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Forage Fish Polychlorinated Biphenyl Monitoring to Support Sediment Cleanup and Source Control in the Anacostia River Watershed, Washington, DC and Maryland, USA

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Abstract

Game fish can bioaccumulate polychlorinated biphenyls (PCBs) from sediments, diet, and water to concentrations requiring the issuance of Fish Consumption Advisories. To reduce impacts, sediment PCB cleanups are often conducted in phases using adaptive management (early action, performance monitoring, and remedy review). Abundant forage fish with high-site fidelity and short lifespans are useful for monitoring focused on the critical food chain pathway for human exposure. In this study, PCBs were monitored in whole-body mummichog (*Fundulus heteroclitus*) and banded killifish (*F. diaphanus*) throughout the Anacostia River and tributaries in Washington, DC and Maryland, USA. Our goal was to evaluate biological, chemical, logistical, and statistical issues for effective and cost-efficient monitoring. The objectives were to: 1) compare total PCBs across locations, focusing on Lower Beaverdam Creek (LB), known to have the highest tributary PCB loading; 2) interpret patterns in homologs; and 3) provide monitoring recommendations. From 2018 through 2022, we collected 2148 fish (185 composite samples) at 22 locations. Of the tributaries, fish from LB had the highest total PCBs (313 parts per billion (ppb) to 2500 ppb), with Northwest and Northeast Branches lowest at 30.2 ppb to 95.8 ppb. LB fish had the highest percentages of lower chlorinated homologs, indicating recent or current releases. Elevated PCBs in the tidal Anacostia below the mouth of LB added to the weight of evidence implicating LB as a major source of loading. Linear models identified lipid as a significant covariate. Recommendations on analytical methods, sampling strategy, reference locations, seasonality and scheduling were provided.

Introduction

Polychlorinated biphenyls (PCBs), widely used industrial chemicals, were banned in the United States (in 1979), and across the globe because of their persistent, bioaccumulative, and toxic properties (<https://www.epa.gov/pcbs/learn-about-polychlorinated-biphenyls#what>). In the United States, PCBs and mercury are the two most common chemicals responsible for Fish Consumption Advisories (FCAs) issued to protect public health (Cleary et al. 2021). Despite the bans, PCBs are still widely detected worldwide in water, sediments, and biota (Ngoubeyou et al. 2022). Ongoing PCB sources to water include releases from wastewater treatment plants and from activities such as metal and waste recycling (Lombard et al. 2026). Fish accumulate PCBs from ingestion of contaminated prey and sediments, and from respiration through the gills. PCBs released from hazardous waste sites accumulate in sediments, which are the primary media targeted for remediation of such sites (U.S. Environmental Protection Agency (US EPA) 2005). Sediment remediation is often conducted in phases using an adaptive management framework consisting of early action, performance monitoring, and remedy review (Department of Energy and Environment (DOEE) 2020).

Many hazardous waste and watershed cleanups have used both game and forage fish to monitor the success of cleanup and source control actions. Remedial action objectives (RAOs) generally target tissue concentrations in game fish so that FCAs can be relaxed or lifted. Forage fish are advantageous for monitoring progress towards this objective for several reasons. First, many species have excellent site fidelity which allows comparisons across locations on a finer scale than with many game species. Second, forage fish are usually short-lived (often 3 to 4 years) facilitating before and after monitoring of remedies. Third, these fish are often available in large numbers, enabling rapid collection of replicate samples (Yeardley 2000). Fourth, forage fish are a key component of the diet of game fish and therefore provide an important food web bioaccumulation link between environmental media being cleaned up and the fish consumed by humans. Also, anglers do not usually treasure forage fish and are unlikely to object to their collection.

In this paper, we investigate the use of forage fish as part of a cleanup and source control investigation of the Anacostia River and tributaries in the 456 sq km largely urban watershed in Washington, DC and suburban Maryland (MD), USA (Fig. 1a, b). There are currently FCAs covering many species in the Washington, DC and suburban MD reaches of the Anacostia River, mostly due to PCBs (Pinkney et al. 2023; <https://doee.dc.gov/service/fishdc>; <https://mde.geodata.md.gov/FCA/>). In 2013, DOEE started the Anacostia River Sediment Project (ARSP, <https://www.anacostiasedimentproject.com/>) to address contamination in the 14-km tidal Anacostia River, 44.5 ha Kingman Lake, and 2 km Washington Channel (termed the ARSP Study Area; Fig. 1a, b). The ARSP included a Remedial Investigation/Feasibility Study (RI/FS; (Tetra Tech 2019a, b) to determine the extent and magnitude of contamination and evaluate cleanup options. There are separate studies of areas bordering the Anacostia River with known or suspected releases of hazardous substances or petroleum. DOEE (2020) identified 15 such areas, termed Potential Environmental Contaminant Sites (PECS).

Applying an adaptive management framework, DOEE (2020) issued an Interim Record of Decision (IROD) that identified four Constituents of Concern (COCs): 1) total PCB congeners; 2) dioxin-like PCBs (as toxic equivalents (TEQs, Van Den Berg et al. 2006); 3) dioxins and furans (as TEQs); and 4) chlordane, an organochlorine (OC) pesticide. The IROD focused on the delineation and remediation of 11 Early Action Areas (EAAs) with surface sediment total PCBs greater than 600 µg/kg (parts per billion (ppb)) dry weight. Two of the four IROD RAOs relate to fish contamination. RAO 1 is intended to “reduce risks associated with the consumption of COCs in fish from the tidal Anacostia River by people with the highest potential exposure.” RAO 4 is intended to “reduce risks associated with COCs in surface sediment to levels protective of fish.”

DOEE, Maryland Department of the Environment (MDE), and Prince Georges and Montgomery Counties have been working to control ongoing sources of COCs in the Anacostia tributaries (DOEE 2020). Source control is critical to ensure that cleanup actions are long-lasting and not subject to re-contamination. Two studies quantified the distribution of PCB loadings among the five largest tributary sub-watersheds in decreasing rank order: Northeast Branch (NE), Northwest Branch (NW), Lower Beaverdam Creek (LB), Watts Branch (WB), and Hickey Run (HR). Wilson (2019) found that, in 2017, LB, which

contributed 14 percent of the total sediment load, accounted for 75 percent of the total PCB load. Lombard et al. (2023) deployed passive samplers in the same tributaries in 2016 and 2017. They reported that LB, with 15 percent of total tributary discharge, contributed 77 percent of the total tributary freely dissolved (C_{free}) PCB load. C_{free} provides critical assessment of the thermodynamic driver for pollutant uptake into an aquatic organism, directly through respiratory surfaces like the gills and indirectly through equilibration with diet such as algae and zooplankton (Ghosh et al. 2021).

EAA sediment cleanups are scheduled to begin in 2027. DOEE developed a Baseline/Performance Monitoring Plan (BPMP) to assess the effectiveness of EAA cleanups and actions to reduce ongoing COC inputs (Tetra Tech 2024). BPMP indicators include COCs in game and forage fish, benthic invertebrates, sediments, and water.

Here we present the results of whole-body forage fish sampling conducted in 2018, 2019, 2020, and 2022 that informed the BPMP. The goal is to evaluate critical biological, chemical, logistical, and statistical issues necessary to design an effective and cost-efficient forage fish monitoring program. The objectives are to 1) measure and compare pre-remedial COC concentrations in locations across the ARSP Study Area, Anacostia tributaries (emphasizing LB), and nearby Potomac River; 2) interpret patterns in PCB homologs; and 3) make monitoring recommendations. This paper provides findings for PCBs; other COCs are discussed in Pinkney and Perry (2025).

Methods

Species

We chose two species that are widely distributed in the Anacostia and Potomac watersheds with relatively short lifespans and small home ranges. Mummichogs (MC; *Fundulus heteroclitus*, Fig. 2a, b) are frequently used in monitoring studies of tidal waters of the U.S. East Coast for PCBs (e.g., Black et al. 1998) and PAHs (e.g., Vogelbein et al. 1990). Banded killifish (BK, *F. diaphanus*, Fig. 2c, d) are also widespread in tidal- and non-tidal freshwater rivers, and in ponds and lakes across the U.S. East Coast and Midwest. Both species have lifespans of 3 to 4 year (Abraham 1985; Phillips et al. 2007). This is advantageous

in that whole body contaminant concentrations reflect recent exposure, facilitating comparisons before and after cleanup and source control actions.

For the current study, we estimated a home range of 0.8 km for each species based on the following studies. For MC, Lotrich (1975), studying a tidal creek in Delaware, estimated a summer home range of 36 m, with a few fish moving 375 m. Skinner et al. (2005) reported that 96.6 percent of recaptured mummichogs were found within 200 m of the release point in the Miramichi River Estuary New Brunswick, Canada. McMahon et al. (2005) found that mummichogs in a salt marsh in Maine fed in 6-ha (600 m x 100 m) area. Haas et al. (2009) cited these studies and their own work in a Massachusetts estuary and stated that most mummichogs move about 1 km or less within a season. For BK, Valley et al. (2010) estimated median displacement distances of 228 m (2005) and 355 m (2006) in Square Lake, Minnesota. Pinkney and McGowan (2006) compared concentrations of DDT isomers in BK collected from the 78-ha Quantico embayment of the tidal Potomac River and nearby Virginia creeks. They found a similar, elevated ratio of p,p'-DDD: p,p'-DDE concentrations in embayment sediments and BK which was not observed in the creeks. They concluded that there was site fidelity for BK in the embayment which is in the shape of a half circle with a radius of 0.8 km.

Sampling Procedures

Fish were collected by teams from the U.S. Fish and Wildlife Service Chesapeake Bay Field Office (USFWS CBFO) in coordination with the DOEE Fisheries Research Branch using beach seines, baited minnow traps, and backpack and boat electroshocking. Fish handling and euthanasia were approved by the National Park Service Institutional Animal Care and Use Committee. During collection, fish were maintained in 20-L buckets with site water. Those of the proper size were transferred to a 4-L bucket and euthanized by immersion in a $>0.5 \text{ mg L}^{-1}$ solution of tricaine methanesulfonate (MS-222, Tricaine-S, 99.5 %, Western Chemical, Ferndale, WA), buffered to pH 7.0 to 7.5 with sodium bicarbonate (American Veterinary Medical Association (AVMA) 2020). MC and BK were double wrapped in aluminum foil in plastic bags, stored on ice during transport and kept in the CBFO freezer at -20°C .

At the end of the sampling year. BK or MC from a single location were thawed, rinsed with deionized water, blotted dry, weighed to the nearest 0.1 g, and measured for total length (mm). Single species composite samples of ≥ 20 g wet weight were prepared for three separate length classes. For each species, the large (L) class ranged from 66 mm to 84 mm and jumbo (J) class was ≥ 85 mm. The small (S) class was 45 mm to 65 mm (MC) and 50 mm to 65 mm (BK). A *collection* was defined as all samples of one species from one location in one year. In 2018 and 2019 samples were mixed sex. In 2020, separate male and female composite samples were prepared to evaluate sex as a possible covariate. Based on 2020 analyses (see Results), mixed sex samples were used thereafter.

Sampling Locations

We sampled the ARSP Study Area and the six largest Anacostia tributaries in terms of discharge (Fig. 1a, b; see Table 1 for global positioning system (GPS) data, location descriptions, and fish sampling methods per location). In addition, nearby Potomac River locations were sampled to provide regional context and identify suitable reference areas. Fish were collected in September and October 2018, June through August 2019, June through November 2020, and September and October 2022. Locations were added to expand spatial coverage and repeated to investigate temporal patterns. The only area where multiple attempts failed to yield samples was the Washington Channel, (Fig. 1a).

The five largest tributaries in terms of discharge (NW, NE, LB, HR, and WB) were sampled near USGS gages, close to the Lombard et al. (2023) passive sampler locations. Nash Run (NR), was sampled in 2020, about 100 m from its mouth in Kenilworth Marsh. In LB, we sampled at two locations near contaminant sources under investigation by MDE (2021, 2023). These are the Joseph Smith & Sons Scrap Yard adjacent to LB1 and the Pennsy Drive industrial area about 0.25 km upstream of LB2. In 2020 and 2022 we sampled location LB1.5, about 2.2 km downstream of LB2 and 1.4 km upstream of LB1. Lombard et al. (2023) reported that passive samplers deployed at this location in 2016 and 2017 had lower PCBs than those at LB1 or LB2. In 2022, we added Lower Beaverdam Mouth (LBM), on the east shoreline of the Anacostia River about 100 m downstream of the LB mouth (Fig. 1b).

146 Anacostia River locations span the 14 km length of the tidal river (Fig. 1a, b; Table 1). Pepco Cove (PC), a 1.5-ha inlet
147 of the Anacostia, was sampled from a mudflat along the south shoreline. The Cove is within Operable Unit 2 of the PEPCO
148 Benning Road Facility, a PECS with PCBs as the primary COC (DOEE 2025). Three locations were within Kingman Lake, a
149 44.5 ha shallow area created in 1920 resulting from the construction of Kingman Island with sediments dredged from the
150 Anacostia. In 2018, minnow traps were deployed along a 0.45-km reach from East Capitol Bridge northward (KL) to the
151 Heritage Island foot bridge but yielded few samples. In 2020, KL was replaced with a beach area suitable for seining on
152 Heritage Island near the foot bridge (KH). In 2020 and 2022, we seined along a mudbank on the western shore of Kingman
153 Lake at the northern area of Langston Golf Course (KG, Fig. 1b).

154 From the Potomac River, in 2019 and 2020, we sampled two locations within the 5.1 km reach from Key Bridge to
155 Chain Bridge used as a sediment background location in the RI (Tetra Tech 2019a). These were KB, about 0.8 km upstream of
156 Key Bridge, and P4 (Fletchers Cove) about 1.6 km downstream of Chain Bridge. In 2019 and 2020, we seined from a beach at
157 the upstream tip of Theodore Roosevelt Island (P3), about 0.4 km downriver of Key Bridge. In 2020, we sampled Dyke Marsh
158 (DM), a National Park Service freshwater tidal wetland about 3.5 km downriver of the DC border at the Woodrow Wilson
159 Bridge (Table 1, Fig. 1a).

160 **Analytical Methods and Quality Control**

161 Samples were shipped on dry ice with chain of custody to SGS AXYS Analytical Services, Ltd. (Sidney, British Columbia, Canada),
162 through contracts administered by the USFWS Analytical Control Facility (ACF). Two analyses for PCB congeners are discussed
163 here. In Low Resolution (Low-Res) Gas Chromatography/Mass Spectrometry (GC/MS), the MS operates at a resolution of 1 atomic
164 mass unit (amu). With High-Resolution (High-Res) GC/MS the MS operates at a resolution (>8000 amu) substantially more specific
165 for PCBs. Here we analyzed all samples for lipid and moisture content by weight and for 209 PCB congeners by Low-Res using SGS
166 AXYS Method MLA 007. This method is considered a precursor to Method 1628 (US EPA 2021), which was improved by adding
167 surrogates and applying correction factors to lessen the effects of co-elution of some higher chlorinated congeners (B. Chandramouli,

168 SGS AXYS, pers. comm.). Typical detection limits for MLA 007 were 0.1 parts per billion (ppb = ng/g) for congeners with 3 to 9
169 chlorines and 0.5 ppb for congeners with 1, 2, or 10 chlorines. Method MLA 007 was chosen as a lower cost alternative to High-Res
170 Method 1668 (US EPA 2010), which has detection limits of 0.5 to 1.0 parts per trillion (ppt). To compare the methods, in 2022, 18
171 samples were analyzed by both procedures.

172 Quality control/quality assurance procedures included the analysis of standard reference materials, procedural blanks, internal
173 standards, surrogates, and matrix spikes. Three duplicates were prepared at the SGS AXYS laboratory from samples with double the
174 usual weight to calculate relative percent difference (RPD). ACF prepared a QA review and EcoChem, Inc. (Seattle, WA) conducted a
175 full US EPA (2017) Stage 4 data validation.

176 **Data Analysis**

177 Total PCBs were calculated as the sum of all 209 congeners, including some that co-eluted (see US EPA (2021) for list), with non-
178 detected congeners treated as zeros. For dioxin-like PCB congeners, TEQs were calculated by multiplying the concentration by its
179 Toxic Equivalent Factor (TEF). The sum of these products is the total PCB TEQ (Van Den Berg et al. 2006). SGS AXYS noted that
180 TEQ calculated from Method 007 should be considered upper bound estimates that will likely result in TEQs being overestimated
181 relative to Method 1668 (B. Chandramouli, SGS AXYS, pers. comm.). Laboratory results were reported as wet weight ppb to three
182 significant digits and pairs of duplicates were reported as single mean concentrations.

183 The percent contribution of individual homologs to total PCBs was explored as an indicator of unique PCB sources or
184 tributary-specific patterns. The summed percentage of mono-through trichloro-homologs (MDT) was used as a measure of lower
185 chlorinated congeners. Since lower chlorinated congeners are more water-soluble, more volatile, and more likely to biodegrade (Beyer
186 and Biziuk 2009), their presence in water, sediments, and biota after decades of losses provides evidence of current or recently
187 mobilized sources. Although some enrichment of lower chlorinated PCBs has been observed at some sites through microbial
188 dechlorination in sediments (Magar et al. 2005), the anaerobic conditions for dechlorination are typically found in deeply buried
189 sediments from which PCB mobilization is slow.

We developed linear models (Zuur et al. 2009) to evaluate covariates for comparisons of total PCBs across collections and locations. Separate one-way models were prepared for each species due to the lack of co-occurrence of both species at all locations. The adjusted means were presented along with standard error, and upper and lower 95th percentile confidence limits. The standard error of the predicted value is a composite of all the residuals from the model as accumulated in the mean squared error term and is justified by the homogeneous variance assumption. The confidence intervals are for predicted values from the fitted linear model.

Lipid and length were the initial covariates investigated (see Gewurtz et al. 2011; Sadraddini et al. 2011). Relationships were explored by plotting the location adjusted residuals of total PCBs against length residuals and percent lipid residuals. Lipid content (as a percent) was centered by subtracting the mean by species for ease of interpretation of model parameters. In 2020, we collected separate male and female composite samples so that sex (Madenjian 2011) could also be evaluated as a covariate. Using the same linear model approach, analysis was restricted to the 13 BK and 9 MC collections with both male and female samples. We then prepared separate length and lipid residual plots for males and females to evaluate lipid and length relationships more closely.

Linear models were fitted using the `lm()` [linear model] function of the R statistical program (R Core Team 2021). For each species, the linear model included collection as a factor variable and percent lipids as a covariate. Following the linear model analysis, adjusted mean concentrations for pairs of locations were compared using a t-statistic computed as a linear function of the parameter vector (Rao 1973). The null hypothesis was that the difference between collections equals zero. For multiple comparison tests we used Holm's method (Holm 1979) for controlling the family-wise false positive rate. Collection comparisons were presented using compact letter display tables, in which groups with different letters are significantly different at $p < 0.05$.

We used Rosner's test (Gilbert 1987) to identify outliers. We computed the studentized residuals (Zuur et al. 2009) from the linear models of the entire data base. Observations with studentized residuals greater than 2.5 were identified as potential

outliers. We present the model results both with outliers included and removed to evaluate their effects on model parameters.

We developed provisional risk categories for total PCBs in BK and MC using a color scheme from highest to lowest: red, orange, yellow, green, and gray (Table 2). The categories were relevant to RAO 1, with the assumption that game fish would accumulate part of their PCB body burden by eating forage fish. We used the MDE meals per month PCB fish thresholds as the basis (A. Laliberte, MDE, pers. comm.). Then, following recommendations of Mandar Bokare (AECOM, personal communication), we used a whole body: fillet ratio of 2.5 derived from largemouth bass (*Micropterus nigricans*) data that was applied in the Hudson River Remedial Investigation/Feasibility Study (TAMS Consultants, Inc. 2000). A game fish: forage fish ratio of 2 was applied based on Bokare's modeling of Anacostia River largemouth bass data. Combining these ratios yielded the equation: whole body forage fish PCB threshold equals 0.8 divided by MDE game fish fillet PCB threshold.

Results

Collections, Duplicates, and Data Validation

We obtained 60 collections consisting of 296 composite samples of a total of 2148 fish (Table S1). There were 36 BK collections resulting in 185 composite samples, containing a total of 1462 individuals. There were 24 MC collections, resulting in 111 samples and 686 individuals. BK were collected from the tributaries, mainstem Anacostia, Kingman Lake, and Potomac River. MC were collected from all areas except the Potomac River, A1, 11A, NW, and LB1.5.

Relative percent differences for the three duplicates were 4.0 percent to 5.7 percent, which satisfies the ACF threshold of 8.64 percent for acceptability (D. Jones, ACF, pers. comm.). In its Data Validation Reports, EcoChem Inc. (Seattle, WA) concluded that all samples were acceptable according to US EPA (2017) guidance (see reports in Pinkney and Perry 2025).

Raw Data: Total PCBs and TEQ

For the entire database, there was an 85-fold range in collection mean total PCB concentrations (Figs. 3 – 6; Table S1), from 29.3 ppb to 2500 ppb. The highest concentrations were in forage fish from LB and lowest in those from P4, NE, NW, and BL. There were 21

cases in which both species were sampled from the same location in the same year. In 20 of these, mean total PCBs were higher in BK, with BK:MC ratios ranging from 0.87 to 3.5 (mean = 1.7 ± 0.7).

In the six tributaries, there was a 46-fold range in mean total PCBs in BK (Fig. 3) and a 28-fold range in MC (Fig. 5, Table S1). Fish from LB had the highest mean total PCBs of all tributaries, with substantial differences across years and within LB. In general, concentrations were highest at LB1 and LB2. In the two years with collections from LB1.5 (2020 and 2022), different patterns were evident. In 2020, BK mean total PCBs concentrations were highest at LB2 (834 ppb), lower at LB1 (588 ppb), and still lower at LB1.5 (327 ppb). In 2022, BK from LB2 were highest again (2500 ppb), but those from LB 1 (389 ppb) and LB1.5 (464 ppb) had similar concentrations. In 2022, BK mean total PCBs at LBM (500 ppb) were also similar to BK concentrations at LB1 and LB1.5. The other tributaries had lower forage fish total PCBs. WB and NR had mean total PCBs ranging from 272 ppb to 354 ppb. The lowest mean total PCBs were in collections from NW and NE, 30.2 ppb to 95.8 ppb, with HR slightly higher at 127 ppb.

In the Anacostia River, the two most upstream locations (DC and BL), had mean collection total PCBs ranging from 29.3 ppb to 119 ppb. Mean total PCB concentrations in both species were higher in all other Anacostia River locations (193 ppb to 537 ppb), except for MC at A2 in 2020 (105 ppb). There were moderately high total PCB concentrations in Kingman Lake forage fish (181 ppb to 376 ppb; Fig. 4, 6; Table S1). In the Potomac River (where only BK were available), the lowest mean total PCBs were from P4 (29.3 ppb in 2019 and 42.3 ppb in 2020). Mean total PCBs from Dyke Marsh was 95.5 ppb. Total PCBs at KB and P3 were more variable with higher concentrations in 2020 vs. 2019. (Fig. 4, Table S1).

The Low-Res analysis yielded higher total PCBs than the High-Res analysis in 15 of the 18 samples. The ratio of Low-Res to High-Res total PCBs had a mean of 1.10 ± 0.14 , ranging from 0.86 to 1.47 (Table 3). For these 18 samples, dioxin-like PCBs were 7.3 ± 1.1 percent of total PCBs in the High-Res samples and 7.0 ± 1.2 percent in the Low-Res samples. For all 296 project samples, dioxin-like PCB congeners comprised a mean of 6.7 ± 1.6 percent of total PCBs, ranging from 3.0 to 15.1 percent. PCB total TEQs were higher in the Low-Res analyses in 17 of the 18 samples. The Low to High ratios averaged 9.1 ± 3.2 with a range from 0.1 to 16.1.

Raw Data: PCB homologs

In the tributaries, LB and NR had the highest MDT contributions to total PCBs. The range at LB was from 11.1 percent to 39.5 percent, whereas at NR the MDT contribution was 17.0 percent in MC and 23.4 percent in BK (Figs. 7, 8; Table S2). The greatest MDT contribution was in forage fish from LB2 at 20.4 percent to 39.5 percent, with LB1 at 15.2 percent to 29.7 percent, and LB1.5 from 11.1 percent to 25.7 percent. In the other tributaries, the MDT contribution was 1.7 percent to 6.3 percent.

At LBM, just below the mouth of LB, the MDT contributions to total PCBs were 25.0 percent in BK and 17.5 percent in MC (Figs. 7, 8; Table S2). In all other mainstem Anacostia locations, MDT homologs were ≤ 13.6 percent of total PCBs (Figs. 9, 10; Table S2). In Kingman Lake collections, the MDT range was 6.0 percent to 12.4 percent of total PCBs. In Potomac River collections, MDT homologs contributed 0.2 percent to 3.1 percent.

Model-based Results

Final statistical model formulation and outlier inclusion was based on sequentially testing the effects of the outliers and individual predictor variables. Based on lipid-adjusted total PCBs results, we identified three BK and two MC outliers. Running models with and without outliers had minimal effects on covariate p values (Table S3), thus, outliers were retained in the final models. For BK, the two statistically significant covariates were collection ($p < 0.0001$) and lipid content ($p = 0.006$). For MC, collection ($p < 0.0001$) and lipid content ($p = 0.047$) were also statistically significant. The initial analysis produced slightly negative coefficients for the median length covariate which indicated an inverse relation to total PCBs. For BK, it appeared to be driven by a negative relationship between fish length and lipids. For MC, there was a U-shaped relationship between length and lipid. Because of the apparent confounding between length and lipids, and inconsistencies between species, length was not included as a covariate. Percent lipid content was a significant variable for all models and model-based results were reported on a lipid-adjusted basis.

Model-based comparisons of the 21 cases in which both species were collected at the same location in the same year yielded results similar to those with raw data. In 20 cases, lipid-adjusted mean total PCBs were higher in BK, with the BK:MC ratios in all 21 ranging from 0.95 to 2.9 (mean = 1.5 ± 0.5 ; Table 6).

277 As described above, in 2020 separate male and female composite samples were obtained to investigate sex as a covariate. For
278 BK, sex was a statistically significant covariate ($p=0.008$). The relationship, however, was not consistent across locations, with males
279 higher at ten locations (M:F total PCB mean ratio of 1.8 ± 0.7) and females higher at three (F:M mean ratio of 1.5 ± 0.2). For MC, sex
280 was not significant ($p = 0.62$), with males higher at five locations and females higher at four. Based on the lack of consistent results,
281 we resumed mixed sex sampling in 2022.

282 Multiple comparison results provided resolution on the geographical patterns and magnitude of spatial and temporal
283 differences in total PCBs. For BK, there were six statistical groups (A through F), with only LB2BK22 (adjusted mean total PCBs of
284 2472 ppb; Table 4) in the highest (Group A). The next highest group (Group B) included LB1BK18 (1113 ppb) and LB2BK18 (919
285 ppb, which was also in Group C). LB2BK20 was in Group C only with an adjusted mean of 825 ppb. Many of the remaining
286 collections were in multiple statistical groups across wide concentration ranges.

287 For MC, there were four statistical groups with LB2MC22 (adjusted mean of 847 ppb) in the highest (Group A) and no other
288 groups (Table 5). Other collections in Groups A and B were LB1MC18 (733 ppb) and LB2MC18 (718 ppb). LB2MC20 was in Group
289 B only (609 ppb) and PCMC18 was in Groups B and C (480 ppb). Nine collections were in Group D only, with adjusted mean total
290 PCBs ranging from 19.2 ppb to 190 ppb (Table 5).

291 Using the thresholds (Table 2), red category (model-based total PCBs >400 ppb) forage fish were found in LB and several
292 mainstem locations. At LB2, both BK and MC were in the red category in all three sampling years (Table 6). At LB1, both species
293 were in the red category in 2018 and orange category (>200 ppb and ≤ 400 ppb) in 2020 and 2022. One LB1.5 collection (LB1.5BK22)
294 was red at 456 ppb, with all other LB1.5 collections orange. At LBM, BK was red (496 ppb) and MC was orange at 300 ppb. Several
295 Pepco Cove collections were red (PCMC18 at 480 ppb and PCBK20 at 496 ppb) with others orange. The only other mainstem
296 collections in the red category were in the most downstream reach: A1BK18 (442 ppb) and 11ABK22 (498 ppb). Green and gray
297 category fish (≤ 50 ppb) were from the Potomac River (P4BK19 and P4BK20), Northwest Branch (NWBK22), and Northeast Branch
298 (NEKB22 and NEMC22).

Discussion

Anacostia Tributaries and the LB Plume

Our finding of elevated total PCBs in LB forage fish compared with NW, NE, HR, and WB is consistent with studies with caged freshwater mussels (*Elliptio complanata*) and passive samplers deployed in surface water (Lombard et al. 2023); surface sediment (Johnson Company 2019); and surface water and suspended sediment concentrations and loadings (Wilson 2019). Our ranking of mean total PCBs in MC and BK from the five largest tributaries was LB>WB>HR>NE=NW. This nearly matched the ranking in suspended sediment total PCBs in 2017 provided by Wilson (2019) with HR and WB reversed. Similar tributary rankings were reported by Lombard et al. (2023) who found that surface water total C_{free} PCBs, were 7 to 77 times greater in LB vs. NE and NW. In 2016 and 2017 deployments of caged mussels, the ranking in total PCBs was LB>WB>HR=NE=NW (Lombard et al. 2023).

The same pattern of enrichment of MDT homologs in LB forage fish compared with NW, NE, HR, and WB (Table S2) was observed in the caged mussels and passive samplers (Lombard et al. 2023; Table S4). In mussels deployed in LB, MDT ranged from 5.4 percent to 19.3 percent of total PCBs vs. 0.1 percent to 2.5 percent in NW, NE, HR, and WB. For passive samplers, MDT in LB was 49.3 percent to 76.5 percent compared to 4.4 percent to 16.2 percent in the other four tributaries. LB2, with the highest total PCBs in forage fish, also had the highest MDT contribution (19.3 percent in caged mussels and 76.5 percent in passive samplers). At LB2, passive samplers had more dichlorobiphenyl homologs (40.4 percent of total PCBs) than trichlorobiphenyls (36.1 percent) in contrast to all other locations. This is further evidence of recent releases of PCBs at LB2.

In LB, total PCBs in BK and MC were consistently highest at LB2 compared to LB1 and LB1.5. In 2018 and 2020, both LB1 and LB2 were clearly higher than LB1.5, which matched the pattern in surface water passive samplers and mussels reported by Lombard et al. (2023), indicating two potential PCB source areas. In 2022, however, PCBs were even higher in forage fish from LB2 but were similar and lower in LB1 and LB1.5 forage fish. In 2022, PCBs in forage fish likely resulted from high inputs in the LB2 area which flowed downriver affecting LB1.5 and LB1. It is uncertain whether the lower 2022 LB1 mean total PCB concentrations

were driven by the March 2022 removal of > 50 ppm PCB stream bank soils at the Joseph Smith and Sons site (MDE 2023). Future monitoring will allow managers to evaluate more fully the success of source control actions within LB.

PCBs appear to be transported out of LB into the mainstem Anacostia where they accumulated in BK and MC at LBM. This is based on: 1) MDE (2021) water column PCB concentration data collected from LB about 500 m upstream of the mouth; 2) the similarity in homolog distribution at LBM with those in LB; and 3) the similar concentrations in 2022 in BK and MC at LB1 and LBM. In a 2019 survey of water column data along LB, MDE (2021) reported the highest surface water total PCB concentration (119 ng L⁻¹) was collected ~400 m downstream of LB1, which is about 500 m upstream of the confluence with the Anacostia River. This concentration is 185 times the MDE human health standard of 0.64 ng L⁻¹ (https://www.epa.gov/system/files/documents/2024-07/md_wqs_0022_051624.pdf).

Further evidence of the contribution of inputs from LB to PCBs in the mainstem and possibly into tidal NR is provided in the Ghosh et al. (2024) report of passive sampler deployments in 2021 through 2022. The samplers spanned the ARSP Study Area and overlapped with many of the fish collection areas. There was a total PCB plume in the river (approximately 4-6 ng L⁻¹) that traveled about 200 m downstream of the LB confluence and mixed with cleaner upstream mainstem water. Within Kenilworth Marsh (Fig. 1b), total PCB concentrations decreased moving away from the Anacostia and towards the tidal NR fish collection area. The MDT contribution was about 50 percent in upstream Anacostia River passive samplers collected at BL and DC. In both LB and LBM samplers, the MDT contribution was about 70 to 75 percent. The contribution decreased to about 60 percent in Kenilworth Marsh and the NR fish collection area. Further downriver, the MDT contribution in the lower portion of the Anacostia (near the 11th St. Bridge) was again about 50 percent. Thus, both concentration and homolog data are consistent with a PCB plume from LB affecting the area immediately downstream and extending into Kenilworth Marsh and the NR fish collection area.

The homolog and PCB concentration data in abiotic media and biota serve as a weight of evidence (Menzie et al. 1996) implicating LB as having current sources of bioavailable PCBs. The similarity in homolog patterns in forage fish from LBM with those from the three LB locations suggests that releases from LB play a role in contaminant uptake in the mainstem Anacostia. This

342 explanation is consistent with the Ghosh et al. (2023) study that used a linked food chain and mass balance model to evaluate
343 scenarios for reducing PCB inputs into the tidal Anacostia River. They calculated that the greatest impact on PCB concentrations from
344 source control in LB would be in non-migratory fish in the reach from Bladensburg downstream to the mouth of Kenilworth Marsh.
345 This reach includes the LBM location.

346 **Anacostia River, Kingman Lake, Potomac River**

347 The lower total PCBs in fish from the two most upstream locations, BL and DC, likely reflect the low PCB loadings from NE and NW
348 (Wilson 2019, Lombard et al. 2023). Several years of sampling at PC indicated moderately high total PCBs, which can serve as a
349 baseline for the upcoming cleanup of the Pepco facility (DOEE 2025). Within Kingman Lake, KG was the location where both species
350 could be readily collected in large numbers. The 2020 and 2022 total PCB concentrations were in a moderate range of 192 ppb to 313
351 ppb. This location would be suitable for pre- and post-remedial monitoring of the Kingman Lake EAAs.

352 For the Potomac River, continued sampling of BK from Dyke Marsh, a protected area would provide regional context for the
353 monitoring. By far the lowest mean total PCB concentrations were near Fletchers Cove (P4), the most upstream location. Both sides of
354 this area of the river have forested buffers and are protected natural areas. In contrast, the downstream sampling areas (KB and P3),
355 which had higher and more variable total PCBs are more urbanized.

356 **Uncertainties**

357 In any monitoring program, there is a conflict between limiting effort (i.e., labor and analytical costs) and obtaining additional samples
358 to enhance the ability to detect statistically significant differences across space and time. Uncertainties can arise due to differences in
359 sampling windows in relation to spawning. Mummichogs spawn on a lunar cycle in late spring through early September in Maryland
360 tidal waters (L. Yonkos, University of Maryland, pers. comm.). The spawning behavior of banded killifish in mid-Atlantic streams has
361 not been documented in the literature. Although most collections were made in the fall, there was some summer sampling in 2019 and
362 2020. Thus, it is possible that seasonal differences may have affected PCB concentrations. Additional uncertainty could be the result

of fish moving between locations, despite the home range-based spacing in the sampling design. Indeed, (Haas et al. 2009) theorized that whereas most MC move on the order of 1 km, some individuals may move as much as 6 km to 8 km.

We are uncertain as to why, in nearly all cases, both raw and lipid-adjusted total PCB concentrations were higher in BK vs. MC. In future years samplers should obtain both species at all locations where they overlap. Documenting BK and MC stomach contents may help determine whether different diets may be a possible contributor.

Uncertainties are present in the development of the provisional forage fish thresholds. Both ratios are based on largemouth bass data and would likely vary across species. Species-specific whole body to fillet ratios could be determined using mass balance. Individual fish of key game species would be analyzed in two portions: 1) whole body minus fillet, and 2) fillet; and then the whole-body concentration would be reconstructed by multiplying concentrations by weight. Similarly, collections of forage and game fish in specific areas could be used to develop species-specific food chain ratios.

Monitoring Recommendations

Species and Sex

MC and BK are appropriate species because of their small home range, ≤ 4 -yr lifespan, and wide distribution throughout the Anacostia mainstem, tributaries, Kingman Lake, and Potomac River. Washington Channel was the only ARSP area with few MC or BK. Although few fish were observed in Washington Channel, we did note the presence of juvenile bluegill (*Lepomis macrochirus*) and pumpkinseed (*L. gibbosus*). Intensive sampling should evaluate whether there are adequate numbers available for the BPMP. Because sex was not a significant covariate that resulted in consistent differences between males and females, we conclude that the added sampling effort to collect single sex composite samples is not warranted.

Background/Reference Areas

We recommend NW and NE as tributary reference areas for monitoring the progress of source control actions in LB. The low inputs of PCBs from NW and NE are reflected in the low concentrations at BL, which serves as an upstream reference for the Anacostia River. For the Potomac River, our results for Fletchers Cove (P4) clearly indicate its suitability as a reference area since total PCB

385 concentrations in both years were less than 50 ppb. These recommendations are consistent with those of TetraTech (2024) in the
386 BPMP.

387 **Low-Res vs. High-Res PCB Analysis**

388 Throughout the project, we used the Low-Res method for PCB congeners because of cost considerations. One advantage of the High-
389 Res method, much lower detection limits, is not important in analyzing fish tissues in this urban area in which there were few non-
390 detects for congeners that contribute the most to total PCBs. The roughly 10 percent higher total PCBs in the Low-Res method can be
391 viewed as a safety factor for decision making related to FCAs. The higher TEQ results for the Low-Res method is largely due to about
392 a tenfold higher mean concentration of congener PCB 126 (Low-Res mean = 2.83E-1 ppb vs. High-Res mean = 2.50 E-2 ppb). This
393 overestimation would be reduced if the newer Low-Res Method 1628 were to be used instead of Method MLA 007 (B. Chandramouli,
394 SGS AXYS, pers. comm). Whether FCAs are based on total PCBs or TEQ will drive the choice of Low-Res vs. High-Res analysis.
395 Options for future monitoring are to use the US EPA Method 1628 Low-Res procedure, switch to the High-Res method, or analyze a
396 subset of samples with both methods.

397 **Monitoring Schedule**

398 The schedule would depend on the timing of remediation and source control actions, keeping in mind the 3- to 4-year lifespan of BK
399 and MC. For example, if a sediment cleanup at an EAA is completed in the summer of 2027, pre-remedial monitoring should occur in
400 fall 2026 and post-remedial monitoring in fall 2031 and fall 2032 to coincide with 5-year reviews. It was much easier to collect forage
401 fish in early- to mid-fall compared to later in the season when water temperatures drop approaching winter. We suggest avoiding
402 spring and summer to minimize possible variation related to spawning. Thus, we recommend a 2-month sampling window from mid-
403 September to mid-November.

404 **Assessing Fish Health**

405 In addition to their use in bioaccumulation monitoring, there is a long history of using forage fish to monitor indicators of fish health,
406 which would apply to the DOEE (2020) IROD RAO 4. Endpoints include the prevalence and severity of neoplastic lesions (Vogelbein

et al. 1990); reproductive (Black et al. 1998) and developmental compromise (Boudreau et al. 2004); immunotoxicity (Fournier et al. 1998); toxicant-altered gene expression (Van Veld et al. 1997); acquired contaminant tolerance (Ownby et al. 2002); among others (Burnett et al. 2007).

Liver histopathology has been widely used to monitor health status of numerous fish species, in some cases before and after remedial actions (e.g., Vogelbein et al. 1990; Myers et al. 2008; Blazer et al. 2009; Pinkney et al. 2019). Adult MC have been used in fish health studies because of their propensity to develop liver tumors and pre-neoplastic lesions following chronic environmental exposure to industrial and urban pollutants (Vogelbein and Unger 2006). In fact, MC is the only species known to exhibit neoplasia in natural populations that is also regularly used in laboratory studies (Burnett et al. 2007). In liver tumor surveys with MC in the Elizabeth River, Virginia (Vogelbein et al. 1990, Vogelbein and Unger 2006) and Hershey Run, Delaware (Pinkney and Harshbarger 2006), there was significantly elevated prevalence in creosote–contaminated areas relative to background locations. Although no published BK liver tumor surveys have been identified, this species may be valuable to include in future fish health studies (V. Blazer, USGS, pers. comm.). Whereas ranges of BK and MC overlap from North Carolina to Maine in coastal and estuarine tidal waters, BK are also found in freshwater streams and lakes throughout the Great Lakes region (Fuller 2020; Fuller and Neilson 2019), expanding the potential reach of monitoring programs.

Forage fish can also be monitored for reproductive health through assessing gonad maturation and histopathology (e.g. Blazer et al. 2013). Additional techniques include computer-assisted semen analysis (CASA, Kime et al. 2001) and analysis of sperm DNA integrity (quantified via gel electrophoresis Comet assay (Cabrita et al. 2014).

Conclusions

Forage fish monitoring provides a critical link between environmental media (sediment and water) concentrations and bioaccumulation in fish consumed by humans and top predators in the aquatic ecosystem. In addition, forage fish often have small home ranges and short lifespans that allow more accurate assessment of environmental recovery post remedy at contaminated sites. As

428 such, these fish species provide a unique monitoring opportunity that can complement other media monitoring and inform decisions at
429 a site undergoing adaptive management.

430 We found that BK and MC are suitable species for forage fish contaminants monitoring across nearly all locations in the
431 Anacostia watershed and nearby Potomac River. In cases where both species were collected at the same location in the same year, BK
432 was nearly always higher with an average BK:MC ratio of 1.5. The highest total PCB concentrations were in forage fish collected
433 from LB1 and LB2. In the six tributaries, we report a large range in total PCBs (46-fold in BK, 28-fold in MC), consistent with studies
434 with passive samplers and caged mussels. These findings were also consistent with proximity to ongoing sources in and near LB and
435 showed a continued elevation of total PCBs into the mainstem Anacostia. Suitable reference areas were P4 in the Potomac River, BL,
436 NW, and NE.

437 Through this multi-year study, we addressed issues related to the use of forage fish that may be applicable to other geographic
438 areas. We found that mixed sex samples can be used to speed and simplify the collection and data analysis, although that finding
439 should be verified for new watersheds. We recommend a two-month sampling window from mid-September through mid-November
440 when fish are widely available and spawning status is not a concern. For studies in which total PCBs are the driver for FCAs rather
441 than TEQ, the Low-Res method is a cost-effective alternative to the High-Res method. Finally, using logistic regression, we identified
442 lipid as a covariate that must be considered for spatial and temporal comparisons.

443 **References**

- 444 Abraham B (1985) Species profiles: life histories and environmental requirements of coastal fishes and invertebrates (mid-Atlantic):
445 Mummichog and striped killifish. U.S. Army Corps of Engineers
- 446 American Veterinary Medical Association (AVMA) (2020) AVMA guidelines for euthanasia in animals —2020 edition. American
447 Veterinary Medical Association (AVMA), Schaumburg, IL

448 Beyer A, Biziuk M (2009) Environmental fate and global distribution of polychlorinated biphenyls. In: Whitacre DM (ed) Rev
 449 Environ Contam Toxicol. Springer US, Boston, MA, Vol. 201, pp 137–158

450 Black DE, Gutjahr-Gobell R, Pruell RJ, et al (1998) Reproduction and polychlorinated biphenyls in *Fundulus heteroclitus* (Linnaeus)
 451 from New Bedford Harbor, Massachusetts, USA. Environ Toxicol Chem 17:1405–1414. [https://doi.org/10.1897/1551-](https://doi.org/10.1897/1551-5028(1998)017%3C1405:RAPBIF%3E2.3.CO;2)
 452 5028(1998)017%3C1405:RAPBIF%3E2.3.CO;2

453 Blazer VS, Rafferty SD, Baumman PC, et al (2009) Assessment of the “fish tumors or other deformities” beneficial use impairment in
 454 brown bullhead (*Ameiurus nebulosus*): II. liver neoplasia. J Great Lakes Res 35:527–537. <https://doi.org/10.1016/j.jglr.2009.08.011>

455 Blazer VS, Pinkney AE, Jenkins JA, et al (2013) Reproductive health of yellow perch *Perca flavescens* in selected tributaries of the
 456 Chesapeake Bay. Sci Tot Environ 447:198–209. <https://doi.org/10.1016/j.scitotenv.2012.12.088>

457 Boudreau M, Courtenay SC, Maclatchy DL, et al (2004) Utility of morphological abnormalities during early-life development of the
 458 estuarine mummichog, *Fundulus heteroclitus*, as an indicator of estrogenic and antiestrogenic endocrine disruption. Environ Toxicol
 459 Chem 23:415–425. <https://doi.org/10.1897/03-50>

460 Burnett KG, Bain LJ, Baldwin WS, et al (2007) *Fundulus* as the premier teleost model in environmental biology: opportunities for
 461 new insights using genomics. Comp Biochem Physiol, Part D: Genomics and Proteomics 2:257–286.
 462 <https://doi.org/10.1016/j.cbd.2007.09.001>

463 Cabrita E, Martínez-Páramo S, Gavaia PJ, et al (2014) Factors enhancing fish sperm quality and emerging tools for sperm analysis.
 464 Aquaculture 432:389–401. <https://doi.org/10.1016/j.aquaculture.2014.04.034>

465 Cleary BM, Romano ME, Chen CY, et al (2021) Comparison of recreational Fish Consumption Advisories across the USA. Curr Envir
 466 Health Rpt 8:71–88. <https://doi.org/10.1007/s40572-021-00312-w>

467 Department of Energy and Environment (DOEE). (2020) Interim Record of Decision (ROD). Early Action Areas in the main stem,
 468 Kingman Lake, and Washington Channel. Anacostia River Sediment Project, Department of Energy and Environment, Washington,
 469 DC

470 Department of Energy and Environment (DOEE) (2025) Proposed plan PEPCO Benning Road Facility Operable Unit 2 (OU 2).
 471 Washington, DC

472 Fournier M, Lacroix A, Voccia I, Brousseau P (1998) Phagocytic and metabolic activities of macrophages from mummichog naturally
 473 exposed to pulp mill effluents in the Miramichi River. *Ecotox Environ Saf* 40:177–183. <https://doi.org/10.1006/eesa.1997.1629>

474 Fuller P (2020) *Fundulus heteroclitus* (Linnaeus, 1766). U.S. Geological Survey, Nonindigenous Aquatic Species Database

475 Fuller P, Neilson M (2019) *Fundulus diaphanus* (Lesueur, 1817): U.S. Geological Survey, Nonindigenous Aquatic Species Database

476 Gewurtz SB, Bhavsar SP, Fletcher R (2011) Influence of fish size and sex on mercury/PCB concentration: Importance for fish
 477 consumption advisories. *Environment International* 37:425–434. <https://doi.org/10.1016/j.envint.2010.11.005>

478 Ghosh U, Bokare M, Gobas FAPC (2021) Deconvoluting thermodynamics from biology in the aquatic food web model. *Environ*
 479 *Toxicol Chem* 40:2145–2155. <https://doi.org/10.1002/etc.5106>

480 Ghosh U, Bokare M, Pinkney A (2023) Development of a linked PCB mass balance and food web model to assess effectiveness of
 481 management options in the Anacostia River. University of Maryland Baltimore County, U.S. Fish and Wildlife Service, University of
 482 Maryland College Park.

483 Ghosh U, Lombard N, Cheung L, et al (2024) Anacostia River monitoring with passive samplers and mussels to refine conceptual
 484 model and define baseline conditions: report year 2. University of Maryland Baltimore County, U.S. Fish and Wildlife Service.

485 Gilbert R (1987) Statistical methods for environmental pollution monitoring. Van Nostrand Reinhold, New York

486 Haas HL, Freeman CJ, Logan JM, et al (2009) Examining mummichog growth and movement: are some individuals making intra-
 487 season migrations to optimize growth? *J Exp Mar Biol Ecol* 369:8–16. <https://doi.org/10.1016/j.jembe.2008.09.027>

488 Holm S (1979) A simple sequentially rejective multiple test procedure. *Scandinavian Journal of Statistics* 6:65-70.

489 Johnson Company (2019) NPS tributary sediment sampling study report, Anacostia River Sediment Project site, National Capital
 490 Region.

491 Kime DE, Van Look KJW, McAllister BG, et al (2001) Computer-assisted sperm analysis (CASA) as a tool for monitoring sperm
 492 quality in fish. *Comp Biochem Physiol, Part C: Toxicol Pharmacol* 130:425–433. [https://doi.org/10.1016/S1532-0456\(01\)00270-8](https://doi.org/10.1016/S1532-0456(01)00270-8)

493 Lombard NJ, Bokare M, Harrison R, et al (2023) Codeployment of passive samplers and mussels reveals major source of ongoing
 494 PCB Inputs to the Anacostia River in Washington, DC. *Environ Sci Technol* 57:1320–1331. <https://doi.org/10.1021/acs.est.2c06646>
 495 Lombard NJ, Needham TP, Fadaei Khoei H, et al (2026) Using long-term trends of PCBs in fish caught in Maryland waters to assess
 496 the effectiveness of anagement actions. *ACS EST Water* 6:419–428. <https://doi.org/10.1021/acsestwater.5c01099>
 497 Lotrich VA (1975) Summer home range and movements of *Fundulus heteroclitus* (Pisces: Cyprinodontidae) in a tidal creek. *Ecology*
 498 56:191–198. <https://doi.org/10.2307/1935311>
 499 Madenjian CP (2011) Sex effect on polychlorinated biphenyl concentrations in fish: a synthesis. *Fish and Fisheries* 12:451–460.
 500 <https://doi.org/10.1111/j.1467-2979.2011.00414.x>
 501 Magar VS, Johnson GW, Brenner RC, et al (2005) Long-term recovery of PCB-contaminated sediments at the Lake Hartwell
 502 Superfund Site: PCB dechlorination. 1. end-member characterization. *Environ Sci Technol* 39:3538–3547.
 503 <https://doi.org/10.1021/es048622y>
 504 Maryland Department of Environment (MDE) (2021) Lower Beaverdam Creek PCB investigation. Baltimore, MD
 505 Maryland Department of Environment (MDE) (2023) PCB source trackdown in Maryland: June 2023 update.
 506 McMahon KW, Johnson BJ, Ambrose WG (2005) Diet and movement of the killifish, *Fundulus heteroclitus*, in a Maine salt marsh
 507 assessed using gut contents and stable isotope analyses. *Estuaries* 28:966–973. <https://doi.org/10.1007/BF02696024>
 508 Menzie C, Henning MH, Cura J, et al (1996) Special report of the Massachusetts weight-of-evidence workgroup. A weight-of-
 509 evidence approach for evaluating ecological risks. *Hum Ecol Risk Assess* 2:277–304. <https://doi.org/10.1080/10807039609383609>
 510 Myers MS, Anulacion BF, French BL, et al (2008) Improved flatfish health following remediation of a PAH-contaminated site in
 511 Eagle Harbor, Washington. *Aquatic Toxicol* 88:277–288. <https://doi.org/10.1016/j.aquatox.2008.05.005>
 512 Ngoubeyou PSK, Wolkersdorfer C, Ndibewu PP, Augustyn W (2022) Toxicity of polychlorinated biphenyls in aquatic environments –
 513 a review. *Aquatic Toxicol* 251:106284. <https://doi.org/10.1016/j.aquatox.2022.106284>

514 Ownby DR, Newman MC, Mulvey M, et al (2002) Fish (*Fundulus heteroclitus*) populations with different exposure histories differ in
 515 tolerance of creosote-contaminated sediments. *Environ Toxicol Chem* 21:1897. [https://doi.org/10.1897/1551-](https://doi.org/10.1897/1551-5028(2002)021%3C1897:FFHPWD%3E2.0.CO;2)
 516 [5028\(2002\)021%3C1897:FFHPWD%3E2.0.CO;2](https://doi.org/10.1897/1551-5028(2002)021%3C1897:FFHPWD%3E2.0.CO;2)

517 Phillips EC, Ewert Y, Speares PA (2007) Fecundity, age and growth, and diet of *Fundulus diaphanus* (Banded Killifish) in Presque Isle
 518 Bay, Lake Erie. *Northeastern Naturalist* 14:269–278. [https://doi.org/10.1656/1092-6194\(2007\)14%5B269:FAAGAD%5D2.0.CO;2](https://doi.org/10.1656/1092-6194(2007)14%5B269:FAAGAD%5D2.0.CO;2)

519 Pinkney AE, Harshbarger JC, Rutter MA, Sakaris PC (2019) Trends in liver and skin tumor prevalence in Brown Bullhead (*Ameiurus*
 520 *nebulosus*) from the Anacostia River, Washington, DC, and nearby waters. *Toxicol Pathol* 47:174–189.
 521 <https://doi.org/10.1177/0192623318823150>

522 Pinkney A, Griffin O, Wetzell R (2023) Contaminant concentrations in fish tissue collected from the waters of the District of
 523 Columbia. U.S. Fish and Wildlife Service, Chesapeake Bay Field Office, Annapolis, MD

524 Pinkney AE, Harshbarger JC (2006) Tumor prevalence in mummichogs from the Delaware Estuary watershed. *J Aquat Animal Health*
 525 18:244–251. <https://doi.org/10.1577/H05-053.1>

526 Pinkney AE, McGowan PC (2006) Use of the p,p'-DDD: p,p'-DDE concentration ratio to trace contaminant migration from a
 527 hazardous waste site. *Environ Monit Assess* 120:559–574. <https://doi.org/10.1007/s10661-005-9103-7>

528 Pinkney A, Perry E (2025) Polychlorinated biphenyls and organochlorine pesticide concentrations in whole body Mummichog and
 529 Banded Killifish from the Anacostia River Watershed: 2018-2022. U.S. Fish and Wildlife Service, Chesapeake Bay Field Office,
 530 Annapolis, MD

531 R Core Team (2021) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna,
 532 Austria

533 Rao C (1973) Linear statistical inference and its applications, 2nd edn. John Wiley & Sons, New York

534 Sadraddini S, Azim ME, Shimoda Y, et al (2011) A Bayesian assessment of the PCB temporal trends in Lake Erie fish communities. *J*
 535 *Great Lakes Res* 37:507–520. <https://doi.org/10.1016/j.jglr.2011.06.005>

536 Skinner MA, Courtenay SC, Parker WR, Curry RA (2005) Site fidelity of mummichogs (*Fundulus heteroclitus*) in an Atlantic
 537 Canadian estuary. *Water Qual Res J* 40:288–298. <https://doi.org/10.2166/wqrj.2005.034>

538 TAMS Consultants, Inc. (2000) Hudson River PCBs reassessment RI/FS. Phase 3 Report: Feasibility Study.

539 Tetra Tech (2019a) Remedial investigation report. Anacostia River Sediment Project, Washington, D.C. Tetra Tech, Chantilly, VA.

540 Tetra Tech (2019b) Focused Feasibility Study report, Early Action Areas in main stem, Kingman Lake, and Washington Channel,
 541 Anacostia River Sediment Project, Washington DC. Tetra Tech, Chantilly, VA.

542 Tetra Tech (2024) Anacostia River Sediment Project, baseline and performance monitoring work plan. Chantilly, VA

543 U.S. Environmental Protection Agency (US EPA) (2005) Contaminated sediment remediation guidance for hazardous waste sites

544 U.S. Environmental Protection Agency (US EPA) (2010) Method 1668C chlorinated biphenyl congeners in water, soil, sediment,
 545 biosolids, and tissue by HRGC/HRMS.

546 U.S. Environmental Protection Agency (US EPA) (2017) National functional guidelines for organic Superfund methods data review.

547 U.S. Environmental Protection Agency (US EPA) (2021) Method 1628 Polychlorinated Biphenyl (PCB) congeners in water, soil,
 548 sediment, biosolids, and tissue by Low-resolution GC/MS using selected ion monitoring.

549 Valley RD, Habrat MD, Dibble ED, Drake MT (2010) Movement patterns and habitat use of three declining littoral fish species in a
 550 north-temperate mesotrophic lake. *Hydrobiologia* 644:385–399. <https://doi.org/10.1007/s10750-010-0207-x>

551 Van Den Berg M, Birnbaum LS, Denison M, et al (2006) The 2005 World Health Organization reevaluation of human and mammalian
 552 Toxic Equivalency Factors for dioxins and dioxin-like compounds. *Toxicol Sci* 93:223–241. <https://doi.org/10.1093/toxsci/kfl055>

553 Van Veld PA, Vogelbein WK, Cochran MK, et al (1997) Route-specific cellular expression of Cytochrome P4501A (CYP1A) in Fish
 554 (*Fundulus heteroclitus*) following exposure to aqueous and dietary benzo[a]pyrene. *Toxicol Appl Pharmacol* 142:348–359.
 555 <https://doi.org/10.1006/taap.1996.8037>

556 Vogelbein W, Fournie J, VanVeld P, Huggett R (1990) Hepatic neoplasms in the mummichog *Fundulus heteroclitus* from a creosote-
 557 contaminated site. *Cancer Res* 50:5978-5986.

558 Vogelbein W, Unger M (2006) Liver carcinogenesis in a non-migratory fish: the association with polycyclic aromatic hydrocarbon
 559 exposure. Bulletin of the European Association of Fish Pathologists 26:11–20

560 Wilson T (2019) Sediment and chemical contaminant loads in tributaries to the Anacostia River, Washington, District of Columbia,
 561 2016-17. USGS Scientific Investigations Report 2019-5092.

562 Yeardley RB (2000) Use of small forage fish for regional streams wildlife risk assessment: relative bioaccumulation of contaminants.
 563 Environ Monit Assess 65:559–585. <https://doi.org/10.1023/A:1006304132068>

564 Zuur A, Ieno E, Walker N, et al (2009) Mixed effects models and extensions in ecology with R. Springer, New York

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568 **Author contributions:** AEP: obtained funding, developed the sampling design, collected samples, interpreted results and was the lead
 569 author on all versions of the manuscript. LRY helped determine the sampling locations, facilitated the collections and wrote sections
 570 of the results and discussion. ESP: conducted the statistical analyses and wrote up the results and interpretation. NJL and UG
 571 contributed to the evaluation of the PCB homolog data and the interpretation of the results culminating in sections of the results and
 572 discussion. All authors read and approved the submissions.

573 **Data Availability:** The datasets generated during and/or analyzed during the current study are available from the corresponding
 574 author on reasonable request.

575 Table 1. Location descriptions (T=tidal, NT=non-tidal) and sampling methods. (S=Seine, M=Minnow traps, BPS: backpack
 576 electroshock, BS=Boat electroshock. GPS in decimal degrees using WGS 84 grid.

Location, City, State	Code	Sampling Years and Notes
Anacostia River		

Bladensburg, MD	BL	2018: Gravel bar about 200 m long, accessed from west side of River downstream of foot bridge (S) [38.9363, -76.9404], and inlet north of foot bridge on east side of River (M) [38.9373, -76.9404]; 2020: gravel bar only (S) (T):
Colmar Manor, MD	DC	2020: Muddy beach, east side of River about 10 m wide across from mouth of Dueling Creek (S) (T) [38.9216, -76.9386]
Lower Beaverdam Mouth, DC	LBM	2022: Sandy beach, east side of River about 100 to 200 m downstream of mouth of Lower Beaverdam Creek (S) (T) [38.9157, -76.9448]
Pepco Cove, DC	PC	2018, 2019, 2020: Muddy beach about 30 m wide on south edge of Pepco Cove closest to River (S) (T) [38.9012, -76.9604]
Above CSX Br., DC	A2	2018, 2020: Sand bar about 10 m long. east side of River, upstream of CSX Bridge (S) (T) [38.8843, -76.9667]
Bet. 11 th & Sousa Bridges, DC	11A	2019, 2020, 2022: Sand bar about 20 m long, east side of River between 11 th St. and Sousa Bridges (S) (T) [38.8732, -76.9854]
Poplar Point, DC	A1	2018: Sand bar about 20 m long, east side of River near Poplar Point (S) (T) [38.8693, -76.9970]
Kingman Lake		
Kingman Lake, DC	KG	2020, 2022: Mudbank about 100 m long along western shore of Lake adjacent to Langston Golf Course (S) (T) [38.9077, -76.9592]
Kingman Lake, DC	KH	2020: Beach area on Heritage Island adjacent to footbridge (S) (T) [38.8940, -76.9666]
Kingman Lake, DC	KL	2018: Set and retrieved baited minnow traps from East Capitol Bridge to Heritage Island footbridge (M) (T) [From: 38.8903, -76.9659 to 38.8937, -76.9653]
Anacostia Tributaries		

Northwest Branch, Hyattsville, MD	NW	2018, 2022: Downstream of USGS gage (BPS) (NT) [From: 38.9510, -76.9609 to 38.9534, -76.9616]
Northeast Branch, Riverdale, MD	NE	2018, 2020, 2022: Downstream of USGS gage (BPS) (T) [From: 38.9554, -76.9287 to 38.9569, -76.9279]
L. Beaverdam Creek, Hyattsville, MD	LB2	2018, 2020, 2022: Country Club Road, near Route 202 and Cabin Branch (BPS) (NT) [From 38.9284, -76.8944 to 38.9271, -76.8976]
L. Beaverdam Creek, Hyattsville MD	LB1.5	2020, 2022: Three beach areas near Jesse Warr Recreation Center (S) (NT) [38.9138, -76.9174]
L. Beaverdam Creek, Capitol Heights, MD	LB1	2018, 2020, 2022: Upstream of USGS gage and near Route 201 (BPS, M) (T) [From: 38.9161, -76.9318 to 38.9158, -76.9315]
Nash Run, DC	NR	2020: About 100 m from mouth of Nash Run into Kenilworth Marsh near footbridge (S) (T) [38.9122, -76.9460]
Watts Branch, DC	WB	2018: Upstream of USGS gage (BPS) (NT) [38.8989, -76.9403]
Hickey Run, DC	HR	2018: Downstream of USGS gage (BPS, M) (T) [38.9115, -76.9639]
Potomac River		
Fletchers Cove, DC	P4	2019, 2020: Mud bar about 100 m long, at river edge of Fletchers Cove (S) (T) [38.9181, -77.1030]
Theodore Roosevelt Isl., DC	P3	2019, 2020: sand bar, upstream tip of island (S) (T) [38.8998, -77.0667]
Key Bridge, DC	KB	2019, 2020: about 0.8 km upstream of Key Bridge (S) (T) [VA side: 38.9024, -77.0794; DC side: 38.9049, -77.0791]

Dyke Marsh,
Alexandria, VA DM 2020: channels in Dyke Marsh (BS) (T) [38.7617, -77.0480]

577

578 Table 2. Rationale for proposed forage fish thresholds based on game fish fillet thresholds
 579 provided by A. Lalibert, MDE (pers. comm.). The whole-body forage fish threshold is based on a
 580 food chain multiplier of 2 and a whole body/fillet ratio of 2.5 (M. Bokare, UMBC, pers. comm.).
 581 Forage fish thresholds are rounded to multiples of 50.

Whole body Forage fish Threshold (ppb)	Color	Estimated game fish fillet (ppb)	MDE meal thresholds (ppb)	General population advisory (meals/month)
≤25	Gray	≤20	≤21.25	8 (unlimited)
>25 to ≤50	Green	>20 to ≤40	≤42.5	4
>50 to ≤200	Yellow	>40 to ≤160	>42.5 to <169.9	1-3
>200 to ≤400	Orange	>160 to ≤320	>170 to <339.9	1 every other month
>400	Red	>320	>340	Do not consume

582

583 Table 3. Comparison of total PCBs and PCB TEQ using Low-Resolution Gas Chromatography/Mass Spectrometry (Low-Res) and
584 High-Resolution Gas Chromatography/Mass Spectrometry (High-Res) methods. All samples are from 2022.

Sample ID	High-Res PCBs			Low-Res PCBs				Low:High		
	Tot PCB High- Res	Tot dioxin-like PCBs	Dioxin- like PCBs	Tot PCB TEQ	Tot PCB Low-Res	Tot dioxin- like PCBs	Dioxin -like PCBs	Tot PCB TEQ	tot PCB	tot TEQ
	ppb wet wt	ppb wet wt	% of total	ppb wet wt	ppb wet wt	ppb wet wt	% of total	ppb wet wt		
11ABK22L01	532	33.8	6.3	6.79E-03	605	36.3	6.0	6.04E-02	1.14	8.9
11ABK22S05	422	26.4	6.2	5.28E-03	457	28.0	6.1	4.64E-02	1.08	8.8
KGBK22L02	173	12.6	7.3	1.99E-03	255	17.3	6.8	3.19E-02	1.47	16.1
KGBK22S01	345	23.7	6.9	4.04E-03	373	25.0	6.7	4.66E-02	1.08	11.5
KGMC22L02	221	16.9	7.6	2.68E-03	211	15.1	7.2	2.72E-02	0.95	10.2
KGMC22S02	190	13.4	7.0	2.56E-03	209	14.4	6.9	2.67E-02	1.10	10.4
LB1BK22L02	289	20.2	7.0	2.52E-03	386	25.5	6.6	3.13E-02	1.34	12.4
LB1BK22S01	419	24.9	6.0	4.08E-03	503	29.8	5.9	4.52E-02	1.20	11.1
LB1MC22L01	267	16.4	6.1	2.83E-03	314	17.2	5.5	2.39E-02	1.18	8.4
LB1MC22L02	315	22.4	7.1	3.17E-03	335	22.3	6.7	3.38E-02	1.06	10.7
LBM22BKS02	519	33.4	6.4	6.17E-03	537	31.4	5.8	3.76E-02	1.03	6.1
LBMMC22L0	205	16.5	8.1	2.89E-03	226	16.7	7.4	2.24E-02	1.10	7.7
LBMMC22S0	313	23.3	7.4	4.15E-03	379	26.7	7.1	3.34E-02	1.21	8.0
NEBK22L01	58.9	4.19	7.1	1.29E-03	59.9	4.29	7.2	1.20E-02	1.02	9.3
NEBK22S01	192	14.2	7.4	2.97E-03	196	14.6	7.4	2.87E-02	1.02	9.7
NEMC22L01	43.8	3.31	7.5	1.37E-03	37.6	2.89	7.7	1.09E-02	0.86	8.0
NWBK22J02	27.9	2.83	10.1	7.87E-04	26.9	2.89	10.7	5.21E-03	0.96	6.6
NWBK22L01	31.6	2.92	9.2	8.67E-04	32.5	2.83	8.7	1.20E-04	1.03	0.1
min	27.9	2.83	6.0	7.87E-04	26.9	2.83	5.5	1.20E-04	0.86	0.1
max	532	33.8	10.1	6.79E-03	605	36.3	10.7	6.04E-02	1.47	16.1
median	244	16.9	7.1	2.89E-03	314	17.3	6.8	3.00E-02	1.08	9.1
mean	254	18.1	7.3	3.13E-03	301	18.5	7.0	2.91E-02	1.10	9.1
std dev	157	9.3	1.1	1.71E-03	172	10.4	1.2	1.55E-02	0.14	3.2

Table 4. Comparison of model-based lipid-adjusted mean concentrations of total PCBs (ppb wet weight) for banded killifish using Holm multiple comparison test with experiment-wise error rate of $p = 0.05$. N: number of samples; Adj: adjusted, SD: Standard Deviation; SE: standard error; LB: lower bound and UB: upper bound 95th percentile confidence intervals for the mean.

Collection ^a	N	Mean $\pm$ SD	Adj. Mean	SE	LB Est.	UB Est.	Group ^b
LB2BK22	1	2500	2472	94.8	2285	2659	A
LB1BK18	3	1110 $\pm$ 131	1113	54.4	1005	1220	B
LB2BK18	5	913 $\pm$ 145	919	42.2	836	1003	BC
LB2BK20	5	834 $\pm$ 239	825	42.2	742	909	C
LB1BK20	6	588 $\pm$ 133	650	44.5	563	738	CD
11ABK22	10	537 $\pm$ 73	498	33.0	433	563	D
PCBK20	7	476 $\pm$ 123	496	36.4	424	568	D
LBMBK22	4	500 $\pm$ 32	469	48.4	373	564	DE
LB1.5BK22	8	464 $\pm$ 96	456	33.4	390	522	D
A1BK18	2	458 $\pm$ 21	442	66.9	310	574	DE
LB1BK22	10	389 $\pm$ 93	383	29.9	324	442	DE
LB1.5BK20	6	327 $\pm$ 110	379	42.7	295	463	DE
KLBK18	1	376	342	95.0	154	530	DEF
NRBK20	2	354 $\pm$ 32	333	67.0	201	466	DE
PCBK19	4	313 $\pm$ 155	322	47.2	228	415	DE
KGBK22	5	313 $\pm$ 81	304	42.3	220	387	DE
A2BK18	4	320 $\pm$ 66	301	47.6	207	395	DE
WBBK18	2	272 $\pm$ 38	274	66.6	143	406	DEF
KHBK20	2	295 $\pm$ 76	272	67.1	140	405	DEF
A2BK20	4	224 $\pm$ 93	244	39.2	167	322	EF
11ABK20	6	196 $\pm$ 92	240	41.6	158	322	EF
KGBK20	4	224 $\pm$ 45	238	47.4	144	332	EF
11ABK19	13	193 $\pm$ 40	219	27.7	164	273	E
P3BK20	6	238 $\pm$ 236	213	39.5	135	291	E
DMBK20	6	95.5 $\pm$ 45.6	156	44.1	68.6	243	EF
BLBK18	6	119 $\pm$ 50	142	39.3	64.2	219	EF
P3BK19	1	89.5	131	95.4	-57.2	320	DEF
KBBK20	5	167 $\pm$ 62	127	44.5	39.1	215	EF
BLBK20	6	103 $\pm$ 40	114	38.7	37.8	191	EF
NEBK18	3	76.1 $\pm$ 9.5	73.5	54.4	-34.1	181	EF
KBBK19	8	48.8 $\pm$ 22.6	67.8	34.0	0.6	135	EF
NWBK18	4	54.0 $\pm$ 4.3	63.0	47.2	-30.3	156	EF
P4BK19	3	29.3 $\pm$ 16.1	44.6	54.7	-63.5	153	EF
NEBK22	7	95.8 $\pm$ 58.6	25.7	43.6	-60.5	112	EF
P4BK20	4	42.3 $\pm$ 2.6	18.6	47.9	-76.0	113	EF
NWBK22	10	65.2 $\pm$ 52.8	5.0	36.8	-67.6	77.7	F

^aCollection: all samples of one species collected in one year.

^bGroups with different letters are significantly different at $p < 0.05$.

Table 5. Comparison of model-based lipid-adjusted mean concentrations of total PCBs (ppb wet weight) for mummichogs using Holm multiple comparison test with experiment-wise error rate of $p = 0.05$. N: number of samples; Adj: adjusted, SD: Standard Deviation; SE: standard error; LB: lower bound and UB: upper bound 95th percentile confidence intervals for the mean.

Collection ^a	N	Mean $\pm$ SD	Adj. Mean	SE	LB Est.	UB Est.	Group ^b
LB2MC22	5	855 $\pm$ 161	847	31.2	785	909	A
LB1MC18	5	726 $\pm$ 79	733	31.2	671	795	AB
LB2MC18	5	723 $\pm$ 134	718	31.1	656	780	AB
LB2MC20	6	650 $\pm$ 63	609	34.9	539	678	B
PCMC18	4	468 $\pm$ 106	480	35.1	410	549	BC
LB1MC22	6	367 $\pm$ 39	356	28.8	298	413	C
LB1MC20	6	313 $\pm$ 66	348	33.3	282	415	C
PCMC20	4	296 $\pm$ 88	320	36.6	247	393	CD
LBMMC22	6	294 $\pm$ 72	300	28.4	244	357	CD
NRMC20	6	308 $\pm$ 83	291	29.4	232	349	CD
WBMC18	3	314 $\pm$ 79	288	42.1	204	371	CD
PCMC19	3	252 $\pm$ 67	275	41.5	192	358	CD
KLMC18	3	233 $\pm$ 34	245	40.4	164	325	CD
KGMC20	6	192 $\pm$ 47	208	29.4	150	267	CD
A2MC18	4	206 $\pm$ 30	191	35.4	121	261	CD
KHMC20	6	181 $\pm$ 41	190	28.6	134	247	D
KGMC22	5	207 $\pm$ 13	179	33.8	112	246	D
A2MC20	2	109 $\pm$ 62	134	50.5	34.0	235	D
DCMC20	8	98.9 $\pm$ 17.0	119	26.5	66.8	172	D
HRMC18	5	127 $\pm$ 34	84.9	37.4	10.6	159	D
BLMC20	4	29.3 $\pm$ 13.2	68.3	39.7	-10.5	147	D
NEMC18	2	47.9 $\pm$ 7.0	58.2	49.2	-39.6	156	D
BLMC18	4	77.2 $\pm$ 18.6	57.9	35.9	-13.5	129	D
NEMC22	3	30.2 $\pm$ 7.6	19.2	40.3	-61.0	99.4	D

^aCollection: all samples of one species collected in one year.

^bGroups with different letters are significantly different at $p < 0.05$.

Table 6. Provisional risk categories^a based on lipid-adjusted mean total PCB concentrations (ppb wet weight) and species ratio of adjusted means.

Location/year^b	BK adjusted mean/ Category	MC adjusted mean/ Category	BK:MC ratio^c
LB2/22	2472/RED	847/RED	2.9
LB1/18	1113/RED	733/RED	1.5
LB2/18	919/RED	718/RED	1.3
LB2/20	825/RED	609/RED	1.4
PC/18	NO FISH	480/RED	
LB1/20	650/RED	348/ORANGE	1.9
11A/22	498/RED	NO FISH	
PC/20	496/RED	320/ORANGE	1.6
LBM/22	469/RED	300/ORANGE	1.6
LB1.5/22	456/RED	NO FISH	
A1/18	442/RED	NO FISH	
LB1/22	383/ORANGE	356/ORANGE	1.1
NR/20	333/ORANGE	291/ORANGE	1.1
KL/18	342/ORANGE	245/ORANGE	1.4
WB/18 ^c	274/ORANGE	288/ORANGE	0.95
LB1.5/20	379/ORANGE	NO FISH	
PC/19	322/ORANGE	275/ORANGE	1.2
KH/20	272/ORANGE	190/YELLOW	1.4
A2/18	301/ORANGE	191/YELLOW	1.6
KG/22	304/ORANGE	179/YELLOW	1.7
KG/20	238/ORANGE	208/ORANGE	1.1
DC/20	NO FISH	119/YELLOW	
A2/20	244/ORANGE	134/YELLOW	1.8
HR/18	NO FISH	85.0/YELLOW	
11A/20	240/ORANGE	NO FISH	
P3/20	213/ORANGE	NO FISH	
11A/19	219/ORANGE	NO FISH	
BL/18	142/YELLOW	57.9/YELLOW	2.5
BL/20	114/YELLOW	68.3/YELLOW	1.7
DM/20	156/YELLOW	NO FISH	
KB/20	127/YELLOW	NO FISH	
NE/18	73.5/YELLOW	58.2/YELLOW	1.3
P3/19	131/YELLOW	NO FISH	
NE/22	25.6/GREEN	19.2/GRAY	1.3
KB/19	67.8/YELLOW	NO FISH	
NW/18	63.0/YELLOW	NO FISH	
P4/19	44.6/GREEN	NO FISH	
P4/20	18.6/GRAY	NO FISH	
NW/22	5.01/GRAY	NO FISH	

^a Gray: ≤25 ppb, Green: > 25 and ≤50 ppb, Yellow: >50 ppb and ≤200 ppb; Orange: >200 ppb and ≤400 ppb, Red: >400 ppb

^b Location/year: BK and MC collections from one location in the same year.

^c Mean ± SD BK:MC ratio: 1.5 ±0

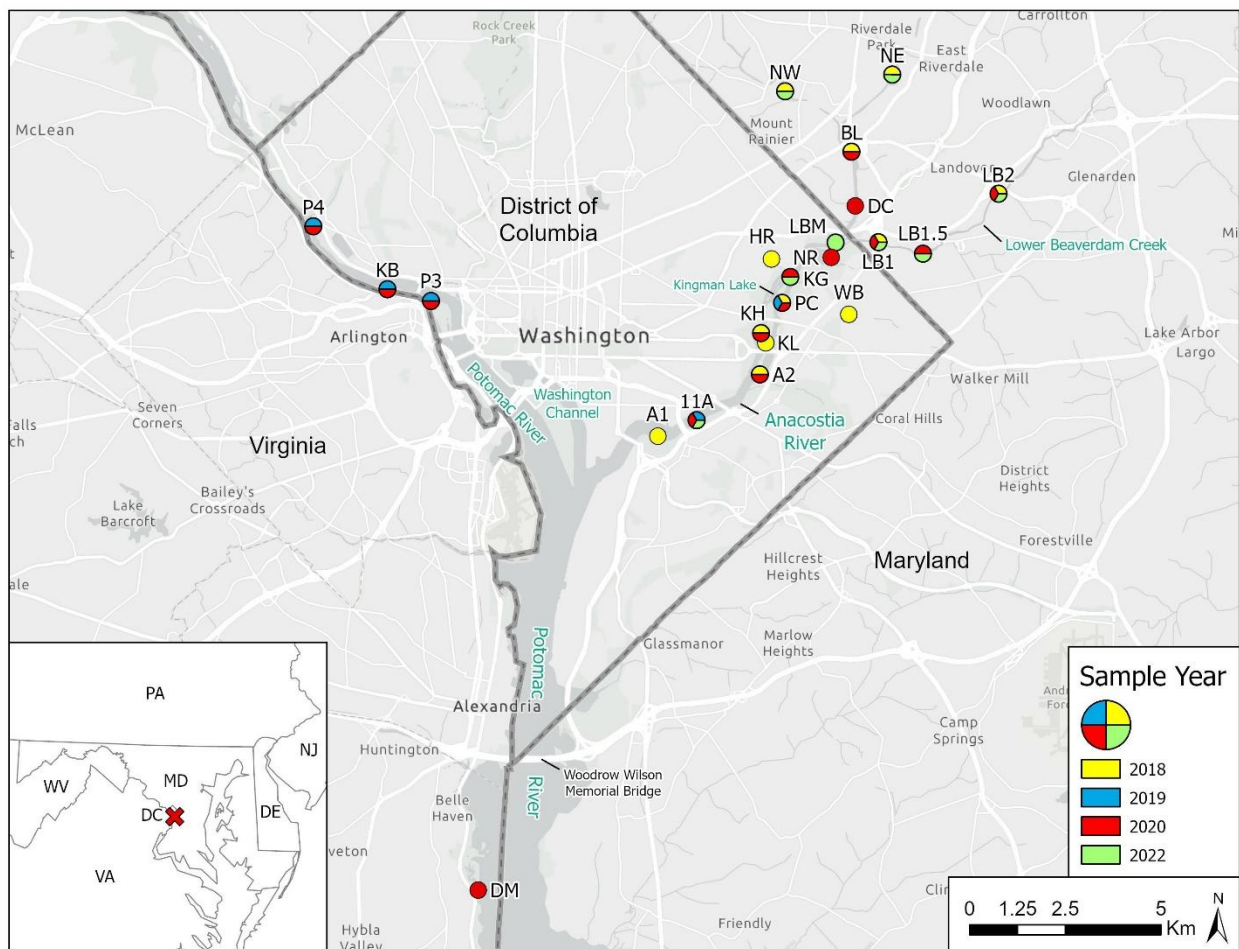


Fig. 1a. Sampling locations across the entire project area. See Table 1 for GPS data, descriptions and abbreviations.

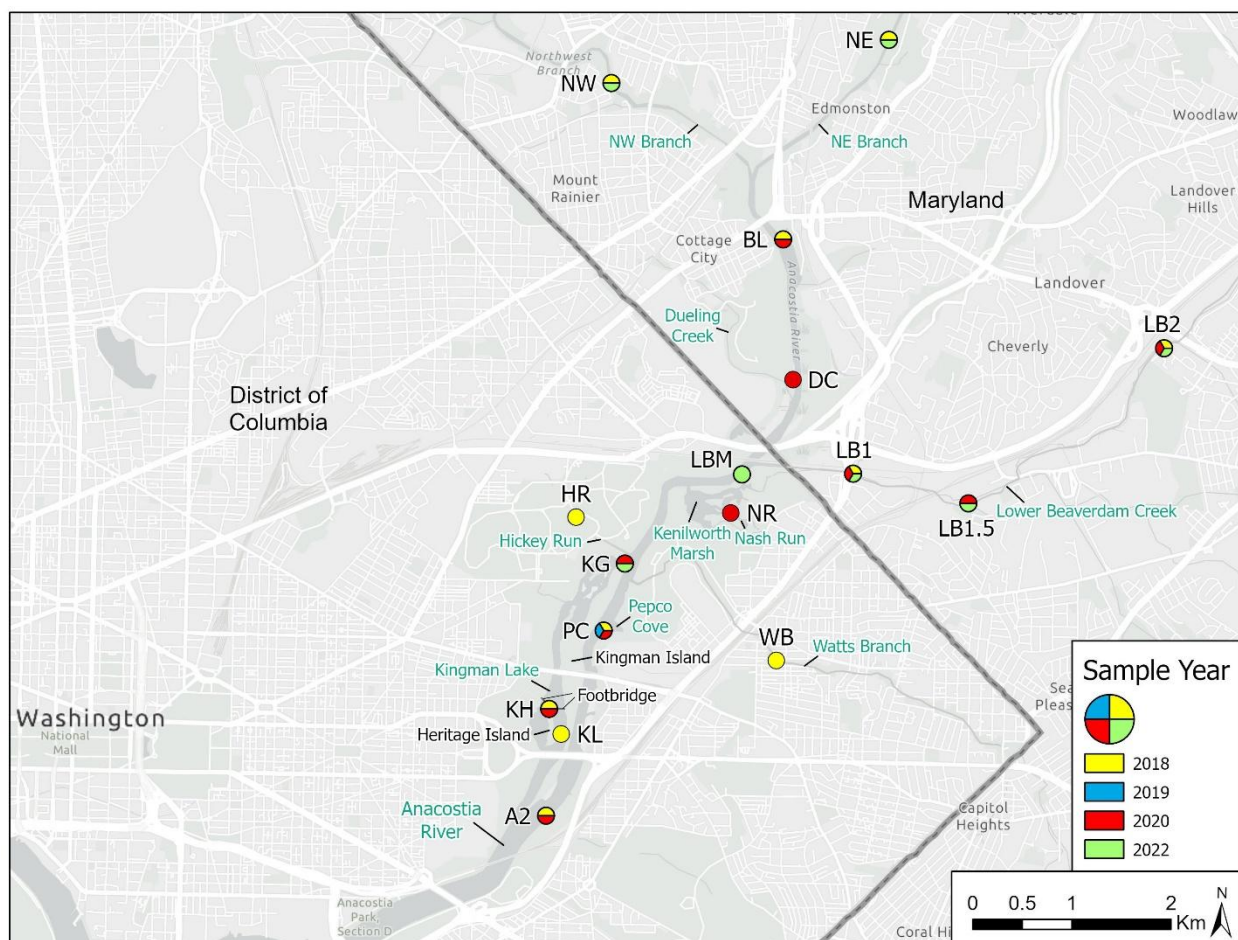


Figure 1b. Closeup of sampling locations in the Anacostia River tributaries, Kingman Lake and nearby mainstem. See Table 1 for GPS data, location descriptions and abbreviations.



Fig 2. Mummichog, MC, *F. heteroclitus*: a) female; b) male. Banded killifish, BK, *Fundulus diaphanus*: c) female; d) male.

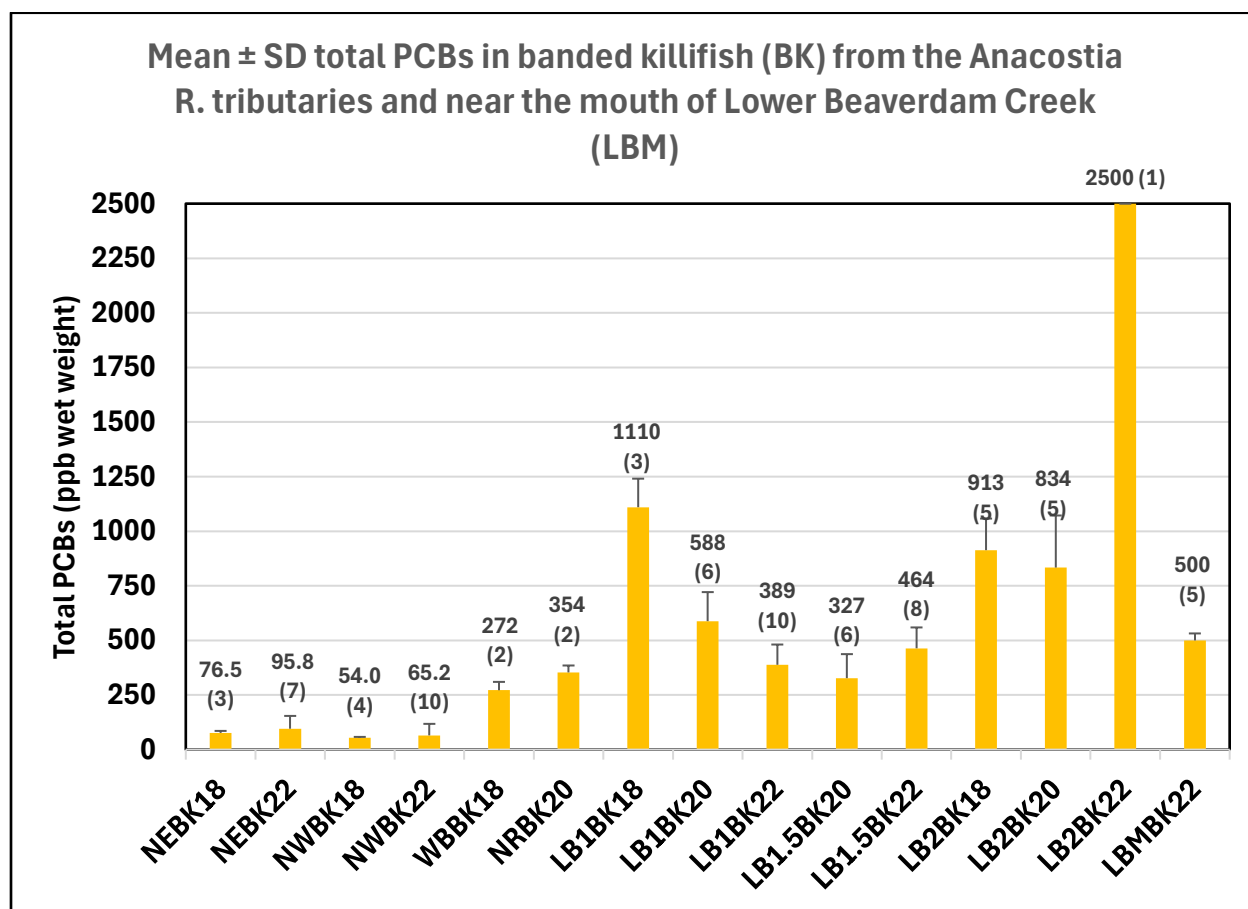


Fig. 3. Mean total PCBs (ppb wet wt., mean + standard deviation) in collections of banded killifish (BK) from the Anacostia River tributaries and near the mouth of Lower Beaverdam Creek (LBM). Numbers of samples in parentheses. See Table 1 for abbreviations.

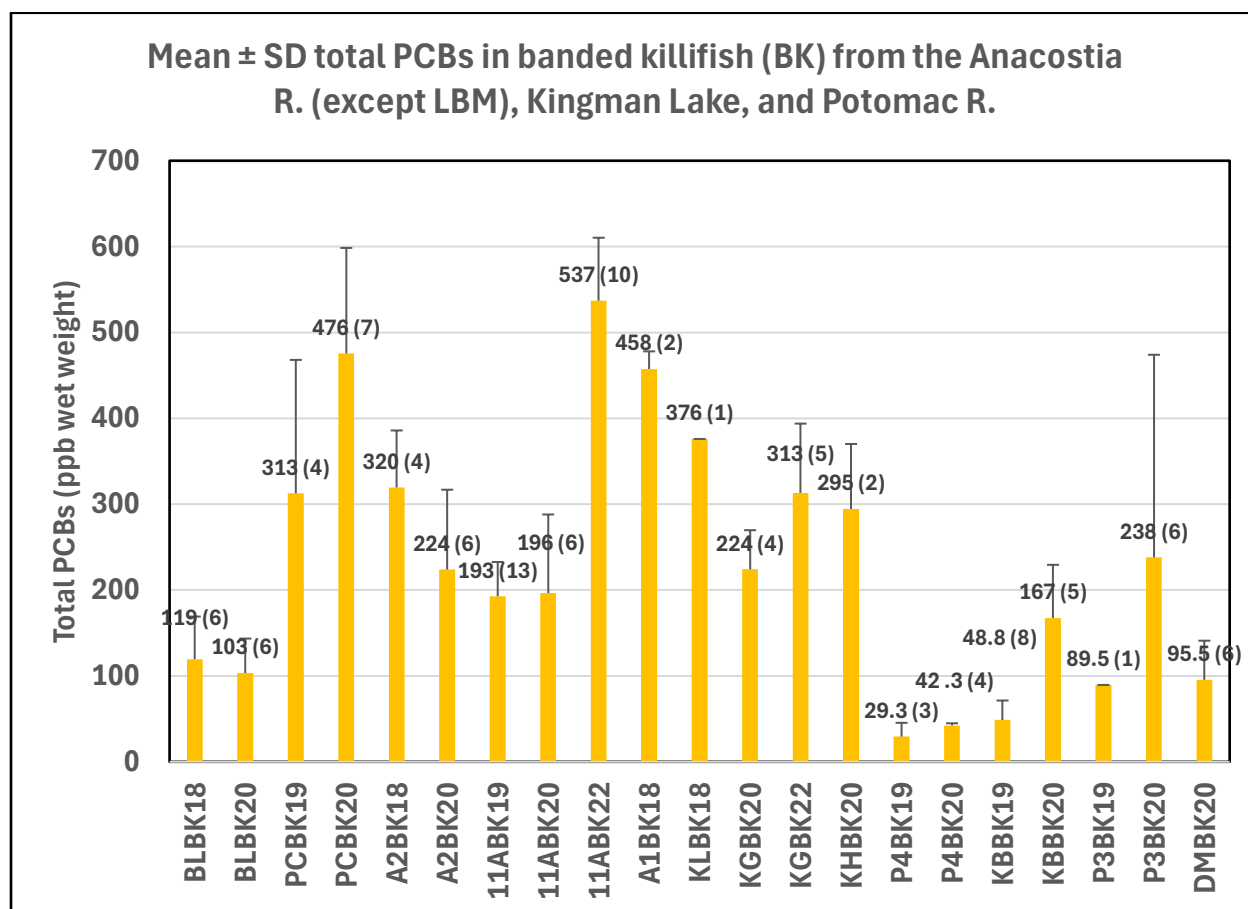


Fig. 4. Mean total PCBs (ppb wet wt., mean + standard deviation) in collections of banded killifish (BK) from the Anacostia R. (except LBM), Kingman Lake, and the Potomac R. Numbers of samples in parentheses. See Table 1 for abbreviations.

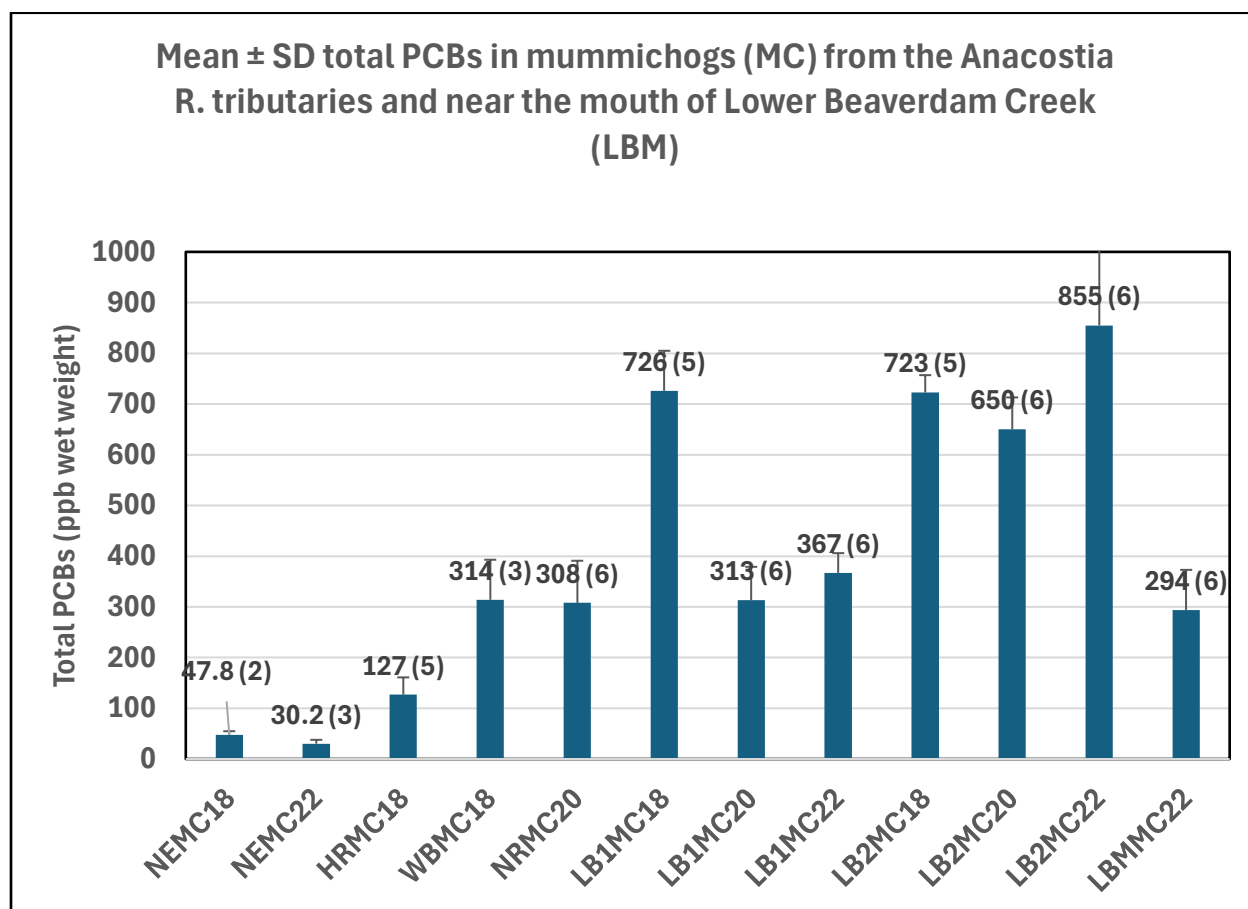


Fig. 5. Mean total PCBs (ppb wet wt., mean + standard deviation) in collections of mummichogs (MC) from the Anacostia River tributaries and near the mouth of Lower Beaverdam Creek (LBM). Numbers of samples in parentheses. See Table 1 for abbreviations.

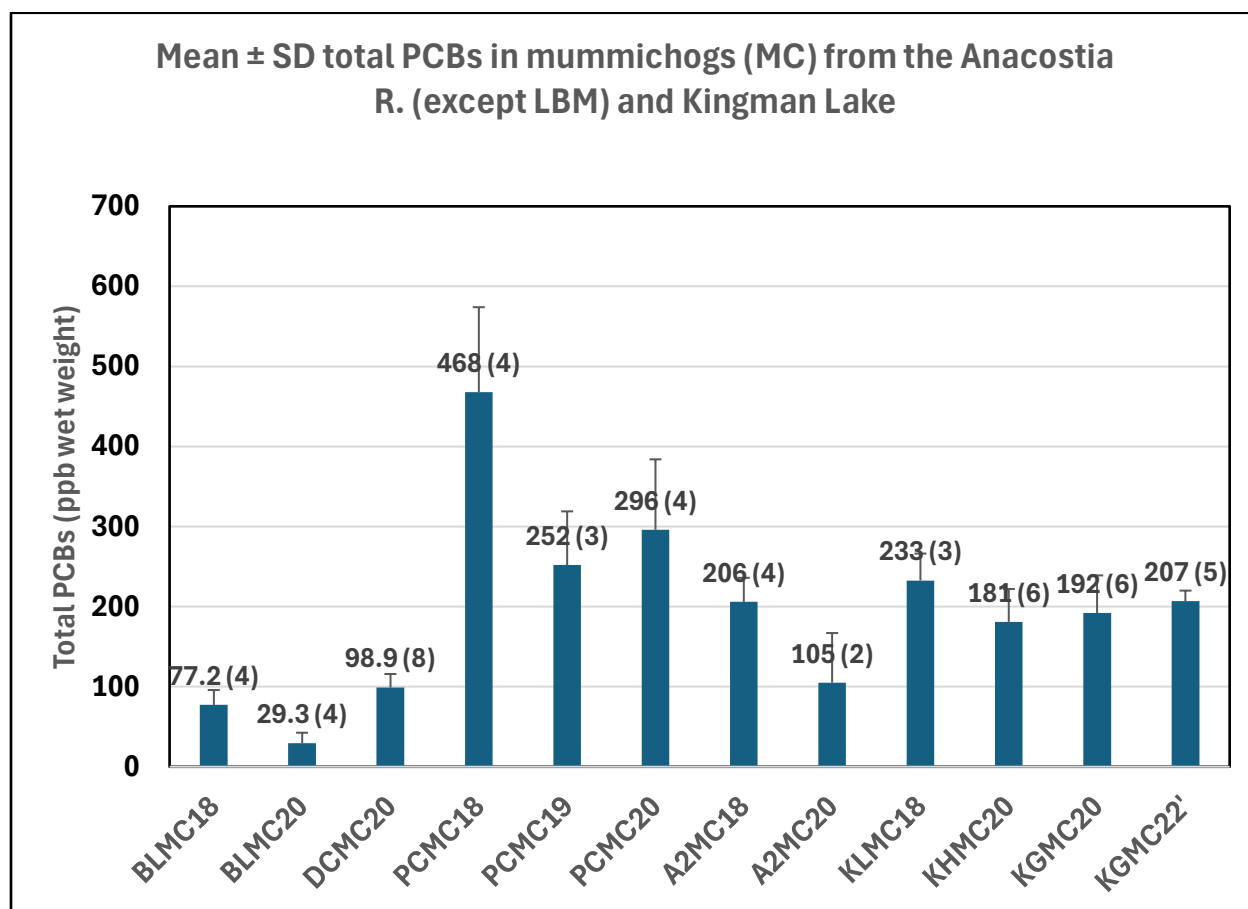


Fig. 6. Mean total PCBs (ppb wet wt., mean + standard deviation) in collections of mummichog from the Anacostia River (except LBM) and Kingman Lake. Numbers of samples in parentheses. See Table 1 for abbreviations.

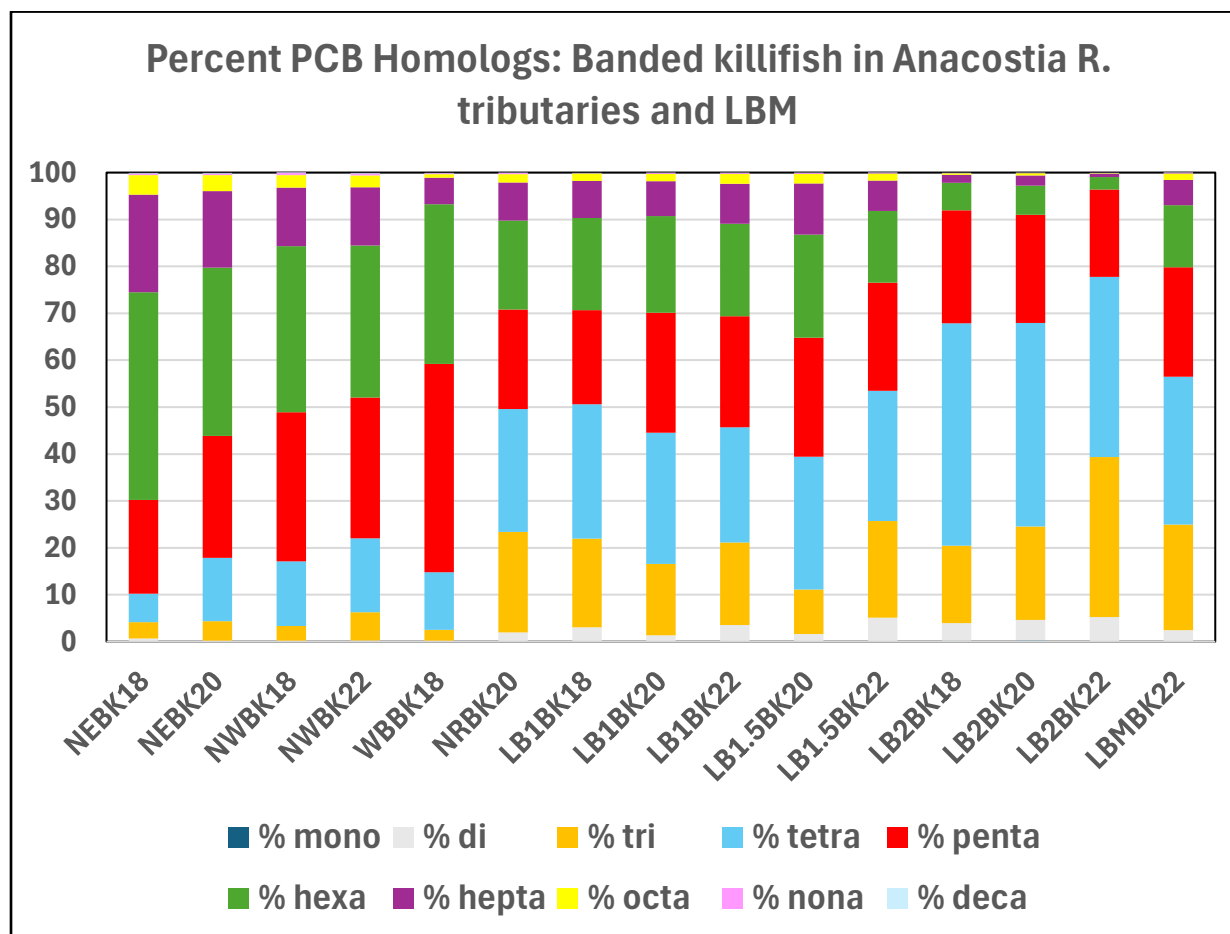


Fig. 7. Mean PCB homolog distribution (% of total) in collections of banded killifish (BK) from Anacostia River tributaries and near the mouth of Lower Beaverdam Creek (LBM); mono-, nona-, and deca- were minimal. See Table 1 for abbreviations.

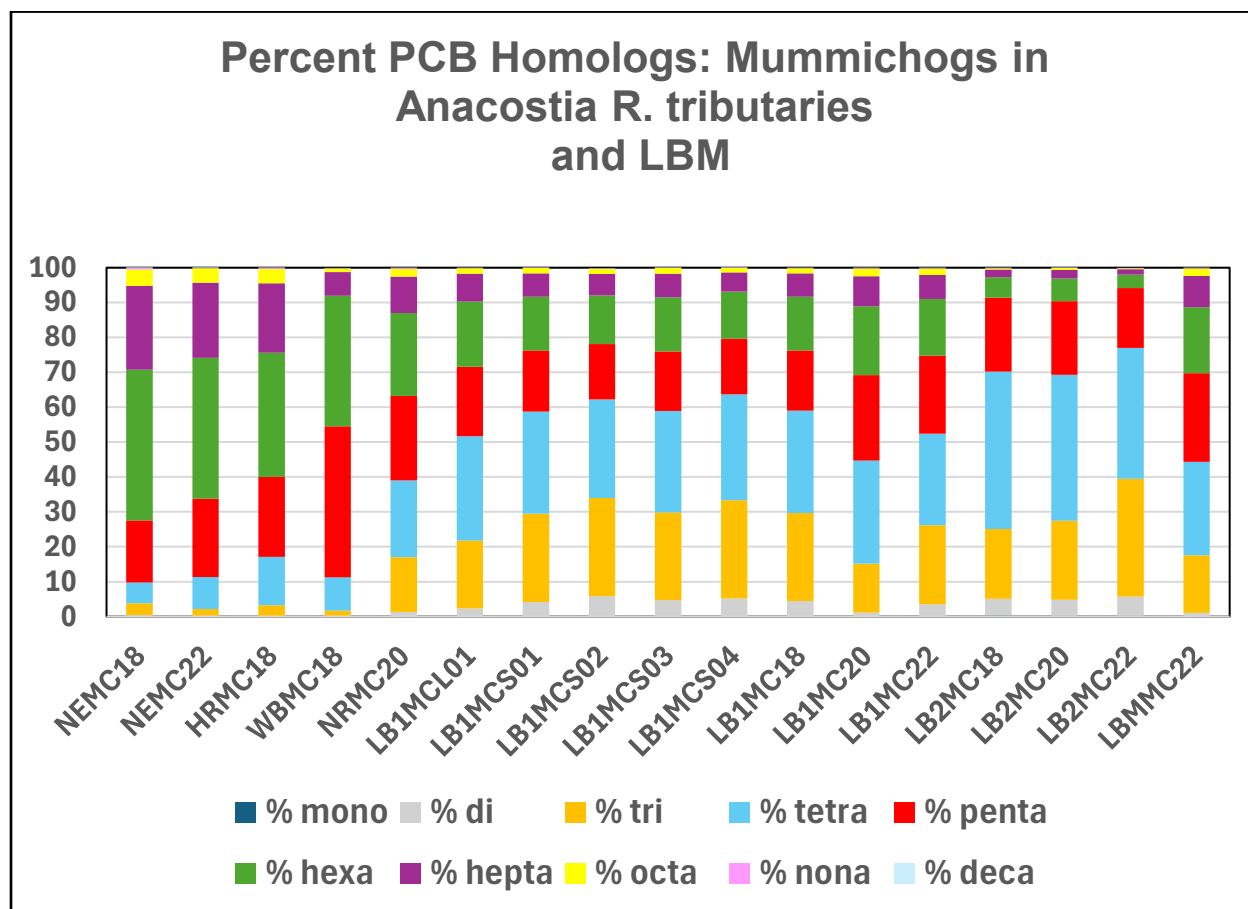


Fig. 8. Mean PCB homolog distribution (% of total) in collections of mummichog (MC) from Anacostia River tributaries and near the mouth of Lower Beaverdam Creek (LBM): mono-, nona-, and deca- were minimal. See Table 1 for abbreviations.

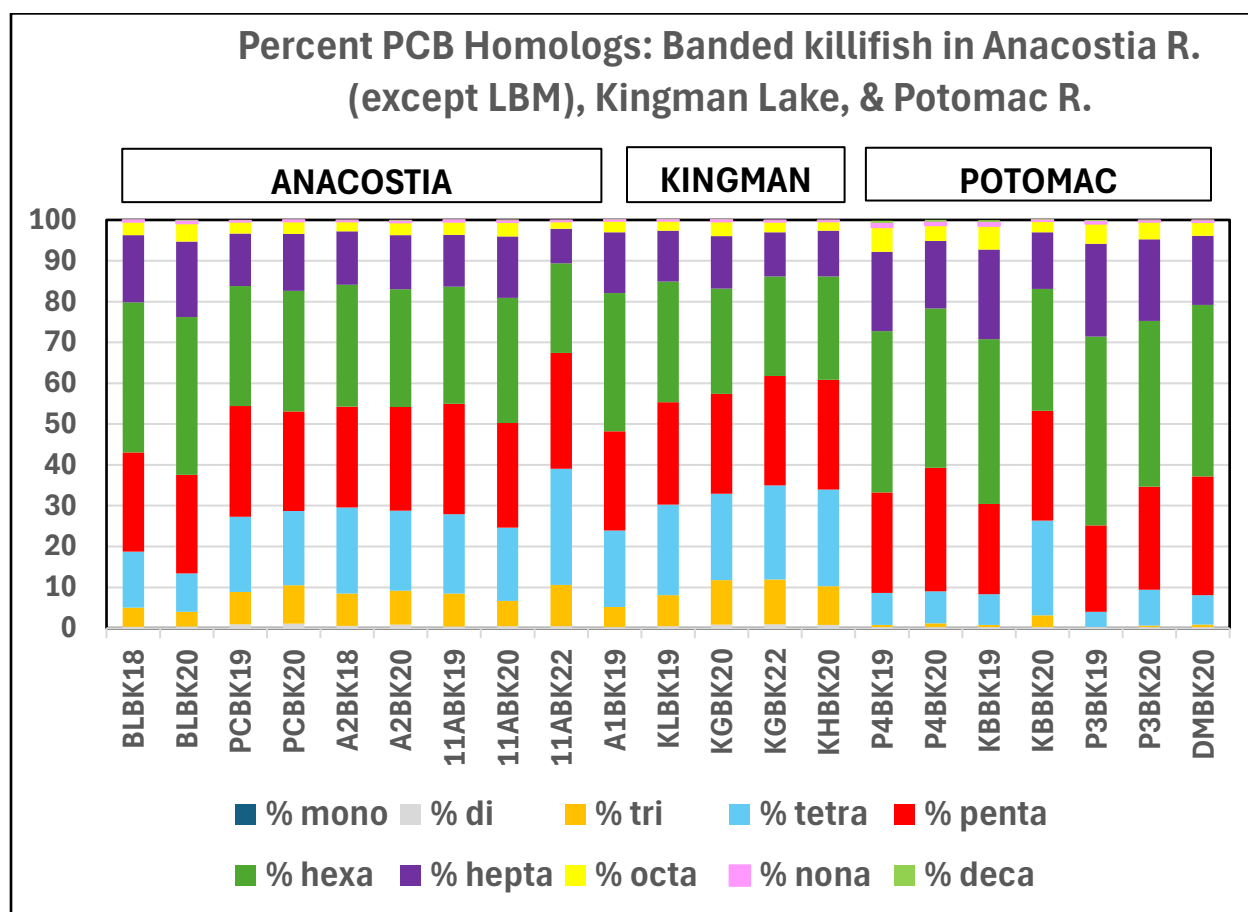


Fig. 9. Mean PCB homolog distribution (% of total) in collections of banded killifish (BK) from Anacostia River (except LBM), Kingman Lake, and Potomac River: mono-, nona-, and deca- were minimal. See Table 1 for abbreviations.

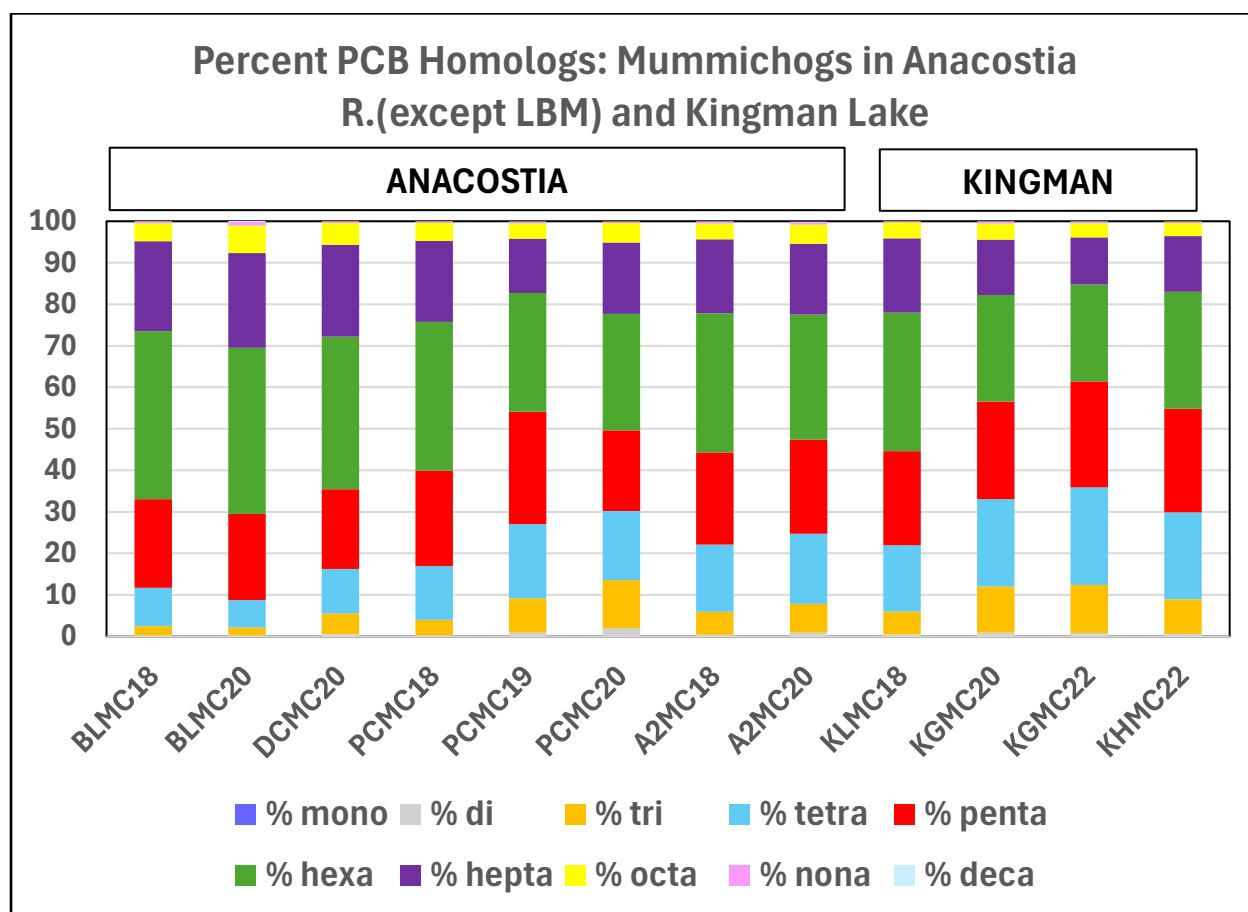


Fig. 10. Mean PCB homolog distribution (% of total) in collections of mummichog (MC) from the Anacostia River (except LBM) and Kingman Lake; mono-, nona-, and deca- were minimal. See Table 1 for abbreviations.

Supplementary Files

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