

LEAD CONCENTRATIONS AND REPRODUCTION IN HIGHWAY-NESTING BARN SWALLOWS

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ABSTRACT.—Lead concentrations in the carcasses and stomach contents of adult and nestling Barn Swallows (*Hirundo rustica*) collected within the right-of-way of a major Maryland highway were greater than those found in Barn Swallows nesting within a rural area. Lead concentrations in the feathers of adults from the highway colony were also greater than those of rural adults, but concentrations in the feathers of nestlings from the two locations were similar. Activity of δ -aminolevulinic acid dehydratase in red blood cells was lower in highway-nesting adults and their young than in their rural counterparts, although hemoglobin concentrations and hematocrits did not differ. The number of eggs, nestlings, and body weights of the latter at 16–18 days of age were similar in the two colonies, as were body weights of adults from the two areas. These results suggest that contamination of roadside habitats by lead from automotive emissions does not pose a serious hazard to birds that are aerial feeders.

Birds inhabiting highway rights-of-way (30.4 million ha in the United States [Smith 1976]) may be exposed to lead from automotive exhaust through inhalation or ingestion of contaminated food or grit. Twenty-two to 58% of the lead emitted from motor vehicles (106–118 thousand metric tons in the United States in 1975 alone [Provenzano 1978]) accumulates in the soil or vegetation within highway rights-of-way (Ward et al. 1975, Little and Wiffen 1978). Accumulation varies with traffic density (Wheeler and Rolfe 1979) and distance from the road surface (Little and Wiffen 1978, Ward et al. 1979). Lead concentrations as high as 6,835, 1,180, and 682 ppm dry weight have been reported in soils, vegetation, and invertebrates, respectively (Williamson and Evans 1972, Little and Wiffen 1978).

Lead concentrations have been measured in urban and rural pigeons (*Columba livia*; e.g., Hutton 1980, Hutton and Goodman 1980, Ohi et al. 1981), doves (Siegfried et al. 1972), and songbirds (Getz et al. 1977). However, the effects of this metal on their survival and reproduction have not been assessed, although dietary concentrations of lead as low as 1 ppm reduced egg production in Japanese Quail (*Coturnix coturnix japonica*, Edens et al. 1976).

The objective of our study was to compare lead concentrations, hematological parameters, and reproductive success in two colonies of Barn Swallows (*Hirundo rustica*), one within the right-of-way of a major highway (Baltimore-Washington Parkway), the other within a nearby rural area (reference colony), to de-

termine if reproduction in birds that are aerial feeders is adversely affected by lead from motor vehicle emissions. We selected Barn Swallows for study because they frequently use highway bridges and culverts as nest sites (Jackson and Burchfield 1975, Barr 1979). The Parkway was selected as a study site because concentrations of lead had been determined in soil and grass (Chow 1970), invertebrates (Gish and Christensen 1973, Beyer and Moore 1980), and small mammals (Clark 1979) collected there. Analyses of stomach contents and guano samples from bats collected close to this Parkway suggested that some species of flying insects in the area contained high concentrations of lead (Clark 1979).

METHODS

The Parkway colony was located beneath a bridge above the Patuxent River (39°4'10"N, 76°49'53"W). According to the Maryland Department of Transportation, the average daily traffic volume (ADT) on the bridge in 1979 was 37,925 vehicles per day. The right-of-way was bordered by eastern deciduous forest; the median at each end of the bridge was covered by short grass. The reference colony was located in two adjacent barns on a small farm in rural Howard County, Maryland (39°12'50"N, 76°58'18"W), about 0.4 km from a lightly traveled secondary road (ADT = 330 vehicles per day in 1979; Howard County Public Works). The surroundings of the reference colony were typical of colonies on farms in eastern Maryland, consisting of pasture, crop-

land (corn fields), and a large water impoundment. This colony was selected as the reference colony because of its accessibility, distance from a major road, and similarity to the Parkway colony in the number of nests present.

During May, June, and July 1979, we monitored reproductive activity in the two colonies. Forty nests in each colony, containing first clutches, were checked every two days until hatching to determine clutch size, and checked again 16–18 days after the eggs hatched. At this time, nestlings were counted and collected. Two were randomly selected from each of 14–21 nests in each colony in order to determine the δ -aminolevulinic acid dehydratase (ALAD, E.C. 4.2.1.24) activity (a sensitive indicator of lead exposure in birds [Hoffman et al. 1981, Ohi et al. 1981]) and hemoglobin content of red blood cells (RBC), hematocrits, and lead concentrations in stomach contents, carcasses, and feathers (a lead accumulator [Nezel and Vogt 1976]). To make our selection random, we removed entire broods from their nests, assigned a number to each nestling within a brood, and chose the nestlings to be sacrificed using a random number table. The remaining nestlings were anesthetized by placing them in a plastic bag with cotton lightly moistened with Metafane^R (Pitman-Moore, Inc., Washington Crossing, NJ. [Use of trade names or names of suppliers is for identification purposes only and does not constitute endorsement by the federal government.]), and returned to their nests. We anesthetized the nestlings so that they would not fledge prematurely when replaced in their nests. We tried to collect nestlings at each colony on the same dates. After most of the young from these first clutches had fledged, we mist-netted 10–13 adults of each sex at each location to determine the ALAD activity and hemoglobin content of RBCs, hematocrits, and lead concentrations in the parts enumerated above.

We weighed all birds collected for blood and lead analyses and obtained a 400- μ l blood sample from each bird by decapitating it. Blood samples were kept on ice prior to analyses. We determined ALAD activity in duplicate within 3 h of collection with methods described by Burch and Siegel (1971), using 50 μ l of blood from nestlings because of their high ALAD levels, but 100 μ l from adults. We defined a unit of ALAD activity as the increase in absorbance (at 555 nm, 1.0-cm light path) per ml RBCs per h at 38°C. Hematocrits were determined shortly after blood samples were collected using the microhematocrit method (Davidsohn and Nelson 1974:115). Additional blood was used to determine hemoglobin concentrations with the cyanomethemoglobin method (Hycel, Inc., Houston, TX).

The entire plumage, bill, feet, and gastrointestinal tract were removed from each specimen. Stomach contents of nestlings in each colony were pooled for lead analysis; those of adults in each colony were pooled by sex. Stomach contents and carcasses were then frozen (-15°C) until analyzed for lead. Feathers (entire plumage) were thoroughly washed in 0.1 % Eastman^R Triton X-100 (Eastman Kodak Co., Rochester, NY) to remove lead particles adsorbed to the surface. Remiges and rectrices were cut into pieces to facilitate washing. Feathers were mechanically agitated in the surfactant for 30 min, drained, and then rinsed thoroughly in deionized water. This process was repeated five times for each sample. (Microscopic examination of selected samples of feathers before and after washing indicated that the majority of the dirt particles had been removed.) Afterwards, feathers were dried in an oven at 90°C for 48 h. Samples of feathers, as well as carcasses and stomach contents, were then transferred to separate chemically-clean bottles, weighed, dried for at least 48 h at 90°C , and then reweighed. Twenty milliliters of concentrated HNO_3 was then added to each sample and it was allowed to stand for at least one week with mixing at least once a day. Samples were then transferred to 100-ml Kjeldahl flasks, additional HNO_3 was added, and the contents reduced to a volume of 3 ml by heating. Carcass and feather samples were then diluted to 50 g with deionized water, whereas stomach contents were diluted to 10 g.

Lead concentrations in stomach contents, carcasses, and feathers were determined (by the Environmental Trace Substances Research Center, University of Missouri, Columbia) with a furnace-atomic absorption spectrophotometer (Perkin-Elmer Model 403 with a HGA-2100 graphite furnace) and an AS-1 auto sampler. Background correction and the method of standard additions were used. The lower limit of reportable residue was 0.01 ppm. High values were checked by flame-atomic absorption spectrophotometry and agreement was satisfactory. Lead concentrations (ppm) are expressed on a dry weight basis unless noted otherwise. Averages of lead concentrations are presented as geometric means because the data were not normally distributed.

We netted an additional 23 adults from the Parkway colony, individually color-marked their feathers, and then released them in order to estimate the number of birds within the colony that actually foraged within the right-of-way. (Some swallows from the Parkway colony had been seen foraging outside of the right-of-way.) We observed swallows from two locations on each side of the Parkway, one on each side of the bridge, between 06:00 and

TABLE 1. Blood ALAD (units), hemoglobin (g/dl), and hematocrit (%) in Barn Swallows from the right-of-way of the Baltimore-Washington Parkway and a nearby rural area (reference). Values in the table are $\bar{x} \pm SD (n)$.*

Blood parameter	Age	Sex	Parkway colony	Reference colony	Difference (%)
ALAD	Adult	Male	109 \pm 29 (10)a	157 \pm 51 (10)b	-30.6
	Adult	Female	96 \pm 26 (13)a	146 \pm 66 (9)b	-34.2
	Nestlings**	—	249 \pm 51 (19)c	289 \pm 62 (12)c	-13.8
Hemoglobin	Adult	Male	15.0 \pm 1.5 (10)a	15.9 \pm 2.3 (9)a	-5.7
	Adult	Female	14.7 \pm 1.4 (13)a	15.2 \pm 1.1 (10)a	-3.3
	Nestlings**	—	11.2 \pm 2.1 (21)b	11.3 \pm 2.7 (14)b	0.9
Hematocrit	Adult	Male	49.6 \pm 5.2 (10)a	51.9 \pm 2.7 (10)a	-4.5
	Adult	Female	50.3 \pm 2.8 (13)a	52.9 \pm 4.8 (10)a	5.2
	Nestlings**	—	40.7 \pm 2.9 (21)b	41.9 \pm 4.5 (14)b	-2.9

* For each blood parameter, values followed by different letters differ significantly ($P < 0.05$, two-way ANOVA with Bonferroni mean separation [Neter and Wasserman 1974: 473-482]).

** Each observation (n) is an average for two 16- to 18-day-old nestlings randomly selected from each nest in the sample (n).

08:00 (east side) and 18:00 and 20:00 (west side) for 3 consecutive days and identified the color-marked individuals that foraged within the right-of-way during each observation period. We believed these data would facilitate interpretation of blood and lead analyses from the Parkway colony where results may have been affected by differences in foraging behavior. We did not conduct a comparable study at the reference colony because the swallows there appeared to restrict their foraging to the fields and impoundment near the barns.

RESULTS

Activity of RBC ALAD was 30-34% lower in adults from the Parkway, and nearly 14% lower in their nestlings compared to levels in their rural counterparts (Table 1). These results are consistent with our observations on the foraging behavior of individually color-marked swallows from the Parkway colony: we saw 15 (62.5%) of the 23 marked swallows foraging within the right-of-way. ALAD levels in nestlings from both colonies were about twice the levels in adults. However, differences in hematocrits and hemoglobin concentrations between swallows from the two colonies were not significant ($P > 0.05$; Table 1). In contrast,

hematocrits and hemoglobin concentrations in nestlings from both colonies were significantly lower than those of adults.

Lead concentrations in stomach contents and carcasses of adults and nestlings from the Parkway colony were greater than those in the same parts of swallows from the reference colony (Table 2). Concentrations in pooled stomach contents of Parkway nestlings and adults were 1.5-24 times those of their rural counterparts. Concentrations in the carcasses of Parkway adults (range = 3.4-8.0 ppm in males and 2.7-21 ppm in females) and adults from the reference colony (range = 2.4-5.7 ppm in males and 1.7-13 ppm in females) were about six times those of 16- to 18-day-old nestlings. Differences in carcass lead concentrations between the sexes were not statistically significant at either colony. Lead concentrations in the carcasses of Parkway nestlings (range = 0.36-5.4 ppm) were about twice those in nestlings from the reference colony (range = 0.50-10 ppm).

Lead concentrations in feathers of Parkway adults (range = 49-102 ppm in males and 30-115 ppm in females) were about three times those in feathers of adults from the reference colony (range = 19-36 ppm for males and 13-

TABLE 2. Lead concentrations (ppm dry weight) in stomach contents, carcasses, and feathers of Barn Swallows within the right-of-way of the Baltimore-Washington Parkway and a nearby rural area (reference). Values in the table are geometric mean (95% confidence interval) [n].*

Part of bird	Age	Sex	Parkway colony	Reference colony
Stomach contents**	Adult	Male	4.8 (-) [1]	0.20 (-) [1]
	Adult	Female	6.8 (-) [1]	1.8 (-) [1]
	Nestlings	—	3.2 (-) [1]	2.3 (-) [1]
Carcasses	Adult	Male	5.1 (4.2-6.3) [10]a	4.0 (3.4-4.8) [10]b
	Adult	Female	8.8 (6.4-12) [13]a	4.5 (3.0-6.6) [10]b
	Nestlings***	—	1.5 (1.3-1.8) [21]c	0.69 (0.68-1.4) [14]d
Feathers	Adult	Male	67 (55-82) [10]a	24 (21-28) [10]b
	Adult	Female	54 (43-68) [13]a	19 (16-22) [10]b
	Nestlings***	—	2.5 (2.3-2.7) [21]c	2.3 (2.0-2.6) [14]c

* For each part of the bird, values followed by different letters differ significantly ($P < 0.05$, two-way ANOVA with Bonferroni mean separation).

** Pooled samples.

*** Each observation (n) is an average for two 16- to 18-day-old nestlings randomly selected from each nest in the sample (n).

TABLE 3. Comparison of reproductive success of Barn Swallows nesting within the right-of-way of the Baltimore-Washington Parkway and a nearby rural area (reference colony) with that reported for other colonies.

Reference	No. of nests	No. of eggs laid	Mean clutch size	Mean no. of young fledged/nest	Productivity (%) ^a
Parkway colony, 1979 ^b	40	197	4.9	4.0 ^c	81.2
	53	263	5.0	—	—
Reference colony, 1979 ^b	40	181	4.5	1.3 ^c	27.2
	55	251	4.6	—	—
Barr (1979)	136	625	4.6	3.1	68.0
McGinn and Clark (1978) ^d	—	—	4.7	—	76.3
Altemus (1977)	14	61	4.4	3.6	96.1
Snapp (1976) ^d	376	1,750	4.7	3.5	74.2
Samuel (1971)	94	430	4.6	3.7	81.2
Mizuta (1963)	47	245	5.2	—	—
	65	—	—	4.3	—
Adams (1957) ^d	441	1,906	4.3	3.1	71.9

^a Percentage of eggs laid from which young fledged.

^b First row = data for nests monitored until the young fledged; second row = data for all nests in the colony.

^c Number of 16- to 18-day-old young in nest.

^d Includes second clutches.

24 ppm for females; Table 2). Differences between the sexes were not statistically significant in either colony. Feathers of Parkway adults contained about 20 times more lead than feathers of their nestlings (range = 1.7–4.3 ppm), whereas those from adults in the reference colony contained about 10 times the lead in feathers of their young (range = 1.4–5.4 ppm). However, the lead content of the feathers of nestlings from the two colonies was similar.

Reproductive success in the Parkway colony was similar to that reported for other colonies of Barn Swallows (Table 3). However, the productivity of the reference colony was unusually low, owing to predation of eggs and nestlings in the barns. Body weights of adults and 16- to 18-day-old nestlings from the Parkway were similar to those of their rural counterparts (Table 4). Nestlings were significantly heavier than the adults in both colonies.

DISCUSSION

Results of the blood and lead analyses for the two colonies of Barn Swallows indicate that the lead intake (dietary and respiratory) of Parkway adults and nestlings was greater than that of their rural counterparts. However, we did not observe any overt signs of lead poisoning (e.g., weight loss, paralysis in wings and legs, or loss of vision; Osweiler and Van Gelder 1978) in swallows from the Parkway colony. Nor did we detect any impairment of reproduction similar to that reported in other avian species exposed to lead under laboratory conditions. For example, dietary lead has decreased eggshell quality and resulted in shell-

less or soft-shelled eggs by interfering with calcium metabolism (e.g., Edens et al. 1976). Japanese Quail fed 1–100 ppm lead experienced delayed sexual maturity, reduced fecundity, and lower hatchability of their eggs (Edens et al. 1976). Dietary concentrations of 500 ppm or more inhibited growth in several avian species (e.g., Damron et al. 1969, Edens et al. 1976). Levels of lead in the stomach contents of our Parkway swallows were, however, generally below those known to affect avian reproduction. Therefore, it is probably not surprising that the clutch sizes, productivity, and body weights of Parkway adults, as well as the body weights of their nestlings, were similar to those of birds from the reference colony or other colonies of Barn Swallows (Table 3). Subtle lead-induced physiological and behavioral abnormalities not detected in our study may nevertheless reduce the survival of juvenile and adult swallows in the Parkway colony. Lead exposure has been shown to alter nerve function (Hunter and Haigh 1977), levels of brain enzymes (Dieter and Finley 1979), behavior (Barthalmus et al. 1977), and the immune response (Truscott 1970) in birds.

We found a 31–34% difference in the RBC ALAD activity of adult Barn Swallows in the two colonies, and a 14% difference between levels in their nestlings. Although cadmium and zinc also accumulate in roadside and urban environments (Gish and Christensen 1973, Hutton and Goodman 1980) and inhibit RBC ALAD activity, they only inhibit the activity of this enzyme at levels 10–100 times those of lead (Davis and Avram 1978). Low concentrations of cadmium and zinc actually stimulate ALAD activity in RBCs (Davis and Avram 1978). Since concentrations of lead in soils within the right-of-way of the Parkway were as much as 440 times that of cadmium and three times that of zinc in 1970 (Gish and Christensen 1973), the differences we noted in RBC ALAD activity between the two colonies probably resulted from differences in the birds' exposure to lead, and not other metals.

We do not know the biological implications of the differences in the RBC ALAD activity we found between the two swallow colonies. This enzyme is necessary for the synthesis of heme, which is incorporated into hemoglobin within erythrocytes and into mitochondrial cytochromes (Sassa et al. 1975). Inhibition of ALAD in cells of critical organs could jeopardize metabolic and detoxifying processes (Sassa et al. 1975). However, the difference in RBC ALAD that we observed was not as pronounced as that reported in birds in which other lead-induced physiological abnormalities have occurred. For example, the activity

TABLE 4. Body weights (g) of Barn Swallows from within the right-of-way of the Baltimore-Washington Parkway and a nearby rural area (reference). Values in the table are $\bar{x} \pm SD (n)$.*

Age	Sex	Parkway colony	Reference colony
Adult	Male	17.9 \pm 0.6 (10)a	17.8 \pm 1.1 (10)a
Adult	Female	18.8 \pm 1.1 (13)a	18.0 \pm 1.8 (10)a
Nestlings**	—	19.5 \pm 1.2 (21)b	19.5 \pm 1.2 (14)b

* Values followed by different letters differ significantly ($P < 0.05$, two-way ANOVA with Bonferroni mean separation).

** Each observation (n) is the average weight for two 16- to 18-day-old nestlings randomly selected from each nest in the sample (n).

of RBC ALAD diminished 75% in Mallard ducks (*Anas platyrhynchos*) after ingestion of lead shot; nerve cell damage in brain tissue was apparent (Dieter and Finley 1979). It declined 80% in Bald Eagles (*Haliaeetus leucocephalus*) that ingested lead shot; hematocrits and hemoglobin concentrations were reduced 20–25%, and changes in serum chemistry indicated that the birds' kidneys and livers were damaged (Hoffman et al. 1981). Four of the five eagles eventually died, and the fifth became blind; cardiovascular and renal lesions attributable to lead poisoning were present in the birds at necropsy (Pattee et al. 1981). The activity of the enzyme was inhibited 96% in pigeons from downtown London, as was that of succinate dehydrogenase in their kidneys (67%) and livers (33%; Hutton 1980). Results of these studies and the absence of detectable differences in hemoglobin and hematocrit levels in the two populations in the present study suggest that the differences among the latter were not biologically significant.

Lead concentrations (ppm dry weight) in the stomach contents of Barn Swallows from the Parkway colony (3.2–6.8) were generally lower than those found in invertebrates collected adjacent to highways: earthworms, for example, contained 26–500 ppm, woodlice 380–682, millipedes 43–82, ants 15–45, beetles 9–16, grasshoppers 3.4–3.8, tent caterpillars 7.1–7.4, mantids 4–15, damsel flies 8–9, sucking insects 10–16, chewing insects 10–27, and predatory insects 20–31 (Williamson and Evans 1972, Giles et al. 1973, Gish and Christensen 1973, Price et al. 1974, Goldsmith and Scanlon 1977, Ash and Lee 1980, Beyer and Moore 1980, Robel et al. 1981). Terrestrial invertebrates apparently accumulate lead in proportion to their proximity to the soil and the amount of soil or litter that they ingest. Lead concentrations are highest in detritivores and lowest in herbivorous insects. Birds, such as Barn Swallows, which feed on small flying insects would be expected to accumulate less dietary lead than birds and small mammals that feed on terrestrial invertebrates. Comparison of lead concentrations (ppm dry weight) in stomach contents of Barn Swallows from the Parkway colony with those in adult and nestling Euro-

pean Starlings (*Sturnus vulgaris*) from nest boxes within the median of the Parkway (84–94 ppm; C. E. Grue, unpubl. data) support this conclusion. Furthermore, the stomach contents of bats (*Eptesicus fuscus* and *Myotis lucifugus*) believed to forage within the right-of-way of this Parkway contained lead concentrations (3.8–26 ppm wet weight) that were considerably lower than those of shrews (240 ppm wet weight) collected in the same area (Clark 1979), but greater than comparable values for the adult Parkway swallows in our study (1.0–1.9 ppm wet weight).

Concentrations of lead in carcasses and feathers were higher in Barn Swallows from the Parkway than in the reference group. Similar results have been reported for these and other parts of birds inhabiting urban areas, highway rights-of-way, and other areas contaminated with lead (e.g., Getz et al. 1977, Hutton 1980, Hutton and Goodman 1980, Ohi et al. 1981; for an exception, see Udevitz et al. 1980). To our knowledge, the only other study in which lead concentrations in avian carcasses are reported as we have done is that of White et al. (1977), who found that urban European Starlings contained a geometric mean (GM) of 0.89 ppm lead on a wet weight basis. Lead concentrations (ppm wet weight) in carcasses of adult Barn Swallows from the Parkway colony (GM = 1.9 ppm for males and 3.5 ppm for females) were two to four times greater than this value.

Lead concentrations in carcasses of female Barn Swallows from both colonies exceeded those in the carcasses of males, but these differences were not statistically significant. Such differences between sexes have, however, been reported in other species of birds (Finley et al. 1976, Finley and Dieter 1978, Hutton and Goodman 1980, Kendall and Scanlon 1981) and small mammals (e.g., Clark 1979). In birds, the differences appear to be associated with the increased calcium requirement of females during egg-laying, because lead accumulation in bone is enhanced by calcium mobilization (Finley et al. 1976, Finley and Dieter 1978). That female birds accumulate more lead in their bones than males may explain why lead concentrations in the feathers of male Barn

Swallows exceeded those of females in both colonies.

Concentrations of lead in the feathers of rural adult Barn Swallows (Table 2) were similar to those found in the feathers of ground-foraging songbirds from rural areas (\bar{x} = 6.4–36 ppm; Getz et al. 1977). However, lead concentrations in feathers of adult swallows from the Parkway colony were about three times those of adults from the reference colony, whereas concentrations in feathers of urban ground-foraging songbirds were about 3–35 times those of their rural counterparts (Getz et al. 1977). These data support the hypothesis that birds which forage on the ground are exposed to more lead than aerial feeders. Our finding that lead levels in tissues of nestling Barn Swallows from both colonies were $\frac{1}{6}$ – $\frac{1}{20}$ th of those found in adults indicates that lead is present in nestlings from colonies in both contaminated and uncontaminated sites, and that it accumulates in tissues over time. A similar conclusion was reached by Tansy and Roth (1970) for urban pigeons.

Concentrations of lead in the carcass were about 15 times greater in the bats that apparently foraged on flying insects above the Parkway right-of-way (GM = 32–47 ppm *wet* weight; Clark 1979) than in adult Barn Swallows in the same area. Several factors may account for this difference. Bats, for example, fed on insects that contained 4–13 times more lead than those consumed by our swallows. Differences in prey, physiology, and the duration of lead exposure may also have been important. Furthermore, the use of unleaded gasoline increased between 1976, when the bats were studied, and 1979, when we did our study. Differences in the accumulation of lead by hair and feathers may also have been a factor, as may the fact that Barn Swallows molt twice a year, whereas bats appear to molt only once (Quay 1970).

Our results suggest that lead from motor vehicle emissions does not pose a serious hazard to birds that feed on flying insects within highway rights-of-way. Comparable data for ground-feeding birds are needed to adequately assess the effects of lead from motor vehicles on birds inhabiting these areas.

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