Environmental Toxicology and Chemistry—Volume 37, Number 9—pp. 2467–2474, 2018
Received: 28 January 2018 | Revised: 23 March 2018 | Accepted: 21 June 2018

Environmental Toxicology

Riparian Spiders Indicate the Magnitude and Sources of Polychlorinated Biphenyl Contamination at a Large Contaminated Sediment Site

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Abstract: We investigated polychlorinated biphenyl (PCB) contamination at the Ashtabula River (northeast OH, USA) area of concern following remedial dredging using araneid and tetragnathid spiders. The PCB concentrations remain elevated in the area of concern compared with reference conditions. Patterns of contamination were strikingly similar between taxa, but were higher in tetragnathids at the most contaminated sites. Spider PCB homolog distributions identified 2 PCB sources to the area of concern. Based on these findings, we recommend situations where these taxa can be used alone, in concert, or combined into a composite "spider" sample to assess environmental contamination. *Environ Toxicol Chem* 2018;37:2467–2474. Published 2018 Wiley Periodicals Inc. on behalf of SETAC. This article is a US government work and, as such, is in the public domain in the United States of America.

Keywords: Aquatic–riparian linkages; Source identification; Bioaccumulation; Polychlorinated biphenyls; Environmental forensics; Environmental fate

INTRODUCTION

Decades of manufacturing and waste-management practices at industries in the Ashtabula River watershed in northeast Ohio (USA) resulted in the discharge or release of hazardous substances, including polychlorinated biphenyls (PCBs). In 1985, the Ashtabula River was designated as a Great Lakes (USA/Canada) area of concern (US Environmental Protection Agency 2014a), in part due to elevated PCB concentrations observed in the sediment and fish. Areas of concern are geographical areas within the Great Lakes Basin that show signs of severe environmental degradation, as defined under the Great Lakes Water Quality Agreement between the United States and Canada. A cleanup effort in 2003 at the Ashtabula River area of concern focused on the removal of PCBs and other contaminants at the Fields Brook Superfund site (US Environmental Protection Agency 2014a). Fields Brook is a tributary draining into the lower Ashtabula River, and is a known source of PCBs to the Ashtabula River area of concern (US Environmental Protection Agency 2014a). Subsequent studies in the Ashtabula River area of concern indicated that contaminant impacts were still present, and in 2007 the US Environmental Protection Agency (USEPA) led dredging efforts that removed approximately 420 500 m^3 of contaminated sediments along a 1.7-km-long reach of the lower Ashtabula River (US Environmental Protection Agency 2010; Figure 1) under the Great Lakes Legacy Act. Few studies since 2006 have investigated the present state of the PCB contamination in the lower Ashtabula River (but see Meier et al. 2015), and none have addressed the potential export of PCBs to the adjacent terrestrial food web.

Spiders have been used as sentinels of aquatic pollution for various contaminants including mercury (Cristol et al. 2008; Schipper et al. 2008; Pennuto and Smith 2015), metals (Otter et al. 2013; Kraus et al. 2014), and PCBs (Walters et al. 2008, 2010; Raikow et al. 2011; Kraus et al. 2017). Prior work has focused on 2 web-building riparian spider taxa from the families Tetragnathidae and Araneidae because they are proven sentinels of contaminant export from aquatic ecosystems to terrestrial food webs (Walters et al. 2010; Kraus et al. 2017). Many tetragnathid spiders are obligate riparian taxa specializing in the consumption of aquatic insects, which they catch in weak, horizontal orb webs. In contrast, araneid spiders are distributed from riparian to upland habitats and build stronger, vertical orb webs in which they catch aquatic and terrestrial insects.

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Published online 21 June 2018 in Wiley Online Library (wileyonlinelibrary.com).
DOI: 10.1002/etc.4216

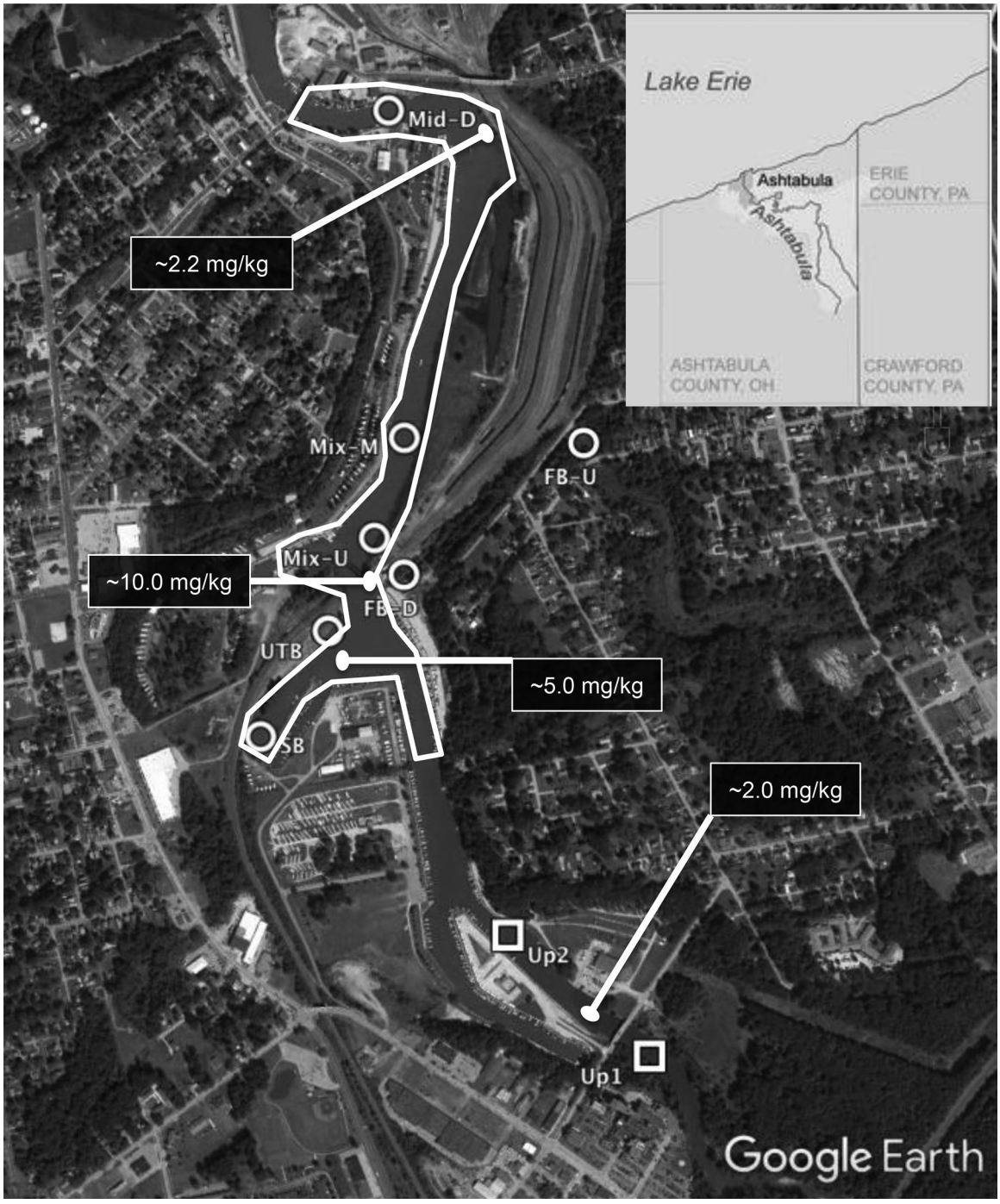

FIGURE 1: Map of sampling locations. Concentrations displayed are for total polychlorinated biphenyls (ΣPCBs; dry mass) in sediment obtained from Meier et al. (2015) during their 2011 sampling. The white polygon delineates the area dredged by the US Environmental Protection Agency in 2007. The area of concern boundaries include the entire mainstem Ashtabula River and Harbor below the bridge crossing near site upstream1 (Up1). The sampling locations were as follows: Up1, Up2 = upstream sites; SB = Strong Brook; UTB = Upper Turning Basin; FB-U and -D = Fields Brook upstream and downstream; Mix-U, -M, and –D = upper, middle, and downstream mixing sites.

In near-shore environments (e.g., within 5 m of the water's edge), both taxa demonstrate strongly aquatic diets, as indicated by a natural abundance of carbon and nitrogen isotopes and by the accumulation of aquatic contaminants in spider tissues (Walters et al. 2008, 2010; Raikow et al. 2011; Speir et al. 2014; Jackson and Sullivan 2017). Even though tetragnathid and araneid spiders are increasingly used as indicators of aquatic contaminants, it remains to be seen whether these 2 taxa tell a similar story about PCB contamination in the environment. That is, are they reliable indicators of one another such that they can be combined in a single statistical analysis or can their tissues can be pooled into a single composite sample for chemical analyses? In 2 prior studies, their PCB concentrations were highly correlated and they ordered sites similarly along a

contamination gradient; however, in one case PCBs in araneids were approximately 2× higher than in tetragnathids (Walters et al. 2010), and in another case, concentrations between these taxa closely approached 1 to 1 (Kraus et al. 2017).

The PCBs are a mixture of up to 209 unique congeners based on the number and location of chlorine atoms on the molecule. They bioaccumulate in the aquatic food web and can be exported from the aquatic to the terrestrial food web (Sullivan and Rodewald 2012). They can cause adverse health effects, including cancer, in animals (National Research Council 2001). The PCB homologs are groups of PCB congeners that have an equal number of chlorines associated with their chemical structure. Through an understanding of the chemical differences between individual congeners and homologs, it is possible to identify source-specific contaminant signatures within and among field sites (Moon et al. 2012; Saba and Su 2013). These types of environmental forensics investigations are most commonly addressed by applying multivariate statistical approaches to the distributions of chemical signatures measured in different matrices such as water, sediment, or biota (Johnson and Ehrlich 2002).

Our goal was to investigate PCB contamination in the waterways associated with the Ashtabula River area of concern using tetragnathid and araneid spiders. Our specific objectives were first to assess the magnitude of PCB exposure in riparian spiders throughout the area of concern. Second, we evaluated whether PCB concentrations in the 2 taxa varied similarly among sites to determine whether different riparian spiders respond in the same fashion to variation in ambient environmental contamination (Beeby 2001). Finally, we applied a multivariate analysis of spider PCB signatures to investigate whether multiple, distinct sources of PCBs existed within the area of concern.

MATERIALS AND METHODS

Sampling locations

We sampled 9 sites associated within the Ashtabula River area of concern (Figure 1). This included 2 sites, Strong Brook and Fields Brook, that were previously thought to be sources of PCBs to the rest of the Ashtabula area of concern (US Environmental Protection Agency 2014a); 2 sites upstream of these putative sources (Up1, Up2); and 4 sites downstream of the Strong and Fields Brooks where we hypothesized that mixing of PCBs from these potential sources could occur (Mix-upstream, upper mixing site; Mix-downstream, downstream mixing site; Mix-middle, middle mixing site; and a site locally known as the Upper Turning Basin, which is maintained for shipping traffic). We sampled a downstream site in Fields Brook (Fields Brook-downstream) near its confluence with the Ashtabula River and an upstream site (Fields Brook-upstream) outside the legally defined boundaries of the area of concern.

Spider collection

We collected spiders in July 2011 using methods similar to those described in Kraus et al. (2017). We first established a transect running perpendicular to flow at each sampling site. We divided the shoreline at each site into 4 quadrants, with 1 quadrant upstream and 1 downstream of the transect on each bank. Each quadrant sampled included 50 m of shoreline. Riparian spiders were then sampled by hand at night using headlamps. Sampling was limited to habitats overhanging the water surface and extending up to 2 m inland from the shoreline. We targeted 2 families of orb-weaving spiders, Tetragnathidae and Araneidae, which are easily identified in the field. Our goal was to collect 4 replicate samples (~15–25 individuals) per taxon at each site; however, we did not achieve this level of replication at some sites due to low spider biomass (Table 1).

PCB and lipid analysis

We homogenized all spider samples prior to extraction using stainless steel or titanium implements. The homogenized sample was aliquoted for extraction into a Teflon® bottle, fortified with surrogate standards, and extracted 3 times with methylene chloride using Tissuemizer™ techniques. We then dried the combined extracts over anhydrous sodium sulfate and removed interfering compounds using an alumina column followed by high-performance liquid chromatography. We exchanged the solvent to *n*-hexane, concentrated the extract to 0.5 mL, and added internal standards. We analyzed the extracts for 122 congeners following USEPA method 8270D (US Environmental Protection Agency 2014b) using gas chromatography–mass spectrometry in the selective ion monitoring mode. All target compounds were quantified using the internal standard method, and all results are reported in ng/g (parts/billion) wet weight. We calculated the total PCBs and homologs by summing measured concentrations for all congeners and for individual homolog groups, respectively. The average method detection limit was 0.04 (± 0.01) ng g^{-1}, and the reporting detection limit was 2.33 (± 1.51) ng g^{-1} among all congeners in all samples. All sample results below the reporting detection limit were assigned a value of 0. Method blanks and laboratory control samples were run at least once per batch (20 samples). Laboratory control samples consisted of extraction solvent spiked with a known volume of PCB at a known stock concentration. Method blanks and laboratory control samples were extracted and analyzed identically to field samples, and laboratory control samples were run in duplicate. Method blanks were composed of a clean reference matrix, using a weight or volume similar to that of field samples. Tissue method blanks contained 1 g of corn oil spiked with 1 mL of calibration standard solution. Method blanks were below the detection limit in all cases. Mean recovery of laboratory control samples was 75.1% (± 12.2, 1 standard deviation) among all samples and congeners. Relative percentage difference between laboratory control sample duplicates was 14.0% (± 13.2). Surrogate spikes of PCB34 and PCB152 were run for each sample, and recovery averaged 68.6% (± 13.4). We determined the percentage of lipid (as total extractable organics) in the tissue samples by drying an aliquot of pre-alumina sample extract from the solvent extraction at 40 °C for approximately 5 min. The percentage of

TABLE 1: Polychlorinated biphenyl (PCB) homolog group (percentage of total PCBs) and percentage of lipid content (mean ± standard deviation) by site and spider taxa

			Homolog group (%)										
	Site	*n*	Mono	Di	Tri	Tetra	Penta	Hexa	Hepta	Octa	Nona	Deca	% Lipid
Araneidae	Up1	1	0.0	0.0	0.0	0.0	8.9	32.1	39.7	15.3	2.7	1.3	4.1 ± —
	Up2	4	0.0	0.0	3.4	14.8	15.6	28.5	24.7	9.5	2.4	1.1	3.7 ± 0.5
	SB	3	0.0	0.0	1.3	5.1	11.0	38.1	34.6	8.9	0.8	0.2	3.6 ± 0.4
	UTB	4	0.0	0.0	4.0	16.2	15.4	31.5	25.9	5.9	0.6	0.4	4.2 ± 0.2
	FB-U	2	0.0	0.0	5.2	45.4	33.3	8.9	4.5	1.7	0.5	0.5	5.4 ± 1.9
	FB-D	4	0.0	0.0	5.7	39.3	31.4	12.2	7.9	2.5	0.5	0.5	5.9 ± 2.3
	Mix-U	4	0.0	0.0	6.0	23.2	18.2	21.4	21.5	7.6	1.3	0.8	4.1 ± 0.8
	Mix-M	4	0.0	0.0	3.8	16.6	17.1	26.1	24.4	9.1	1.7	1.1	3.0 ± 0.2
	Mix-D	4	0.0	0.0	3.6	19.8	20.5	25.8	20.7	7.1	1.4	1.1	3.8 ± 0.1
Tetragnathidae	Up1	4	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	2.8 ± 0.2
	Up2	2	0.0	0.0	4.4	26.7	22.9	25.9	17.3	2.0	0.5	0.3	3.9 ± 0.3
	SB	4	0.0	0.0	1.7	7.7	10.9	38.2	32.8	8.1	0.5	0.1	3.4 ± 0.4
	UTB	4	0.0	0.0	4.9	21.7	17.0	28.0	22.4	5.2	0.5	0.3	3.0 ± 0.4
	FB-U	4	0.0	0.0	3.5	45.7	36.3	8.2	3.7	1.4	0.5	0.6	3.3 ± 1.1
	FB-D	4	0.0	0.0	5.7	48.4	31.9	7.9	4.0	1.4	0.4	0.4	2.9 ± 0.5
	Mix-U	5	0.0	0.0	8.5	36.3	22.6	14.2	13.3	3.9	0.6	0.6	3.7 ± 0.4
	Mix-M	3	0.0	0.0	5.8	28.9	21.8	19.0	17.2	5.4	0.9	1.0	4.1 ± 0.6
	Mix-D	2	0.0	0.0	3.4	25.5	27.3	23.4	16.2	3.2	0.5	0.6	4.9 ± 0.2

n = sample size; Up1, Up2 = upstream sites; SB-U and -D = Strong Brook upstream and downstream; UTB = Upper Turning Basin; FB-U and -D = Fields Brook upstream and downstream; Mix-U, -M, and –D = upper, middle, and downstream mixing sites.

lipid (wet wt) was determined by a gravimetric analysis of the extract residue following evaporation.

Statistical analysis

We investigated spatial differences in total PCB concentrations using one-way analysis of variance with Tukey's post hoc tests. We modeled the relationship between araneid and tetragnathid PCB concentrations using linear regression to determine their degree of correlation and calculated Spearman's rho (a nonparametric rank correlation procedure) to determine whether they ranked sites similarly along a gradient of PCB contamination. We used JMP Pro 9 (SAS Institute) for statistical analyses with significant differences defined by p values <0.05. We evaluated spatial patterns in PCB homolog composition with nonmetric multidimensional scaling ordination using PC-ORD 6.0 (MjM Software) following the procedure recommended by McCune and Mefford (1999). This approach determines the appropriate dimensionality and statistical significance of results while seeking to avoid local minima. We used the Sørenson (Bray–Curtis) coefficient as the distance measure in the analysis. We ran the nonmetric multidimensional scaling using the mean relative abundance of PCB homolog groups (as a proportion of ΣPCB) for each spider taxon at each site.

Preliminary inspection of the nonmetric multidimensional scaling ordination indicated that sites clustered into 3 distinct groups with unique spider PCB homolog signatures. We applied the multiple response permutation procedure, a nonparametric technique for quantifying differences between or among groups (McCune et al. 2002), to test the hypothesis that sites within these groups had different homolog signatures. We ran the multiple response permutation procedure test using the ranked transformed Bray–Curtis (Sorenson) distance matrix (McCune et al. 2002). The procedure first tests for differences among all groups (an omnibus test), followed by post hoc pairwise comparisons if the omnibus test is significant. We evaluated the multiple response permutation procedure results based on p values to test for significance and the A statistic, a measure of chance corrected for within-group agreement. The A statistic is a measure of effect size that is independent of sample size. Possible values of A range from 0 (no separation between groups) to 1 (complete separations between groups). Values of $A > 0.3$ are relatively high (McCune et al. 2002), indicating large separation among groups.

We excluded sampling location Up2 from all tetragnathid analyses because PCBs were all below method detection limits at this site. All PCB congener data and lipid data are provided in Walters et al. 2018 (see *Data Availability* statement).

RESULTS

Mean concentrations for total PCBs ranged from 24.6 to 574.6 ng/g wet weight and from below detection limit to 991.8 ng/g wet weight for araneid and tetragnathid spiders, respectively. Total PCB concentrations were significantly different among sites for araneid ($F_{(8,29)} = 23.48$; $p < 0.0001$) and tetragnathid spiders ($F_{(7,27)} = 28.91$; $p < 0.0001$; Figure 2). The highest concentrations of PCBs in both taxa were at sampling locations Fields Brook-upstream and Fields Brook-downstream in Fields Brook, and these were significantly higher than those at all other sites with the exception of araneids at Strong Brook (Figure 2).

Mean total PCB concentrations in araneid and tetragnathid spiders were significantly and positively correlated across all sampling locations ($R^2_{(8)} = 0.95$, $p < 0.0001$; Figure 3). Araneids and tetragnathids also ranked sites nearly identically along a gradient in PCB contamination (Spearman's $r = 0.98$). The

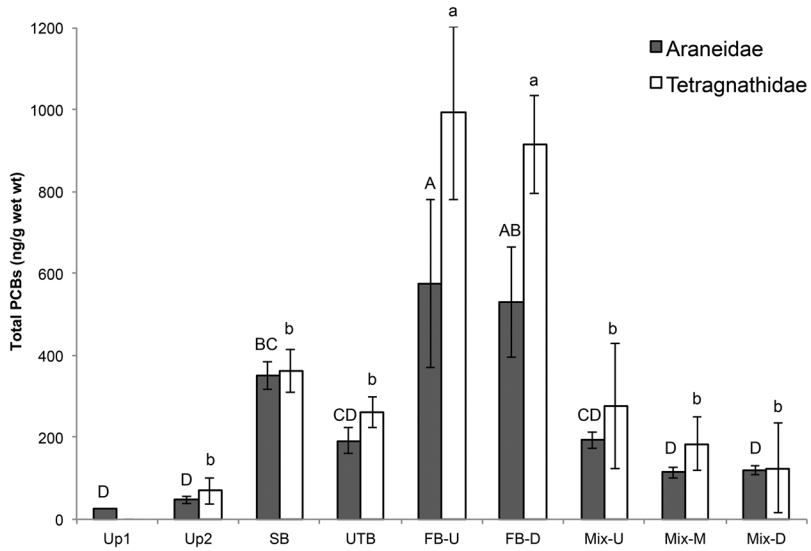

FIGURE 2: Total polychlorinated biphenyl (PCB) concentrations (mean ± 1 standard deviation) in araneid and tetragnathid spiders. Significant difference between sampling locations are indicated by different uppercase letters (araneid) and lowercase letters (tetragnathid; $p < 0.05$, by analysis of variance). The sampling locations were as follows: Up1, Up2 = upstream sites; SB = Strong Brook; UTB = Upper Turning Basin; FB-U and -D = Fields Brook upstream and downstream; Mix-U, -M, and -D = upper, middle, and downstream mixing sites.

magnitude of PCB contamination between spider taxa was similar (i.e., approaching 1:1; Figure 3) except for the sites in Fields Brook where tetragnathid concentrations were 1.7 times higher than araneid concentrations.

The nonmetric multidimensional scaling analysis resulted in a 2-dimensional solution with a final stress of 2.8, indicating a robust ordination with a high degree of interpretability (McCune et al. 2002). The nonmetric multidimensional scaling analysis revealed 2 highly distinct PCB signatures contrasting location Strong Brook with locations Fields Brook-upstream and Fields Brook-downstream in Fields Brook (Figure 4). The PCB homolog composition in spiders from sites Fields Brook-upstream and Fields Brook-downstream was dominated by tetra- and pentachlorinated PCBs (~70–80%), whereas spiders at location

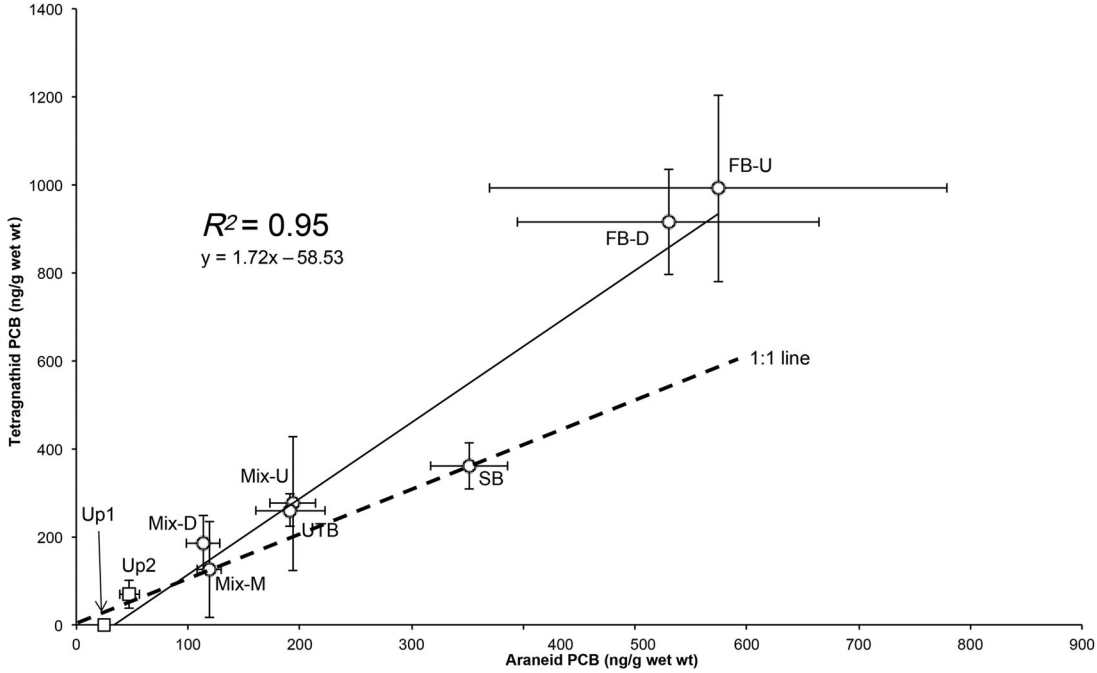

FIGURE 3: Relationship between polychlorinated biphenyl (PCB) concentrations in araneid and tetragnathid spiders. Data are presented as mean ± standard deviation for each sampling location. The solid line is the regression model; the dashed line represents the 1:1 relationship. The sampling locations were as follows: Up1, Up2 = upstream sites; SB = Strong Brook; UTB = Upper Turning Basin; FB-U and -D = Fields Brook upstream and downstream; Mix-U, -M, and -D = upper, middle, and downstream mixing sites.

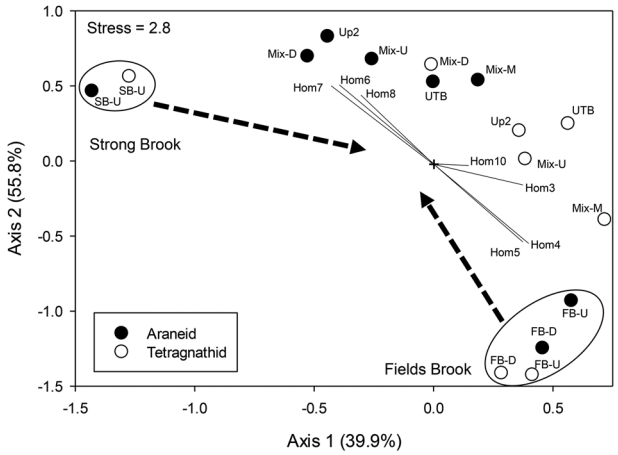

FIGURE 4: Nonmetric multidimensional scaling plots of spider taxa by sites in polychlorinated biphenyl (PCB) homolog (Hom) space. Point labels correspond to sites mapped in Figure 1. Spider data are overlaid with PCB homolog groups, which are driving the point distributions. Sampling locations Strong Brook and Fields Brook-upstream/Fields Brook-downstream are circled, indicating distinct PCB signatures. Sampling locations in the mainstem of the Ashtabula River have intermediate homolog profiles, indicating mixing (dashed arrows) of these 2 sources. Plotted homolog groups are correlated with either axis at $p < 0.01$. Vectors associated with homologs (e.g., Hom3) indicate the direction and magnitude of these correlations. The plus symbol marks the centroid of the plot. Numbers in parentheses indicate the amount of variance explained by each axis. The sampling locations were as follows: Up1, Up2 = upstream sites; SB-U and -D = Strong Brook upstream and downstream; UTB = Upper Turning Basin; FB-U and -D = Fields Brook upstream and downstream; Mix-U, -M, and -D = upper, middle, and downstream mixing sites.

Strong Brook were dominated by hexa- and heptachlorinated PCBs (~70%; Table 1). Other sampling locations had homolog profiles that were intermediate between these endpoints, indicating mixing of these 2 primary PCB sources (Figure 4). This was also true for Up2, which is subject to seiche effects (i.e., upstream river flow) from Lake Erie (USA/Canada). The homolog signatures among groups were significantly different and showed a high degree of separation (multiple response permutation procedure omnibus test; $p < 0.0001$, $A = 0.53$). Spiders at Strong Brook, Fields Brook, and mixing sites each had distinct homolog signatures (multiple response permutation procedure pairwise comparisons; $p < 0.02$; A values of 0.30–0.42 among comparisons). Homolog signatures were most distinct between spiders at Strong Brook and Fields Brook ($A = 0.42$). Apart from the strong spatial patterns in homolog composition, we also observed a more subtle difference in homolog composition between taxa within a site. Araneids tended to have a greater proportion of the higher homolog groups than tetragnathids, as evidenced by the separation of these taxa along axis 2.

DISCUSSION

Total PCB concentrations were significantly higher than upstream site concentrations at sites Fields Brook-upstream, Fields Brook-downstream, and Strong Brook in araneid spiders and at Fields Brook-upstream and Fields Brook-downstream in tetragnathid spiders (Figures 1 and 2). These are 2 distinct areas (Fields Brook, represented by Fields Brook-upstream and Fields Brook-downstream, and Strong Book, represented by Strong Brook) that drain into the Ashtabula River near its termination into Lake Erie. Our results generally support the findings of Meier et al. (2015). They investigated sediment and macroinvertebrate PCB concentrations at multiple locations in the Ashtabula area of concern, including sites that overlapped with sampling locations of the present study (Fields Brook-downstream, Upper Turning Basin, Up1, and Mix-downstream). In 2011, Meier et al. (2015) measured the highest concentrations of PCBs (~900 ng/kg wet wt in macroinvertebrates and ~10 mg/kg dry wt in sediment) at the site corresponding to Fields Brook-downstream and the second highest concentrations from the site corresponding to Upper Turning Basin (~300 ng/kg wet wt in macroinvertebrates and ~5 mg/kg dry wt in sediment). Likewise, we measured the highest PCB concentrations in spiders within the boundary of the area of concern at site Fields Brook-downstream. In addition, Meier et al. (2015) noted that toward the end of their study a second source of PCBs was identified as originating at Strong Brook and that further remediation was performed in that portion of the river in 2013. The results of the present study, combined with results from Meier et al. (2015), indicate that Fields Brook and Strong Brook were sources of PCBs to the rest of the Ashtabula Harbor as late as 2011. Further testing is needed to assess the effectiveness of the 2013 remediation at Strong Brook. Our finding that PCBs remain high in Fields Brook upstream of its confluence with the Ashtabula River (Fields Brook-upstream) indicate that Fields Brook itself could still be contributing PCBs to the Ashtabula River area of concern, if PCBs remaining in that portion of the tributary are transported downstream and deposited into the Ashtabula River itself.

Accumulation of PCBs in araneid and tetragnathid spiders indicated that they responded similarly to local-scale variation in ambient environmental contamination across the area of concern. The PCB concentrations in araneid and tetragnathid spiders were strongly correlated with one another and were nearly identical in ranking the sites from least to most contaminated (i.e., Spearman's $r = 0.98$). The ΣPCB concentrations between the taxa approached a 1 to 1 relationship, suggesting that factors influencing PCB accumulation (e.g., diet, uptake, and elimination) were also similar between taxa. This relationship did not hold at the Fields Brook sites. In the present study we measured the highest PCB concentrations in both taxa, yet concentrations in tetragnathids were approximately double those in araneids. It is possible that lower concentrations in araneids relative to tetragnathids are due to dietary differences for araneids. Fields Brook is a small stream (<5 m wetted width) completely enclosed by the forest canopy. Tetragnathid spiders build weak webs and are specialized consumers of weak-flying aquatic insects, whereas araneids build more robust webs capable of capturing stronger flying terrestrial prey (Kraus et al. 2017). It is possible then that araneids in this more forested environment consumed relatively more terrestrial prey than did tetragnathid spiders.

The PCB homolog distributions further support the idea that PCBs from 2 distinct sources were present in 2011 and were mixing in the main stem of the Ashtabula River downstream of the Fields Brook confluence (Figures 1 and 4). Araneid and tetragnathid homolog distributions had unique PCB signatures at Fields Brook and Strong Brook. This finding supports earlier documentation of contamination at the site showing that PCBs were entering the area of concern from the Fields Brook Superfund site (US Environmental Protection Agency 2014a) and that additional industrial activities (improper storage of electrical transformers) were introducing PCBs into Strong Brook (Leppert Associates 2007). Spiders at Fields Brook (Fields Brook-upstream and Fields Brook-downstream) had PCB distributions composed mainly of less chlorinated homolog groups (e.g., tetra- and pentachlorinated PCBs), whereas those at Strong Book contained more chlorinated homolog groups (e.g., hexa- and heptachlorinated PCBs). The source of the Fields Brook contamination likely originated upstream of the Fields Brook-upstream sampling location, which is well outside the legally proscribed boundary of the Ashtabula River area of concern. Mixing of PCBs from these 2 sources was evident downstream of the Fields Brook confluence based on homolog profiles of spider tissues. Evidence of mixing upstream of the Fields Brook confluence was also observed at Up2, possibly due to upstream transport of PCBs from Strong Brook and Fields Brook during seiche events (which we occasionally observed while working at this site).

The homolog profiles were more enriched (i.e., favoring heavier homolog groups) in araneid spiders than in tetragnathid spiders, and we observed similar trends in homolog distributions between these taxa at the Manistique River and Harbor area of concern on Lake Michigan (USA; D.M. Walters, unpublished data). These patterns could be related either to differences in diet or to differences in uptake and excretion between these taxa. It is unlikely that dietary differences are driving these patterns, as there is little reason to suspect that the trajectory of PCB exposure (uptake of PCBs by aquatic insect larvae, retention of PCBs in adult aquatic insects through metamorphosis, and trophic transfer of PCBs from adult insects to spiders) is different between these spider taxa. We propose that taxa-specific physiological processes led to differential uptake and excretion of PCB congeners between spider taxa, and testing this hypothesis will require controlled laboratory feeding studies.

CONCLUSIONS

Many contaminants, including PCBs, are exported from aquatic ecosystems to terrestrial food webs via emerging adult aquatic insects (Sullivan and Rodewald 2012). Riparian spiders are effective sentinels that bridge aquatic and terrestrial ecosystems (Beeby 2001; Walters et al. 2010; Speir et al. 2014; Kraus et al. 2017). In the present study, variation in the magnitude of spider PCB concentrations indicated 2 PCB contamination hotspots where PCBs were known historically to enter the Ashtabula River via tributary inputs. Quantitatively (linear regression analysis) and qualitatively (Spearman's rank correlation), these taxa provided similar information on the degree of PCB contamination across the broader area of concern, yet their concentrations did diverge markedly at the 2 sites within Fields Brook. These results indicate that either taxa can be used individually, to save on analytical chemistry costs (rather than sampling both taxa at a site), or that their tissues could be combined into a single composite sample depending on the project goal. For example, if the goal is to map areas of relatively high environmental contamination or contaminant bioavailability, then either taxa or a composite of the 2 should suffice. However, if the goal is to monitor long-term changes in contaminant concentrations across a site, then a single taxa should be selected a priori for that assessment. Finally, our results highlight the application of spider tissue contaminant profiles to identify contaminant sources using environmental forensic techniques. Prior studies using these techniques have focused more on contaminant profiles in aquatic media such as sediments and fish tissues.

Acknowledgment—We thank A. Mucha and S. Cieniawski (US Environmental Protection Agency [USEPA]) for contributing to different aspects of the present study including study design and project management. This manuscript was improved by helpful comments from R. Yeardley and T. Luxton. The present study was supported by a USEPA Great Lakes Restoration Initiative grant to D.M. Walters and by the USEPA Office of Research and Development.

Disclaimer—This research was subjected to US Geological Survey and US Environmental Protection Agency review and approved for publication. Any use of trade, firm, or product names is for descriptive purposes only and does not imply endorsement by the US Government.

Data availability—The following information is available: PCB congener data by sample, as well as lipid and total homolog and total PCB data by sample (totals by sum of congeners). Data used in the present study are reported in Walters et al. (2018) and are available at this link: https://doi.org/10.5066/P9SX2FCX.

REFERENCES

Beeby A. 2001. What do sentinels stand for? *Environ Pollut* 112:285–298.

Cristol DA, Brasso RL, Condon AM, Fovargue RE, Friedman SL, Hallinger KK, Monroe AP, White AE. 2008. The movement of aquatic mercury through terrestial food webs. *Science* 320:335.

Jackson BK, Sullivan SMP. 2017. Ecosystem size and flooding drive trophic dynamics of riparian spiders in a fire-prone Sierra Nevada river system. *Can J Fish Aquat Sci* 75:308–318.

Johnson GW, Ehrlich R. 2002. State of the art report on multivariate chemometric methods in environmental forensics. *Environ Forensics* 3:59–79.

Kraus JM, Schmidt TS, Walters DM, Wanty RB, Zuellig RE, Wolf RE. 2014. Cross-ecosystem impacts of stream pollution reduce resource and contaminant flux to riparian food webs. *Ecol Appl* 24:235–243.

Kraus JM, Gibson PP, Walters DM, Mills MA. 2017. Riparian spiders as sentinels of polychlorinated biphenyl contamination across heterogeneous aquatic ecosystems. *Environ Toxicol Chem* 36:1278–1286.

Leppert Associates. 2007. On-site PCB impact characterization and remediation final report. Prepared for Clean Harbors PPM, Coffeeville, KS, USA.

McCune B, Mefford MJ. 1999. *Multivariate Analysis of Ecological Data*, Ver 4.10. MjM Software, Gleneden Beach, OR, USA.

McCune B, Grace JB, Urban DL. 2002. *Analysis of Ecological Communities*. MjM Software Design, Gleneden Beach, OR, USA.

Meier JR, Lazorchak JM, Mills M, Wernsing P, Baumann PC. 2015. Monitoring exposure of brown bullheads and benthic macroinvertebrates to sediment contaminants in the Ashtabula River before, during, and after remediation. *Environ Toxicol Chem* 34:1267–1276.

Moon HB, Choi M, Choi HG, Kannan K. 2012. Severe pollution of PCDD/Fs and dioxin-like PCBs in sediments from Lake Shihwa, Korea: Tracking the source. *Mar Pollut Bull* 64:2357–2363.

National Research Council. 2001. *A Risk-Management Strategy for PCB-Contaminated Sediments*. National Academy Press, Washington, DC, USA.

Otter RR, Hayden M, Mathews T, Fortner A, Bailey FC. 2013. The use of tetragnathid spiders as bioindicators of metal exposure at a coal ash spill site. *Environ Toxicol Chem* 32:2065–2068.

Pennuto CM, Smith M. 2015. From midges to spiders: Mercury biotransport in riparian zones near the Buffalo River Area of Concern (AOC), USA. *Bull Environ Contam Toxicol* 95:701–706.

Raikow DF, Walters DM, Fritz KM, Mills MA. 2011. The distance that contaminated aquatic subsidies extend into lake riparian zones. *Ecol Appl* 21:983–990.

Saba T, Su S. 2013. Tracking polychlorinated biphenyls (PCBs) congener patterns in Newark Bay surface sediment using principal component analysis (PCA) and positive matrix factorization (PMF). *J Hazard Mater* 260:634–643.

Schipper AM, Wijnhoven S, Leuven R, Ragas AMJ, Hendriks AJ. 2008. Spatial distribution and internal metal concentrations of terrestrial arthropods in a moderately contaminated lowland floodplain along the Rhine River. *Environ Pollut* 151:17–26.

Speir SL, Chumchal MM, Drenner RW, Cocke WG, Lewis ME, Whitt HJ. 2014. Methyl mercury and stable isotopes of nitrogen reveal that a terrestrial spider has a diet of emergent aquatic insects. *Environ Toxicol Chem* 33:2506–2509.

Sullivan SMP, Rodewald AD. 2012. In a state of flux: The energetic pathways that move contaminants from aquatic to terrestrial environments. *Environ Toxicol Chem* 31:1175–1183.

US Environmental Protection Agency. 2010. Field study on environmental residuals: Ashtabula River, Vol I. Final Report. EPA/600/R-10/126. Washington, DC.

US Environmental Protection Agency. 2014a. Third five-year review report for Fields Brook Superfund Site Ashtabula County, Ohio. Region 5. Chicago, IL.

US Environmental Protection Agency. 2014b. Method 8270D: Semivolatile organic compounds by gas chromatography/mass spectrometry. *Test Methods for Evaluating Solid Wastes: Physical/Chemical Methods*. EPA SW-846. Washington, DC.

Walters DM, Fritz KM, Otter RR. 2008. The dark side of subsidies: Adult stream insects export organic contaminants to riparian predators. *Ecol Appl* 18:1835–1841.

Walters DM, Mills MA, Fritz KM, Raikow DF. 2010. Spider-mediated flux of PCBs from contaminated sediments to terrestrial ecosystems and potential risks to arachnivorous birds. *Environ Sci Technol* 44:2849–2856.

Walters DM, Otter RR, Kraus JM, Mills MA. 2018. Polychlorinated biphenyl concentrations and lipid content in riparian spiders at the Ashtabula River Area of Concern, USA. USGS Data Release. US Geological Survey, Reston, VA, USA. DOI:10.5066/P9SX2FCX.

