METAL RESIDUES IN BAT COLONIES, JACKSON COUNTY, FLORIDA, 1981-1983

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Abstract.—From 1970 to 1982 metals escaped from a battery salvage plant at Steel City, Jackson County, Florida. Chemical analyses of bat guano showed that cadmium was elevated at two nearby caves and at one more distant cave used by both endangered gray bats and southeastern bats. Further chemical analyses showed that cadmium was elevated in livers (2.4 times) and kidneys (3.3 times) of southeastern bats from nearby Judges Cave relative to southeastern bats from a Gainesville roost. The maximum concentration in kidneys from the Judges Cave colony (2.9 ppm) was less than minimum concentrations (3.41, 8.5, and 44 ppm) associated with various harmful effects in other mammalian species. We conclude that the bat populations have not been harmed by metal pollution.

Since 1970, wastes containing heavy metals have been escaping from a battery salvage plant at Steel City, Jackson County, Florida (Fig. 1). The plant ceased operations in January 1980, but pollutants continued to escape from the property for two more years. Heavy metals were thought to have contaminated Little Dry Creek and possibly portions of the Chipola River (Fig. 1). Damage to vegetation, fish, and shellfish has been described (Lynch 1981, Becker 1981). Concern developed for major colonies of the endangered gray bat (*Myotis grisescens*) and the southeastern bat (*M. austroriparius*) at Judges and Geromes Caves (Fig. 1) because these bats feed, in part, on insects whose larvae develop in the Chipola River. Geromes Cave is surrounded by peanut fields and pesticide contamination also seemed possible. Estimates of bat numbers at Geromes Cave declined between 1970 and 1981 (Wenner 1984). Our objective was to measure and evaluate contaminant levels in these bat colonies.

METHODS

Chemical analysis of a single guano sample from beneath a bat colony can provide an accurate indication of the pesticide contamination of the bats that deposited it (Clark et al. 1982). Other data at hand show a similar correspondence for lead. We have assumed the same is true for zinc and cadmium. On 22 October 1981, one of us (ASW) collected one guano sample each from Judges and Geromes Caves. A third location, Sneads Cave (Fig.

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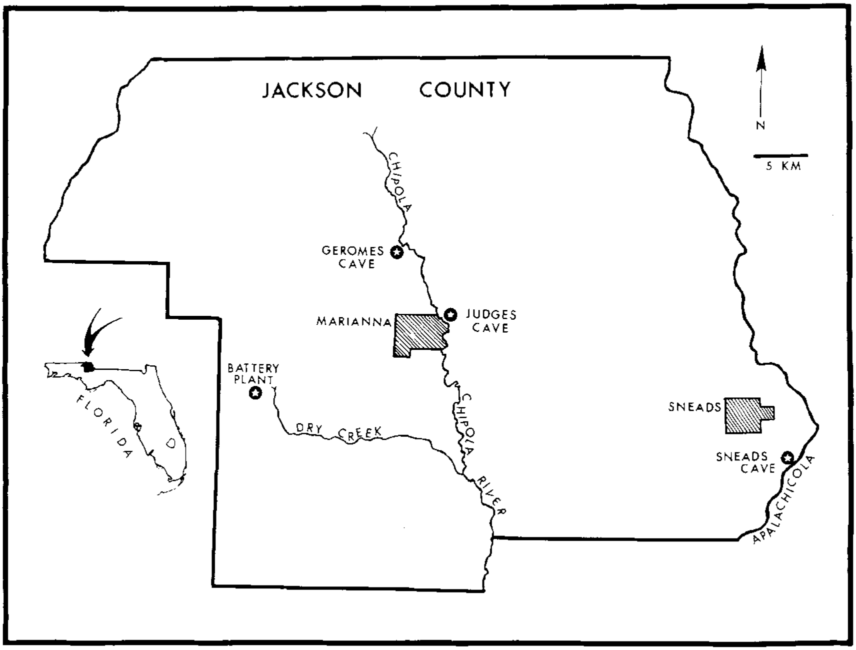

Figure 1. Locations of collection sites and battery salvage plant in Jackson County, Florida

1), was also sampled the same day as a "control" because its location is more distant from the metal contamination. Because Sneads Cave is surrounded by intensive agriculture, we also checked for possible exposure of the bats to pesticides. These guano samples, which may represent either species, or both, were analyzed at the Patuxent Wildlife Research Center for organochlorine pesticides and PCB's (polychlorinated biphenyls) and for lead, chromium, zinc, and cadmium. Because these results indicated that Sneads Cave did not have lower levels of metal contamination than Judges and Geromes Caves and thus might not be a suitable control site, a sample of big brown bat (*Eptesicus fuscus*) guano was collected at Patuxent in May 1982 and analyzed for lead, chromium, zinc, and cadmium. We have assumed that metal levels in guano from big brown bats not exposed to pollution sources would be similar to levels in the Florida samples if the Florida bats were also not exposed.

Comparisons of the samples suggested that three of the four metals (chromium, zinc, and cadmium) were elevated at both Judges and Sneads Caves. On this basis, 10 southeastern bats (1 male, 9 females) were collected at Judges Cave on 9 July 1983. As controls, another 10 southeastern bats (males) were collected beneath a highway bridge in Gainesville, Florida, on 27 October 1983. Bats were frozen and later shipped to Patuxent where livers and kidneys were removed for chemical analysis. Livers were analyzed for lead, chromium, zinc, and cadmium. Kidneys, because of their small size, were analyzed only for cadmium.

Judges and Geromes Caves are on the Chipola River about 11.3 and 16.1 km from the nearest contaminated part of that river (Fig. 1). Sneads Cave is on the Apalachicola River about 27.4 km from the contaminated Chipola River (Fig. 1).

Judges Cave, the most important gray bat cave in Florida, is a maternity cave in which 50,000 to 100,000 bats (10% gray, 90% southeastern) bred in 1983 when the cave was last checked. Geromes Cave is considered to be the second most important gray bat cave in Florida. In 1970 it contained 2,000 gray and 15,000 southeastern bats, but in 1982 only 200 bats (species ratio unknown) raised young there. Sneads Cave contained 100 male gray bats and 35,000 southeastern bats in 1970. In 1981, the number of bats was larger but the ratio of gray to southeastern bats was not determined. Data on the status and management of these caves and their bat populations were summarized by Wenner (1984).

Guano samples (6-12 g each) were desiccated at room temperature for 30 days with calcium sulfate. Samples for pesticide and PCB analyses were analyzed for *p,p'*-DDE, *p,p'*-DDD, *p,p'*-DDT, dieldrin, heptachlor epoxide, oxychlordane, *cis*-chlordane, *trans*-nonachlor, *cis*-nonachlor, endrin, toxaphene, and PCB's (quantified as Aroclor 1260). Samples were ground with anhydrous sodium sulfate and extracted in a Soxhlet apparatus. Pesticides and PCB's were separated by silica gel chromatography (Kaiser et al. 1980). Chemicals were quantified by electron capture gas liquid chromatography using a 1.5/1.95% SP 22-50/SP-2401 column. Average percentage recoveries from spiked Mallard (*Anas platyrhynchos*) tissue ranged from 85-103%, except *trans*-nonachlor was 75%. Residue levels were not corrected for percent recovery. The lower limits of reportable residues were 0.1 ppm (parts per million) for pesticides and 0.5 ppm for PCB's. Metal analyses of guano samples followed previously described procedures (Haseltine et al. 1981). The lower limits of reportable residues were 0.1 ppm for lead, cadmium and zinc, and 0.05 ppm for chromium.

For metal analyses of tissues, the entire organ (liver or both kidneys) was dissolved in 1 ml of concentrated nitric acid in a 15 ml polypropylene tube. Then the tube was placed in boiling water for 1 hr after which 0.5 ml of hydrogen peroxide was added and the tube was filled to 5 ml with distilled, deionized water. Zinc was determined by flame atomic absorption spectrophotometric (AAS) analysis using a Perkin-Elmer 5000 under the manufacturer's standard operating conditions. Lead, cadmium and chromium were determined by stabilized temperature Platform Furnace AAS techniques (Slavin et al. 1983). A Perkin-Elmer Zeeman 5000 equipped with a model 500 furnace controller, an AS-40 autosampler, and a Data System 10 with the HGA Graphics software were used for all determinations. Ammonium phosphate was the matrix modifier for lead and cadmium analyses; magnesium nitrate was the modifier for chromium. Standards were made in 5% nitric acid. Peak area measurements yielded characteristic masses of 11.6 pg for lead, 0.38 pg for cadmium, and 2.9 pg for chromium. Recoveries averaged 98% and residues were not corrected for percent recovery. The lower limits of reportable residues were 0.50 ppm for zinc, 0.025 ppm for lead, 0.0025 ppm for cadmium, and 0.05 ppm for chromium in livers and 0.025 ppm for cadmium in kidneys.

For comparisons with our wet weight data, we converted ppm dry weight values from two studies (Andrews et al. 1984, Hunter and Johnson 1982) to ppm wet weight by division by four on the basis of a dry/wet ratio in small rodent kidneys of about 1:4 (Linder pers. comm.). Because of positive skewness, residue data were log-transformed for statistical testing. Geometric means are given for residue data.

RESULTS

Guano samples contained only small amounts of some organochlorines. DDE was about 0.80 ppm in all three samples. Dieldrin at 0.13 ppm was found only in guano from Judges Cave and PCB at 0.81 ppm was present only at Sneads Cave.

In contrast, comparisons with the Patuxent sample indicate that levels of chromium, zinc, and cadmium were elevated in guano from Judges and Sneads Caves, and that cadmium also was elevated at Geromes Cave (Table 1). Lead levels at Geromes Cave were similar to lead in the Patuxent colony of big brown bats, a level that other data at hand indicate is only slightly elevated.

Analyses of tissues showed zinc in livers of bats from Judges Cave to be slightly (1.1 times) but significantly (t=2.63, $0.05>p>0.01$) elevated compared to the Gainesville Colony (Table 2). Cadmium levels in livers and kidneys of bats from Judges Cave were significantly elevated by 2.4 times (t=4.51, $p<0.001$) and 3.3 times (t=3.48, $0.01>p>0.001$) (Table 2).

Levels of cadmium in liver and kidney were positively correlated in both the Gainesville bats (r=0.837, $0.001>p>0.001$) and the bats from Judges Cave (r=0.852, $0.01>p>0.001$). The amount in the liver exceeded that in the kidney in most (6 of 10) Gainesville bats, whereas the amount in the kidney exceeded that in the liver in most (8 of 10) bats from Judges Cave.

Chromium was found at low levels (0.05, 0.06 ppm) in livers of only two Gainesville bats, and it was not found in livers of any bats from Judges Cave. Lead occurred in livers of five Gainesville bats (0.05, 0.26, 0.29, 0.41, and 0.58 ppm) and four Judges Cave bats (0.15, 0.17, 0.23, and 0.23 ppm). Overall lead seems higher among the Gainesville bats, probably due to the proximity of the highway.

Table 1. Concentrations of metals (ppm dry weight) measured in samples of bat guano.

Cave or Location	Metal			
	Lead	Chromium	Zinc	Cadmium
Judges	3.4	2.7	640.	2.2
Geromes	6.1	0.83	390.	1.9
Sneads	3.9	5.0	530.	2.3
PWRC[1]	7.1	0.54	340.	0.30

[1]Patuxent Wildlife Research Center.

Table 2. Concentrations of metals[1] (ppm wet weight) in 10 southeastern bats from each of two Florida locations.

	Liver		Kidney
Sex	Zinc	Cadmium	Cadmium
	Judges Cave		
F	30.	0.85	2.9
F	27.	0.59	1.5
F	31.	0.82	1.1
F	29.	0.49	0.80
F	35.	0.36	0.22
F	29.	0.56	0.67
M	35.	0.80	1.4
F	29.	0.70	1.0
F	32.	0.66	1.4
F	34.	0.49	0.28
Geom. mean	31.0	0.612	0.889
95% conf. int.	29.1-33.0	0.503-0.745	0.508-1.56
Range	27-35	0.36-0.85	0.22-2.9
	Gainesville		
M	30.	0.35	0.24
M	29.	0.34	0.27
M	28.	0.18	0.17
M	30.	0.75	2.1
M	28.	0.19	0.19
M	30.	0.17	0.15
M	26.	0.14	0.15
M	28.	0.41	0.31
M	29.	0.29	0.24
M	27.	0.14	0.24
Geom. mean	28.5	0.256	0.266
95% conf. int.	27.5-29.4	0.173-0.378	0.154-0.460
Range	26-30	0.14-0.75	0.15-2.1

[1]Because lead and chromium were not found in most bats, these residues are not presented here but are discussed in text.

Discussion

The guano data (Table 1) suggest that bats at Sneads Cave are as contaminated as those from Judges Cave even though Sneads is 16.1 km farther from the apparent source. This may result from a high rate of interchange of bats between these caves.

Amounts of DDE, dieldrin, and PCB's measured in guano were far below levels associated with harmful effects in bats (Clark 1978, Clark et al. 1982). Similarly, the lead concentration in guano from Judges Cave was far below that in stable populations of big brown and little brown bats (*M. lucifugus*) in Maryland (Clark 1979). Chromium did not accumulate in livers, and zinc was only slightly higher than in Gainesville bats. Cadmium is the contaminant most likely to affect these bats adversely.

The average level of cadmium (arithmetic average for purposes of comparison with literature values, 1.127±0.243 ppm, range 0.22-2.9 ppm) in bat kidneys from Judges Cave was similar to values (1.30±0.22 ppm, Andrews et al. 1984) for field voles (*Microtus agrestis*) from a mine waste area in England and to values (1.8±0.2 ppm) from woodmice (*Apodemus sylvaticus*) exposed to smelter emissions in England (Hunter and Johnson 1982). However, cadmium in kidneys of field voles from the smelter site (5.8±0.9 ppm, Hunter and Johnson 1982) was higher and cadmium in common shrews (*Sorex araneus*) from both sites (39.5±3.8 ppm, Andrews et al. 1984; 48.2±7.2 ppm, Hunter and Johnson 1982) was still higher than cadmium in bat kidneys. Thus the cadmium levels found at Judges Cave are not exceptional relative to those at other contaminated sites. Because these two English studies indicated that carnivory resulted in higher cadmium accumulation than did herbivory, the bat results may be more properly compared to the shrews than to the mice.

The question of whether cadmium levels found in bats at Judges Cave were high enough to damage the population was not answered. In a comparable study with male Norway (laboratory) rats (*Rattus norvegicus*), the lowest kidney concentration of cadmium associated with renal tubular lesions was 44 ppm (Kawai et al. 1976), well above both the mean (0.889 ppm) and the maximum (2.9 ppm) found among the Judges Cave bats.

Significantly reduced survivorship in young of enclosed populations of meadow voles (*Microtus pennsylvanicus*) was associated with kidney concentrations of cadmium that averaged as low as 3.41 ppm (Maly 1984). The experimental plots had been treated with sewage sludge that contained cadmium but no cause-effect relationship was proven. Studies with laboratory rats have shown that young were more sensitive (by LD_{50} tests) to cadmium than were adults (Kostial et al. 1978).

Male laboratory rats showed focal testicular necrosis, reduced spermatogenesis, and reduced fertility associated with about 8.5 ppm cadmium in the kidneys (Kotsonis and Klaassen 1977). However, Barlow and Sullivan (1982: 141) stated that testicular damage ". . . can occur in the absence of any other signs of Cd toxicity to other organs with much higher Cd contents than the testes." This implies that testicular damage may occur when kidney levels are less than 8.5 ppm. Nonscrotal mammalian species are generally insensitive to cadmium compounds (Barlow and Sullivan 1982: 141), however Dixit and Lohiya (1974) showed the rat-tailed bat (*Rhinopoma kinneari*) to be an exception, thus we expect that other bats may also be sensitive.

Nine of 10 bats from Judges Cave were females. Because Buhler et al. (1981) showed that female rats assimilate cadmium at a higher rate and retain more than do males, the levels of cadmium found in male bats in the contaminated colonies could be less than our data suggest. However, lead in mammals has also been found at higher levels in females than males except for the big brown bat where concentrations in males were significantly higher than females by about 1.5 times (Clark 1979). Thus our data may actually underestimate the accumulation in males. Another consideration is that testicular damage by cadmium may be prevented by simultaneous intake of zinc (Parizek 1957), and such intake of zinc obviously has occurred at Judges Cave.

The most recent population estimates (1981-83) indicated that bat numbers have remained stable or increased since 1970-71 at Judges and Sneads Caves but have decreased at Geromes Cave (Wenner 1984). Because metal levels in guano were lowest at Geromes, it seems unlikely that these contaminants have had any detrimental impact. This agrees with the overall picture gained from the literature in which the kidney concentrations of cadmium associated with harmful effects are higher than those found in these bats.

If cadmium concentrations in the contaminated colonies should increase, the first affects would probably be reduced male fertility or death of young. In a colony of tens of thousands, a contaminant that affects only 1% will effect hundreds of bats. With the basic plan for protection and management (Wenner 1984) in place, non-intrusive methods should be used to conduct regular censuses of these colonies and to look for die-offs. If numbers decline or dead are found, chemical analyses can be repeated to determine whether contaminants were involved.

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