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Heavy Metal and Organochlorine Compound Concentrations in Tissues of Raccoons from East-Central Michigan

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Organochlorine (OC) pesticides and related compounds and heavy metals are persistent contaminants in the environment (Fleming et al., 1983; and Korschgen, 1970). Bioconcentration and biomagnification are well reported for organochlorine compounds (Hernandez et al., 1985; and Korschgen, 1970) and heavy metals (Mirenda, 1986). These compounds have a great potential for causing wildlife mortality or serious behavioral, reproductive, carcinogenic, teratogenic, and mutagenic effects, along with specific organ toxicity (Eisler, 1985; Hernandez et al., 1985; and Mirenda, 1986).

Fleming et al. (1983) reported that the levels of OC pesticides and polychlorinated biphenyls (PCB's) have decreased dramatically in the environment during the last decade. Even though the use of some of these compounds has been either banned or highly restricted since the mid to late 1970's, concentrations of OC pesticides and PCB's, from both present and past use, remain high enough to reduce recruitment and survival in selected wildlife populations. Heavy metals reach environmentally unsafe levels in many areas mainly through human sources (Ernst et al., 1983).

The pervasive nature of toxic substances in the environment necessitates some knowledge for potential exposure of wildlife species. Without baseline values of contaminant loads for selected indicator species it is impossible to determine when abnormal or pathological conditions exist in wild populations. The purpose of this study was to provide baseline values for selected environmental contaminants in the raccoon (Procyon lotor), a potential indicator species for wildlife and to see if heavy metal accumulation was related to age or sex.

MATERIALS AND METHODS

The study took place on the Shiawassee National Wildlife Refuge which is located in the east-central portion of Michigan's Lower Peninsula just south-west of Saginaw. The Refuge is comprised of approximately 3,642 ha, which includes the following habitat types: bottomland hardwood forests (1,700 ha), cropland (688 ha),

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marsh (526 ha), moist soil land management (243 ha), grassland (202 ha), and rivers and streams (162 surface ha). The quality of waters entering the Refuge is generally poor. Most of the rivers pass through municipalities and areas of heavy industry as well as agricultural land. Industrial and wastewater discharge along with agricultural run-off have contributed to deteriorating water quality.

Soil samples were taken from three sites on 11 February, 1986 for the analysis of heavy metals, OC and organophosphorus (OP) pesticides, and PCB's. The samples were stored at -15° C and sent to Hazleton Laboratories America, Inc.(3301 Kinsman Blvd., P.O. Box 7545, Madison, Wisconsin 53707) for analysis. Method references for the analysis of heavy metals, OC and OP compounds in soils are described in Herbert (1988).

Raccoons were trapped with Tomahawk (Tomahawk Live Trap Co., Tomahawk, Wisconsin) double-door live-traps throughout August of 1985 and from June to September, 1986 at three sites on the Refuge. Captured raccoons were euthanized with UTHOL (sodium pentabarbitol, 1ml/4.55 kg). The liver, kidneys, brain and fat sample were removed and individually triple-wrapped in aluminum foil, placed in a ziplock bag and stored at -15° C until analyzed. Sample preparation and atomic absorption spectrophotometry procedures for cadmium, lead, mercury, and zinc were those of Herbert (1988). Selenium analysis on liver tissue and analysis of the 1986 fat samples for OC compounds were performed by Hazleton Labs using the methods described in Herbert (1988).

Scheffe's test, Tukey's studentized range test, and one-way ANOVA (SAS Institute Inc., 1985) were used to test for differences between age, and sex for heavy metals and OC compounds. A simple linear regression was performed to determine if selenium and mercury liver concentrations were correlated.

RESULTS AND DISCUSSION

Concentrations of OP's, OC's, and PCB's, along with mercury and cadmium, for the 1986 set of soil samples were below detection limits for all three sites. Mean (± s.d.) concentrations of lead, selenium, and zinc in soils from the three sites were 3.04 ± 0.48, 0.15 ± 0.03, and 25.0 ± 6.5 mg/kg, respectively.

A total of 25 raccoons (15 female, 10 male) was captured during the summers of 1985 and 1986. Mean concentrations of heavy metals in raccoon tissues showed that cadmium levels in liver and kidney tissues differed significantly ($p < 0.05$) between adult and juvenile raccoons but no differences were found in brain tissues (Tables 1 and 2). Mercury levels were significantly different ($p < 0.05$) between adults and juveniles for liver, kidney, and brain tissues (Table 2). No differences were found between adult and juvenile raccoons for lead, selenium, or zinc concentrations. No differences in cadmium, lead, mercury,

Table 1. Heavy metal concentrations (mg/kg wet weight) in liver, kidney, and brain tissues of raccoons from the Shiawassee National Wildlife Refuge, Michigan, 1985-86. Values are reported as mean ± standard deviation (n = 25).

Metal	Liver	Kidney	Brain
Cd	0.79 ± 0.50	3.48 ± 3.42	0.25 ± 0.05
Pb	0.24 ± 0.17	0.20 ± 0.11	0.09 ± 0.13
Hg	1.18 ± 0.87	0.27 ± 0.17	0.04 ± 0.03
Se	2.29 ± 0.72[a]	---	---
Zn	44.4 ± 10.6[b]	18.6 ± 2.90	12.1 ± 0.89

[a] n=17
[b] n=24

selenium, or zinc concentrations were found between sexes ($p > 0.05$). Metal concentrations for male and female raccoons were reported as pooled values because no significant differences were found. Cadmium and mercury values were shown in Table 2 as adult values, while the values for lead, selenium, and zinc were reported in Table 1. A simple linear regression analysis was performed to determine if selenium and mercury liver concentrations were correlated. No significant correlation was found ($y = 0.31x + 1.10$; $r = 0.22$, $df = 15$).

Adipose samples from raccoons caught in 1986 were analyzed for OC insecticides and PCB's. DDE and Lindane (gamma BHC) were the only OC's along with PCB's which had levels above detection limits (Table 3). No significant differences between sexes were found for DDE, Lindane, or PCB's.

The results from the 1986 soil analyses suggested that no serious contamination existed in the soils on the refuge. Quarles et al. (1974) considered 16 ppm lead to be a normal background soil concentration while Eisler (1985) reported a background level of 0.2 ppm for selenium in soils. Soil concentrations of zinc from a contaminated site were 240-7,700 ppm (Chupp and Dalke, 1964). All of the contaminants tested for in soils were either below detection limits or below the normal background levels reported in the literature. Despite the concern over the poor water quality of the rivers entering the refuge, there appeared to be no accumulation of contaminants in the soils on the refuge. The absence of high concentrations of xenobiotics in the soils may have been the result of the large volumes of water which inundated the refuge each spring and autumn. This would in essence flush the refuge and dilute any contaminants which remained suspended in the water column.

Table 2. Heavy metal levels (mg/kg wet weight) in adult and juvenile raccoons from the Shiawassee National Wildlife Refuge, Michigan, 1985-86. Values are reported as means ± standard deviation (n = 7 for juveniles and 18 for adults).

Metal	Age	Liver	Kidney	Brain
Cd	Juvenile	0.25 ± 0.10*	0.40 ± 0.28*	0.23 ± 0.03
	Adult	1.00 ± 0.43	4.68 ± 3.33	0.27 ± 0.05
Pb	Juvenile	0.29 ± 0.23	0.23 ± 0.14	0.16 ± 0.23
	Adult	0.23 ± 0.14	0.19 ± 0.09	0.06 ± 0.06
Hg	Juvenile	0.09 ± 0.06*	0.07 ± 0.07*	0.01 ± 0.00*
	Adult	1.60 ± 0.63	0.35 ± 0.13	0.05 ± 0.02
Se	Juvenile	2.82 ± 0.10 (n = 2)		
	Adult	2.22 ± 0.74 (n = 15)		
Zn	Juvenile	45.3 ± 10.1	19.2 ± 3.53	12.2 ± 0.99
	Adult	44.1 ± 11.2[a]	18.4 ± 2.69	12.1 ± 0.87

[a] n = 17
* $p < 0.05$

Cadmium levels in the liver and kidney (Table 1) were higher than values from uncontaminated sites reported in the literature. Wren (1984) reported cadmium concentrations in raccoon liver and kidney tissues of 0.23 ± 0.09 and 1.22 ± 0.41 ppm, respectively. Raccoons from a Florida site had kidney concentrations of 2.48 ± 1.66 ppm (Hoff et al., 1977). Raccoons from east-central Michigan had cadmium concentrations in liver and kidney tissues which were above environmental background levels. One possible way in which raccoons were bioaccumulating cadmium may have been through food-chain biomagnification. Cadmium levels in soil were below detection limits, therefore it can be assumed that cadmium accumulation was chronic, not acute. Mirenda (1986) showed that cadmium accumulation in specific organs of the crayfish (Orconectes virilis) was directly related to cadmium concentrations in the aquatic system. Because raccoons depend upon aquatic systems for their food source (such as crayfish) they are highly susceptible to ingesting organisms which have accumulated high levels of cadmium via biomagnification or bioconcentration. Cadmium levels in this study were highest in the kidneys, followed by the liver. These findings agreed with other studies which reported the kidney as being the major storage site for cadmium (Wren, 1984; and Erickson and Lindzey, 1983).

Raccoons from this study had lead liver concentrations (Table 1) which were below those reported from a study by Hoff et al. (1977) in which raccoons from an uncontaminated site in Florida

Table 3. Concentrations (mg/kg) of PCB, DDE, and Lindane in adipose tissue of adult raccoons from the Shiawassee National Wildlife Refuge, Michigan, 1986.

Specimen	Sex	PCB	DDE	Lindane
1	M	5.96	1.00	0.18
2	F	2.90	0.39	0.03
3	M	26.3	0.89	0.07
4	F	24.6	0.98	0.06
5	M	9.16	0.29	< 0.01
6	M	9.50	0.05	< 0.01
7	F	3.80	0.14	0.02
8	M	23.0	0.92	0.03
9	M	19.8	0.23	0.02
X ± s.d.		13.8 ± 9.44	0.54 ± 0.39	0.04 ± 0.05

had lead liver concentrations of 0.47 ± 0.22 ppm. Raccoons from a contaminated site in Connecticut had lead liver concentrations of 6.2 ± 5.4 ppm (Ditters and Nielsen, 1978). Zinc and Selenium concentrations reported in this study (Table 1) were consistent with values reported in the literature. Wren (1984) reported zinc concentrations in raccoon liver and kidney tissues of 34.4 ± 2.0 and 29.5 ± 10.5 ppm, respectively, while Hoff et al. (1977) reported a kidney value of 75.88 ± 16.54 ppm. Both of these studies were at sites considered to be uncontaminated. Thus it appeared that zinc concentrations in raccoon tissues were somewhat variable which seems logical since zinc is an essential micronutrient and its concentrations in the body would be dependent upon the physiological state of the organism. Selenium liver concentrations in raccoons were 2.8 ± 1.2 ppm (Wren, 1984). The results from this study suggested that there is a homeostatic concentration range for seleium in the bodies of healthy raccoons. Mercury concentrations in raccoons from this study were below values given in the literature with respect to liver, kidney, and brain (Table 1). Wren (1984), Sheffy and St. Amant (1982), and Cumbie (1975) reported liver mercury concentrations in raccoons of 4.5 ± 3.5, 2.01 ± 0.93, and 3.54 ± 1.5 ppm, respectively. Kidney mercury levels were 1.1 ± 0.4, and 1.36 ± 0.4 ppm (Wren, 1984; Sheffy and St. Amant, 1982, respectively). Mercury levels in brain tissue were < 0.02 ppm (Sheffy and St. Amant, 1982). Sites from these three studies were basically considered uncontaminated, however, they were located in areas where mercury occurred at naturally high levels in soil and rock. The results from this study agreed with those of Sheffy and St. Amant (1982) in showing the liver to be the major storage site for mercury, followed by the kidney and brain.

Cadmium and mercury both showed age-related accumulation in this study. Adult raccoons had significantly higher cadmium levels in liver and kidney tissues than juveniles (Table 2). These findings agreed with those of McKinnon et al. (1976) and Erickson and Lindzey (1983) in which age-related variability of cadmium concentrations was shown in gray squirrels (Sciurus carolinensis) and muskrats (Ondatra zibethicus), respectively. Adult raccoons also had significantly higher mercury levels in liver, kidney, and brain tissues than juveniles (Table 2). No difference in lead concentrations was found between adults and juveniles. This supported the work of Sanderson and Thomas (1961) which showed no age-related accumulation of lead in raccoons. No differences in selenium or zinc levels were found between adults and juveniles. No sex-related differences in accumulation were found for any of the metals. Sanderson and Thomas (1961) also found no difference in lead accumulation between sexes. It appeared that sex was not a major determining factor for heavy metal accumulation in raccoons.

Selenium is known to provide some protection from methyl mercury poisoning in Japanese quail (Coturnix coturnix) (Stoewsand et al., 1974) and marine mammals (Koeman et al., 1975). Wren (1984) found a significant correlation ($r = 0.96$) between liver selenium and mercury levels in mammals. However, samples of raccoons, beaver (Castor canadensis), and otters (Lutra canadensis) were combined for a sample size of 12 and the effects of any species variability were ignored. The data from this study showed no correlation between liver selenium and mercury concentrations in raccoons ($r = 0.22$, $df = 15$).

Mean concentrations of DDE and Lindane in adipose tissue from raccoons in this study were similar to those reported in the literature (Table 3). Nalley et al. (1975) reported p,p-DDE levels in raccoons from Collier County, Florida of 0.66 ± 0.70 ppm and levels of p,p-DDE and Lindane in raccoons from Sarasota County of 0.83 ± 0.97 and 0.05 ± 0.04 ppm, respectively. No PCB's were found in raccoons from either county. PCB levels in raccoons from this study were considerably high, ranging from 2.90-26.3 ppm with a mean of 13.8 ± 9.44 ppm (Table 3). These results along with those of Montz et al. (1982) suggested that raccoons were relatively resistant to PCB exposure.

This study provided baseline data for the accumulation of heavy metals and OC compounds in various tissues of raccoons along with age and sex-related accumulation trends. This study also supported the idea that the raccoon would be a good biological indicator organism for a variety of contaminants (Bigler et al., 1975). Raccoons from this study accumulated both cadmium and PCB's to fairly high organ levels without showing any obvious deleterious effects. It has been shown that raccoons were relatively resistant to the toxic effects of lead (Sanderson and Thomas, 1961; and Ditters and Nielsen, 1978). Being resistant to the effects of contaminants is an important quality for an indicator organism because the organism is able to accumulate a compound above environmental levels assurring an unbiased sample. Sensitive animals show mortality at low levels and dead animals are not readily censused. Therefore those animals which are trapped are those with low concentrations of the compound which leads to the assumption that no contamination exists.

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