a range of sub-lethal adverse effects in exposed zebrafish embryos including yolk and pericardial edema, tail and head malformations, reduced heart rate, and blood stasis (Zezza et al., 2019). The brominated flame retardant, PBDE-209 exceeded thresholds at HAR, FWM, and MAN, pointing to a variety of potential pathways or sources including wastewater treatment plants and industrial effluents. Bisphenol S exceeded thresholds once at each of MAN, FSA and FNA (north arm), suggesting diffuse or unclear pathways rather than a single point of discharge. Similar to Tier 1 pharmaceutical metformin, diltiazem and venlafaxine showed the highest frequency of threshold exceedances at FSA (three times each), with diltiazem exhibiting additional threshold exceedances at MAN (three), also located in the south arm. This pattern of Priority pharmaceuticals further implicates the large WWTP in the south arm of the Fraser River as a likely contributor to elevated PPCP-related risk to juvenile Chinook.

Tier 3 Priority contaminants included two PPCPs (cocaine and theophylline), two PAHs (indeno[1,2,3-Cd]Pyrene and retene), one pesticide (methoprene) and one PCB (PCB-126). Cocaine, a central nervous system stimulant drug, has been shown to cause oxidative damage in zebrafish exposed to environmentally relevant levels (De Farias Araujo et al., 2025) while theophylline, a pharmaceutical used to treat the symptoms of asthma and other respiratory illnesses, induced cardiac changes including heart rate abnormality in zebrafish embryo (Basnet et al., 2017). PAHs, including indeno[1,2,3-Cd]Pyrene and retene, generally occur as complex mixtures and are released into the environment through the incomplete combustion of fuels,

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wood burning (including forest fires), and use in the wood preservative industry (e.g. creosote). Fish toxicity has been observed with many PAHs mixtures, and individual PAHs such as retene have been associated with a range of adverse effects in exposed fish including cardiovascular anomalies (Scott et al., 2011). Environmentally relevant concentrations of PAHs in Washington resulted in reduced biomass and lipid stores in juvenile Chinook, increasing their potential for mortality during their first winter (Meador et al., 2006). Methoprene, a registered insect growth regulator in Canada used for flea, tick, and mosquito control, has been associated with reduced fry length, weight, and altered stress hormone responses in exposed fathead minnows (Ross et al., 1994). PCB-126 exposure has led to pancreatic islet abnormalities in zebrafish (Timme-Laragy et al., 2015) and morphometric changes including reduced body and eye size in sturgeon larvae (Chambers et al., 2012), reinforcing its developmental toxicity across species. Although threshold exceedances of Tier 3 Priority contaminants did not show consistent spatial or temporal patterns, their documented toxicological effects underscore their potential to contribute to sublethal impacts in juvenile Chinook across the Lower Faser River. Lastly, several of the Tier 1, 2, and 3 Priority contaminants in this study (bisphenol A, caffeine, metformin, venlafaxine, and theophylline) were also recognized as priority contaminants in a recent evaluation of emerging concerns (in water, fish, and wastewater treatment plant effluent from an urban estuary in Washington State (James et al., 2023). This highlights the pervasiveness of these contaminants and their associated risks to salmon as well as other aquatic species in urban estuaries.

Interestingly, caffeine (Priority Tier 1) and theophylline (Priority Tier 3) are both antagonists of the adenosine receptor (Biaggioni et al., 1991). Theophylline, a methylxanthine drug commonly used to treat asthma and other respiratory diseases, is chemically similar to caffeine (also a methylxanthine) and likewise, is found in plants products that also contain caffeine (e.g. cacao beans, green tea, coffee, chocolate; Basnet et al., 2017; Menacherry et al., 2022). The shared receptor activity of these Priority contaminants underscores the importance of considering potential mixture toxicity when conducting contaminant assessments in complex aquatic environments.

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The concept of mixture toxicity was central to the classification of Watchlist contaminants, which represented contaminants with the greatest potential to contribute to additive toxic effects despite not exceeding individual thresholds. The Watchlist framework identified contaminants ranked in the 95th percentile of site's average contaminant BRRs. Those at the higher end of the BRR distribution are likely the next group of analytes to potentially pose increased risk to biota in the receiving environment, despite not exceeding threshold levels. The significance of assessing mixture toxicity, rather than focusing solely on individual contaminants, has been demonstrated in studies on pesticide mixtures (Laetz et al., 2009), and structurally dissimilar contaminants from various chemical classes (Walter et al., 2002). While PPCPs made up half of the Priority contaminants, pesticides, PAHs, and flame retardants were the dominant chemical classes among Watchlist contaminants (22, 26, 30%, respectively based on the total number of Watchlist contaminants). When considering potential additive toxicity among Watchlist and Priority contaminants, PAHs for example, can have multiple modes of actions that can activate or inhibit several receptor signaling pathways that other compounds may be affecting. For example, many, but not all PAHs activate the aryl hydrocarbon receptor (AhR). Priority and Watchlist contaminants in this study that bind to and activate the AhR include Indeno[1,2,3-Cd]Pyrene (Tier 3 Priority), benz[a]anthracene, benzo[b]fluoranthene (Larsson et al., 2014), pyrene (Incardona et al., 2005; all Tier 1 Watchlist), and dibenz[a,h]anthracene(Larsson et al., 2014; Tier 2 Watchlist). Furthermore, other halogenated aromatic hydrocarbons (e.g. PCBs, dioxins) may also bind and activate AhR. The AhR is involved in xenobiotic metabolism and activation and may cause a variety of adverse effects including hepatocellular damage, immune suppression, and teratogenesis (Stevens et al., 2009). The presence of multiple PAHs and other AhR activating contaminants suggest that both benchmark and detailed assessments of site toxicity should consider additive (or synergistic) impacts of contaminants on overall aquatic health. While this framework aimed to address risk related with mixture toxicity, the lack of screening benchmarks or water quality guidelines for 160 of the 289 detected contaminants in water (due to limited toxicity data) highlights the uncertainty in risk screening related to the gaps in available toxicological data.

4.3. Site differences in total BRR and contaminant BRR profiles

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Contaminant risk and mixture effects at each site were assessed using total BRR and BRR-based contaminant profiles. Together, these methods offered distinct yet complementary insights into contaminant risk. Significant site variation in $BRR_{wqbench}$ values highlighted elevated contaminant risk at FSA, with a 17-fold higher average risk relative to HAR (a semi-urban site; Figure 3a). Across all contaminant risk profiles (water $BRR_{wqbench}$, water and tissue $BRR_{toxEval}$), PPCPs were consistently identified as the dominant drivers of total mixture risk, particularly at estuary sites FSA and FNA. Caffeine, metformin, and diltiazem were identified as the dominant PPCPs based on $BRR_{wqbench}$ (Figure 3a), while $BRR_{toxEval}$ identified bisphenol A and metformin as the greatest contributors (Figure 3b). In tissue samples, Bisphenol A remained a prominent contributor, apart from norgestimate, an oral contraceptive, which accounted for the highest proportion of risk in the FSA profile (Figure 3c). Additionally, PAHs, flame retardants and PFAS were also identified as significant contributors to total contaminant risk across all sites. Collectively, these findings suggest that these contaminant classes may pose significant additive risk throughout the Lower Fraser River to Chinook health.

4.4. Study limitations

To aid in the interpretation of results, it is important to consider the limitations of this study. CAS numbers were used to link detected contaminants to ecotoxicological data and screening benchmarks. While CAS numbers are intended to ensure consistency in chemical identification, discrepancies may arise due to the use of different CAS numbers across databases or deprecated numbers. Databases (e.g. ECOTOX) may not regularly update previous records with current CAS numbers, therefore some relevant toxicity data may not have been included. Furthermore, to broaden the list of analytes with a screening benchmark, we did not focus on Chinook or other salmonids and instead relied on values derived from various organisms, including invertebrates, fish, and plants. Lastly, in vitro bioactivity data (e.g., data found in ToxCast), though useful in prioritization/screening of contaminants, is not a direct surrogate for in vivo effects (Schaupp et al., 2023), therefore use of in vitro bioactivity SBs should be interpreted cautiously. Furthermore, while this study measured a broad range of organic contaminants (more than 595), there were however some limitations in the scope of contaminants evaluated. For instance, natural and synthetic hormones such as 17α-ethinyl

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oestradiol, commonly associated with WWTP effluent were not included. Synthetic oestrogens in particular have been shown to affect reproduction, and population levels in fish (Marlatt et al., 2022). Other suggested contaminants include a wider range of lipid regulators (pharmaceuticals), UV filters and preservatives, solvents, surfactants and rubber-tire related compounds (including 6PPD-Quinone). Given their potential biological effects and widespread occurrence in aquatic environments, expanding future monitoring programs to include a broader range of hormones and other underrepresented or emerging contaminants is critical for achieving a more comprehensive understanding of ecological risks.

4.5. Broader implications and future research

Of the Priority contaminants identified in this study, 10 of 16 have been linked to WWTP effluent (Meador et al., 2016 and Paíga et al., 2019 [Priority PPCPs], Qian et al., 2021 [Priority Bisphenols], and Samara et al., 2006 [PCBs and PBDE-penta]). There are five WWTP in the region, including one upriver of the Fraser's bifurcation, one in the south arm upriver of MAN and one at the mouth of the river, at the end of the south arm (near FSA; Metro Vancouver, 2025). Many PPCPs are only partially degraded or sorbed during conventional secondary treatment, resulting in their discharge to receiving environments (Lin et al., 2024). Overall, the identified Priority contaminants suggest that WWTP effluent from up to three plants discharging into the Lower Fraser along with additional urban, agricultural, and industrial pathways (as indicated by Priority contaminants such as endrin and methoprene (pesticides), and Indeno[1,2,3-Cd]Pyrene and retene [PAHs]), are contributing to contaminant related risk to the health of juvenile Chinook and their aquatic environment. Further study of these contaminants and their relationship to local WWTP effluents and industrial and non-point sources should be pursued.

Based on contaminant data collected in the Fraser River estuary between 1993 and 1994, the primary organic contaminants of concern were pyrene, benzo(a)pyrene, and phenanthrene, which exceeded provincial water quality criteria in one or more samples (Shaw & Tuominen, 1999). Other organic contaminants including chlorophenols, PCBs, and organochlorine pesticides were near or below detection limits and thus were considered of limited concern at the time of reporting. Although the historical dataset is limited, it suggests that the risk posed

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by PAHs may have shifted since the 1990s. In the present study, neither pyrene, benzo(a)pyrene, or phenanthrene exceeded current water quality guidelines or were identified as Priority contaminants. However, PAHs in water remain contaminants of concern as two PAHs (indeno[1,2,3-Cd]pyrene and retene) were classified as Priority contaminants, and six PAHs (benz[a]anthracene, benzo(g,h,i)perylene, benzo[b]fluoranthene, pyrene, dibenz[a,h]anthracene, fluoranthene were classified as Watchlist contaminants (Tables 1 and 2). Evaluating trends in other organic contaminants detected in the Lower Fraser River remains challenging, as many compounds were not measured in earlier publicly available studies, and analytical methods have changed considerably over the past two decades.

To complete a holistic understanding of contaminant risk to juvenile Chinook and overall aquatic health in the Lower Fraser River and its watershed, assessments of contaminants in sediment, in addition to a comprehensive analysis of contaminants in small tributaries of the Lower Fraser River, should be evaluated. Sediment contamination may affect juvenile Chinook both directly and indirectly. Direct exposure may occur if sediment contaminants redissolve or resuspend into the water column (Barbee et al., 2008). Sediment contaminants may also be taken up by benthic invertebrates, serving as a dietary exposure route for juvenile Chinook (Anzalone et al., 2022; Johnson et al., 2007) and/or may affect Chinook prey availability and quality. Also, integrating an assessment of metals in the Lower Fraser River would provide a more comprehensive understanding of overall contaminant related risk to juvenile Chinook. In 1990's, total aluminum, total copper and total iron were identified as exceeding water quality guidelines or criteria. Given these historical exceedances, a current evaluation of metal concentrations is warranted to determine whether these risks persist. Also, mixture toxicity could be further explored using whole effluent testing. Furthermore, the Fraser River delta (part of the Lower Fraser basin) contains 40 tributary streams that serve as critical spawning and rearing habitats for Pacific salmon and due to their relatively low flow rates, these streams are particularly vulnerable to contaminant inputs, including those from non-point sources (Nener & Wernick, 1997). Further investigation within these tributaries is recommended to evaluate organic contaminant related risks and potential impacts on salmon health.

4.0 Conclusion

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This study provides a comprehensive characterization and prioritization of organic contaminants in the Lower Fraser River and its estuary, highlighting the potential risks to juvenile Chinook salmon. The wide range of contaminant classes detected in juvenile Chinook and their habitat (i.e., water) suggests that individuals outmigrating through the Lower Fraser River are exposed to complex mixtures of contaminants with potential for associated effects. By leveraging environmental quality guidelines and multiple screening benchmarks based on in vivo and in vitro data from multiple databases, this study was able to identify 16 Priority and 23 Watchlist contaminants that may be posing a risk to at-risk Fraser River Chinook populations. Also, the prioritization framework highlighted the importance of evaluating contaminants across multiple matrices and consideration of potential additive (mixture) toxic effects. These findings serve as an important screening of organic contaminant risk in the Lower Fraser River and serve as a foundation for future conservation and management strategies of Chinook populations and their habitat as well as the Endangered Southern Resident killer whales that depend on them.

Figure 1. Map of study sites along the Lower Fraser River and estuary, British Columbia. Sampling sites include an upriver tributary (HAR; natal river for Harrison Chinook salmon), a site in the main stem of the Lower Fraser (FWM), two sites in the south arm (MAN, FSA), and one site in the north arm (FNA).

Alt text: Map of the Lower Fraser River and estuary in British Columbia showing five sampling sites. HAR is located on the Harrison River; FWM is on the Lower Fraser main stem; MAN and FSA are on the south arm; and FNA is on the north arm. Major water bodies and municipalities within Metro Vancouver are labeled to provide geographic context.

Figure 2. Contaminant coverage in water samples among three screening-benchmark sources (wqbench, toxEval, Posthuma et al. 2019) used in this study. The Venn diagram shows the number of detected water contaminants with screening benchmarks unique to each source and shared across pairwise and three-way intersections.

Alt text: Venn diagram illustrating how many detected water contaminants have screening benchmarks across three sources: wqbench, toxEval, and Posthuma et al. (2019). Each circle represents one source and the numbers within the circles indicate contaminants with screening benchmarks unique to that source and those shared in pairwise or three-way overlaps.

Figure 3. Total average Biological Response Ratios (BRRs) across Lower Fraser River sites based on water (A) $BRR_{wqbench}$; (B) $BRR_{toxEval}$ and juvenile Chinook tissue (C) $BRR_{toxEval}$. BRRs were calculated using benchmark values from wqbench and TOXCAST to characterize contaminant risk profiles. Individual contaminants with BRRs ranking in the 95th percentile at one or more sites are shown, while remaining detected contaminants with BRR values are grouped as "Other." Sampling sites span a gradient from semi-urban (HAR) to heavily urbanized (FWM,

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MAN, FSA, FNA). Tissue-based BRRs (C) were assessed at HAR, FSA, and FNA; MAN was excluded due to insufficient sample sizes across contaminant classes.

Alt text: Three stacked bar charts compare total average Biological Response Ratios (BRRs) across Lower Fraser River sites. Panel A shows water‑based BRRs using wqbench benchmarks; Panel B shows water‑based BRRs using toxEval benchmarks; Panel C shows tissue‑based BRRs using toxEval benchmarks. Each bar represents a sampling site, and colored segments indicate contaminants that rank in the 95th‑percentile BRR at one or more sites, with all remaining contaminants grouped as "Other." The charts illustrate differences in contaminant contributions and total BRR levels across sites ranging from semi‑urban to heavily urbanized areas, with tissue BRRs shown only for HAR, FSA, and FNA.

Note: Pharmaceutical and personal care products (PPCP); polycyclic aromatic hydrocarbon (PAH); per‑ and polyfluoroalkyl substances (PFAS)

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Table 1. Water and tissue sampling site locations, number of samples collected, and summary of detected analytes collected during Harrison stock Chinook outmigration seasons of 2020 and 2021. Analytes from all contaminant classes measured (alkylphenols, dioxins and furans, polyaromatic hydrocarbons (PAHs), flame retardants, polychlorinated biphenyls (PCBs), per- and polyfluoroalkyl substances (PFAS), pesticides, chlorinated paraffins (tissue only), and pharmaceuticals and personal care products) were detected.

Site Code	Location description	Number of samples		Number of contaminants detected per sample (min–max)		Number of contaminants detected ≥1 time(s) (per site)	
		water	tissue	water	tissue	water	tissue
HAR	Mouth of Harrison River	3	3	71–103	205–246	157	259
FWM	Main arm, upriver from bifurcation	5	Not sampled	81–146	Not sampled	219	Not sampled
MAN	South arm, downriver of wastewater treatment plant	5	4	74–136	188–204	204	239
FSA	South arm, near river mouth	5	5	95–159	250–268	206	326
FNA	North arm, near river mouth	5	6	85–158	247–279	219	339

Note: Water samples were analyzed individually, while tissue samples were comprised of pools of 23 to 87 whole Chinook composite samples collected across the entire sampling period.

Water samples: HAR was collected in 2021; FWM, MAN, FSA, and FNA were collected in 2020 (2 samples per site) and 2021 (3 samples per site).

Tissue samples: HAR (3 composites) was collected in 2021; MAN included 1 composite from 2020 and 3 from 2021; FSA included 2 composites from 2020 and from 2021; FNA included 3 composites from 2020 and 3 from 2021.

See Table S1 (online supplementary material) for site coordinates.

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Table 2. Priority contaminants in the Lower Fraser River. Contaminants that exceeded threshold value(s) and the number of exceedances per site in water and Chinook samples collected at four urban sites and one semi-urban site (HAR) in the Fraser River. Biological response ratios (BRRs) were derived by dividing measured concentration by contaminant benchmark values. Screening benchmarks were derived from four data sources: wqbench (based on ECOTOX data), toxEval (based on ToxCast data), Environmental quality guidelines, and Posthuma et al. 2019 (based on HC5 values derived from species sensitivity distributions). Priority Tier indicates level from high, medium-high and medium (1 to 3) and is based on frequency of threshold exceedances across and within sites.

Priority Tier	CAS #	Contaminant	Class	$BRR_{wqbench}$					$BRR_{toxEval}$					BRR_{EQG}					$BRR_{Posthuma}$
				HAR	FWM	MAN	FSA	FNA	HAR	FWM	MAN	FSA	FNA	HAR	FWM	MAN	FSA	FNA	all sites
Water																			
1	80-05-7	Bisphenol A	PPCP	<T	<T	<T	<T	<T	2	2	2	2	2			<T			<T
1	58-08-2	Caffeine	PPCP	ND	1	1	3	1	ND	<T	<T	<T	<T			No SB			<T
1	72-20-8	Endrin	Pesticide	ND	1	1	1	2	ND	<T	<T	<T	<T	ND	<T	<T	<T	<T	<T
1	657-24-9	Metformin	PPCP	ND	<T	3	4	1	ND	<T	4	4	1			No SB			No SB
1	NA	PCB-total	PCB			No SB					No SB			<T	1	<T	4	<T	No SB
2	80-09-1	Bisphenol S	PPCP	ND	ND	1	1	1	ND	ND	<T	<T	<T			No SB			No SB
2	42399-41-7	Diltiazem	PPCP	ND	ND	3	3	ND	ND	ND	<T	<T	ND			No SB			No SB
2	93413-69-5	Venlafaxine	PPCP	ND	ND	1	3	1			No SB					No SB			No SB
2	1163-19-5	PBDE-209	FR	1	2	1	<T	<T			No SB					No SB			No SB
3	50-36-2	Cocaine	PPCP	ND	1	1	ND	ND			No SB					No SB			No SB
3	193-39-5	Indeno[1,2,3-Cd]Pyrene	PAH			No SB			<T	<T	<T	1	<T			No SB			<T
3	40596-69-8	Methoprene	Pesticide	ND	ND	ND	ND	1			<SB					<T			<T
3	57465-28-8	PCB-126	PCB			No SB					No SB			ND	1	ND	ND	1	No SB
3	483-65-8	Retene	PAH	<T	<T	1	<T	<T			No SB					No SB			No SB
3	58-55-9	Theophylline	PPCP			No SB			ND	<T	1	<T	<T			No SB			No SB
Tissue																			
1	NA	PBDE-penta	FR			No SB					No SB			<T	NA	4	2	6	No SB

Number of samples: Water-HAR: 3, all other sites: 5; Tissue-HAR:3, MAN:4, FSA:5, FNA:6

No SB = Screening benchmark not derived due to unavailability of toxicological data, therefore BRR could not be calculated

<T = BRR(s) did not exceed threshold; ND = Not detected; NA = Not applicable

Pharmaceutical and personal care products (PPCP); polychlorinated biphenyls (PCB); flame retardant (FR); polycyclic aromatic hydrocarbon (PAH)

Priority Tier 1: High-Contaminant exceeded threshold(s) at 4+ sites; or 1+ sites had 4+ sample exceedances

Priority Tier 2: Medium-high-Contaminant exceeded threshold(s) at 3 sites; or 1+ sites had 3+ sample exceedances

Priority Tier 3: Medium-Contaminant exceeded threshold(s) at up to 2 sites, with up to 2 sample exceedances at each site

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Table 3. Watchlist contaminants in the Lower Fraser River based on site-level Benchmark Response Ratios (BRRs). These contaminants did not exceed thresholds but were ranked in the 95th percentile at one or more sites, indicating potential elevated mixture-related risk. The Watchlist tiers were classified as either Tier 1 or Tier 2. This categorization was determined by the prevalence of the contaminant across study sites.

	Watchlist Tier	Contaminant	CAS #	Class	HAR	FWM	MAN	FSA	FNA
Water	1	Benz[a]anthracene	56-55-3	PAH	95th P	95th P	95th P	95th P	95th P
	1	Benzo(g,h,i)perylene	191-24-2	PAH	95th P	95th P	95th P	95th P	95th P
	1	Benzo[b]fluoranthene	205-99-2	PAH	95th P	95th P	95th P	95th P	95th P
	1	Endosulphan*	115-29-7	Pesticide	95th P	95th P	95th P	95th P	95th P
	1	Metolachlor	51218-45-2	Pesticide	NA	95th P	95th P	95th P	95th P
	1	Pyrene	129-00-0	PAH	95th P	95th P	95th P	95th P	95th P
	1	TBEP	78−51−3	Flame retardant	95th P	95th P	95th P	95th P	95th P
	1	TBP	126−73−8	Flame retardant	95th P	95th P	95th P	95th P	95th P
	2	Dibenz[a,h]anthracene	53-70-3	PAH	NA	95th P	NA	NA	95th P
	2	Fluoranthene	206-44-0	PAH	95th P	NA	NA	95th P	NA
	2	PBDE-penta (total)	none	Flame retardant	NA	95th P	95th P	NA	NA
	2	TCPP	13674−84−5	Flame retardant	95th P	NA	NA	95th P	95th P
	2	TEP	78-40-0	Flame retardant	95th P	95th P	95th P	NA	NA
	2	TPP	115-86-6	Flame retardant	95th P	NA	95th P	NA	NA
Tissue	1	4,4'-DDE	72-55-9	Pesticide	95th P	NS	IS	95th P	95th P
	1	4-NP	104-40-5	Alkyl Phenol	95th P	NS	95th P	95th P	95th P
	1	DEET	134-62-3	PPCP	95th P	NS	95th P	NA	NA
	1	Metolachlor	51218-45-2	Pesticide	NA	NS	NA	NA	95th P
	1	PBDE-47	5436-43-1	Flame retardant	95th P	NS	95th P	95th P	95th P
	1	PFDA	335-76-2	PFAS	95th P	NS	95th P	95th P	95th P
	1	PFOS	1763-23-1	PFAS	95th P	NS	95th P	95th P	95th P
	2	4,4'-DDD	72-54-8	Pesticide	95th P	NS	95th P	95th P	NA
	2	Dieldrin	60-57-1	Pesticide	95th P	NS	95th P	NA	95th P
	2	Thiabendazole	148-79-8	PPCP	95th P	NS	95th P	95th P	NA

95th P = contaminant is in the 95th percentile in one or more BRR type at site

NA = contaminant not in 95th percentile under any BRR type at site; NS = no sample collected; IS = insufficient sample for analysis of all contaminant classes

Polycyclic aromatic hydrocarbon (PAH); pharmaceutical and personal care products (PPCP); per- and polyfluoroalkyl substances (PFAS)

Tier 1: contaminant is in 95th percentile at 4 or 5 sites; Tier 2: contaminant in in 95th percentile at 2 or 3 sites

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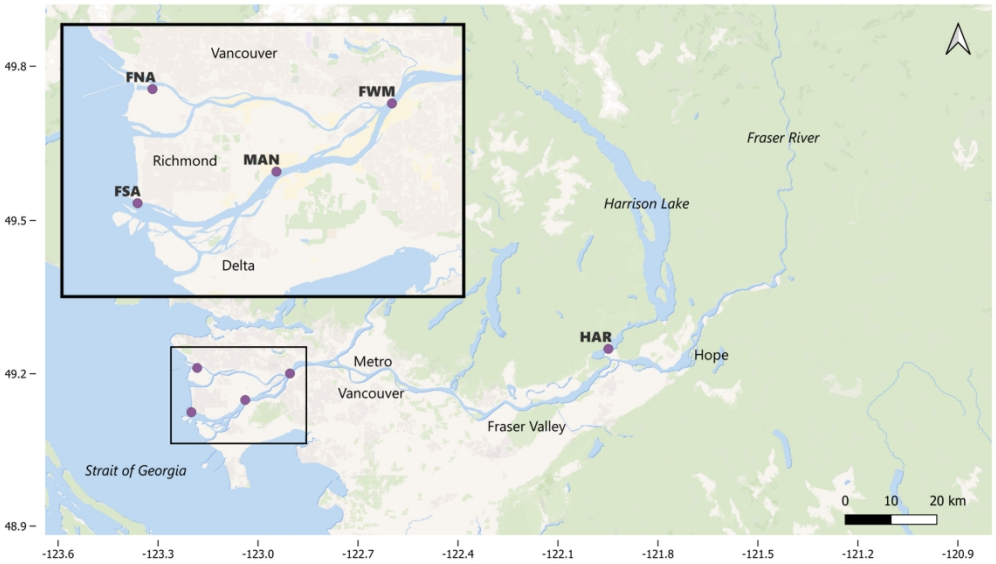

Figure 1. Map of study sites along the Lower Fraser River and estuary, British Columbia. Sampling sites include an upriver tributary (HAR; natal river for Harrison Chinook salmon), a site in the main stem of the Lower Fraser (FWM), two sites in the south arm (MAN, FSA), and one site in the north arm (FNA).

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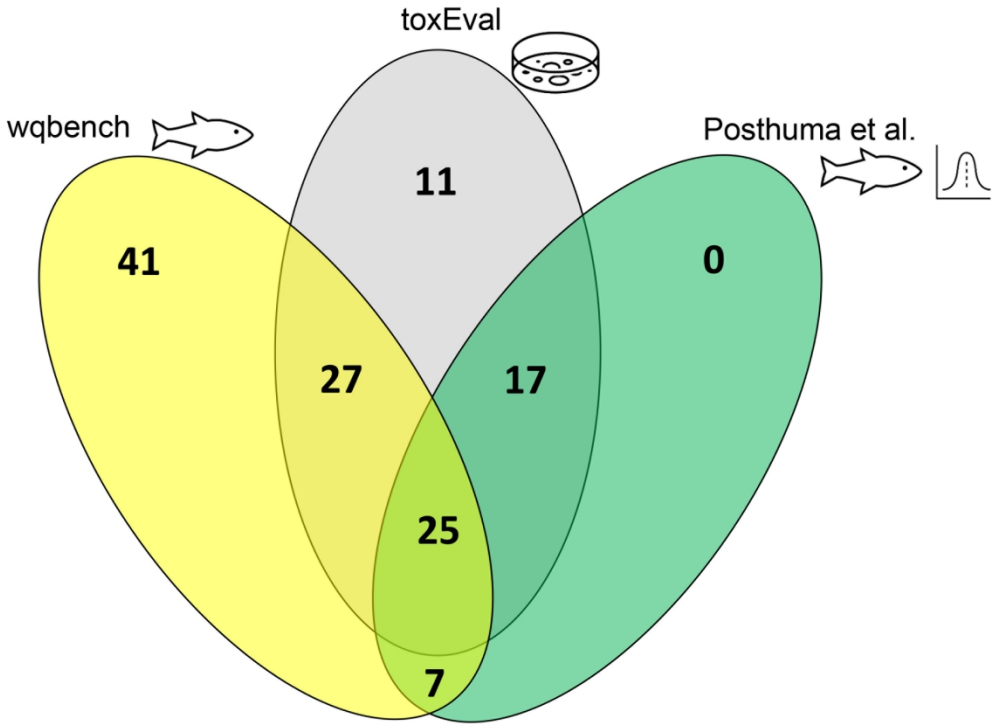

Figure 2. Contaminant coverage in water samples among three screening-benchmark sources (wqbench, toxEval, Postuma et al. (2019)) used in this study. The Venn diagram shows the number of detected water contaminants with screening benchmarks unique to each source and shared across pairwise and three-way intersections.

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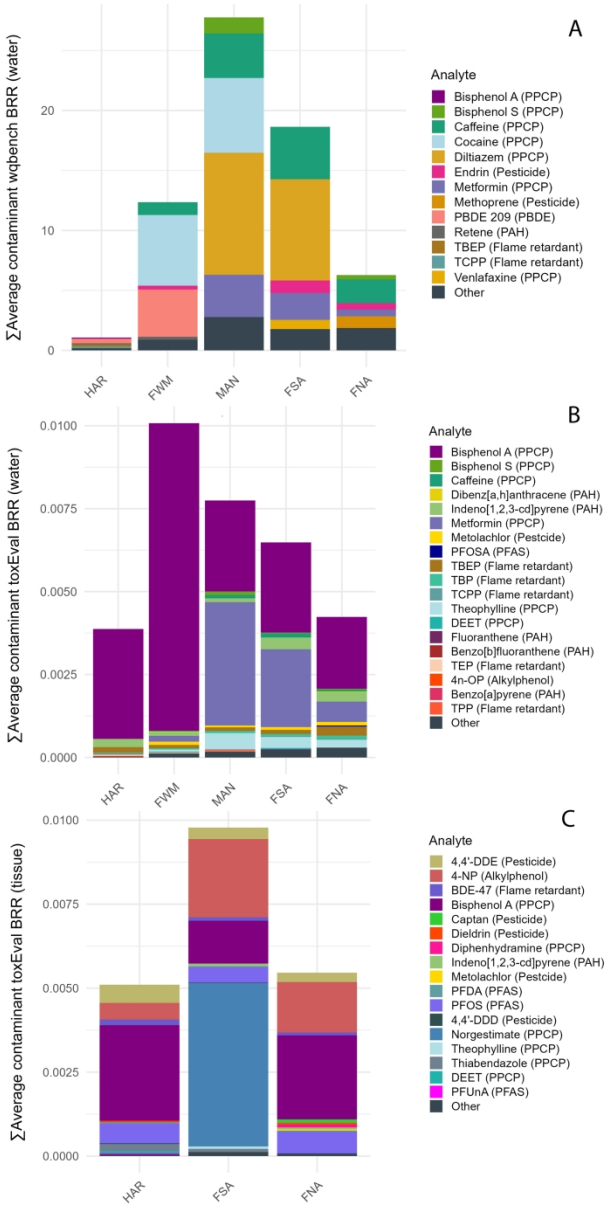

Figure 3. Total average Biological Response Ratios (BRRs) across Lower Fraser River sites based on water (A) BRRwqbench; B) BRRtoxEval and juvenile Chinook tissue (C) BRRtoxEval. BRRs were calculated using benchmark values from wqbench and TOXCAST to characterize contaminant risk profiles. Individual contaminants with BRRs ranking in the 95th percentile at one or more sites are shown, while remaining detected contaminants with BRR values are grouped as "Other." Sampling sites span a gradient from semi-urban (HAR) to heavily urbanized (FWM, MAN, FSA, FNA). Tissue-based BRRs (C) were assessed at HAR, FSA, and FNA; MAN was excluded due to insufficient sample sizes across contaminant classes.

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