

# LEVELS IN BIOTIC COMPARTMENTS

## LEVELS OF PCDD/DFS AND DIOXIN-LIKE PCBs IN BLOOD OF BLACK AND TURKEY VULTURES FROM SAVANNAH RIVER SITE, SOUTH CAROLINA, USA

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### Introduction

Polychlorinated dibenzo-*p*-dioxins (PCDDs), dibenzofurans (PCDFs) and dioxin-like polychlorinated biphenyls (dioxin-like PCBs) have become widespread environmental pollutants that can now be found in the most remote parts of the world<sup>1</sup>. PCDDs, PCDFs and PCBs show a complex spectrum of biological and toxicological properties, with qualitative and quantitative differences depending on the congeneric structure and wild birds. Particularly, birds belong to top of the food chain showed considerable bioaccumulation of PCDD/DFs and dioxin-like PCBs<sup>2</sup>. There are several incidences of population declines have put forward for predator birds such as bald eagles, white-tailed sea eagles, golden eagles and colonial fish-eating water birds from most part of the world. However, there is no study focussed on accumulation profiles and toxic effects of 2378-PCDD/DFs and dioxin-like PCBs in vultures that belongs to higher trophic level that mostly feeds on carrion's, terrestrial animals and small mammals and expected to bio-accumulate greater levels of these chemicals. Considering the lack of information on accumulation of PCDDs, PCDFs and dioxin-like PCBs in blood of black and turkey vultures from Savannah River Site at South Carolina, USA, we measured accumulation profiles of 2378-chlorine substituted PCDD/DFs and dioxin-like PCBs in these two species. TEQs were calculated using the World Health Organization (WHO) toxic equivalency factors (TEFs) for birds<sup>3</sup>. Species-specific accumulation and current status of TEQ were also discussed.

### Materials and Methods

#### *Sample collection*

Blood (approximately 5-9 ml) of black vulture and turkey vulture were collected using clean micro syringe during either walk-in cage trap or rocket net at the Savannah River Site near Aiken of South Carolina, USA in the years 2000 and 2001 (Table 1). Notably, all the individuals were caught near the R-Reactor, an abandoned nuclear reactor that serves as vulture major roost. Prior to collect the blood, bird weight was measured and their age was determined based on the color and degree of wrinkling of the heads and then released. The collected blood was heparinized immediately and transported to the laboratory for chemical analysis and stored at -20°C until analyzed.

#### *Analysis*

The whole blood samples were ground with sodium sulphate and then extracted using Soxhlet apparatus for 15 hrs using dichloromethane as an extract solvent. Extracts were then subjected to

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concentrate to 20 ml using Kuderna-Danish (K-D) concentrator. The aliquots of extract were used for fat determination. While in the remaining extracts a total of sixteen  $^{13}\text{C}_{12}$ -labelled 2378-chlorine-substituted PCDD/DFs and 14 of  $^{13}\text{C}_{12}$ -labelled dioxin-like PCBs (including 4 non-, 8 mono- and 2 di-ortho PCBs) were spiked and further subjected to sulfuric acid treatment, moisture removal and sequence of silica gel, alumina and silica gel impregnated carbon column separations. Details of the analytical procedures have been reported in our earlier study<sup>4</sup>. Identification and quantification of 2378-substituted congeners of PCDD/DFs and dioxin-like PCBs (non-, mono- and di-ortho-substituted congeners) was performed using Hewlett Packard 6890 Series high-resolution gas chromatography interfaced with a Micromass Autospec - Ultima high-resolution mass spectrometer.

## Results and Discussion

### PCDD/DFs

Concentrations of PCDDs were greater in both the species with the ranges of 360-750 and 380-1000 pg/mL fat in black vulture and turkey vulture, respectively (Table 1). PCDFs were much lower than PCDDs in black vulture than in turkey vultures. In general, species-wise, the turkey vultures contained greater accumulation levels than the black vultures. Notably, PCDFs compositions were slightly higher in turkey vultures than in black vultures. The difference accumulation profiles suggested different ecology and feeding habits of these two species. On the whole, levels of PCDD/DFs were one magnitude lower in black vulture and 2-3 times lower in turkey vulture than the bald eagles from Michigan<sup>4</sup> and bald eagles from British Columbia of Canada<sup>5</sup>.

The results of homologue profiles suggested the enrichment of OCDD in black vultures and penta-CDD and OCDD in turkey vultures (Figure 1). Among PCDFs, there is no specific difference between species except presence of TCDF in turkey vultures, which is, absent in other species. Collectively, HxCDF and HpCDF enriched greatly in vulture blood. OCDD was prevalent in bald eagle blood plasma and homologue profiles in this study were closely related with pattern in bald eagles of Great Lakes, USA. The relatively great proportion of OCDD in blood plasma has been suggested to be due to binding of this congener to serum proteins.

Sample ID	Fat (%)	Age	Bird weight	Date of collection	PCDDs	PDCFs	NO-PCBs	MO-PCBs	DO-PCBs	TEQs	
BV-04	3.67	A	2.2 kg	10/11/00	610	170	760	42000	88000	100	130760
BV-14	4.84	J	2.4 kg	6/28/01	390	61	510	34000	33000	57	67510
BV-16	3.93	A	2.3 kg	7/27/01	360	42	320	26000	36000	46	62320
BV-19	4.04	A	2.4 kg	7/27/01	590	73	1700	30000	37000	200	68700
BV-22 (05)*	2.89	A,A	2.2 kg; 2.2 kg	9/19/2001 ; 10/11/00	750	74	670	120000	270000	96	390670
TV-04	2.62	J	1.9 kg	6/28/01	390	350	500	150000	230000	140	380500
TV-06	2.74	A	1.7 kg	6/28/01	1000	240	590	83000	270000	650	353590
TV-07	3.32	A	1.9 kg	7/27/01	930	220	650	66000	200000	630	266650
TV-09	1.63	J	2.2 kg	7/27/01	880	430	730	41000	41000	460	82730
TV-12	1.43	A	2.1 kg	7/27/01	380	160	700	92000	120000	260	212700

\*Captured twice; all the samples were collected from vultures of Savannah River site of USA.

BV and TV respectively, black vulture and turkey vulture; A and J respectively adult and juvenile.

2378-PCDDs; 2378-PCDFs; non-ortho PCBs; mono-ortho PCBs; di-ortho PCBs and toxic equivalency;

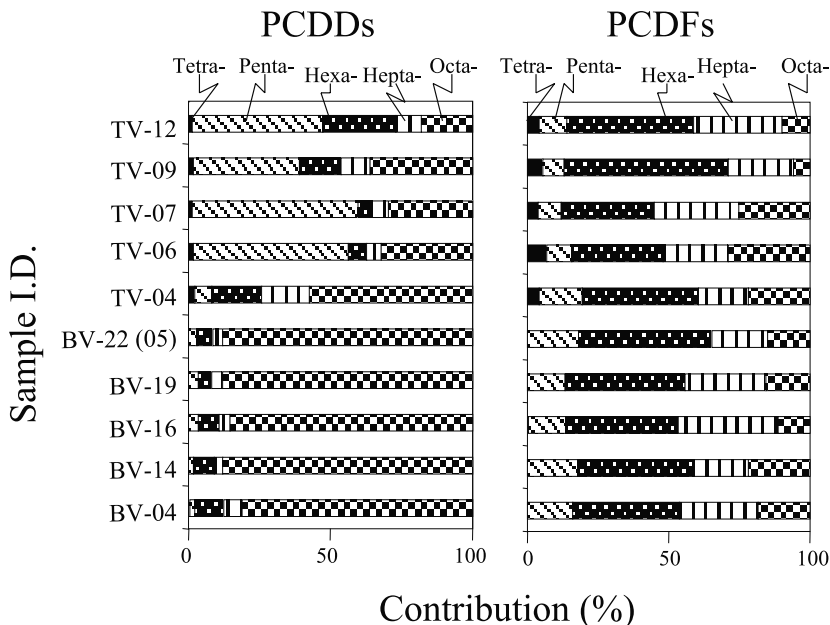
and concentrations represents pg/mL fat weight basis; The values rounded.

### PCBs

Concentrations of dioxin-like PCBs were in the ranges of 62-391 ng/mL fat in black vultures and 83-381 ng/mL fat in turkey vultures (Table 1). Four non-ortho PCBs contributed very less to sum of

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dioxin-like PCBs with 0.51-2.5% and 0.13-0.88% in black vulture and turkey vulture, respectively. While 2 congeners of di-*ortho* PCBs had greater contribution with 49-69% in former and 50-76% in latter species and remaining contribution is shared by 8 mono-*ortho* PCB congeners. The observed PCB levels were one to two-order magnitude less than bald eagles from Michigan, USA<sup>4</sup> and Canadian British Columbia<sup>5</sup>. Overall, CB-180, 170 and 118 were predominant congeners in vulture blood. Interestingly, non-*ortho* PCB accumulation pattern found to be different with CB-126>CB-77>CB-169>CB-81 in black vulture and CB-77>CB-126>CB-169>CB-81 in turkey vulture. Again these results suggested that different feeding habits, ecology, movements, age, gender variation in between species taken into a great account.



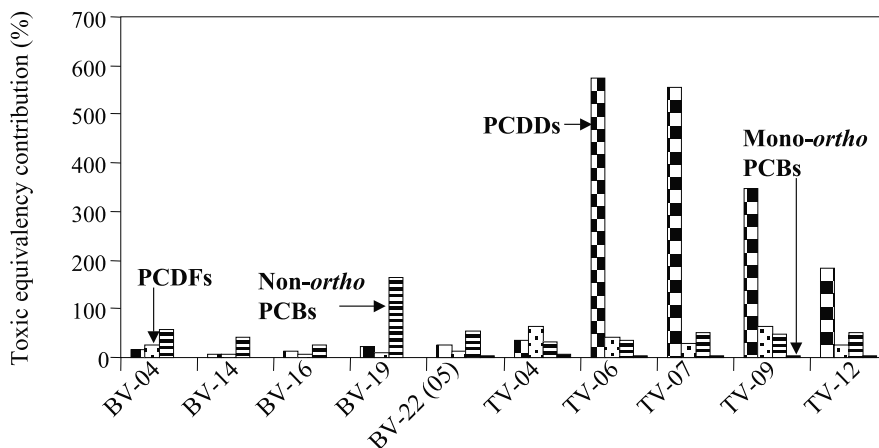
**Figure 1.** Homologue Profiles of PCDD and PCDF in blood of black and turkey vultures from Savannah, USA.

### Toxic Equivalents

Toxic equivalency (TEQs) concentrations of PCDD/DFs and dioxin-like PCBs were in the ranges of 46-200 pgTEQ/mL fat in black vultures and 140-650 pgTEQ/mL fat in turkey vultures (Table 1). Greater contribution by non-*ortho* PCBs was noticed in black vulture followed by PCDDs, PCDFs and mono-*ortho* PCBs with slight difference between individuals (Figure 2). Whereas, PCDDs showed greater toxic contribution in turkey vulture followed by non-*ortho* PCBs, PCDFs and mono-*ortho* PCBs with slight difference between individuals (Figure 2). In general, 12378-PeCDD, 23478-PeCDF, 2378-TCDD/DF contributed to greater toxicity in either species. While, non-*ortho* PCBs IUPAC Nos. 126, 77 and 81 contributed to 50% of total TEQ contribution. Altogether, the TEQ levels in both the species were less than that experiences any toxic threat.

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The age difference was not pronounced in either species. One individual juvenile black vulture contained lesser PCDDs, PCDFs mono- and di-*ortho* PCBs when compare to other adults. Considerably, one adult black vulture blood collected in 2000 had comparatively greater levels than in other 3 individual adults. Similarly, two juvenile turkey vultures showed almost similar concentration levels with those of 3 adult turkey vultures. The age and temporal difference is not withstanding due to small sample size. Further, large number of sample analysis is warranted to see any specific trend of PCDD/DF and dioxin-like PCBs accumulation and their intrinsic effects to vultures.



**Figure 2.** Toxic equivalency (TEQ) contribution by PCDD/DFs a dioxin-like PCBs in blood of black and turkey vultures from Savannah, USA.

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