Analysis of Metals and Polychlorinated Biphenyl (PCB) Residues in Beaver Tissue Samples Collected January 27 and February 1, 2000 from PGDP Ditches

Wesley J. Birge

David J. Price

FINAL REPORT SUBMITTED

February 7, 2001

Submitted to

Jon Maybriar

Division of Waste Management Kentucky Department for Environmental Protection

INTRODUCTION

Over the past two years, we have received requests to analyze samples of avian and mammalian tissues. These were "collections of opportunity" by Division of Waste Management and Fish and Wildlife personnel and intended to be used for preliminary screening and prioritizing further studies. Tissues from three beaver samples were collected in and around PGDP and were sent to us for analysis. Precise location of the beaver collections were not given at that time. The tissues included the kidneys, liver and fat. Samples of each of the two kidneys were analyzed for both PCBs and metals. Duplicate assays for each of liver and fat tissue samples were also analyzed for PCBs and metals. Three Aroclors (*i.e.* 1248, 1254, and 1260) and 8 metals (*i.e.* Ag, Be, Cd, Cr, Cu, Ni, Pb and Zn) were analyzed.

More recently, we received tissue samples from two additional beavers (numbers 4 and 5). We also received from Gaye Brewer (*personal comm.*) details and collection locations on all five beavers, including one pregnant female (*i.e.* beaver # 5). This study takes on added significance because beavers # 2 and 3 were from the Western Kentucky Wildlife Management Area in Ballard County. Tissues form these animals contained little or no PCB or metal contamination and provide a reference base for comparisons with beaver tissues taken from the PGDP area. Descriptions of the five beavers provided by Gaye Brewer are attached at Appendix I.

1

METHODS

Tissue Digestions for Metal Assays

Tissues selected for analysis were prepared according to modified procedures described by Shaw *et al.* (1998), Hogstrand *et al.* (1996), and U.S. EPA (1997). All chemicals used were ACS grade or better and all acids were TraceMetal grade. Tissue samples were wet-weighed and placed in 50-mL Hot-Block® digestion tubes. The samples were digested with 1:1 TraceMetal grade HNO₃ (3.0 mL/g wet weight) and heated to 95° C for 15 min. The samples were allowed to cool to room temperature and 5.0 mL of TraceMetal grade concentrated HNO₃ were then added and the sample was heated to 95° C for 30 min or until a volume of 5.0 mL was reached. Once complete digestion was achieved, 2.0 mL of nano-pure water and 3.0 mL of 30% H₂O₂ were added to each sample followed by heat-instilling. Thirty percent H₂O₂ was added, followed by heating until no effervescence was observed. The samples were then heated to obtain a final volume of 5.0 mL and then filtered through a Gelman Sciences Type A/E glass fiber filter to remove suspended particulates. The filters were rinsed with 0.5 % HNO₃ prior to use and filtrates were taken to a final volume of 100.0 mL.

Metal Determinations

Eight metals, including silver (Ag), beryllium (Be), cadmium (Cd), chromium (Cr), copper (Cu), lead (Pb), nickel (Ni), and zinc (Zn), were analyzed by atomic absorption spectrophotometry (AAS), using graphite furnace atomization techniques. The instrument was a Varian AAS (Model Spectra AA-20), equipped with a GTA-96 graphite furnace. All gases used were ultra pure carrier grade. Calibration curves were based on five standards. The instrument was programmed to take three readings per sample and

average the absorbance. Instrument blanks ($0.5 \% HNO_3$) and check standards were processed with all samples. Sample concentrations were then corrected for deviations from the standards and sample weights were factored into the calculation of final values.

PCB Tissue Extraction

PCBs in tissues were extracted and analyzed using standard U.S. EPA methods (Watts, 1980; U.S. EPA, 1997; Erickson, 1997). The samples were ground with 20-40g anhydrous sodium sulfate and the powder extracted with petroleum ether in a Soxhlet apparatus for 5-h. The extracts were concentrated to near dryness in a Roto-evaporator (Buchi Model RE121). Reconstituted samples (10.0 mL in iso-octane) were then cleaned of interferences as described below and then analyzed by gas chromatography. Lipid and pesticide clean-up was performed by eluting a 2.0 mL sample through a column of 100 g activated 100-200 mesh Florisil® (100 °C/24 h) with 180 mL 6% ethyl ether in petroleum ether and evaporated to 2.0 mL (Erickson, 1997; U.S. EPA, 1997, SW-846 Method 3620B, Florisil cleanup). Fat samples required further cleanup and were eluted through an alumina/silica gel column (4-g activity IV alumina 10%; 2-g fully activated silica gel) with 10 mL hexane as described by Erickson (1997). Elemental sulfur was then removed by shaking with 2-propanol (2 mL) and tetrabutylammonium sulfite (2 mL), adding ultra-pure water (8 mL), and reshaking. The organic extract was removed and mixed with 2.0-mL concentrated sulfuric acid (Jensen et al., 1977; U.S. EPA, 1997, SW-846 Method 3660B, sulfur cleanup). A 4 µL sub-sample was then analyzed by gas chromatography.

PCB Determinations

Samples were analyzed for Aroclors 1248, 1254, and 1260 according to SW-846

Method 8082 (Polychlorinated biphenyls by gas chromatography, U.S. EPA, 1997). Analysis were performed using a Hewlett-Packard (HP) Model 5890A gas chromatograph equipped with an electron capture detector and an HP Model 7673A Automatic Sampler. Samples were analyzed using a 60m X 0.53mm ID SPB-5 (0.5µm film) fused silica megabore column (Supelco, Inc.) with ultra-high purity helium and nitrogen as carrier and makeup gases, respectively. The temperature program was set at 160 °C (6 min)-10 °C/min-235 °C (0 min)-0.9 °C/min-260 °C (10 min): Injector temperature, 280 °C; Detector temperature, 300 °C. PCB peak heights were quantified using an HP Model 3396A integrator. Aroclor concentrations were calculated from heights of 6 to 9 peaks for Aroclors 1248 and 1260 and 4-6 peaks for Aroclor 1254. Five external standards were used for calibration curves and for every tenth sample either a solvent blank or a standard was analyzed. Statistical quantitation of peak heights was determined by multiple-peak linear regression analysis with Lotus-123® software. The Lotus program regresses data from PCB standards to the sample being analyzed. Each peak selected for each Aroclor class was statistically analyzed (e.g., standard deviation; standard error; relative deviation). Chromatographs and bench records for all PCB assays will be maintained as given below under quality assurance.

Quality Assurance

Permanent bench records were kept of all assays and annotated as required under Good Laboratory Practices (Federal Register, 40 CFR, Part 160, August 17, 1989). All printouts and graphic recordings were filed and are open for inspection. These bench records will be archived within two years after the close of the project but retrievable upon request. Chain of Custody was maintained for the samples collected.

RESULTS

Polychlorinated biphenyls were not detected in beaver # 2 and only Aroclor 1254 at 0.018 µg/g (*i.e.*, ppm) was found in one of two fat samples for beaver # 3 (Table 1). Therefore, there was little or no PCB contamination in beavers from the Western Kentucky Wildlife Management Area (WKWMA) in Ballard County. Consequently, these were used as a reference base. However, beaver # 1, an immature female taken from the Little Bayou Creek area at PGDP, contained PCB residues in all tissue samples analyzed and 1248 was the predominant Aroclor. Aroclor 1248 concentrations in µg/g (ppm) ranged from 0.028-0.032, 0.043-0.049, and 0.699-0.773 in kidney, liver and fat, respectively. Maximum values for total PCB per tissue, given in the same order, were 0.049, 0.059, and 0.915 μ g/g. Values below detection were not included. Fat samples contained much higher PCB residues, which ranged close to $1 \mu g/g$. Beavers # 4 and 5 (Appendix I) were collected near the effluent outfall of 001 on Big Bayou Creek. Tissue samples from each animal contained significant concentrations of PCBs (Table 1). As for Beaver # 1, PCB residues increased in the order of kidney to liver to fat. Maximum total PCB concentrations were 0.14 and 0.58 µg/g in liver and fat from beaver # 4, and ~0.03 and 0.51 μ g/g in liver and fat from beaver # 5, a pregnant female. Unfortunately,

fetal tissues were not available for analysis. It should be noted that, as herbivores, beavers are not subject to the same potential for PCB biomagnification up the food chain as are carnivores. Therefore, the exposure to PCBs would mostly constitute water and plant intake. As reported in other studies (Birge *et al.*, 1998), essentially all categories of wildlife collected at PGDP have been found to be contaminated with PCB (Figure 1). These findings further support the premise that PCB pollution is pervasive and widespread in the vicinity of PGDP. More monitoring of wildlife at PGDP, WKWMA and other reference areas is recommended.

Of the eight metals analyzed in tissues from beavers # 1, 2, and 3, only beryllium (Be) was undetected (Table 2). With a few exceptions (*e.g.* liver Cu, Pb, Ni, Zn), beaver # 1 contained greater tissue metal residues. This trend was particularly evident in the kidney and fat samples. Highest metal concentration in all tissues were for Cu and Zn, each of which is an essential element. The highest concentrations in kidney tissue of beaver # 1, taken from the vicinity of Little Bayou Creek, were 6.57 and 51.38 μ g/g for Cu and Zn, respectively.

As to non-essential metals, tissue residues were observed for Cd and Pb. In kidney tissues, Cd concentrations ranged from 1.55 μ g/g in beaver # 3 to 3.78 μ g/g in beaver # 1, whereas Pb concentrations were in the range of 0.53 to 1.00 μ g/g overall. Detectable values (μ g/g) for Pb ranged up to 0.89 in liver tissue. Ni residues appeared somewhat elevated, with maximum concentrations (μ g/g) of 1.26 (beaver # 1), 0.90 (beaver # 2), and 1.00 (beaver # 1) in kidney, liver and fat samples, respectively.

Mean concentrations for tissue metals are given in Table 3. Concentrations $(\mu g/g)$ in increasing order for beavers # 1, 2 and 3 generally were Be (not detected), Ag,

Pb, Cr, Ni, Cd, Cu and Zn for kidney; and Be, Ag, Cr, Cd, Pb, Ni, Cu and Zn for liver and fat. The highest mean values that may indicate concern for biological effects were 2.92 μ g/g Cd and 0.78 μ g/g Pb in kidney from beaver # 1.

Beavers # 4 and 5, as noted above, were from the area of the 001 effluent outfall. This is the sector of Big Bayou Creek shown to contain highest degree of metal contamination (Birge *et al.*, 1992). Compared to the WKWMA beavers, kidney tissue residues generally were somewhat higher in beavers #4 and/or 5 for Ag, Be, Cd, and Zn. The same trend was noted for Ag, Be, Cd, Cr and Zn residues in liver tissues. Compared to the beavers from the WKWMA, tissue samples from beavers #4 and 5 (*i.e.* 001 outfall) contained higher residues of Ag, Be, Cd, and Zn in kidney and liver tissues (Table 3). Metal and PCB contamination in beavers appear sufficient to warrant further investigations.

In summary, PCBs were substantial in beavers from the PGDP area, whereas it was undetected or minimal in beavers from WKWMA. Relative to PCB pollution at PGDP, these results support earlier findings for other species of wildlife (Price and Birge 1998a and 1998b; Figure 1). The overall pattern of metal contamination also was greater, especially for Ag, Be, Cd, and Zn. A literature search for "normal" metal concentrations in beavers from unpolluted areas should be undertaken. The metal residues do correlate with and further support the patterns of environmental metal pollution observed at PGDP (Birge *et al.*, 1998; Birge *et al.*, 1992).

7

		Sample	USFWS	Wt. of	Wt. of		Aroclor conce	entration (µg	/g)
Name	Date	No. ^a	ld No.	Organ (g)	Tissue (g)	1248	1254	1260	Total
BEAV1	1/27/00	KD1	0100008	28.739	4.980	0.028	<0.016	0.006*	0.028
BEAV1	1/27/00	KD2	0100008	26.188	3.845	0.032	<0.021	0.017	0.049
			MEAN	27.464	4.413	0.030	N.D.	0.012	0.039
BEAV2	02/01/00	KD1	0200001	43.563	3.910	<0.020	<0.020	<0.020	<0.020
BEAV2	02/01/00	KD2	0200001	48.339	4.728	<0.017	<0.017	<0.017	<0.017
			MEAN	45.951	4.319	N.D.	N.D.	N.D.	N.D.
BEAV3	02/01/00	KD1	0200002	46.815	4.624	<0.017	<0.017	<0.017	<0.017
BEAV3	V3 02/01/00 KD2	KD2	0200002	47.393	6.591	<0.012	<0.012	<0.012	<0.012
			MEAN	47.104	5.6075	N.D.	N.D.	N.D.	N.D.
BEAV4	03/08/00	KD1	0300006	453.6	4.832	<0.041	0.081	0.016*	0.081
BEAV4	03/08/00	KD2A	0300006	See KD1	6.363	<0.031	<0.031	0.008*	0.008*
BEAV4	03/08/00	KD2B	0300006		5.254	<0.038	<0.038	<0.038	<0.038
			MEAN	453.6	5.809	N.D.	N.D.	0.008*	0.008*
BEAV5	03/15/00	KD1	0300007	226.8	6.231	<0.032	0.136	0.023*	0.136
BEAV5 BEAV5	03/15/00 03/15/00	KD2A KD2B	0300007 0300007	See KD1	3.620 4.310	<0.055 <0.046	<0.055 <0.046	<0.055 <0.046	<0.055 <0.046
			MEAN	226.8	3.965	N.D.	N.D.	N.D.	N.D.

Table 1. PCB concentrations in beaver tissue samples collected by U.S. Fish and Wildlife Services at the Paducah Gaseous Diffusion Plant and the Western Kentucky Wildlife Management Area.

^a Designations KD and LIV are kidney and liver samples, respectively. Samples designated 1 and 2 are separate samples, whereas samples A and B are duplicates from the same tissue sample.

* PCBs detected but values below minimum quantitation limit (MQL).

		Sample	USFWS	Wt. of	Wt. of		entration (µg	/g)	
Name	Date	No. ^a	ld No.	Organ (g)	Tissue (g)	1248	1254	1260	Total
BEAV1 BEAV1	01/27/00 01/27/00	LIV1A LIV1B	0100008 0100008	351.47	7.912 8.098		<0.010 <0.010	0.010 0.008*	0.059 0.043
			MEAN	351.47	8.005	0.046	N.D.	0.009	0.051
BEAV2	02/01/00	LIV1A	0200001	598.17	6.687	<0.012	<0.012	<0.012	<0.012
BEAV3 BEAV3		LIV1A LIV1B	0200002 0200002	532.50	7.053 4.815	<0.011 <0.017	<0.011 <0.017	<0.011 <0.017	<0.011 <0.017
			MEAN	532.5	5.934	N.D.	N.D.	N.D.	N.D.
BEAV4 BEAV4	03/08/00 03/08/00	LIV1A LIV1B	0300006 0300006	907.2	5.427 5.073	<0.037 <0.039	0.144 <0.039	0.028* 0.029*	0.144 0.029*
			MEAN	907.2	5.250	N.D.	0.144	0.029*	0.144
BEAV5 BEAV5		LIV1A LIV1B	0300007 0300007	907.2	4.329 5.262	<0.046 <0.038	<0.046 <0.038	<0.046 0.028*	<0.055 0.028*
			MEAN	907.2	4.800	N.D.	N.D.	0.028*	0.028*

Table 1, continued. PCB concentrations in beaver tissue samples collected by U.S. Fish and Wildlife Services at the Paducah Gaseous Diffusion Plant and the Western Kentucky Wildlife Management Area.

^a Designations KD and LIV are kidney and liver samples, respectively. Samples designated 1 and 2 are separate samples, whereas samples A and B are duplicates from the same tissue sample.

* PCBs detected but values below minimum quantitation limit (MQL).

		Sample	USFWS	Wt. of	Wt. of		Aroclor conce	entration (µg	ı/g)
Name	Date	No. ^a	ld No.	Organ (g)	Tissue (g)	1248	1254	1260	Total
BEAV1	01/27/00	FAT1A	0100008	21.641	2.949	0.773	<0.027	0.142	0.915
BEAV1	01/27/00	FAT1B	0100008		2.648	0.699	<0.030	0.160	0.859
			MEAN	21.641	2.799	0.736	N.D.	0.151	0.887
BEAV2 BEAV2	02/01/00 02/01/00	FAT1A FAT1B	0200001 0200001	88.68	5.629 5.412	<0.028 <0.030	<0.028 <0.030	<0.028 <0.030	<0.028 <0.030
			MEAN	88.68	5.521	N.D.	N.D.	N.D.	N.D.
BEAV3	02/01/00	FAT1A	0200002	127.03	3.295	<0.024	<0.024	<0.024	<0.024
BEAV3	02/01/00	FAT1B	0200002		7.405	<0.011	0.018	<0.011	0.018
			MEAN	127.03	5.350	N.D.	0.018	N.D.	0.018
BEAV4	03/08/00	FAT1A	0300006	113.4	4.326	<0.046	0.341	0.200	0.541
BEAV4	03/08/00	FAT1B	0300006		4.577	<0.044	0.356	0.219	0.575
			MEAN	113.4	4.452	N.D.	0.349	0.210	0.559
BEAV5	03/15/00	FAT1A	0300007	113.4	3.568	<0.056	0.366	0.146	0.512
BEAV5	03/15/00	FAT1B	0300007		4.225	<0.047	<0.047	0.173	0.173
			MEAN	113.4	3.897	N.D.	0.366	0.160	0.526

Table 1, continued. PCB concentrations in beaver tissue samples collected by U.S. Fish and Wildlife at the Paducah Gaseous Diffusion Plant and the Western Kentucky Wildlife Management Area.

^a Designations KD and LIV are kidney and liver samples, respectively. Samples designated 1 and 2 are separate samples, whereas samples A and B are duplicates from the same tissue sample.

* PCBs detected but values below minimum quantitation limit (MQL).

		Wt. Of Tissue			Me	etal Concer	ntration (µg/	g)		
Sample Date	e Number ^a	(g)	Ag	Ве	Cd	Cr	Cu	Pb	Ni	Zn
BEAV#1 01/27,		1.512	0.015*	<0.13	2.06	0.14	3.70	0.57	0.75	20.94
BEAV#1 01/27,		0.618	0.027*	<0.32	3.78	1.05	6.57	1.00	1.26	51.38
BEAV#2 02/01,		1.169	0.013*	<0.17	1.71	<0.09	3.85	0.54	0.76	27.61
BEAV#2 02/01,		1.107	0.009*	<0.18	1.73	<0.09	4.25	0.53	0.71	10.31
BEAV#3 02/01,		1.077	0.009*	<0.19	1.63	0.81	3.76	0.53	0.81	11.37
BEAV#3 02/01,		1.482	0.013*	<0.13	1.55	0.50	4.59	0.60	0.87	20.54
BEAV#4 03/08	/00 KD1	0.836	0.040	0.03	2.63	0.14	1.81	0.44	0.40	72.46
BEAV#4 03/08,		0.658	0.041	0.03	2.82	0.13	1.95	0.47	0.36	59.78
BEAV#4 03/08,		0.392	0.038	0.04	2.75	0.29	2.33	0.49	0.33	128.26
BEAV#5 03/15	/00 KD1	0.694	0.029	0.03	1.53	0.15	2.34	0.43	0.34	59.94
BEAV#5 03/15,		0.426	0.037	0.02	1.33	0.14	2.58	0.34	<0.23	52.12
BEAV#5 03/15,		0.972	0.028	0.03	1.33	0.09	2.03	0.38	0.41	47.19

Table 2. Metal concentrations in beaver tissue samples collected by U.S. Fish and Wildlife at the Paducah Gaseous Diffusion Plant and the Western Kentucky Wildlife Management Area.

^a Designations KD and LIV are kidney and liver samples, respectively. Samples designated 1 and 2 are separate samples, whereas samples A and B are duplicates from the same tissue sample.

* Metals detected but values below minimum quantitation limit (MQL), qualitative use only.

		Wt. Of Tissue			Me	etal Concer	itration (µg	/g)		
Sample Da	ate Number ^a	(g)	Ag	Be	Cd	Cr	Cu	Pb	Ni	Zn
BEAV#1 01/2		1.200	0.023*	<0.17	0.22	0.10	3.75	0.58	0.78	10.42
BEAV#1 01/2		1.885	0.011*	<0.11	0.24	0.15	3.62	0.55	0.87	17.97
BEAV#2 02/0	01/00 LIV1A	1.287	0.011*	<0.16	0.11	0.09	3.47	0.59	0.90	23.64
BEAV#3 02/0		1.132	0.013*	<0.18	0.11	<0.09	5.68	0.89	0.87	27.98
BEAV#3 02/0		1.150	0.004*	<0.17	<0.09	0.14	0.94	<0.09	<0.26	2.05
BEAV#4 03/0		0.526	0.041	0.03	0.16	0.19	2.31	0.39	0.24	83.41
BEAV#4 03/0		0.636	0.041	0.03	0.15	0.19	4.13	0.41	<0.16	85.62
BEAV#5 03/1		0.774	0.026	0.02	0.08	0.12	3.98	0.41	0.21	53.62
BEAV#5 03/1		0.724	0.029	0.02	0.09	0.20	3.25	0.32	<0.14	59.22

Table 2, continued. Metal concentrations in beaver tissue samples collected by U.S. Fish and Wildlife Services at the Paducah Gaseous Diffusion Plant and the Western Kentucky Wildlife Management Area.

^a Designations KD and LIV are kidney and liver samples, respectively. Samples designated 1 and 2 are separate samples, whereas samples A and B are duplicates from the same tissue sample.

* Metals detected but values below minimum quantitation limit (MQL), qualitative use only.

			Wt. Of Tissue			Ме	tal Concen	tration (µg	J/g)		
Sample	Date	Number ^a	(g)	Ag	Be	Cd	Cr	Cu	Pb	Ni	Zn
BEAV#1 0 [°] BEAV#1 0 [°]			0.921 1.327	0.030* 0.002*	<0.22 <0.15	0.12 <0.08	0.15 0.17	4.86 0.78	0.77 <0.08	1.00 <0.23	2.39 1.47
BEAV#2 02 BEAV#2 02			2.956 2.142	0.005* 0.005*	<0.07 <0.09	<0.03 <0.05	0.07 0.07	0.61 0.87	<0.03 0.06	<0.10 <0.14	1.12 1.98
BEAV#3 02 BEAV#3 02			0.877 1.916	0.013* 0.002*	<0.23 <0.10	<0.01 <0.05	0.14 0.06	1.39 0.45	<0.11 <0.05	<0.34 <0.16	3.35 0.73
BEAV#4 03 BEAV#4 03			0.649 0.830	0.022 0.014	<0.02 <0.01	0.02 0.17	0.19 0.10	0.39 0.23	0.06 0.06	<0.15 <0.12	2.15 1.07
BEAV#5 03 BEAV#5 03			0.830 0.746	<0.012 <0.013	<0.01 <0.01	<0.01 <0.01	0.10 0.14	0.26 0.27	0.04 0.04	<0.12 0.20	1.83 1.77

Table 2, continued. Metal concentrations in beaver tissue samples collected by U.S. Fish and Wildlife Services at the Paducah Gaseous Diffusion Plant and the Western Kentucky Wildlife Management Area.

^a Designations KD and LIV are kidney and liver samples, respectively. Samples designated 1 and 2 are separate samples, whereas samples A and B are duplicates from the same tissue sample.

* Metals detected but values below minimum quantitation limit (MQL), qualitative use only.

			Wt. Of Tissue			Mean	Metal Cond	centration (na/a) _a		
Sample	Date	Tissue	(g)	Ag	Be	Cd	Cr	Cu	Pb	Ni	Zn
BEAV#1 0	12700	Kidney	1.065	0.021	N.D.	2.92	0.59	5.13	0.78	1.01	36.16
BEAV#2 02	20100	Kidney	1.138	0.011	N.D.	1.72	N.D.	4.05	0.53	0.74	18.96
BEAV#3 02	20100	Kidney	1.280	0.011	N.D.	1.59	0.65	4.18	0.57	0.84	15.95
BEAV#4 03	30800	Kidney	0.629	0.040	0.03	2.73	0.19	2.03	0.47	0.36	86.83
BEAV#5 0	31500	Kidney	0.697	0.031	0.03	1.40	0.13	2.32	0.38	0.37	53.08
BEAV#1 0	12700	Liver	1.543	0.017	N.D.	0.23	0.13	3.69	0.56	0.83	14.20
BEAV#2 02	20100	Liver	1.287	0.011	N.D.	0.11	0.09	3.47	0.59	0.90	23.64
BEAV#3 02	20100	Liver	1.141	0.009	N.D.	0.11	0.14	3.31	0.89	0.87	15.02
BEAV#4 0	30800	Liver	0.581	0.041	0.03	0.16	0.19	3.22	0.40	0.24	84.52
BEAV#5 0	31500	Liver	0.749	0.027	0.02	0.09	0.16	3.62	0.36	0.21	56.42
BEAV#1 0	12700	Fat	1.124	0.017	N.D.	0.12	0.16	2.82	0.77	1.00	1.93
BEAV#2 02	20100	Fat	2.549	0.005	N.D.	N.D.	0.07	0.74	0.06	N.D.	1.55
BEAV#3 02	20100	Fat	1.397	0.007	N.D.	N.D.	0.10	0.92	N.D.	N.D.	2.04
BEAV#4 03	30800	Fat	0.740	0.018	N.D.	0.09	0.14	0.31	0.06	N.D.	1.61
BEAV#5 03	31500	Fat	0.788	N.D.	N.D.	N.D.	0.12	0.26	0.04	0.20	1.80

Table 3. Metal concentrations in beaver samples collected by U.S. Fish and Wildlife at the Paducah Gaseous Diffusion Plant and the Western Kentucky Wildlife Management Area.

^a N.D. indicates not detected.

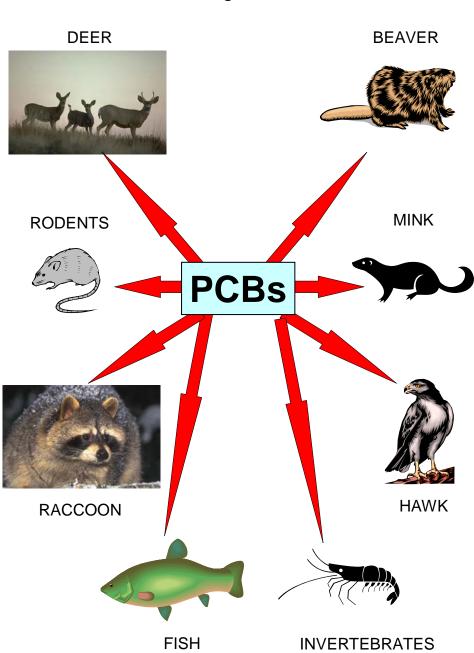


Figure 1

APPENDIX I

Data for Beavers # 1 – 5

Beaver #1 (DWM Sample # 0100008) was taken from Little Bayou Creek approximately 100 yds. South of the culvert that is NE of the C755 trailer complex north of McCaw Rd. Beaver #1 was an immature female that weighed 25.1 lbs. She was about 8-9 months old and measured 95 cm from the tip of the nose to the tip of the tail. It was difficult to find good fat deposits on her due to apparently low subcutaneous and abdominal fat. There were no obvious abnormalities of the internal organs and no obvious parasites except lice.

Beaver #2 (DWM sample # 0200001) was taken from the Ballard Co Wildlife Mgt. Area. Beaver #2 was a mature female, possibly going into estrus. She weighed 39.0 lbs and was 109 cm long. More fat was apparent than on the first beaver. There were no obvious abnormalities of the internal organs. Organ weights: ovaries - 9.62 g. Other organs too heavy for balance.

Beaver #3 (DWM sample #0200002) was also taken from the Ballard Co. WMA. Beaver #3 was an adult male that weighed 42 lbs and measured 114 cm in length. The baculum measured 42 mm from the tip to the front of the gland. Plenty of fat was found on this individual. Abdominal fat was collected adjacent to the kidneys. There were no obvious abnormalities of the internal organs. Organ weights: tail fat – 126.58 g. testes – 42.0 g. abdominal fat – 33.21 g. kidneys – 95.13 g. Liver too heavy for balance.

Beaver #4 (DWM sample # 030006) was taken from 001 outfall. Beaver #4 was an adult male that weighed 40.3 lbs and measured 111 cm in total length. The baculum weighed 4.16 gm and measured 98 mm from the tip to in front of the gland. There were no noticeable lesions on the beaver. Organ weights: testes – 30 g. kidneys – 113 g. liver – 592 g. tail fat – 46 g.

Beaver #5 (DWM sample # 030007) was taken from 001 outfall. Beaver #5 was a pregnant mature female. She weighed 37.2 lbs and measured 96 cm in total length. She had more subcutaneous fat than any of the others.. She carried 3 kits that were retained for analysis. She appeared to be an older animal because her rib bones were thicker. There were no apparent abnormalities except that her left front leg was broken off from the trapping. Organ weights: liver – 564 g. ovaries – 4.0g. tail fat – 68 g. kidney 100 g. fetus #1 – 440 g. fetus #2 – 432 g. fetus #3 – 441 g.

All carcasses were retained for further testing.

From Gaye Brewer (June 29, 2000).

REFERENCES

Birge, W.J., D.J. Price, and M.D. Kercher. 1998. Metal Body Burden in Stoneroller Minnows (*Campostoma anomalum*) from the Bayou Creek System. April 10, 1998. Submitted to Jon Maybriar, Division of Waste Management.

Birge, W.J., D.J. Price, M.D. Kercher, D.P. Keogh and J.A. Zuiderveen. 1992. *Biological Monitoring Program for the Paducah Gaseous Diffusion Plant.* Annual Report for the Study Period October, 1990 through December, 1992. University of Kentucky. Lexington, KY.

Erickson, M.D. 1997. Analytical Chemistry of PCBs, 2nd edition. CRC Press, Boca Raton, FL. pp.667.

Federal Register. 1989. Good Laboratory Practice Standards. 40 CFR Part 160. August 17, 1989. Washington, DC.

Hogstrand, C., F. Galvez, and C.W. Wood. 1996. Toxicity, silver accumulation and metallothionein induction in freshwater trout during exposure to different silver salts. *Environ. Toxicol. Chem.* 15:1102-1108.

Jensen, S., Renberg, L., Reutergådh, L. 1977. Residue analysis of sediment and sewage sludge for organochlorines in the presence of elemental sulfur. *Anal Chem.* 49(2):316-318.

Price D. J. and W. J. Birge. 1998a. Analysis of Polychlorinated Biphenyls (PCB) in Redtailed Hawk Blood, Mink and Deer Liver and Kidney. March 1998. Submitted to Jon Maybriar, Division of Waste Management.

Price, D.J. and W.J. Birge. 1998b. Analysis of Metals in Red-tailed Hawk Blood from PGDP. Final Report, May 28, 1998. Submitted to Jon Maybriar, Division of Waste Management.

Shaw, J.R., C.M. Wood, W.J. Birge, and C. Hogstrand. 1998. Toxicity of silver to the marine teleost (*Oligocottus maculosus*) effects of salinity and ammonia. *Environ. Toxicol. Chem.* 17(4):594-600.

U.S. EPA. 1997. Test methods for evaluating solid wastes, SW-846, Final Update 3. Office of Solid Waste and Emergency Response, Washington, DC.

Watts, R.R. *ed.* 1980. Analysis of pesticide residues in human and environmental samples. A compilation of methods selected for use in pesticide monitoring programs. EPA/600/8-80/033. U.S. EPA, Research Triangle Park, NC. Section 5, A, 1.