BIOMAGNIFICATION FACTORS OF PCDD/DFS FOR GREAT BLUE HERON AND BELTED KINGFISHER RESIDING IN THE TITTABAWASSEE RIVER FLOODPLAIN, MI, USA

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Introduction

The great blue heron (*Ardea herodias*) (GBH) and belted kingfisher (*Ceryle alcyon*) (KF) were selected as species of interest in an ecological risk assessment being performed on the Tittabawassee River, Michigan, USA. The trophic status of both the GBH and KF, along with their strong site fidelity and territoriality, make them ideal species to investigate bioaccumulative contaminants. The study area, which includes 38 km of river from the upstream boundary at the city of Midland, MI to the confluence of the Tittabawassee and Shiawassee Rivers, has previously been shown to contain elevated concentrations of polychlorinated dibenzofurans (PCDFs) and polychlorinated dibenzo-*p*-dioxins (PCDDs) in soils, sediments, and biota in comparison to reference areas upstream of Midland[1,2]. The site-specific mixture of dioxin-like compounds in the study area is predominately furan congeners, specifically 2,3,7,8 TCDF (TCDF) and 2,3,4,7,8 PeCDF (4-PeCDF)[1,2]. To understand the movement of this site-specific mixture of furans and dioxins up the food chain, comparisons of the biomagnification factors (BMFs) of different congeners were made within a species and also between species. Differences in congener-specific BMFs may suggest a difference in the half-lives between various congeners.

Materials and methods

Prey items, which included forage fish, crayfish, and frogs, were collected from six predetermined biological sampling areas (BSAs), four located in the study area and two in upstream reference areas. Selection of prey items was based on site-specific observations and literature-based dietary compositions of GBH and KF. Fish and crayfish were collected in September 2003 and again in May and June of 2004. Electrofishing and seining were used to collect forage fish, which was a targeted class size of <25cm and thus contains multiple species. Crayfish were collected by seining and modified minnow traps. Three species of frogs, green (*Rana clamitans*), northern leopard (*Rana pipiens*), and wood frogs (*Rana sylvatica*), were collected by hand and dip nets in June 2005. Forage fish and crayfish were analyzed as composites from each BSA. Frogs were analyzed individually, with the exception of some wood frogs within a BSA being combined to obtain necessary sample mass.

GBH nestling tissues, including liver, adipose, and blood plasma were collected from GBH rookeries in the study area. Liver and adipose were harvested from nestlings which were opportunistically collected after nestlings fell from nests as a result of weather events or siblicide. Plasma was collected by accessing nests when nestlings were approximately four to five weeks old, lowering individuals to the ground for processing (banding, blood draw, morphological measurements), then replacing individuals back in the nest. Analysis was performed on homogenates of each tissue type individually. KF nestlings were collected from nest burrows located within the study area. After locating an active kingfisher nest burrow, it was excavated and fitted with an access door to allow for nest monitoring and sample collection. Kingfisher nestlings collected for contaminant analysis were sacrificed at fourteen days old. Analysis was performed on whole-body homogenates, after the removal of stomach contents, feathers, beak, and legs.

All samples were collected with the approval and permitting of appropriate state and federal agencies and in accordance with Michigan State University's All-University Committee on Animal Use and Care. Analyses of PCDD, and PCDF, concentrations in samples were conducted at AgriQuality Limited (Lower

Hutt, New Zealand) using EPA method 8290. Concentrations of the seventeen 2,3,7,8 substituted PCDD/DFs were quantified in each tissue type and normalized to 2,3,7,8-dibenzo-*p*-dioxin using WHO avian TEFs[3].

To calculate site-specific BMFs, lipid-normalized analytical concentrations of each of the seventeen 2,3,7,8 substituted PCDD/DF congeners in receptor tissues were divided by those in the diet.

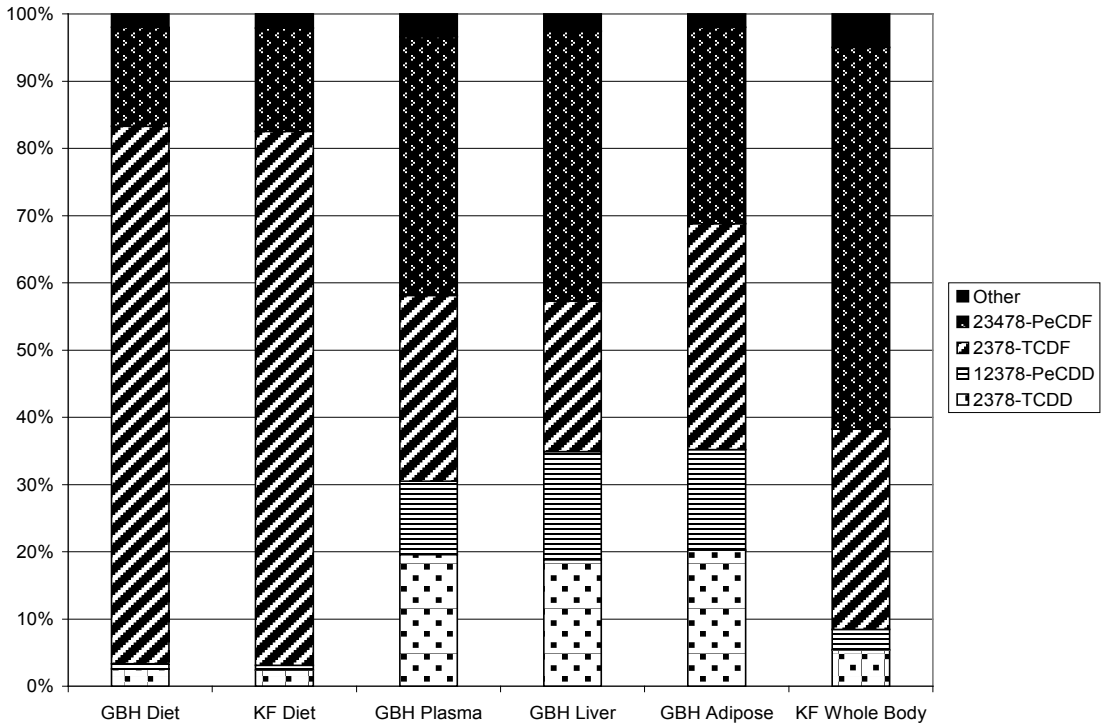
Results and discussion

In the GBH diet, which was comprised of 96% fish, 2% crayfish, and 2% frog, mean total TEQs was $2.1x10^2$ ng/kg (ww). Mean total TEQs in the KF diet, which was comprised of 82% fish, 13% crayfish, and 5% frog, was $2.0x10^2$ ng/kg (ww). Mean total TEQs in GBH and KF tissues ranged from $1.4x10^0$ to $1.2x10^2$ ng/kg (ww) (Table 1). All dietary items and receptor tissues were collected from within the study area. Due to a lack of site-specific data, dietary composition of the GBH is taken from Alexander[4] as cited in the EPA Wildlife Exposure Handbook[5], while the KF dietary composition was determined through the identification of prey remains collected from nest burrows located within the study area.

Table 1. Sum WHO_{AVIAN} TEQs ng/kg (ww)(1/2DL) from PCDD/DFs in the diet, GBH, and KF collected from the Tittabawassee River study area.

Species	Tissue	Mean TEQ (range)
GBH	Diet (96% fish, 2% crayfish, 2% frog)	210
	Plasma (n=13)	1.4 (0.52 – 2.8)
	Liver (n=9)	4.1 (1.7 – 7.7)
	Adipose (n=9)	120 (64 – 320)
KF	Diet (82% fish, 13% crayfish, 5% frog)	200
	Whole body (n=9)	91 (33 – 180)

Figure 1. Percent contribution of the predominant PCDD/DFs congeners to the sum TEQ, based on WHO_{AVIAN} TEQs (1/2DL). GBH diet is a composite of 96% fish, 2% crayfish, and 2% frog. KF diet is a composite of 82% fish, 13% crayfish, and 5% frog. Each receptor tissue profile represents an average of all individuals analyzed (GBH plasma n=13; GBH liver n=9; GBH adipose n=9; KF whole body n=9).



TCDF accounted for 80% and 79% of the total concentrations of TEQ in the diet of the GBH and KF, respectively, while only contributing 28%, 22%, 34%, and 30% to total TEQ concentrations in GBH plasma, GBH liver, GBH adipose, and KF nestlings, respectively (Figure 1). In most receptor tissues, 4-PeCDF is the predominant congener, accounting for 38%, 40%, and 57% of total TEQ concentrations in GBH plasma, GBH liver, and KF nestlings, respectively (Figure 1). GBH adipose is the exception, with 4-PeCDF contributing only 29% to the total TEQ concentrations.

Table 2. Biomagnification factors of 4-PeCDF, TCDF, TCDD and sum PCDD/DFs in GBH and KF. GBH diet based on a literature-derived diet of 96% fish, 2% crayfish, and 2% frog. KF diet based on a site-specific diet of 82% fish, 13% crayfish, and 5%. BMFs from a previous study included for comparison.

Species	Tissue	Biomagnification Factors			Ratio
		4-PeCDF	TCDF	TCDD	4-PeCDF:TCDF
GBH	Plasma	0.27	0.042	0.75	6.6
	Liver	0.075	0.0084	0.21	9.0
	Adipose	0.081	0.018	0.34	4.4
KF	Whole Body	1.8	0.18	1.1	10
HG[a]	Whole Body	6.6	1.3	32	5.1

[a] Herring gull biomagnification factors based on whole body herring gulls and a diet of alewife collected from Lake Ontario[6].

Although the GBH and KF have relatively equal trophic status, there is a large difference in the magnitude of the BMFs of the various congeners (Table 2). This disparity could be due to distinctly different metabolism and elimination of the congeners by the two species, a misunderstanding of the dietary compositions, or most likely it can be attributed to the difference in foraging range between the two species. GBH from rookeries located along a riverine system in North Dakota were shown to travel a mean distance of 3.1 km from their rookery to foraging locations, with a distance as far as 24.4 km being documented[7]. In contrast, the mean KF territory size during the breeding season is 1.03 km of river length[8]. It is reasonable to assume that GBH breeding in rookeries located within the Tittabawassee River floodplain are spending a portion of their time foraging offsite.

To determine the potential impact of dietary composition, BMFs were generated using a variety of different dietary compositions (Table 3). The low variation between BMFs based on different dietary compositions suggests that dietary composition is not driving the BMF.

Table 3. 2,3,4,7,8-PeCDF biomagnifications factors for GBH tissues using differing dietary compositions.

	BMF of 4-PeCDF		
Dietary Composition	GBH Plasma	GBH Liver	GBH Adipose
70% fish, 20% crayfish, 10% frog	0.19	0.053	0.057
82% fish, 13% crayfish, 5% frog	0.22	0.061	0.066
96% fish, 2% crayfish, 2% frog	0.27	0.075	0.081
100% fish	0.29	0.081	0.087

Induction of EROD and MROD (ethoxyresorufin-*O*-dealkylase and methoxyresorufin-*O*-dealkylase) in GBH liver from the study area were comparable to sample blanks in the assay[9], suggesting that accelerated metabolism and elimination of aryl hydrocarbon receptor (AhR)-active compounds in GBH is not occurring.

Despite the difference in the magnitude of BMFs between GBH and KF, the 4-PeCDF:TCDF BMF ratio remains similar among different species, ranging between 4.4 and 10 (Table 2). Previous studies have also observed a similar ratio in fish-eating birds[6]. This ratio indicates that TCDF is more rapidly eliminated or metabolized than the 4-PeCDF. This is also supported by the ratio of 4-PeCDF:TCDF being 2-fold greater in GBH liver, a metabolically active tissue, than in GBH adipose. A recent laboratory mink feeding study

demonstrated an increase in the metabolism and subsequent depuration of TCDF when co-administered with other aryl hydrocarbon receptor (AhR)-active compounds[10]. The study population of GBH and KF is exposed to a mixture of AhR-active compounds which could accelerate the metabolism of TCDF, leading to the BMF ratio observed.

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