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Bioavailability of Lead from Contaminated Sediment in Northern Bobwhites, *Colinus virginianus*

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Abstract. Sediment from Killarney Lake, Idaho was added to the diet of captive northern bobwhites (*Colinus virginianus*) to determine absorption of Pb from contaminated sediment. The sediment, containing 4,500 $\mu g\ g^{-1}$ Pb dry weight (d.w.), was added to ground poultry ration at 8% dry matter intake (DMI) for 21 days. Concentrations of Pb in blood, liver, and kidneys of each bobwhite were determined and compared to concentrations in untreated control bobwhites. Treated bobwhites showed no significant decline ($P > 0.05$) in food intake or body mass over time. In 90% of treated bobwhites, blood Pb concentrations reached levels associated with clinical Pb poisoning (>0.8 $\mu g\ g^{-1}$ wet weight, w.w.); and all treated bobwhites had elevated liver and kidney Pb concentrations. It was shown that tissue Pb accumulation can occur from ingestion of Pb-contaminated sediment.

Soil is an important component of ingesta of such domestic animals as cattle (Mayland *et al.* 1975; Kirby and Stuth 1980), sheep (Field and Purves 1964; Healy 1967) and swine (Fries *et al.* 1982). Soil ingestion occurs in mule deer (*Odocoileus hemionus*), pronghorns (*Antilocapra americana*), and jackrabbits (*Lepus californicus*) (Arthur and Alldredge 1979; Arthur and Gates 1988). As in the domestic species, the wild herbivores consume considerable quantities of soil. Further, some evidence exists that shorebirds ingest inorganic materials (sand, soil, and/or sediments). Reeder (1951) showed that inorganic materials in the alimentary tracts of eight species of shorebirds ranged from 10% to 60% of total contents. Work by Connor (1993) showed that mallards (*Anas platyrhynchos*) had an average soil intake of 12% DMI and Canada geese (*Branta canadensis*) averaged 5% DMI. Soil and sediment ingested by animals may contain a variety of contaminants and thereby act as vehicles by which animals are exposed to environmental contaminants.

Lead poisoning in waterfowl considered linked to Pb-contaminated sediment ingestion has been reported in mining areas of northern Idaho (Chupp and Dalke 1964; Benson *et al.* 1976; Krieger 1990; Blus *et al.* 1991). Lead concentrations in sediments from the area are very high, ranging from 2,000 to 8,000 $\mu g\ g^{-1}$ (Krieger 1990). However, it is well known that ingestion of Pb artifacts (Pb shot and fishing sinkers) is an important cause of Pb poisoning in waterfowl in many areas (Sanderson and Bellrose 1986). Unfortunately, there is little information on the relative importance of sediment ingestion as a route of contaminant exposure to wildlife. The objective of this study was to determine whether ingestion of Pb-contaminated sediment produces elevated tissue concentrations of Pb in birds, using northern bobwhites (*Colinus virginianus*) as a model.

Materials and Methods

Sediment containing 4,500 $\mu g\ g^{-1}$ Pb (d.w.) was collected from Killarney Lake in northern Idaho, an area with extensive Pb contamination due to mining wastes. The sediment was dried at 55°C, sieved through an 840 μm sieve, and added to ground poultry ration (Southern States Gamebird Starter and Grower Medicated, Richmond) at 8% d.w. Fifteen male northern bobwhites 17 weeks of age were assigned to one of two groups (seven treated, eight untreated controls). Bobwhites were housed individually in stainless steel hanging wire rodent cages (20 cm × 18 cm × 18 cm) and provided with water and food *ad libitum*. Bobwhites were acclimated to the hanging cages and a ground poultry ration for 14 days prior to the study. Controls were fed ground poultry ration and treated bobwhites received the ground poultry ration spiked with 8% sediment for 21 days. Daily food intake by each bobwhite was measured. Blood (0.4–1 ml) was collected the final day of the study from each bobwhite via the jugular vein using a needle with a heparinized syringe. Body mass of each bobwhite was measured on day (D) 0, 7, 14, and 21 of the study. On D 21, all bobwhites were sacrificed via cervical dislocation. Liver and kidneys of each bobwhite were removed, freeze-dried, and prepared for Pb analysis using the methods of Sandell (1950) with the following modifications. Freeze dried samples (0.2–0.5 g) were digested in 4 ml nitric acid at room temperature overnight in acid-washed 50 ml digestion tubes. Perchloric acid (2 ml) was then added to each tube and digested on a block digestor until samples became colorless and tubes contained about 1 ml of liquid. Samples were diluted to 12.5 ml without filtration and analyzed for Pb concentration via ICP spectrometry. Spectrometry was conducted by the VPI&SU Soil Testing and Plant Analysis Laboratory using a Thermo Jarrell Ash ICAP 9000 (upgraded to 61). Quality

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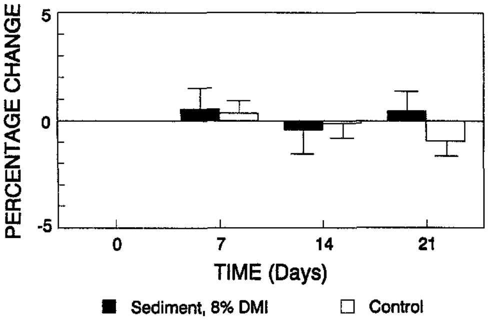

Fig. 1. Percentage change from initial body mass (Day 0) over time in control bobwhites (*Colinus virginianus*) (n = 8) and bobwhites fed Pb-contaminated sediment at 8% of intake (n = 7) for 21 days. Data are expressed as the mean with standard error in the positive direction

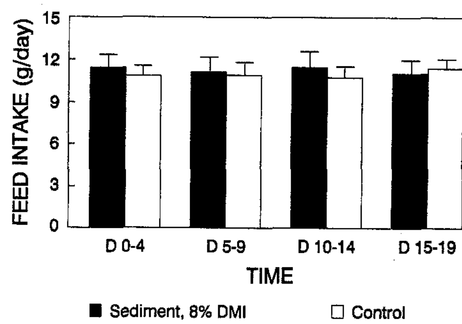

Fig. 2. Daily food intake (g/day) by control bobwhites (*Colinus virginianus*) (n = 8) and bobwhites fed Pb-contaminated sediment at 8% of intake (n = 7) for 21 days. Data are expressed as the mean with standard error in the positive direction

control standards were made from stock solution and run periodically. In addition, one spiked sample was run per 30 test samples analyzed. Blood samples were diluted five times with 0.1 N HNO_3 and analyzed for Pb concentration via ICP spectrometry. The detection limit of the ICP was 0.1 μg g^{-1}.

Statistical Analysis

Statistical analyses were conducted using the Statistical Analysis System (SAS Institute Inc 1987). An ANOVA with repeated measures was used to analyze the effect of time and/or treatment on food intake and percentage change from initial body mass. Statistix Version 3.5 (Analytical Software 1991) was used to conduct a Mann Whitney U-test to analyze the effect of treatment on blood, liver, and kidney Pb concentrations. A value of one half the detection limit was used for Pb concentrations determined to be below the detection limit of the ICP spectrophotometer. An alpha level of 0.05 was used for all statistical inferences.

Results

Body Mass

There was no time × treatment interaction for percentage change from initial body mass over time, but there was a significant time effect ($P = 0.0460$). Overall, mean percentage change in body mass in both treated and control bobwhites did not vary by more than ±1% (Figure 1). There was no significant difference in percentage change from initial body mass between treated bobwhites and control bobwhites on D 7, D 14, or D 21 of the study.

Food Intake

For mean daily food intake during D 0–4, D 5–9, D 10–14, and D 15–19 (Figure 2), there was no time × treatment interaction and no time effect. There was also no significant difference between treated and control bobwhites in mean daily food intake at any time during the study.

Blood Lead

There was a significant difference ($P < 0.001$) between blood Pb of controls and treated bobwhites (Table 1). Analysis of blood revealed controls had a mean (±S.E.) blood Pb concentration of 0.63 (±0.04) μg g^{-1} (w.w.) and a range of 0.50 to 0.75 μg g^{-1}. Treated bobwhites exhibited blood Pb concentrations ranging from 0.65 to 1.75 μg g^{-1} (w.w.) and had a mean concentration of 1.26 (±0.14) μg g^{-1}. All bobwhites had blood Pb concentrations greater than 0.15 μg g^{-1} (w.w.). Of the seven treated bobwhites, six exhibited blood Pb concentrations >0.8 μg g^{-1} (w.w.), which is considered indicative of clinical Pb poisoning in mallards (Scheuhammer 1989).

Liver and Kidney Lead

Liver and kidney Pb concentrations of all controls were below the detection limit of the ICP (<0.1 μg g^{-1}). However, all treated bobwhites had Pb present in the liver and kidneys (Table 1). Liver Pb ranged from 5 to 11 μg g^{-1} (d.w.) with a mean of 7 μg g^{-1}. Mean kidney Pb of treated bobwhites was 30 μg g^{-1} (d.w.) and ranged from 17 to 48 μg g^{-1}.

Discussion

The results of this study demonstrated that northern bobwhites accumulated tissue residues of Pb as a result of ingesting Pb-contaminated sediment in the diet. However, the birds did not exhibit any overt indications of Pb toxicity, and neither body mass nor food intake of bobwhites were affected over the 21-day feeding period. No Pb concentrations were detected in liver or kidneys of controls.

Scanlon *et al.* (1980) suggested up to 10 μg g^{-1} Pb (d.w.) in liver as a "background level" for Pb exposure in wild mallards. Thus, the mean liver Pb concentration of treated bobwhites was not high enough to be considered above "background levels" in wild populations. Longcore *et al.* (1974) suggested Pb levels of 6–20 μg g^{-1} (w.w.) in liver and kidneys as indicative of serious Pb intoxication in mallards. These levels convert to approximately 18–61 μg g^{-1} (d.w.) in liver and 26 to 87 μg g^{-1} in kidneys based on conversion factors developed by Scanlon (1982). Thus, tissue residues in our bobwhites did not indicate

Table 1. Influence of Pb-contaminated sediment ingestion by northern bobwhites (*Colinus virginianus*) on Pb concentrations (mean ± S.E.) in liver, kidneys, and blood

Parameter	Diet	
	Sediment, 8% DMI[a]	Control
N	7	8
Liver, $\mu g\ g^{-1}$ (d.w.)	7.11 ± 0.81^{b}	$<0.10^{c}$
Kidney, $\mu g\ g^{-1}$ (d.w.)	30.33 ± 6.95^{b}	$<0.10^{c}$
Blood, $\mu g\ g^{-1}$ (w.w.)	1.26 ± 0.14^{b}	$0.63 \pm 0.04^{c,d}$

[a] Dry matter intake
[b,c] Row means having different superscripts are significantly ($P < 0.002$) different from one another
[d] Based on seven samples due to loss of one sample

Pb poisoning. However, using 0.8 $\mu g\ g^{-1}$ blood Pb as an indicator of clinical Pb poisoning in waterfowl (Scheuhammer 1989), blood Pb concentrations of treated bobwhites were well above those which are considered indicative of Pb poisoning.

The bobwhites in this study ingested, on average, a total of approximately 83 mg of Pb, or 4 mg per day for the 21-day feeding period. Dieter and Finley (1979) found that mallards dosed with one Pb shot (No. 4, 200 mg) showed no mortality or significant changes in body mass; mean Pb concentrations (w.w.) were 1 $\mu g\ g^{-1}$ in blood and 2 $\mu g\ g^{-1}$ (w.w.) in liver, or 7 $\mu g\ g^{-1}$ (d.w.) (Scanlon 1982). Thus, Pb concentrations in their birds were nearly identical to those in this study. This suggests that Pb ingested chronically in the form of sediment is more available than Pb from an acute dose from Pb shot ingestion. Dieter and Finley (1979), found that at 1 $\mu g\ g^{-1}$ (w.w.) Pb in blood, δ-ALAD enzyme activity in blood was reduced by 75% and by 35–50% in the brain and liver. The authors concluded that δ-ALAD inhibition in cells of critical organs such as the brain, could lead to reduced survival due to impaired motor, visual, and auditory responses. The ultra sensitivity of δ-ALAD to Pb in the blood and high variability in unexposed birds, however, may limit its use as an indicator of debilitating exposure to Pb (Pain 1989). Based on tissue and blood Pb residues and the physical characteristics of the bobwhites in our study, it is difficult to assess the effects of Pb exposure to bobwhites. Bobwhites in this study were provided a well-balanced, high protein diet (26%) and an optimal thermal environment which may have reduced the toxic effects of Pb.

Blus *et al.* (1991) collected blood from 16 tundra swans (*Cygnus columbianus*) from an area of northern Idaho near the area where the sediment for this study was obtained. Blood Pb in the swans ranged from 0.5 to 1.6 $\mu g\ g^{-1}$, yet all appeared healthy. There was a significant ($P < 0.01$) relationship between blood Pb and hemoglobin, hematocrit, protoporphyrin, and ALAD in these swans. Further, Blus *et al.* (1993) found high Pb concentrations in livers (0–14 $\mu g\ g^{-1}$, w.w.) and blood (0–5 $\mu g\ g^{-1}$) of wood ducks (*Aix sponsa*) from the same area of Idaho. Yet, the ducks showed no obvious signs of Pb poisoning or reduced nesting success. The authors suggested that current blood Pb criterion may be too low for most birds due to interspecific differences. It is also possible that Pb from sediments is less toxic to wildlife than Pb from shot when ingested due to the nature of exposure. Lead in sediment is ingested chronically and at low concentrations, possibly enabling animals to adapt to the low doses of exposure. Over long periods of exposure or times of high stress, animals may then succumb to the toxic effects of Pb.

Despite the lack of evidence of deleterious effects of Pb on the bobwhites in this study, it was demonstrated that ingestion of Pb-contaminated sediment resulted in significant accumulation of Pb in tissues and blood. Sediments containing numerous contaminants, if ingested, could be particularly harmful to wildlife. Additional research is needed to determine the effects of contaminated sediments on wildlife. Soil and sediment ingestion must be considered as an important route of exposure to contaminants by wildlife in future risk assessment studies.

References

Analytical Software (1991) Version 3.5, Analytical Software, St. Paul, MN

Arthur WJ, Alldredge AW (1979) Soil ingestion by mule deer in northcentral Colorado. J Range Manage 32:67–71

Arthur WJ, Gates RJ (1988) Trace element intake via soil ingestion in pronghorns and in black-tailed jackrabbits. J Range Manage 41:162–166

Benson WW, Brock DW, Gabica J, Loomis M (1976) Swan mortality due to certain heavy metals in the Mission Lake Area, Idaho. Bull Environ Contam Toxicol 15:171–174

Blus LJ, Henny CJ, Hoffman DJ, Grove RA (1991) Lead toxicosis in tundra swans near a mining and smelting complex in northern Idaho. Arch Environ Contam Toxicol 21:549–555

Chupp NR, Dalke PD (1964) Waterfowl mortality in the Coeur d'Alene River Valley, Idaho. J Wildl Manage 28:692–702

Connor EE (1993) Soil ingestion and lead concentration in wildlife species. M.S. Thesis. Virginia Polytechnic Institute and State University, Blacksburg, VA

Dieter MP, Finley MT (1979) δ-Aminolevulinic acid dehydratase enzyme activity in blood, brain, and liver of lead-dosed ducks, Environ Res 19:127–135

Field AC, Purves D (1964) The intake of soil by the grazing sheep. Proc Nutr Soc 23:xxiv–xxv

Fries GF, Marrow GS, Snow PA (1982) Soil ingestion by swine as a route of contaminant exposure. Environ Toxicol Chem 1:201–204

Healy WB (1967) Ingestion of soil by sheep. Proc New Zealand Soc Anim Prod 27:109–120

Kirby DR, Stuth JW (1980) Soil ingestion rates of steers following brush management in central Texas. J Range Manage 33:207–209

Krieger RI (1990) Toxicity and bioavailability studies of lead and other elements in the Lower Coeur d'Alene River. Idaho BLM Tech Bull 90–3, Boise, ID

Mayland HF, Florence AR, Rosenau RC, Lazar VA, Turner HA (1975) Soil ingestion by cattle on semiarid range as reflected by titanium analysis of feces. J Range Manage 28:448–452

Pain DJ (1989) Haematological parameters as predictors of blood lead and indicators of lead poisoning in the black duck (Anas rubripes). Environ Pollut 60:67–81

Reeder WG (1951) Stomach analysis of a group of shorebirds. Condor 53:43–45

Sandell EB (1950) Colorimetric determinations of trace metals. Wiley-Interscience, NY

Sanderson GC, Bellrose FC (1986) A review of the problem of lead poisoning in waterfowl. Illinois Natural History Survey Special Publication 4, Champaign, IL

SAS Institute Inc (1987) Release 6.04 Edition. SAS Institute Inc, Cary, NC

Scanlon PF (1982) Wet and dry weight relationships of mallard

(*Anas platyrhynchos*) tissues. Bull Environ Contam Toxicol 29:615–617
Scanlon PF, Stotts VD, Oderwald RG, Dietrick TJ, Kendall RJ (1980) Lead concentrations in livers of Maryland waterfowl with and without ingested lead shot present in gizzards. Bull Environ Contam Toxicol 25:855–860
Scheuhammer AM (1989) Monitoring wild bird populations for lead exposure. J Wildl Manage 53:759–765