Dioxin-like toxicity of polychlorinated biphenyls (PCBs) in mink and otter collected from the Hudson River drainage, 1998-2002.

Introduction

Dioxin-like toxicity of polychlorinated biphenyls (PCBs), thought to be responsible for reduction in litter size and the "wasting syndrome" that reduces the kit survival of breeding mink experimentally exposed to PCBs, is associated with a relatively small number of chlorinated biphenyl congeners. Currently, twelve congeners (nonortho-chloro substituted [coplanar] and mono-ortho-chloro substituted biphenyls) are believed to exhibit significant dioxin-like toxicity and have been assigned toxicity equivalency factors (TEFs) by the World Health Organization (2005). These dioxin-like congeners are generally low in concentration compared to many congeners comprising PCBs in the environment. However, congener composition can vary widely, and, so too, the toxigenic potential of PCBs. Consequently, the potential impact of environmentally available PCBs on mink and otter is related to the concentration of total PCBs (TPCBs) accumulated in tissues and their concomitant toxigenic potential (expressed as 2, 3, 7, 8 tetra-chlorinated dibenzo-*p*-dioxin-equivalency quotients [TCDD-EQs]).

Mink (*Mustela vison*) and river otter (*Lontra canadensis*) were collected from the Hudson River drainage (and from some adjacent drainages) during the 1998-2002 trapping seasons. Animals were evaluated by necropsy and livers from selected animals were analyzed for PCBs as Aroclors and percent lipids. Samples $\geq$ 9 µg TPCBs/g lipid as Aroclors were analyzed subsequently for specific congeners by Environmental Protection Agency Method 1668a. Age and gender were determined for all animals. Contents of

digestive tracts of mink and otter (including additional animals collected state-wide) were analyzed for dietary composition.

This analysis evaluates the relative toxigenic potential of hepatic PCBs of mink and otter from the Hudson River drainage. Mink and otter within one home range below the major point-source discharge of PCBs on the main-stem Hudson River are compared with ranch mink experimentally exposed to PCB-contaminated carp (*Cyprinus carpio*) from the Hudson River. The toxigenic potential of hepatic PCBs is also compared for mink and otter greater than one home range or above the major point-source discharge of PCBs. Implications are discussed for these species with regard to sources of exposure to dioxin-like congeners.

Methods

Levels of TPCBs in hepatic samples from mink and otter were categorized by location and level of contamination. Samples were categorized according to capture location: 1) within one home range of the main-stem Hudson River below the point of contaminant discharge or 2) greater than one home range of the main-stem Hudson River (including animals collected from adjacent watersheds) or above the point of contaminant discharge. Distance along the course from the capture point on the tributary (or tributaries with headwaters in close proximity for animals captured in adjacent watersheds) to the main-stem river was used to determine home range (0-6 km and 0-30 km for home range of mink and otter, respectively). Within each location category, samples were categorized as "baseline" if TPCB level was < 9 μg/g lipid as Aroclors or as the sum of congeners; or as "elevated" if TPCB level was ≥ 9 μg/g lipid (congener sum). (Animals subsequently analyzed by congener-specific methods were re-classified as "baseline" if

the congener sum was < 9 µg TPCBs/g lipid.) Total PCB level as Aroclors typically exceeds that determined by congener sum within the same sample by approximately 20%; consequently, all samples categorized as "baseline" had a TPCB level as a congener sum < 9 µg/g lipid.

Tetra-chlorinated (2, 3, 7, 8) dibenzo-*p*-dioxin-equivalency quotients were calculated (TEFs: Word Health Organization 2005) for samples analyzed by congener-specific methods and were evaluated relative to TPCB concentration (congener sum) by regression. Linear regressions were performed for mink and otter within specific location-contaminant level categories. For comparison, similar regressions were performed for hepatic PCBs in ranch mink experimentally fed five exposure levels of PCB-contaminated carp from the Hudson River (Exhibits 6 and 8, TWG Meeting, June 16, 2008).

Results

For animals within one home range of the main-stem Hudson River below the point of contaminant discharge, all animals (with the exception of one mink without a useable liver sample and five mink collected late in the collection period) were analyzed for TPCBs (Table 1). Approximately half the mink and otter within one home range of the contaminated main-stem Hudson River were classified as "baseline" (< 9 µg TPCBs/g lipid) and half as "elevated" (≥ 9 µg TPCBs/g lipid).

Most of the 115 mink and 58 otter that had been selected for analysis of TPCBs from animals greater than one home range from the main-stem Hudson River or above the point of contaminant discharge were classified as "baseline" (Table 2). Only six mink and seven otter were classified as "elevated". Of the "elevated" animals, four mink and

six otter originated outside the Hudson River drainage. Two mink (with marginally elevated levels of 10.8 µg/g lipid, congener sum, and 13.1µg/g lipid, Aroclors) and one otter (with marginally elevated level of 9.8 µg/g lipid, Aroclors) originated within the Hudson River drainage.

Gender-specific regressions for mink and otter within one home range of the main-stem Hudson River below the point of contaminant discharge and classified as "elevated" were compared with regressions for ranch mink experimentally exposed to PCB-contaminated carp from the Hudson River (Fig.1). The TCDD-EQ was considerably greater for field mink than predicted for ranch mink relative to TPCB concentration. No substantial gender difference in the TCDD-EQ-TPCB relationship was evident for ranch mink or otter. However, an evaluation of a gender effect could not be made for field mink due to the low number of females in the sample. The TCDD-EQ for otter was less than predicted for ranch mink relative to TPCB concentration and, likewise, considerably less than for field mink. For both species, males tended to have greater maximum levels of TPCBs and TCDD-EQs than females.

Regressions for mink and otter greater than one home range from main-stem Hudson River or above the point of contaminant discharge and classified as "baseline" were conducted by gender or by ecozone of origin (Fig. 2). The TCDD-EQ for field mink was substantially greater than for otter relative to TPCB concentration. No substantial gender effect was evident for either species. Although the distribution for TPCB concentration varied noticeably with ecozone for both species, the TCDD-EQ-TPCB relationship seemed consistent among ecozones, with TCDD-EQ substantially greater for mink than otter relative to TPCB concentration.

Discussion

Animals with an “elevated” level of TPCBs are largely restricted to within one home range of the main-stem Hudson River below the major point-source discharge of PCBs. However, approximately half the animals from this region were at a “baseline” level of PCB contamination suggesting not all individuals are exposed to high-level contamination likely associated with the major point source. Animals may have migrated from more-distant, less-contaminated regions of the watershed and entered the population with a relatively low contaminant level immediately before sampling. Animals classified as “elevated” were rare beyond one home range of the contaminated main-stem Hudson River or above the point of PCB discharge and were only marginally elevated in PCB contamination if they originated within the Hudson River drainage. Animals classified as “elevated” from adjacent watersheds had substantial contamination, likely due to exposure to PCBs from point sources within those watersheds.

Polychlorinated biphenyls have a substantially greater TCDD-EQ in field mink than otter relative to TPCB concentration. This difference is particularly evident for animals with elevated levels of contamination potentially due to contaminant exposure from the major point-source discharge to the Hudson River but is not limited to animals with high-level contamination. This difference is also evident for “baseline” animals with low-level contamination likely due to exposure to PCBs distributed watershed-wide by aerial deposition. The consistent difference suggests that level and source of contamination are not factors likely to affect the inter-species relationship. Mink, as compared with otter, either may be exposed to dietary sources of PCBs having relatively high TEFs or may preferentially retain high-TEF PCBs physiologically. If the inter-

species difference in TCCD-EQ is due to a physiological difference independent of dietary source one would expect that, when the exposure of mink to contaminants through the diet is altered to be more consistent with that of otter (primarily fish), the inter-species difference would remain unaffected. However, TCDD-EQ for mink experimentally fed carp from the Hudson River was much lower than for field mink relative to TPCB concentration and more consistent with otter suggesting that a potential physiological difference is not likely to account for the difference between mink and otter. Thus, a difference in diet is suspected of contributing to the inter-species difference in the TCDD-EQ-TPCB relationship.

In addition to fish, mammals, amphibians and crayfish contribute to the diet of field mink. For mink collected from New York State, 1998-2002, 41.4%, 26.9%, 26.5%, and 10.8% contained remains of fish, mammals, amphibians and crayfish, respectively, suggesting that mammals and amphibians are important components of the field mink diet. Herbivorous mammals comprised the bulk of mammal species recovered from mink whereas predatory mammals, such as shrews and moles, were recovered infrequently (1.2% of mink contained shrews; moles were not recovered) from mink. Frogs comprise most of the amphibian component of the diet. Of the mink examined, 1.6%, 11.6%, 5.2% and 8.0% contained bullfrogs (*Rana catesbiana*), green frogs (*Rana clamitans*), leopard frogs (*Rana pipiens*) and unidentified frogs in their dietary remains, respectively; whereas, less than 1% contained salamanders. Frogs also comprised a higher percent volume of the dietary remains than many other prey items suggesting that frogs contribute a greater biomass per prey item to the diet.

Floodplain- and wetland-associated PCBs may have a higher TCDD-EQ relative to TPCB concentration than PCBs associated with the aquatic system due to a greater affinity of high-TEF congeners for soil carbon and the loss of other congeners to volatilization and "weathering" in floodplain and wetland environments. Frogs breed and develop as tadpoles in these habitats during periods of flooding. Aquatic exposure to soil-retained and differentiated PCBs afford frogs, especially longer-lived green frogs, an opportunity to bioconcentrate these PCBs. In contrast, herbivorous mammals have limited uptake of PCBs from soils (e.g. muskrats from the Hudson River drainage). Furthermore, predatory mammals, such as shrews and moles, although expected to have a substantial PCB uptake from the terrestrial food web, are relatively rare in the dietary remains of mink. Their sporadic inclusion in the diet would likely produce a variable TCDD-EQ-TPCB relationship. The TCDD-EQ-TPCB relationship (R^2=82%) for male field mink (Fig. 1), in contrast, is more consistent with uptake of PCBs from a staple dietary component with a consistent congener composition. Frogs, given their high frequency and volume in the mink diet and potential uptake of soil-associated PCBs, are a possible source of high-TEF congeners that increase the relative TCDD-EQ of PCBs in field mink.

Although the TCDD-EQ for most otter was lower than predicted for ranch mink relative to TPCB concentration, three male otter exhibited a TCDD-EQ that was greater (Fig. 1). These otter were taken (two otter on the same date) by the same trapper near the mouth of the Moses Kill in an area having floodplain and wetland habitat that may have provided an opportunity for these otter to feed on frogs. In contrast, several otter, captured near the Hudson River but in areas with little wetland or floodplain habitat

(male otter, above 2000 TPCBs ng/g, with the lowest and highest TPCBs), had a TCDD-EQ below that predicted for ranch mink relative to TPCB concentration. Other otters with a low TCDD-EQ relative to TPCB concentration were from small stream and pond systems within the eastern portion of the Hudson River drainage, including several otter captured just beyond one home range from the main-stem Hudson River and potentially contaminated with PCBs from a landfill draining to the Valatie Kill. Consequently, the variation in TCDD-EQ-TPCB relationship observed for otter may be associated with habitat variation that may influence the contamination of prey with dioxin-like congeners and the subsequent availability of prey to otters.

The generally lower TCDD-EQ for otter than predicted for ranch mink relative to TPCB concentration suggests that otter have less exposure to high-TEF congeners than ranch mink fed PCB-contaminated carp from the Hudson River. Although otter feed primarily on fish and, thus, their source of contaminant exposure is generally similar to that of the carp experimentally fed to ranch mink, the fish that comprise the diet of otter are smaller and generally younger than mature carp. Small fish may biconcentrate high-TEF congeners to a lesser extent than carp due age, size and physiological differences or may be less exposed to high-TEF congeners due to habitat difference. Carp feed and spawn in shallow, often temporarily flooded, embayments; whereas, most prey fish of otter are from deeper, more rapidly flowing reaches. Embayments have high sediment deposition and, as for soils, high-TEF congeners may have a high affinity for embayment sediments or previously contaminated soils may erode to form these sediments. Either directly contaminated sediments or sediments indirectly contaminated by eroding soils may increase the exposure of high-TEF congeners to biota, such as carp, that ingest or are

in contact with sediments. Consequently, potential exposure of carp to sediment-associated, high-TEF congeners may be responsible for relatively greater TCDD-EQ in experimental ranch mink than otter relative to TPCB concentration.

The toxigenic quality of PCBs accumulated by mink is possibly influenced by dietary source. Many of the toxic, high-TEF congeners are sequestered on floodplains or in wetlands and are likely transferred to mink through the consumption of frogs that breed and develop in these areas during periods of flooding. Consequently, floodplains and riparian wetlands may be important sources of PCBs that are toxicologically relevant to field mink. Further work on the composition of PCBs retained in floodplains and wetlands and subsequently accumulated by the potential prey of mink is needed to substantiate inferences made in this analysis.

Table 1. Contaminant level categories for hepatic samples of mink and otter analyzed for total polychlorinated biphenyl concentrations (TPCBs). Animals captured within one home range of the main-stem Hudson River below the point of contaminant discharge.

Contaminant level and analysis	Mink (0-6 km)	Otter (0-30 km)
Baseline (< 9 TPCBs μg/g lipid)	15	9
Aroclor only	14	3
Aroclor + Congener	1	6
Elevated (≥ 9 TPCBs μg/g lipid)	12	22[#]
Aroclor only	1*	0
Aroclor + Congener	11	22
Not Analyzed	5	0
Not Available	1	0
Total Sample	33	31

* Small sample size precluded additional congener analysis

[#] Included were two samples marginally greater than 30 km from main-stem Hudson River (35 and 37 km) but substantially elevated in TPCBs (6549 and 2741 ng/g).

Table 2. Contaminant level categories for hepatic samples of mink and otter analyzed for total polychlorinated biphenyl concentrations (TPCBs). Animals captured greater than one home range of the main-stem Hudson River (including animals collected from adjacent watersheds) or above the point of contaminant discharge.

Contaminant level and analysis	Mink (> 6 km)	Otter (> 30 km)
Baseline (< 9 TPCBs µg/g lipid)	109	51
Aroclor only	79	32
Aroclor + Congener	30	19
Elevated (≥ 9 TPCBs µg/g lipid)	6	7[#]
Aroclor only	1*	1
Aroclor + Congener	5	6
Total Sample	115	58

* Small sample size precluded additional congener analysis

[#] Not included were two samples marginally greater than 30 km from main-stem Hudson River (35 and 37 km) but substantially elevated in TPCBs (6549 and 2741 ng/g).

Figure 1. Mink and otter categorized as "elevated" (≥ 9 µg TPCBs/g lipid) and captured within a home range of the contaminated main-stem Hudson River and ranch mink experimentally fed PCB-contaminated carp from the Hudson River

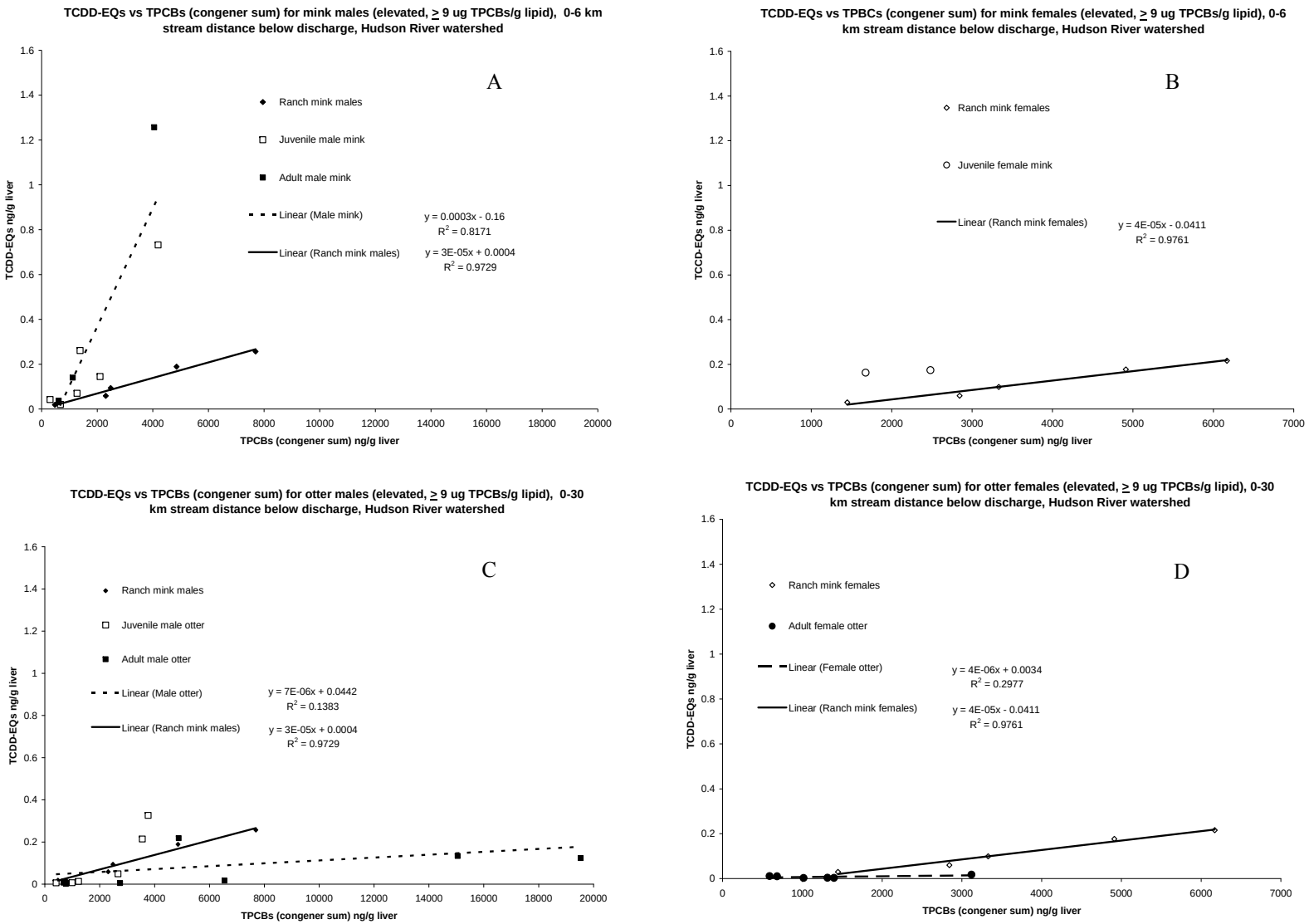
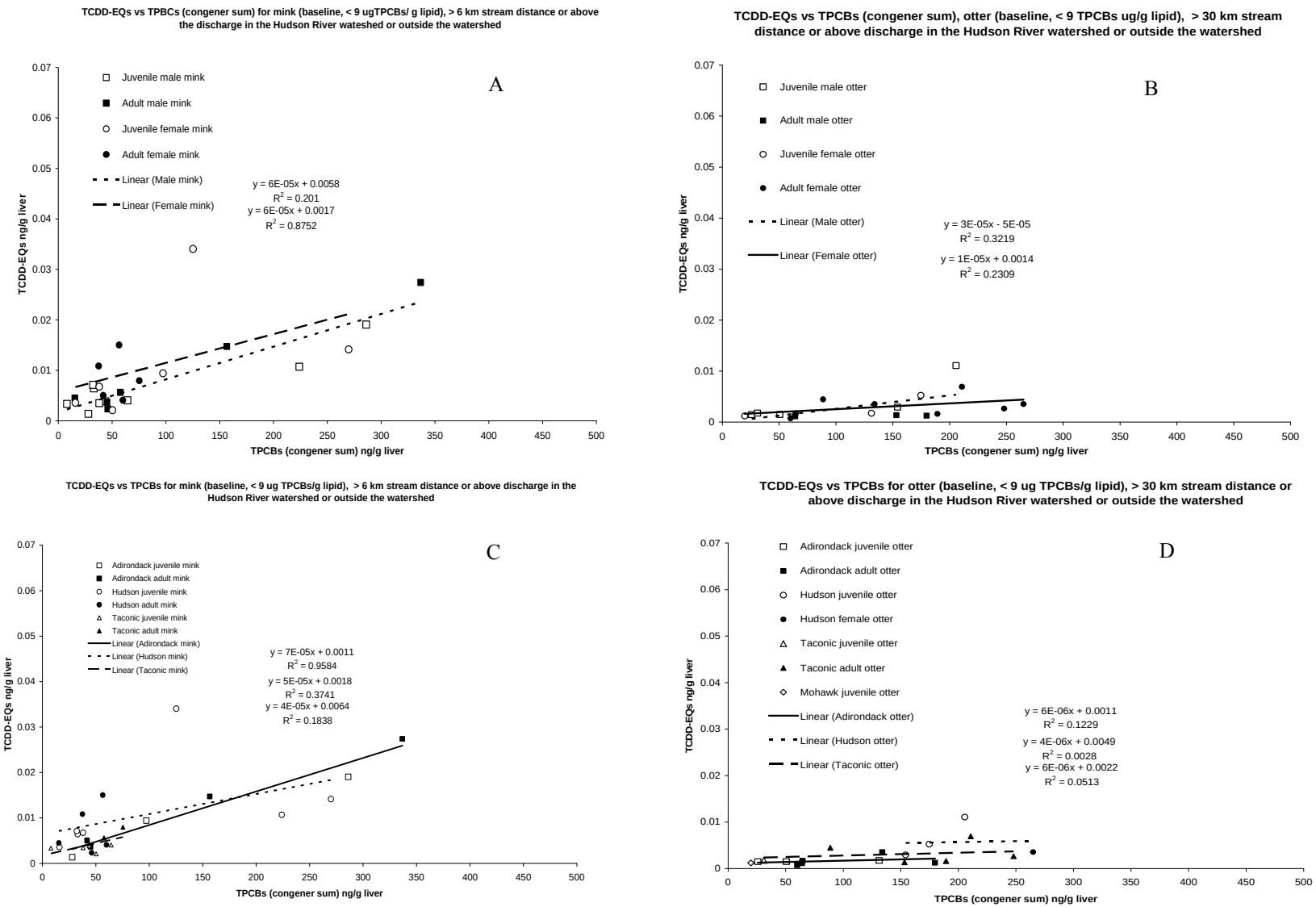
Figure 2. Mink and Otter categorized as "baseline" (< 9 µg TPCBs/g lipid), and captured greater than a home range from the contaminated main-stem Hudson River or above the point of contaminant discharge. Regressions by gender (A, B) and by ecozone (C, D).