Hepatic Burdens of PCB and PCDD/F Congeners in Federally Endangered Shortnose Sturgeon and Atlantic Sturgeon from the Hudson River, New York, USA: Burden Patterns and Potential Consequences in Offspring

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Abstract

Sturgeon populations worldwide are threatened with extirpation but little is known about their tendency to bioaccumulate contaminants and their sensitivities to environmental burdens of these contaminants. Shortnose sturgeon and Atlantic sturgeon, two species that are federally endangered in the U.S., co-occur in the Hudson River (HR) where high sediment levels of polychlorinated biphenyls (PCBs), polychlorinated dibenzo-*p*-dioxins (PCDDs) and polychlorinated dibenzo-*p*-furans (PCDFs) occur. Previous controlled laboratory studies showed that young life-stages of both species are sensitive to toxicities at low levels of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) and PCB126 exposure. The objective here was to measure congener-specific hepatic levels of PCBs and PCDD/Fs in HR specimens in order to determine if *in situ* bioaccumulation of these compounds is sufficiently high to have caused the early life-stage toxicities previously observed. Estimates of hepatic burdens of PCBs and PCDD/Fs were obtained from a small number of specimens of each species collected between 2014 and 2016 and specimens of shortnose sturgeon collected over 30 yr earlier and archived in a museum collection. Several significant patterns emerged. Hepatic levels of legacy PCBs and PCDDs were low in specimens of both species but typically higher in shortnose than Atlantic sturgeon, a pattern consistent with their habitat use in the HR. Hepatic burdens in shortnose sturgeon tended to be higher in archived specimens than in more recently collected ones despite expected reduction in archived specimens due to preservation methods. Several inadvertent PCBs congeners were detected at high levels, including PCB11, but their toxicity to natural populations remains unknown. Levels of select PCDFs congeners, 2,3,7,8-TCDF and 2,3,4,7,8 PeCDF, were elevated in some shortnose sturgeon individuals from the HR. Using Relative Potency (ReP) factors derived from white sturgeon, the observed levels of some hepatic PCDFs in HR shortnose sturgeon may have been sufficiently high to impair recruitment of young life-stages in this ecosystem.

Introduction

Sturgeons (Acipenseridae) are an ancient taxon distributed throughout the Northern Hemisphere that has suffered severe declines in all 27 extant species. All sturgeons are listed on the IUCN Red List, and abundances of 22 of the 27 taxa continue to decrease (www.iucnredlist.org). Furthermore, 15 taxa are listed as "Critically Endangered," the most threatened of all listing categories. Several factors have been proposed to explain these declines including overharvest, habitat loss (dam construction and channel dredging), vessel strikes, impaired water quality, and chemical contamination. Many studies have examined the effects of the first four of these factors on declining population abundances, but few have investigated the role that chemical contaminants may have played in these declines (Chambers et al. 2012; Doering 2016).

Two species of sturgeons inhabit the Atlantic seaboard of North America – shortnose sturgeon (*Acipenser brevirostrum*) and Atlantic sturgeon (*A. oxyrinchus oxyrinchus*). Both are federal status species which protects them from harvest, restricts disturbance to their habitats, and limits their access to researchers. Shortnose sturgeon was listed in 1973 under the U.S. Endangered Species Act (ESA) as endangered throughout its U.S. range. In 2012, Atlantic sturgeon was also listed as endangered throughout its U.S. distribution except for the Gulf of Maine population which is listed as threatened (Federal Register 2012a, 2012b).

Shortnose sturgeon is amphidromous and resident, but with seasonal movements, within its natal estuaries (Altenritter et al. 2018). In the Hudson River (HR), male shortnose sturgeon mature at 3 to 4 yr and females in 6 to 8 yr (Bain 1997). Atlantic sturgeon is anadromous and highly migratory after its 2 to 8 yr estuarine-resident juvenile life-stage (Bain 1997; Caron and Tremlay 1999; Fox and Peterson 2019). The distance of its migrations in Atlantic coastal waters may be thousands of kilometers (Erickson et al. 2011; Wirgin et al. 2015; Rothermel et al. 2020) with returns to natal estuaries for spawning commencing at ages 6 to 8 yr in southern populations and 25 to 30 yr in northern ones (Dadswell 2006). Both species spawn demersal eggs in spring to early summer in freshwater (Hager et al. 2020), and larvae occupy bottom habitats. Lifespans of these sturgeons are long (Jaric et al. 2018) and can extend to at least 60 yr in more northerly populations of both species (Gilbert 1989; Dadswell 2006; Dadswell et al. 1984).

Some of the largest populations of Atlantic Coast sturgeons occur in estuaries noted for high levels of sediment-borne chemical contaminants. For example, the HR is believed to host the largest population of Atlantic sturgeon coastwide (Waldman et al. 2018; Kazyak et al. 2020) and a

robust population of shortnose sturgeon (Bain 1997). The types and sources of contaminants prevalent in the HR include elevated levels of polychlorinated biphenyls (PCBs) released from the 1940s to the mid 1970s from two General Electric electrical capacitor manufacturing facilities located 314 and 318 river kilometers (rkm) upstream from the river's mouth (rkm 0) (Figure 1). As a result, a 318-rkm reach of the HR mainstem is designated a federal Superfund site, the largest in the U.S. Trustees of the HR PCBs Superfund site are charged with determining if PCBs have caused injuries to its resources. Hence, there is particular concern regarding the potential injuries from PCBs on HR sturgeon populations but the ESA protected status precludes the collection of these sturgeons for studies including estimation of contaminant body burdens.

The HR ecosystem is also beset with high levels of polychlorinated dibenzo-*p*-dioxins (PCDDs). The main source of PCDDs was the Diamond Alkali manufacturing facility located next to the Passaic River in Newark, New Jersey, several km upstream from Newark Bay. Newark Bay is contiguous with the HR via the Kill van Kull and New York Harbor (Figure 1). From 1951 to 1969, this manufacturer produced herbicides and defoliants, including Agent Orange. Some product leached into the Passaic River and was subsequently transported downstream to Newark Bay and into the Hackensack River. As a result, the lower Passaic River and part of Newark Bay are designated as a federal Superfund site. The HR Estuary also contains elevated levels of polychlorinated dibenzofurans (PCDFs) from sources including PCB production and incineration of municipal wastes.

Despite the cessation of the release of these contaminants, they continue as a concern for human and ecosystem health. They are highly lipophilic, persistent, bioaccumulative, and biomagnify in food chains which can result in high burdens in piscivorous fishes. In the HR Estuary, high levels of total PCBs were regularly reported in finfishes that are desired for human consumption (Skinner 2011) and for Atlantic tomcod (*Microgadus tomcod*), a sentinel of ecological health (Courtenay et al. 1999; Fernandez et al. 2004). Far less data exist on burdens of PCDD/Fs, but highly elevated hepatic levels were reported in Atlantic tomcod (Courtenay et al. 1999; Fernandez et al. 2004) and in several crustaceans, e.g., blue crab (*Callinectes sapidus*) and American lobster (*Homarus americanus*) (Hauge et al. 1994), from the Passaic River-Hackensack River-Newark Bay complex. Largely because of the endangered status of sturgeons, there are no studies on levels of these contaminants – by class or congeners – in either sturgeon species in the HR Estuary nor on a congener-specific basis in any sturgeon worldwide. To begin filling this data

void, this paper uses all available HR sturgeon samples known to us – N = 13 sturgeon collected from various sources over a three-decade span (1984 to 2016) – and analyzed for PCB and PCDD/F contaminants on a congener-specific basis.

Early life-stages of fishes are often exquisitely sensitive to toxicities from PCBs and PCDD/Fs. But, finfish species (Elonen et al. 1988), and even populations within species (Nacci et al. 2010; Wirgin et al. 2011), vary by orders of magnitude in their vulnerabilities to these contaminants. For example, lake trout (*Salvelinus namaycush*) is the most sensitive vertebrate taxon to TCDD known (Walker et al 2011), while other finfishes (e.g., zebrafish *Danio rerio*) are far less sensitive (Henry et al. 1997).

PCBs and PCDD/Fs exist in nature as heterogeneous mixtures with varying persistence and toxicities among homologue classes and congeners. Coplanar congeners are structurally most similar to dioxin and are most toxic due to their high affinity to bind with the aryl hydrocarbon receptor (AHR) which is known to activate most of the early life-stage toxicities from these compounds. Unlike in mammals, there are at least two AHRs in fishes (AHR1 and AHR2), including in shortnose sturgeon and Atlantic sturgeon (Roy et al. 2018ab). AHR2 is likely more important in mediating toxicities because of its greater expression among most tissues and efficacy in binding TCDD. The most toxic PCDD congener, 2,3,7,8 tetrachlorodibenzo-*p*-dioxin (TCDD), is used for computing the relative toxicities of coplanar PCB and PCDD/Fs congeners via a Toxic Equivalency Factors (TEFs) computation. Recent studies have detected several PCB congeners in environmental samples that are not part of legacy Aroclor mixtures, but are deemed 'inadvertent PCBs' (Vorkamp 2016) originating as byproducts of manufacturing products such as certain pigments and dyes. The toxicities of these compounds are largely unknown (Roy et al. 2019).

Recent controlled laboratory studies investigated the sensitivities of early life-stages of several North American sturgeon species to toxicities from PCBs and PCDD/Fs exposures. Early life-stages of shortnose sturgeon and Atlantic sturgeon were sensitive at low doses of PCB126 and TCDD (at > 0.1 parts per billion (ppb) and > 0.01 ppb, respectively) to cytochrome P4501A mRNA expression (Roy et al. 2011) and to whole-organism, early life-stage measures of fish health (Chambers et al. 2012). Similarly, studies of white sturgeon (*A. transmontanus*) and lake sturgeon (*A. fulvescens*) demonstrated these taxa to be among the more sensitive finfishes to PCDD/Fs early life-stage toxicities (Doering et al. 2014; Doering et al. 2015; Doering 2016; Eisner et al. 2016).

A challenge encountered in such studies on sturgeons is linking the outcomes from laboratory exposure experiments to *in situ* effects due to the near absence of tissue-burden data for these contaminants in environmentally-exposed specimens due to their endangered status. The objective here was to identify and assay the limited number of shortnose sturgeon and Atlantic sturgeon tissue samples from *in situ* collections, then quantify the congener-specific hepatic burdens of PCBs and PCDD/Fs. These data were then used to determine if the *in situ* concentrations of these compounds were sufficiently high to elicit the toxicities observed in young life-stages of these sturgeons in previous laboratory studies. Further, the available tissue samples from HR sturgeons allowed examination of the patterns of relative hepatic burdens between these two sturgeons and over time.

Methods

U.S. ESA listing of both sturgeon species precluded sacrificing additional specimens for this study. Hence, the sample sizes of each species reflect collections from fatalities associated with bridge construction, a small set of non-destructively sampled fish from a previous study, and museum specimens. Despite this constrained sample size, it was sufficient to assess and relate trends in hepatic contaminant burdens to life history and habitats of these taxa, and to compare burdens in environmentally exposed specimens to those eliciting toxicities in controlled laboratory studies.

We analyzed livers from eight shortnose sturgeon and five Atlantic sturgeon from the HR (Table 1). Specimens were obtained from three sources that differed in collection location, time, and method which also affected the status of the specimen for quantifying hepatic contaminant burdens. First, three archived shortnose sturgeon were obtained from the New York State Museum, Albany, New York. These were collected from the HR in the mid 1980s, initially stored in buffered formalin, and subsequently transferred to 95% ethanol. Second, seven live specimens (3 shortnose sturgeon and 4 Atlantic sturgeon) were collected and liver samples were obtained by laparoscopic biopsy. These individuals were collected in the Haverstraw Bay region (rkm 59-63) in September 2014 and April 2016, anesthetized (Matsche 2011), and a plug of liver tissue (≈ 100 mg / individual) obtained as described in Matsche (2013). Third, two recently dead shortnose sturgeon and one Atlantic sturgeon were collected in 2015 and 2016 from within 16 km of the Tappan Zee Bridge (rkm 44) by Allee, King, Rosen, and Fleming (AKRF), Inc. These were

collected as part of a retrieval program for sturgeon fatalities associated with the construction of the Mario Cuomo Bridge (adjacent to and replacing the Tappan Zee Bridge). All three fish had external injuries consistent with vessel strikes. Overall, the eight shortnose sturgeon ranged from 33.3 to 88.3 cm total length (TL), with seven > 46 cm TL and likely adults. The five Atlantic sturgeon ranged from 42.7 to 76.5 cm TL placing four of five in the subadult range (subadults reported by Bain (1997) as 50 to 150 cm TL).

Congener-specific PCB and PCDD/Fs analyses were conducted by AXYS Analytical Services, Sydney, British Columbia, Canada. Liver samples weights ranged from approximately 0.1 g (biopsies) to 25 g (dead and archived specimens). The liver samples were homogenized using dissection scissors to ensure complete extraction. Extraction and chromatographic clean-up procedures were performed in accordance with SGS-AXYS Method MLA-013: Analytical Method for The Determination of: Polybrominated Diphenyl Ethers, PCB Congeners, Chlorinated Pesticides, Technical Toxaphene, Toxaphene Congeners/Parlars and Polychlorinated Dibenzodioxins and Furans using Co-Extraction Techniques. Samples were fortified with isotopically labeled surrogate standards, and Soxhlet extracted in 1:1 Dichloromethane:Hexane for 16 hr. The resulting extract was gravimetrically split to reserve a portion as backup. Each resulting extract was spiked with clean-up standards, subsampled for lipid analysis, and then cleaned using a series of chromatographic columns. The clean-up columns generated separate fractions for the PCDD/F and PCB analyses.

Dioxin/furan analysis procedures were in accordance with USEPA Method 1613, Revision B, as documented in SGS AXYS Method MLA-017: Analytical Method for the Determination of Polychlorinated Dibenzodioxins and Dibenzofurans by EPA Method 1613B, EPA Method 8290/8290A or EPA Method DLM02.2. The fraction for PCDD/Fs analysis was concentrated and spiked with ^{13}C-labeled recovery (internal) standards, for a final volume of 20 µL. The injection volumes were 1 µL for the DB-5 column analysis and 2 µL for the DB-225 confirmation analysis. The DB-5 capillary column (60 m, 0.25 mm i.d., 0.1 µm film thickness) was coupled to a high-resolution mass spectrometer (Waters Micromass Autospec Premier, Milford, MA). The mass spectrometer was tuned to a static mass resolution of $\geq$ 10,000 in the electron impact ionization mode and data acquired in the voltage selected recording mode (SIR). A second instrumental analysis was performed using a DB-225 capillary column (30 m, 0.25 mm i.d., 0.15 µm film thickness) coupled to a mass spectrometer (Waters Micromass Autospec Ultima) to confirm and/or

quantify 2,3,7,8-TCDF and 1,2,3,7,8,9-HxCDD. This column was necessary to resolve non-toxic congeners that co-elute with 2,3,7,8-TCDF and 1,2,3,7,8,9-HxCDD on a DB-5 column. Analyte concentrations were quantified using MassLynx software (Waters, Milford, MA).

PCB congener analyses were in accordance with the USEPA Method 1668, Revision A: Chlorinated Biphenyl Congeners in Water, Soil, Sediment, and Tissue by HRGC/HRMS as documented in SGS AXYS Method MLA-010. The PCB fraction extract was further cleaned using Alumina chromatographic columns. The final extract was reduced in volume and spiked with ^{13}C-labeled recovery (internal) standards prior to instrumental analysis. PCB extracts were concentrated to 20 µL, and 1 µL was injected onto an SPB-Octly column (30 m, 0.25 mm i.d., 0.25 µm film thickness) coupled to a high-resolution mass spectrometer (Waters Micromass Ultima). The mass spectrometer was tuned to a static mass resolution of $\geq$ 10,000 in the electron impact ionization mode and data acquired in the voltage selected recording mode. Analyte concentrations were quantified using Micromass OPUSquan software.

All samples were analyzed in a single batch with associated QC samples consisting of a procedural blank and a lab-generated reference sample known as the Ongoing Precision and Recovery (OPR). The QC samples were prepared using canola oil as the matrix. The liver samples and the QC samples were prepared alongside each other and were subjected to the same analytical procedures. QC samples were evaluated against the analytical method criteria.

PCDD/Fs and PCBs homologue totals are the sum of concentrations of detected congeners at each level of chlorination. Congener peaks that did not meet the method ion abundance ratio criteria were excluded from the homologue totals and Toxic Equivalent (TEQ) calculations. TEQs were calculated per Van den Berg et al. (1998) for World Health Organization (WHO) fish TEFs.

Means ($\pm$ SEM), sample sizes, and the number of samples are reported for concentrations above the level of analytical detectability for each congener within a class, class totals, and summary TEQs. Statistical tests (one-way linear model) were conducted on two null hypotheses, H_0. H_{01} tested for no difference in hepatic burden between time periods of collection (2010's vs. 1984). H_{02} tested for no difference between species (shortnose sturgeon vs. Atlantic sturgeon). For H_{02}, only fish from contemporaneous collections were used (i.e., 2010's). A significance level of $p < 0.05$ was used throughout. Further, a summary of trends is provided in the results due to the small sample sizes ($N = 5$ for the contemporaneously collected specimens of both species, and $N = 3$ for the archived shortnose sturgeon specimens) and the further reduction in power of tests due to

a substantial fraction of contaminant assays falling below detectability levels. Trends were summarized in three ways: 1) The proportion of congeners within a class where the mean hepatic concentration is significant in one of the two possible directions pertinent to the H_0, 2) The proportion of mean directionality regardless of meeting the $p < 0.05$ criterion, and 3) The ratio of the grand mean (mean of mean concentrations) of congener concentrations within each contaminant class. This last assessment of trends provides an assessment of the magnitude of deviation from no difference between the groups being contrasted.

Results

Congener-specific hepatic PCB and PCDD/F concentrations (pg/g ww) were successfully obtained for all 13 sturgeon specimens including the three archived shortnose specimens from the mid 1980's. A number of hepatic concentrations were below analytical detectability (97 of 267 congener-specific assays or 27% of all cases).

Regarding coplanar PCB congeners (both non-ortho and mono-ortho substituted), significantly greater hepatic concentrations were found in contemporaneously collected (2014 to 2016) shortnose sturgeon than in Atlantic sturgeon for 9 of 11 congeners (Table 2). The overall hepatic concentration (ratio of grand means across all individual coplanar PCB congeners) was 5.4 × higher in shortnose sturgeon than Atlantic sturgeon for the 11 congeners tested and 5.8 × higher for total PCBs (Table 2). Hepatic concentrations of individual coplanar PCB congeners did not significantly differ between archived specimens (1980's) and those collected in the 2010's but 7 of 11 congeners (and the total PCB levels) were higher in archived samples. The overall hepatic coplanar PCB congener concentration was 3.4 × higher in archived versus recent shortnose sturgeon specimens and 1.4 × higher for total PCBs.

Of the three inadvertent PCB congeners (PCB5, PCB11, and PCB52), concentrations of PCB11 and PCB52 were high in both species and significantly higher (5 to nearly 7 ×) in contemporary shortnose than Atlantic sturgeon specimens (Table 2). For example, the mean hepatic levels of PCB11 were 398 pg/g ww and 60 pg/g ww, respectively. Similarly, the mean hepatic levels of PCB52 were 346,250 pg/g ww and 64,300 pg/g ww, respectively. PCB11 and PCB52 were over 4 × higher in contemporary than archived shortnose sturgeon specimens.

Concentrations of the two non-ortho substituted coplanar PCB congeners for which levels were detectable (PCB77, PCB81), were 3 to 8 × higher, respectively, in shortnose sturgeon than in

Atlantic sturgeon. The PCB81 value for Atlantic sturgeon may be biased downward as four of five observations were below detectability (Table 2). Concentrations of the two other non-ortho substituted coplanar congeners (PCB126, PCB169) were non-detectable in all specimens but one archived shortnose sturgeon collected near Albany (rkm 232) which had a high PCB126 burden (38.4 pg/g ww). PCB77 was the dominant of the four dioxin-like congers in livers of recent shortnose sturgeon and Atlantic sturgeon (mean = 1,032 and 349 pg/g ww, respectively) and was comparably high in the archived shortnose sturgeon specimens (mean = 969 pg/g ww).

Hepatic burdens of PCDDs were low and variable among all HR groups (Table 3). Hepatic burdens of TCDD, the most toxic PCDD congener, were relatively low and variable with concentrations for individuals ranging from non-detectable to 2.72 pg/g ww (Atlantic sturgeon). The highest group mean (1.71 pg/g ww) was for recent shortnose sturgeon but without significant differences between Atlantic sturgeon and shortnose sturgeon or between contemporary versus archived shortnose sturgeon. The highest mean PCDD concentrations were for OctaCDD in recent and archived shortnose sturgeon (9.3 and 178 pg/g ww, respectively), and in Atlantic sturgeon (8.2 pg/g ww). A tendency was evident for greater levels in archived than recent specimens of shortnose sturgeon (4.8 ×). The concentrations of 1,2,3,4,6,7,8 HeapTACDD were also high, differing significantly between recent and archived shortnose sturgeon (4.9 and 14.2 pg/g ww, respectively), and were also high in Atlantic sturgeon (7.4 pg/g ww) (Table 3).

Mean hepatic burdens of PCDFs varied widely among the 10 congeners with highest concentrations for the most toxic PCDF congener (2,3,7,8 TCDF) in all three HR sturgeon groups (Table 4). Recently collected shortnose sturgeon tended to have higher mean hepatic levels of 2,3,7,8 TCDF (26.2 pg/g ww) than archived ones (20.0 pg/g ww), and contemporary shortnose sturgeon had significantly higher levels than Atlantic sturgeon (11.1 pg/g ww). The hepatic level of 2,3,7,8-TCDF was high (42.4 pg/g ww) in one of two archived shortnose sturgeon specimens collected near Indian Point, New York. Levels of 2,3,4,7,8 PentaCDF were also high in shortnose sturgeon (e.g., 6.3 and 4.9 pg/g ww for recent and archived specimens, respectively) and significantly higher in contemporary shortnose sturgeon than Atlantic sturgeon. Overall, archived specimens tended to have greater hepatic burdens of PCDFs than recent ones (2.9 ×) and shortnose sturgeon had modestly higher PCDFs concentrations than Atlantic sturgeon (1.5 ×).

From these data, TCDD TEQs for PCBs and PCDD/Fs were derived for each specimen using TEFs for fishes in Van den Berg et al. (1998), summary statistics calculated, and contrasts

performed (Table 5). Mean hepatic TCDD TEQs were higher for PCDD/Fs than PCBs for both species (Table 5). Among recently collected specimens, shortnose sturgeon had significantly higher TCDD TEQs for PCBs than did Atlantic sturgeon (means: 1.6 vs. 0.3). Archived shortnose sturgeon specimens tended to have higher TCDD TEQs for PCBs than recent specimens; indeed, the highest two TCDD TEQs for PCBs (8.45 and 3.30) in shortnose sturgeon were both from archived specimens collected near Indian Point (rkm 69) while the lowest value for shortnose sturgeon (0.386) was also observed in an archived specimen but collected much further upstream (rkm 229) near Albany. The interspecific difference in TCDD TEQs for PCDD/Fs was marginally significant with values generally higher in shortnose sturgeon than Atlantic sturgeon (6.58 and 3.48, respectively). The lowest TCDD TEQ for PCDD/Fs (0.799) was in an Atlantic sturgeon collected in 2016 from Haverstraw Bay (rkm 59-63) while the two highest values were in shortnose sturgeon (an archived specimen (9.29) collected near Indian Point and a recent one (9.17) collected at Nyack, New York (rkm 43). Total TCDD TEQs were significantly greater (> 2 ×) in shortnose sturgeon than Atlantic sturgeon (Table 5). Archived and recent specimens of shortnose sturgeon were similar in mean concentration (9.01 vs 8.16, respectively).

TCDD TEQs were calculated for 2,3,7,8 TCDF and 2,3,4,7,8 PeCDF, and their sum based on relative potency (ReP) values developed from AHR2 activation in white sturgeon liver explants (PeCDF=1.4 and TCDF=1.2) reported by Eisner et al. (2016). Among recently collected specimens, shortnose sturgeon had significantly higher TCDD TEQs for 2,3,7,8 TCDF than did Atlantic sturgeon (means: 36.65 vs. 15.58) (Table 6). Archived shortnose sturgeon specimens tended to have lower TCDD TEQs for 2,3,7,8 TCDF than recent specimens but the highest TCDD TEQ in shortnose sturgeon (59.36) was from an archived specimen collected near Indian Point (rkm 69) while the lowest one for shortnose sturgeon (5.50) was from an archived specimen collected near Albany (rkm 229). The values of TCDD TEQs for 2,3,4,7,8 PeCDF were significantly higher in shortnose sturgeon than Atlantic sturgeon (7.51 vs 2.75, respectively). The highest TCDD TEQ for 2,3,4,7,8 PeCDF (0.96) was in an archived shortnose sturgeon (11.92) collected near Indian Point while the lowest one (0.96) was in an Atlantic sturgeon collected in 2016 from Haverstraw Bay (rkm 59-63). TCDD TEQs for the sum of these PCDFs was significantly greater (> 2 ×) in shortnose sturgeon than Atlantic sturgeon (Table 6).

Discussion

Despite the small sample size of sturgeon specimens reported here, which was constrained by the protected status in these taxa in the focal river system, this study provides the first quantification of the congener-specific levels of PCBs and PCDD/Fs in any environmentally exposed sturgeon species worldwide. The results serve as an initial evaluation of the potential effects of these compounds on the success of early life-stages and recruitment to adult populations. Our sources of specimens included a carcass retrieval program, biopsying specimens using laparoscopy, and archived museum specimens. These unique data sources on the bioaccumulation of PCBs and PCDD/Fs by sturgeons in a highly contaminated environment establish a context for prior and future experimental studies of the biological consequences of sturgeon exposure to environmentally relevant levels and classes of contaminants. Further, the results offer important insights about species differences and the role of habitat and life history in risks to contaminants.

The intent of this study was to determine if coplanar PCBs and PCDD/Fs levels in environmentally exposed HR sturgeons were at levels that induced CYP1A mRNA expression (Roy et al. 2011) and early life-stage toxicities in earlier controlled laboratory experiments (Chambers et al. 2012). Activation of the aryl hydrocarbon receptor 2 (AHR2) is required to mediate most such early life-stage toxicities in fishes including sturgeons (Roy et al. 2018a), and CYP1A induction is an indicator of AHR2 activation. The lowest nominal waterborne concentrations of PCB126 and TCDD tested by Roy et al. (2011) (0.01 ppb and 0.001 ppb, respectively) significantly induced CYP1A mRNA expression in shortnose sturgeon and Atlantic sturgeon larvae (Table 7). Furthermore, Chambers et al. (2012) demonstrated that a variety of early life-stage toxicities in larval shortnose sturgeon and Atlantic sturgeon significantly increased in prevalence at nominal waterborne concentrations from 0.01 to 0.1 ppb PCB126 and from 0.001 to 0.01 ppb 2,3,7,8 TCDD (Table 7). These studies suggest that early life-stages of both HR sturgeons are highly sensitive to PCBs and PCDDs toxicities through AHR2 activation.

How do TCDD TEQs for hepatic coplanar PCBs and PCDDs in environmentally exposed, wild HR sturgeons compare to TCDD TEQs that elicited significant CYP1A mRNA induction and early life-stage toxicities in laboratory experiments? Mean total hepatic TCDD TEQs from PCDD/Fs and PCBs in contemporary collections of environmentally exposed sturgeons from the HR were 8.2 for shortnose sturgeon and 3.8 for Atlantic sturgeon (Table 5). These are sufficiently high in both species to induce the significant CYP1A mRNA expression previously observed (Roy et al 2011). The mean hepatic burdens of coplanar PCBs alone expressed as TCDD TEQs (1.6

and 0.3 for shortnose sturgeon and Atlantic sturgeon, respectively) may not be sufficiently high to induce significant CYP1A expression in larval sturgeons. Levels of total TCDD TEQs that elicited the sublethal early life-stage toxicities varied between TCDD TEQ=1 for impaired eye development in shortnose sturgeon to TCDD TEQ=100 for impaired eye development in Atlantic sturgeon (Table 7, Chambers et al. 2012). A mean TCDD TEQ=10 significantly decreased larval length and abbreviated larval life span in laboratory experiments on early life-stages of both sturgeon species (Chambers et al. 2012). Thus, hepatic burdens of TCDD TEQs in wild, environmentally exposed sturgeons are potentially sufficient to activate some AHR-mediated sublethal toxicities in sturgeons from the HR. However, the contribution of hepatic coplanar PCBs to the total TCDD TEQs was probably insufficient to induce toxicities on their own. Levels of TCDD TEQs in environmentally exposed early life-stages of HR sturgeon will remain unknown because of strict limitations on their collection and difficulties collecting them at small sizes.

Two of the PCDFs at relatively high concentrations in HR sturgeons (2,3,7,8 TCDF and 2,3,4,7,8 PeCDF) are included in the WHO listing of TEFs for fishes (Van den Berg et a;/ 1998) with TEFs of 0.05 and 0.5, respectively. The toxicities to either HR sturgeon species of these PCDFs have not been assessed empirically. The relative potencies (ReP, a measure of activity of individual congeners compared to that of TCDD) of these PCDFs were quantified by Doering (2016) and Eisner et al. (2016) using *in vitro* activation of AHR2 in white sturgeon as their endpoint. They report considerably higher ReP values for these PCDFs in white sturgeon (1.4 and 1.2 for 2,3,7,8 TCDF and 2,3,4,7,8 PeCDF, respectively) than the WHO values summarized by Van den Berg et al. (1998). Applying the RePs for these two PCDFs in white sturgeon to hepatic burdens of the same PCDFs in HR sturgeons results in hepatic TCDD TEQs of 71.3 in one archived shortnose sturgeon and 64.9 in one contemporary shortnose sturgeon. The mean hepatic TCDD TEQs for HR sturgeon based on white sturgeon PCDFs RePs was 33.68 for archived and 44.16 for contemporary shortnose sturgeon, and 18.33 for the Atlantic sturgeon.

The hepatic TCDD TEQs burdens based on these sturgeon-specific RePs for PCDFs appear to be sufficiently high in environmentally exposed HR sturgeons to cause the toxicities observed in laboratory experiments. These burdens are sufficient to activate the AHR2 pathway *in vivo* in both HR sturgeon species (Roy et al. 2011) and to elicit the higher-level developmental defects seen in experiments by Chambers et al. (2012) on larval HR sturgeons at nominal waterborne levels of TCDD TEQs (Table 7).

While levels of these PCDFs were high in the livers of adult shortnose sturgeon from the HR, the levels in the young life-stages, which are most vulnerable to their damaging effects, are unknown. The link between hepatic burdens in subadult and adult sturgeons reported here, and the manifestation of toxicities to these PCDF congeners in offspring *in situ*, is likely to lie in the maternal transfer of congeners from female to ova to offspring. Even without direct measure of congener burdens in sturgeons eggs spawned *in situ*, such maternal transfer can be inferred from the reduced levels of hepatic burdens in females compared to males in another HR taxa, Atlantic tomcod (Courtenay et al. 1999) and in white sturgeon (Gundersen et al 2015).

The environmental prevalence of inadvertent PCBs is a growing concern. These are generated in the manufacture of dyes, pigments, and inks by processes that include chlorine and high temperatures (Vorkamp 2016; Heine and Tebilcock 2018). For example, PCB11 has been reported in high concentrations in the HR presumably due to effluents from the manufacturers of yellow paint pigments (Litten 2007). Elevated concentrations of PCB11 has raised concerns for its bioaccumulative properties and toxicity (Rodenburg et al. 2015). This has resulted in national and international regulations that allow inadvertent PCBs to be produced in pigments at maximum and mean concentrations of 50 and 25 ppm, respectively. Initial studies on PCB11 toxicity in zebrafish suggest it to be a partial agonist/antagonist of the AHR pathway that may modify the toxicity of co-occurring coplanar congeners (Roy et al. 2019), and that it is neurotoxic in rodents (Sethi et al. 2018). Hepatic PCB11 was detectable in both HR sturgeons and was 5 × higher in shortnose sturgeon than Atlantic sturgeon (means = 398 and 59.9 pg/g ww, respectively). While these concentrations are far lower than those eliciting AHR activity and inhibiting CYP1A expression in zebrafish, they are concentrations that promoted dendritic growth in rat neurons. Levels of PCB11 in HR sturgeons were significantly lower than those in adult tomcod from nine locales in the mainstem HR (mean 1256 pg/g ww) (Wirgin et al. unpublished data). Toxicities of PCB11 and other inadvertent PCBs remains a priority for resource managers of impacted systems.

Levels of hepatic total and coplanar PCBs in contemporary collections of HR sturgeons were significantly higher (~ 5 ×) in shortnose sturgeon than in Atlantic sturgeon. All of these levels of PCBs were lower than expected based on those reported in sympatric Atlantic tomcod in the HR (Fernandez et al 2004). Consistent with studies in HR tomcod (Courtenay et al. 1999; Fernandez et al. 2004), PCB77 was the predominant non-ortho substituted coplanar PCB congener, whereas levels of PCB126 and PCB169 were usually non-detectable. PCB126, the

congener with the highest TEF, was non-detectable in all but one archived shortnose sturgeon collected near Albany (rkm 229) which had a relatively high concentration (38.4 pg/g ww). In contrast, PCB 126 was detectable and at relatively high levels in juvenile tomcod from nine locales in the mainstem HR (mean: 1,395 pg/g ww; range: 593 to 2,282 pg/g ww) (Fernandez et al. 2004).

The significantly higher levels of hepatic coplanar PCBs in shortnose sturgeon than Atlantic sturgeon for all congeners except for those below detection levels (PCB126 and PCB169) may be due to three factors. First, sediment-borne PCBs levels are higher at more upstream locales in the HR (Farley and Thomann 1998; Farley et al 2006) where shortnose sturgeon reside compared to locales further downstream inhabited by juvenile and subadult Atlantic sturgeon. Second, shortnose sturgeon have a nearly twofold greater embryonic period duration than Atlantic sturgeon at a common temperature. The difference in duration is likely further exaggerated by the fact that shortnose sturgeon spawning earlier in the year at cooler temperatures and have a substantially greater eggs size. Lastly, for the sizes of sturgeon specimens used here, all but one shortnose sturgeon was likely to have been mature and these adult shortnose sturgeon would likely have been older than similarly sized, juvenile Atlantic sturgeon. Hence, these shortnose sturgeon would have been bioaccumulating PCBs for more years than the Atlantic sturgeon specimens.

The relatively low levels of PCBs in sturgeons from the HR contrasts with levels in the only other sympatric HR resident fish species, Atlantic tomcod, for which congener-specific hepatic PCB and PCDD/F data are available (Courtenay et al. 1999; Fernandez et al. 2004). For example, the mean total hepatic PCBs in pools of young-of-the-year (YOY) tomcod collected at nine different locales in the mainstem HR (rkm 0 to 82) in 1998 ranged from 7 to 34 ug/g ww (Fernandez et al. 2004 Supporting Information) compared to values in contemporary specimens of shortnose sturgeon (8.6 ug/g ww) and Atlantic sturgeon (1.48 ug/g ww). Non-ortho substituted coplanar PCBs in tomcod were also dominated by PCB77, but at ~ 30 to 90-fold higher levels than in HR sturgeons (mean level of hepatic PCB77 in pooled YOY tomcod from rkm 59 was 31,674 pg/g ww (Fernandez et al. 2004) compared to means of 349 pg/g ww and 1,032 pg/g ww of PCB77 in recent shortnose sturgeon and Atlantic sturgeon, respectively). PCB126 with the highest TEF was not detectable in any contemporary sturgeon but was detectable at high levels in YOY tomcod in the HR with hepatic concentrations ranging from 593 pg/g ww (rkm 82) to 2,282 pg/g ww (rkm 37).

Although these three species co-occur in the HR, their differences in life histories, temporal use of habitats, and diets may play a role in determining hepatic PCB burdens. Regarding life-history and habitat differences, all three taxa spawn demersal, adhesive eggs. The sturgeons spawn in upriver stretches (mostly upstream of rkm 125) in mid spring (shortnose sturgeon) to late spring-early-summer (Atlantic sturgeon). In contrast, tomcod spawn in early to mid-winter (December-January) and its eggs are retained in the mid-river span (rkm 50 to 90). The seasonal difference in spawning results in a two to four-fold longer embryonic period duration in tomcod (~ 1 month) than in shortnose sturgeon (2 weeks) or Atlantic (1 week) sturgeon. The extent of direct exposure of embryos to contaminated sediments likely scales proportionately with period duration. Sturgeons also differ dramatically from tomcod in lifespan and the proportion of the life spent in early life-stages. First spawning of shortnose sturgeon occurs from 3 to 4 yr (males) and 6 to 8 yr (females) and maximum lifespan is up to 37 yr for the HR population (Greeley 1937, Dadswell et al. 1984) where they remain as river residents throughout their lives. For HR Atlantic sturgeon, maturation occurs at ages of at least 12 and 15 yr (males and females, respectively, Bain (1997) and after up to a decade at sea (Everly and Boreman 1999) with each subsequent spawning season for an individual interspersed by 1 to 5 yr at sea. Maximum lifespan for HR Atlantic sturgeon is at least 30 yr (Gilbert 1989). In contrast, HR tomcod mature in 1 yr and few live longer than 2 yr (McClaren et al. 1988). In the context of life-history scaling, the embryonic period in Atlantic tomcod is a far greater proportion of maximum lifespan (~ 3 to 6%) than is the case for either sturgeon species (< 0.01 % for both sturgeons) by a factor of 300- to 600-fold. Compared to sturgeons, tomcod have a greatly reduced opportunity for contaminant depuration after hatching. Hence, tomcod is far more likely to be influenced by environmental (sediment) exposure experienced during the embryonic period and by maternal transfer of contaminants to offspring. Maternal transfer of PCBs and PCDD/Fs is substantial in fishes, including HR tomcod (Courtenay et al. 1999), but has not been evaluated in HR sturgeons.

Regarding diet differences among the species, sturgeons and tomcod are opportunistic generalist feeders with their diets reflecting the habitat differences of the same life-stages. The diets of early life-stages of both sturgeons are not well known (Dadswell et al. 1984) due to their protected status but some insights into the diets of YOY and yearling HR sturgeon have been obtained from *in situ* specimens using lavage methods (Haley 1998) and from specimens impinged at HR power plants (Carlson and Simpson 1987). The diets of impinged YOY shortnose sturgeon

collected from mid-HR locations above the salt front were dominated by freshwater taxa including midge larvae (Chironomidae), amphipods, and isopods. Molluscs and oligochaetes were uncommon in YOY shortnose diets despite an oligochaete (*Limnodrilus hoffmeisteri*) being a community dominant in freshwater habitats likely used by sturgeons (Simpson et al. 1985). The diets of tomcod in the HR consist almost exclusively of invertebrate prey including calanoid copepods, gammarids, *Neomysis* sp., polychaetes, and *Monoculodes* (Nittel 1976; Grabe 1978; 1980). More information is needed on the amount and type of contaminant in the diets of young sturgeons and tomcod.

There was no evidence of elevated hepatic PCDDs in either of the two HR sturgeons. For example, the mean level of hepatic 2,3,7,8 TCDD was 1.71 pg/g ww in contemporary collections of shortnose sturgeon and 0.63 pg/g ww in Atlantic sturgeon. Low levels of PCDDs, including TCDD, were expected in sturgeons from the mainstem HR because the predominant source of PCDDs for the lower HR Estuary was the Diamond Alkali facility, located adjacent to the lower Passaic River in the western reaches of the Estuary where neither sturgeon species is common (Wilk et al. 1997). Furthermore, 86% to 98% of total PCDDs in soils in the vicinity of Diamond Alkali was 2,3,7,8 TCDD (Umbreit et al. 1986; Wenning et al. 1993). The bioavailability of PCDDs to at least one member of the finfish community at the Diamond Alkali site was revealed by the near-record levels of hepatic PCDDs (mean 673 TCDD TEQs from PCDD), >80% of which was TCDD, in tomcod from Newark Bay and the adjacent Hackensack River (Courtenay et al. 1999; Fernandez et al. 2004). Evidence is lacking for transport of PCDDs from the Passaic River through to Newark Bay and into the Kill van Kull and the mainstem HR. Thus, low burdens of PCDDs in HR sturgeons is consistent with their life history and behavior.

Levels of some PCDFs, particularly those with potentially high TEFs in sturgeons, were greater than expected in shortnose sturgeon and in a single Atlantic sturgeon specimen. The mean level of 2,3,7,8 TCDF in contemporary shortnose sturgeon and Atlantic sturgeon was 26.18 pg/g ww and 11.13 pg/g ww, respectively. Additionally, the mean level of 2,3,4,7, 8 PeCDF was 6.26 pg/g ww and 2.29 pg/g ww in contemporary shortnose sturgeon and Atlantic sturgeon, respectively. The ratios of PCDDs to PCDFs in Atlantic tomcod were greater in tomcod from Newark Bay/Hackensack River than in specimens from the mainstem HR whereas hepatic burdens of PCDFs exceeded that of PCDDs in the HR tomcod (Fernandez et al. 2004). The same pattern in ratios of PCDDs to PCDFs was observed in sturgeons in this study. Specific sources of PCDFs

in the mainstem HR are unknown, but PCDFs have been shown to be a co-contaminant in PCB formulations (O'Keefe et al. 1984).

Conclusions

Despite their bottom-dwelling lifestyle, longevity, and occurrence in the tidal HR estuary where levels of sediment-borne PCBs are high, juvenile and adult HR sturgeons bioaccumulated relatively low levels of hepatic PCBs compared to a sympatric species. The low level of these contaminants in sturgeon may be due in part to their diet and life history pattern. Similarly, hepatic burdens of PCDDs in HR sturgeons were low but this is consistent with their absence from locales where sediment levels of PCDDs are high. In contrast, burdens of select PCDFs were unexpectedly high in HR sturgeons and at levels that activated AHR toxicity in another North American sturgeon species. We suggest that laboratory studies be conducted to evaluate the concentrations of PCDFs that result in toxicity in young life-stages of HR sturgeons.

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Figure Legend

Fig. 1

Left Panel-Map of the western Hudson River Estuary including the 17 mile long Diamond Alkali TCDD Superfund site in Newark on the Passaic River and Newark Bay. Right Panel-Map of the 198 mile long Hudson River PCBs Superfund site on the mainstem Hudson River. Locations of sturgeon collection sites on the mainstem river are indicated by river kilometers (Rkm).

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Figure 1

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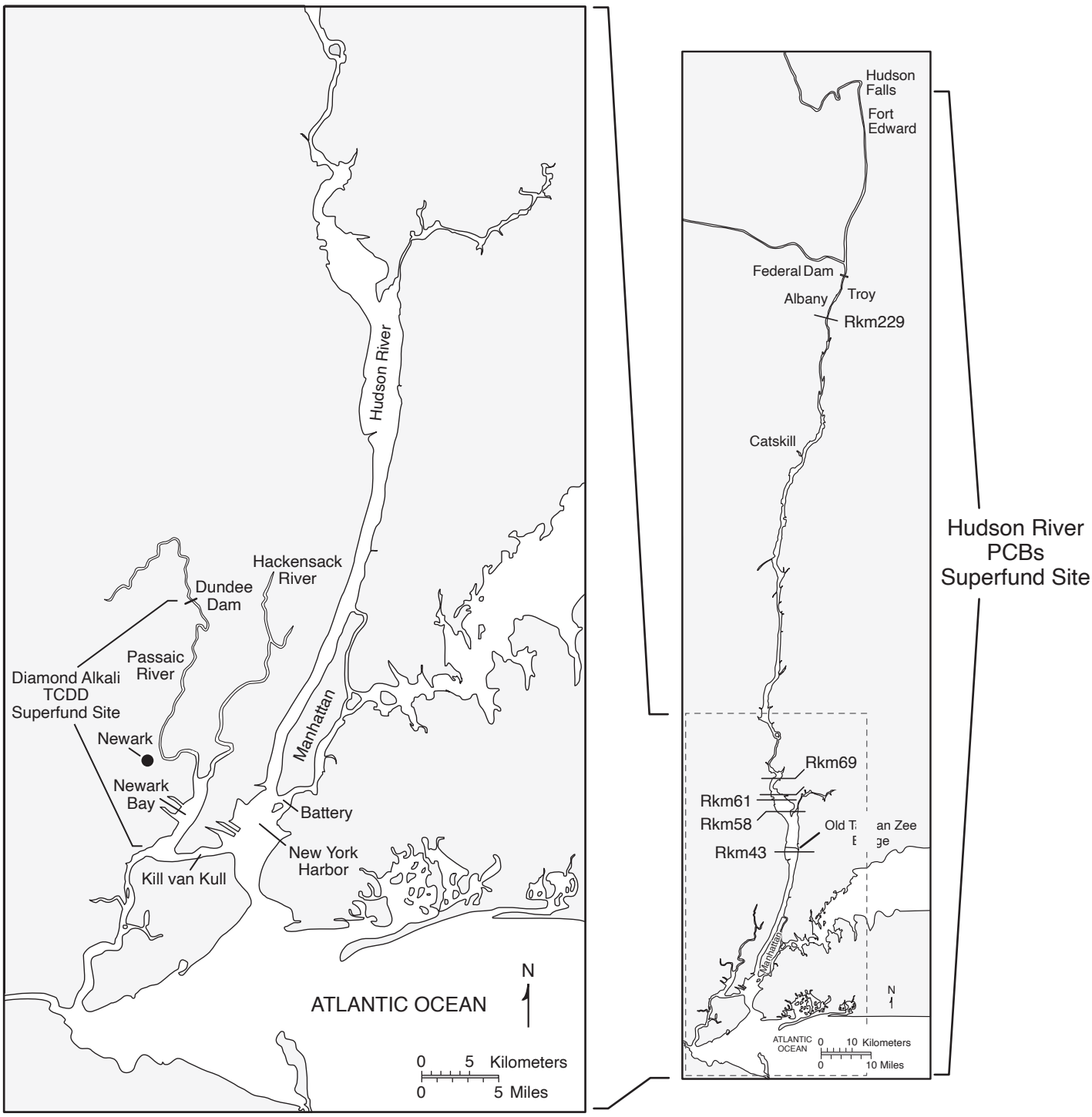

Table 1. Collection details of Hudson River shortnose sturgeon and Atlantic sturgeon used in this study to quantify hepatic PCB and PCDD/F burdens. All fish collected in the 2010's were either live with liver samples taken by biopsy or recent fatalities. Fish from 1984 were museum specimens that had been fixed in formalin and later preserved in ethanol. All locations are in New York. Abbreviations: SS, shortnose sturgeon; AS, Atlantic sturgeon; Rkm, river kilometer (km upstream from the Battery, southern Manhattan, New York); TL, Total Length; WT, total body weight.

Species	Collection			TL (cm)	WT (g)	Specimen status
	Location	Rkm	Date			
SS	Nyack	43	8/13/2015	80.0	3241	fatality
SS	Haverstraw	63	8/28/2016	ND	ND	fatality
SS	Haverstraw	63	9/16/2014	65.5	1070	live
SS	Haverstraw	63	9/16/2014	79.1	3750	live
SS	Haverstraw	63	9/16/2014	88.3	4700	live
SS	Indian Point	69	5/21/1984	46.6	598	preserved
SS	Indian Point	69	3/22/1984	55.6	884	preserved
SS	Albany	229	6/28/1984	33.3	ND	preserved
AS	Nyack	43	8/31/2015	42.7	310	fatality
AS	Haverstraw	58	4/18/2016	74.7	2400	live
AS	Haverstraw	61	4/19/2016	73.4	2700	live
AS	Haverstraw	63	4/18/2016	76.5	2300	live
AS	Haverstraw	63	4/18/2016	63.3	900	live

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Table 2. Descriptive statistics, between-group tests, and trends of differences for 11 dioxin-like PCB congeners, 3 inadvertent PCB congeners, and total PCBs (pg/g-ww) in livers of shortnose sturgeon and Atlantic sturgeon collected from the Hudson River. Type II error is shown for all tests with $p \leq 0.1$. The number of tests that was significant ($p \leq 0.05$) and in the direction specified in the Summary heading (bottom of table), out of all tests conducted on individual congeners and total (all) PCB congeners, also includes observations below levels of detectability. 'Trend' is the number of cases for which the means are in the direction stated in the Summary heading compared to the total number of tests conducted. Trends are also reflected in the overall magnitude of differences between groups (e.g., archived vs. recent, or shortnose sturgeon vs. Atlantic sturgeon) as quantified by the ratio of the grand means (congeners and total PCB) between contrasted groups (N = 2 a minimum for inclusion). Abbreviations: *nd*, non-detectible; *NS*, not significant ($p > 0.05$); *na*, not available.

	Shortnose sturgeon (recent)			Shortnose sturgeon (archive)			Atlantic sturgeon			Shortnose sturgeon (archive vs. recent)			Shortnose sturgeon vs. Atlantic sturgeon		
	Mean	SEM	N (*nd*)	Mean	SEM	N (*nd*)	Mean	SEM	N (*nd*)	F	df	p	F	df	p
Coplanar PCBs															
Non-Ortho Substituted															
77	1,032	123	5	969	534	3	349	274	5	0.02	1,6	NS	5.17	1,8	0.05
81	71.7	9.48	5	210	147	3	8.7	8.74	5 (4)	1.64	1,6	NS	23.81	1,8	< 0.01
126	0	0	5 (5)	12.8	12.80	3 (2)	0	0	5 (5)	1.88	1,6	NS	-	-	-
169	0	0	5 (5)	0	0	3 (3)	0	0	5 (5)	-	-	-	-	-	-
Mono-Ortho Substituted															
105	50,180	7,566	5	10,600	.	1	8,076	2,247	5	4.56	1,4	NS	28.46	1,8	< 0.01
114	4,612	767	5	16,143	9,889	3	635	141	5	2.45	1,6	NS	26.02	1,8	< 0.01
118	96,725	10,389	4	33,100	.	1	18,630	9,809	5	7.50	1,3	NS	29.47	1,7	< 0.01
123	1,028	205	5	7,279	4,493	3	265	151	5	3.60	1,6	NS	8.961	1,8	0.02
156/7	37,760	6,941	5	48,097	27,378	3	6,218	1,592	5	0.22	1,6	NS	19.62	1,8	< 0.01
167	1,294	170	5	1,932	969	3	436	327	5	0.74	1,6	NS	5.42	1,8	0.05
189	3,732	644	5	3,283	1,606	3	714	128	5	0.10	1,6	NS	21.11	1,8	< 0.01
Inadvertent PCBs															
5	3.9	2.53	5 (3)	48.3	32.74	3	0.47	0.47	5 (4)	3.39	1,6	NS	1.75	1,8	NS
11	398	128	5	90.5	32.04	3	59.9	25.49	5	3.18	1,6	NS	6.71	1,8	0.03
52	346,250	23,558	4 (0)	85,100	na	1	64,300	31,326	5	24.52	1,3	0.02	47.01	1,7	< 0.01
Total PCBs	8.60 $x10^6$	1.44 $x10^6$	5	12.17 $x10^6$	7.88 $x10^6$	3	1.48 $x10^6$	1.26 $x10^6$	5	0.35	1,6	NS	22.19	1,8	< 0.01
									Summary:	archive > recent			shortnose > Atlantic		

Significance test:	0 of 13	12 of 13
Trend:	7 of 11	12 of 12
Ratio of grand means:	3.2	5.5

Table 3. Descriptive statistics, between-group tests, and trends of differences in 7 PCCD congeners (pg/g-ww) in livers of shortnose sturgeon and Atlantic sturgeon collected from the Hudson River. Type II error shown for all tests with $p \leq 0.1$. See Table 2 for interpretation of summary on significance tests, trends, and ratios of grand means. Abbreviations: *nd*, non-detectible; *NS*, not significant ($p > 0.05$).

	Shortnose sturgeon (recent)			Shortnose sturgeon (archive)			Atlantic sturgeon			Shortnose sturgeon (recent vs. archive)			Shortnose sturgeon vs. Atlantic sturgeon		
PCCD Congener	Mean	SEM	N (*nd*)	Mean	SEM	N (*nd*)	Mean	SEM	N (*nd*)	F	df	p	F	df	p
2,3,7,8 TCDD	1.7	0.455	5 (1)	0.99	0.521	3	0.63	0.529	5 (3)	1.0	1,6	NS	2.4	1,8	NS
1,2,3,7,8 PenTCDD	0.30	0.184	5 (3)	0.50	0.054	3	0.31	0.176	5 (2)	0.65	1,6	NS	0.01	1,8	NS
1,2,3,4,7,8 HexaCDD	0.22	0.137	5 (3)	1.46	1.302	3 (1)	0.92	0.434	5 (1)	1.7	1,6	NS	2.4	1,8	NS
1,2,3,6,7,8 HexaCDD	1.8	0.528	5 (1)	1.1	0.582	3	2.5	1.434	5 (1)	0.75	1,6	NS	0.22	1,8	NS
1,2,3,7,8,9 HexaCDD	0.32	0.194	5 (3)	0.62	0.154	3	0.92	0.608	5 (2)	1.2	1,6	NS	0.91	1,8	NS
1,2,3,4,6,7,8 HeapTACDD	4.9	0.41	5	14.2	4.00	3	7.4	1.17	5	9.8	1,6	0.02	4.1	1,8	0.08
OctaCDD	9.3	1.70	5	178	111	3	8.2	1.68	5	4.3	1,6	0.08	0.20	1,8	NS
									Summary:	archive > recent			shortnose > Atlantic		
									Significance test:	1 of 7			0 of 7		
									Trend:	5 of 7			2 of 7		
									Ratio of grand means:	4.8			0.9		

Table 4. Descriptive statistics, between-group tests, and trends of differences for 10 PCDF congeners (pg/g-ww) in livers of shortnose sturgeon and Atlantic sturgeon collected from the Hudson River. Type II error shown for all tests with $p \leq 0.1$. See Table 2 for interpretation of summary on significance tests, trends, and ratios of grand means. Abbreviations: *nd*, non-detectible; *NS*, not significant ($p > 0.05$).

	Shortnose sturgeon (recent)			Shortnose sturgeon (archive)			Atlantic sturgeon			Shortnose sturgeon (recent vs. archive)			Shortnose sturgeon vs. Atlantic sturgeon		
PCCD Congener	Mean	SEM	N (*nd*)	Mean	SEM	N (*nd*)	Mean	SEM	N (*nd*)	F	df	p	F	df	p
2,3,7,8 TetraCDF	26.2	4.63	5	20.0	11.54	3	11.1	4.18	5	0.35	1,6	NS	5.8	1,8	0.04
1,2,3,7,8 PentaCDF	1.2	0.42	5 (1)	1.7	0.89	3	1.4	0.81	5 (1)	0.38	1,6	NS	0.06	1,8	NS
2,3,4,7,8 PentaCDF	6.3	0.85	5	4.9	2.66	3	2.3	0.80	5	0.39	1,6	NS	11.6	1,8	< 0.01
1,2,3,4,7,8 HexaCDF	0.25	0.161	5 (3)	0.79	0.300	3	0.94	0.444	5 (1)	3.1	1,6	NS	2.2	1,8	NS
1,2,3,6,7,8 HexaCDF	1.1	0.53	5 (2)	0.48	0.282	3 (1)	1.0	0.55	5 (1)	0.74	1,6	NS	0.01	1,8	NS
1,2,3,7,8,9 HexaCDF	0.17	0.168	5 (4)	0	0	3 (3)	0	0	5 (5)	0.56	1,6	NS	1.0	1,8	NS
2,3,4,6,7,8 HexaCDF	0.18	0.110	5 (3)	0.25	0.142	3 (1)	0.30	0.192	5 (2)	0.16	1,6	NS	0.28	1,8	NS
1,2,3,4,6,7,8 HepaCDF	0.96	0.258	5 (1)	10.5	8.96	3	2.3	1.21	5 (1)	2.1	1,6	NS	1.2	1,8	NS
1,2,3,4,7,8,9 HepaCDF	0.05	0.047	5 (4)	0.49	0.215	3	0.08	0.080	5 (4)	7.0	1,6	0.04	0.13	1,8	NS
OCDF	0.26	0.260	5 (4)	68.5	66.23	3	0.41	0.223	5 (1)	2.0	1,6	NS	0.20	1,8	NS
									Summary:	archive > recent			shortnose > Atlantic		
									Significance test:	1 of 10			2 of 10		
									Trend:	6 of 10			4 of 10		
									Ratio of grand means:	2.9			1.5		

Table 5. Summary statistics and between-group tests and trends of differences for levels of hepatic PCB Toxic Equivalency Quotients (TEQs), PCDD/Fs TEQs, and total TCDD TEQs in livers of shortnose sturgeon and Atlantic sturgeon collected from the Hudson River. TEFs values used are those for fishes from Van den Berg et al. (1998). Type II error shown for all tests with $p \leq 0.1$. Abbreviations: *NS*, not significant; $p > 0.05$.

	Shortnose sturgeon (recent)			Shortnose sturgeon (archive)			Atlantic sturgeon			Shortnose sturgeon (recent vs. archive)			Shortnose sturgeon vs. Atlantic sturgeon		
TEQ	Mean	SEM	N	Mean	SEM	N	Mean	SEM	N	F	df	p	F	df	p
PCBs	1.6	0.27	5	4.0	2.36	3	0.3	0.108	5	2.0	1,6	NS	18.9	1,8	< 0.01
PCDD/Fs	6.6	0.95	5	5.0	2.29	3	3.5	1.12	5	0.60	1,6	NS	4.5	1,8	0.07
Total TEQs	8.2	1.19	5	9.0	4.64	3	3.8	1.21	5	0.05	1,6	NS	6.6	1,8	0.03

Table 6. Summary statistics and between-group tests and trends of differences for TCDD Toxic Equivalency Quotients (TEQs) of 2,3,7,8 TCDF, 2,3,4,7,8 PeCDF and their sum in shortnose sturgeon and Atlantic sturgeon livers collected from the Hudson River. TEQs are based on RePs developed by Eisner et al. (2016) in white sturgeon livers for 2,3,7,8 TCDF (1.4) and 2,3,4,7,8 PeCDF (1.2). Type II error shown for all tests with $p \leq 0.1$. Abbreviations: *nd*, non-detectible; *NS*, not significant, $p > 0.05$.

	Shortnose sturgeon (recent)			Shortnose sturgeon (archive)			Atlantic sturgeon			Shortnose sturgeon (recent vs. archive)			Shortnose sturgeon vs. Atlantic sturgeon		
TEQ	Mean	SEM	N (*nd*)	Mean	SEM	N (*nd*)	Mean	SEM	N (*nd*)	F	df	p	F	df	p
A) 2,3,7,8 TCDF	36.65	4.63	5	27.85	11.54	3	15.58	4.18	5	0.35	1,6	NS	5.8	1,8	0.04
B) 2,3,4,7,8 PeCDF	7.51	0.85	5	5.83	2.66	3	2.75	0.80	5	0.39	1,6	NS	11.6	1,8	< 0.01
Total (A + B)	44.16	5.45	5	33.68	14.20	3	18.33	4.70	5	0.36	1,6	NS	7.0	1,8	0.03

Table 7. Minimal nominal waterborne concentrations (ppb) of PCB126 and 2,3,7,8 TCDD that induced significant early-life toxicities (Chambers et al. 2012) and induction of CYP1A mRNA expression (Roy et al. 2011) in larval shortnose sturgeon and Atlantic sturgeon exposed as embryos under controlled laboratory conditions. Nominal waterborne doses of PCB126 ranged from 0.01 to 1,000 ppb and 2,3,7,8 TCDD from 0.001 to 100 ppb. TEFs were calculated as the ratio between concentrations of 2,3,7,8 TCDD and PCB126 eliciting an effect in each of the measured responses. Abbreviations: *NE*, no effect; *NA*. not applicable.

Early life-stage toxicities	Shortnose sturgeon			Atlantic sturgeon		
	PCB 126	2,3,7,8 TCDD	TEF	PCB 126	2,3,7,8 TCDD	TEF
Embryo survival	$\geq$ 10	$\geq$ 1	0.1	$\geq$ 100	$\geq$ 1	0.01
Embryo period duration	NE	NE	NA	NE	NE	NA
Length (larvae)	$\geq$ 1	$\geq$ 0.01	0.01	$\geq$ 0.1	$\geq$ 0.01	0.1
Development (eye)	$\geq$ 0.01	$\geq$ 0.001	0.1	$\geq$ 1	$\geq$ 0.1	0.1
Larval lifespan (unfed)	$\geq$ 0.1	$\geq$ 0.01	0.1	$\geq$ 0.1	$\geq$ 0.01	0.1
CYP1A mRNA Induction	$\geq$ 0.01	$\geq$ 0.001	0.1	$\geq$ 0.01	$\geq$ 0.001	0.1

Sample characterization table

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PCB data

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