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ILLINOIS NATURAL  
HISTORY SURVEY







# EFFECTS OF INGESTED LEAD-IRON SHOT ON MALLARDS

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## ABSTRACT

Each of nine wild male mallards in each of 12 groups was dosed as follows: 0 shot (control group); 1 No. 4 lead shot; 5 No. 4 steel shot; 1, 3, or 5 No. 4 shot of 36.67 percent lead and 63.33 percent iron; 1, 3, or 5 No. 4 shot of 60.90 percent lead and 39.10 percent iron; or 1, 3, or 5 shot of 64.60 percent lead and 35.40 percent iron.

In an average of 28.5 days, 2.1 percent of the shot was expelled. The weight of shot eroded varied from 21 to 62 percent. The weight eroded per shot per day varied from 1.0 to 2.0 percent. The daily rate of erosion was not significantly affected by the number of pellets in the gizzard and was significantly higher for heavy shot than for light shot.

Ducks in the control group and those dosed with steel pellets had the lowest daily losses of body weight, and the group dosed with one lead pellet each had the highest daily weight loss. Mean weights of surviving ducks on 28 May and 12 June showed significant inverse relationships with the weight of lead dosed. Apparently, ducks given only one lead-iron shot with up to 65 percent lead and ducks given as many as five lead-iron shot with up to 37 percent lead receive substantial protection from lead poisoning by some action of the iron. Although there is evidence that the iron gave some protection in all cases in the present experiment, ducks dosed with three or more lead-iron pellets containing  $\geq 61$  percent lead had significant weight losses.

Packed-cell volume (PCV) and hemoglobin (Hb) determinations support the indications of weight losses regarding effects of lead and iron. Mortality rate, body weight, and PCV are perhaps the most useful measurements for studying lead toxicity in wild captive mallards on a diet of corn. These parameters indicate not only that iron offers a protective value against lead toxicity, but also that the effects of lead are detectable at these doses even in the presence of iron.

A significant negative correlation was found between the amount of lead administered and the weight of the spleen, pancreas, liver, and kidneys. A significant positive linear relationship occurred between the amount of lead administered and the mean weight of adrenals. Active spermatogenesis was seen only in the testes of ducks in the control group and of ducks dosed with steel shot. A granular pigment appeared in both the red and

white pulp of the spleens of ducks dosed with steel pellets, and it appeared in moderate amounts throughout the spleens of all ducks dosed with lead. Liver sections of all ducks, including the controls, were abnormal. Abnormal changes occurred in the liver, spleen, kidneys, and testes with all levels of lead dosage, but the changes were greater for higher lead doses. Hemosiderosis was found in the spleens and livers of all dosed ducks. Intracellular inclusion bodies were found in kidney tubule cells of most ducks dosed with lead.

The ingestion of five steel pellets may benefit ducks, and the ingestion of up to five lead-iron pellets with up to 37 percent lead is relatively nontoxic.

## INTRODUCTION

The poisoning of wild waterfowl by ingested lead shot is a continuing concern (Jordan & Bellrose 1951, Bellrose 1959, Anderson 1975). The U.S. Fish and Wildlife Service (1974. Use of steel shot for hunting waterfowl in the United States. Department of the Interior DES 74-76. 79 p. + 6 appendices.) proposed the gradual elimination of lead shot for hunting migratory waterfowl in some flyways and in certain areas in other flyways, beginning with the fall hunting season in 1976. Although the proposed regulations prohibited the taking of migratory game birds "With a shotgun containing shells loaded with shot composed of any metal other than such material as may be determined by the Director to be nontoxic to migratory waterfowl. . . ." (U.S. Fish and Wildlife Service. 1974. Use of steel shot for hunting waterfowl in the United States. Department of the Interior DES 74-76. 79 p. + 6 appendices.), the preliminary impact statement implied (1974:75) that only soft steel shot and a mixture of lead and iron shot appeared promising as a substitute for lead shot for hunting migratory waterfowl.

Because of reduced toxicity with ingested lead-iron shot in captive waterfowl as reported by Irwin et al. (1974), Dr. Edward L. Kozicky, Director of Conservation, Winchester Group, Olin Corporation, asked the Illinois Natural History Survey to conduct an additional study, using lead-iron shot in captive wild waterfowl.

## METHODS

Wild male mallards (*Anas platyrhynchos*) were trapped on 20 March 1974 at the Union County Refuge in southern Illinois and on the Keokuk Pool on the Mississippi River and were transported to the Illinois Natural History Survey Laboratory, Havana, Illinois. They were held in two pens that were 9.1 x 3.6 x 0.6 m (30 x 12 x 2 feet) (Jordan & Bellrose 1950:157) and were provided duck pellets, cracked corn and water ad libitum

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until 29 April, 15 days before the experiment began. The birds were banded with individually numbered leg bands, randomly assigned to 1 of 12 groups of 10 ducks each, and placed in 12 wire-covered pens 1.2 x 1.2 x 0.8 m (4 x 4 x 2.5 feet). Each pen had running water in a 5-inch rain gutter, a resting platform covering about half of the bottom of the pen, and a board covering half of the top of the pen. Because the rain gutter carrying running water was too small for the ducks to bathe in conveniently, the ducks did not adapt to these smaller pens, and at the start of the experiment they were returned to the two large holding pens. Losses during the conditioning period necessitated reducing the number of birds in the experiment from 10 to 9 per group. At the start of the experiment on 13 May, the ducks were placed on a diet of whole corn and water ad libitum.

The doses were as follows for the ducks in each of the 12 groups: 0 shot (controls); 1 No. 4 lead shot; 5 No. 4 steel shot; 1, 3, or 5 No. 4 shot of 36.67 percent lead and 63.33 percent iron; 1, 3, or 5 shot of 60.90 percent lead and 39.10 percent iron; and 1, 3, or 5 shot of 64.60 percent lead and 35.40 percent iron. Throughout this report, dosage groups are identified by the number of pellets given, the percentage of lead in the pellet, and the percentage of iron in the pellet. Thus, 1-100-0 equals 1 shot of 100 percent lead and 0 percent iron, and 3-37-63 indicates 3 pellets with approximately 37 percent lead and 63 percent iron. The lead-iron shot were prepared by the National Research Council of Canada from a mixture of lead and iron particles. The plans were to use shot composed of 45, 60, and 70 percent lead, but analyses indicated that the above percentages of lead and iron were as close as they came to their objectives. The all-lead and all-steel shot were taken from 12-gauge shotgun shells.

Each dose of shot was weighed before being administered to a duck, and all recovered shot were weighed. When no shot was expelled from the gizzard, we obtained the actual weight lost by the pellets while they were in the gizzard. For ducks that expelled some shot, we used average weights of shot administered to and recovered from each duck. Analysis of a number of shot removed from the gizzards showed no appreciable change in the lead:iron ratio.

The shot were placed in the proventriculus of each duck by inserting a plastic tube passed through the esophagus and were followed by approximately 5 ml of water. Controls were handled in the same manner except that they received no shot.

The ducks were dosed on 13 May 1974, and the experiment ended 12 June 1974. Birds were weighed, and blood samples for packed-cell volume (PCV), hemoglobin (Hb) determinations, and lead analyses were taken on 13 May, 28 May, and 12 June. When a duck died, it was weighed and autopsied as soon as practicable. The pens were checked for dead ducks at least twice daily.

Fresh, wet weights were obtained for testes, thyroids, spleen, pancreas, liver, kidneys, and adrenals. Tissues from the heart, lungs, liver, kidneys, gonads, spleen, and femur of two ducks in each group were preserved in 10-percent buffered neutral formalin for histological study (Luna 1968). Blood samples (at the beginning, middle,

and end of the experiment) and livers from the same two ducks from each group were frozen for determination of lead contents. The ducks chosen for histological study and lead analyses were the second and sixth ducks to die in each group if they were obtained soon enough after death. In groups in which no bird died, or when fewer than two or six ducks had died by the end of the experiment, the second and sixth ducks autopsied at the end of the experiment, or the last duck autopsied if fewer than six remained in the group, were chosen for histological study and lead analyses.

Except for the kidneys, which were stained with Kinyown's carbolfuchsin for acid-fast nuclear inclusion bodies, tissue sections were stained with hematoxylin and eosin. The method of preparing the bone for sectioning and staining did not permit the evaluation of bone marrow activity.

Blood smears were stained with hematoxylin and eosin. The initial sample from each duck was used as the control or base line for evaluating subsequent samples from each duck. Each smear was evaluated by cellular morphology.

White-blood-cell differential counts were performed on individual smears to try to quantitate the effects of the various shot doses. A malaria-like infection of the red blood cells prevented effective use of this datum. This infection influenced white cell differentials and caused a degree of anemia in about 30 percent of the test ducks.

The experiment was terminated on the 30th day, when all surviving ducks were weighed and blood samples were collected. All ducks selected for histological study and lead analyses were killed on the 30th day. For lack of time, the remaining ducks were killed and autopsied on the 31st day. Except for total erosion and daily rate of shot eroded in the gizzard, all determinations were made as if all ducks had been killed on the 30th day.

The ducks were decapitated and allowed to bleed freely. Blood collected for lead analyses was drawn from the wing vein, placed in 2-ml heparinized Vacutainers (TM, Becton, Dickinson and Company, Columbus, Nebraska), and frozen (5°F). Organs collected for lead analyses were placed in individual plastic bags and frozen.

Gizzards and proventriculi were opened and searched for shot. Shot were sometimes found in the proventriculus, but the locations of shot found were not recorded. Shot recovered from each duck were placed in a coin envelope and were weighed later.

One duck escaped on 28 May as the birds were being transferred to carrying crates, and one duck expelled all five pellets administered. These two birds were eliminated from all analyses.

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## RESULTS AND DISCUSSION

### Percentages of Shot Expelled

Of 291 shot administered to 97 ducks, 285 shot were recovered at autopsy (Table 1). None of these ducks lost more than one shot, no treatment group lost more than two shot, and 7 of 11 groups retained all shot. As mentioned earlier, one duck expelled all five shot, presumably shortly after dosing, and was eliminated from the analysis. The total expulsion rate was 2.1 percent of the shot in an average of 28.5 days. There was no significant difference ( $P > 0.05$ ) in the rate of shot expelled among groups.

### Dosage and Rate of Erosion

Because the lead-iron shot were handcrafted, the weight of the shot varied considerably (Table 2). Thus, ducks in the 5-61-39 group were dosed with more lead and more iron than ducks in the 5-65-35 group. Five groups of ducks were given more lead than the group that was dosed with only one lead shot each.

Weight eroded varied from 21.2 percent for the 5-65-35 group to 62.0 percent for the 1-61-39 group (Table 3). The percentage of total weight of shot eroded probably relates to the mortality rate. A higher rate of exposure to lead (as in the 5-65-35 group compared with the 1-61-39 group) might result in earlier death and less total erosion of the shot.

Weight eroded per shot per day varied from 1.0 to 2.0 percent (Table 3), less than expected. The rate of erosion may be correlated with the rate of food consumption (Irwin, J. C., L. H. Karstad, and R. G. Thomson. 1974. Progress report on lead poisoning in waterfowl March 1973 to March 1974. University of Guelph, Guelph, Ontario. 66 p. Mimeographed.), and low air temperatures

Table 1.—Numbers of shot administered and recovered, percentages of shot expelled, and average days to death of ducks.

Dose <sup>a</sup>	Number of Shot		Percent Expelled	Avg. Percent Expelled per Group Daily <sup>b</sup>	Avg. No. of Days to Death <sup>c</sup>
	Given	Recovered			
1-100-0	9	9	0	0.0	26.7
5-0-100	45	45	0	0.0	31.0
1-37-63	9	9	0	0.0	29.6
3-37-63	27	26	4	0.13	28.2
5-37-63 <sup>d</sup>	40	40	0	0.0	27.6
1-61-39 <sup>e</sup>	8	8	0	0.0	30.9
3-61-39	27	25	7	0.26	28.2
5-61-39	45	43	4	1.14	30.8
1-65-35	9	9	0	0.0	30.7
3-65-35	27	27	0	0.0	27.9
5-65-35	45	44	2	0.11	20.7
Total or mean	291	285	2	0.07	28.5

<sup>a</sup> Number of shot given: percent lead (Pb)-percent iron (Fe). Chemical analyses indicated these compositions of pellets:

37-63 = 36.67 percent Pb and 63.33 percent Fe  
 61-39 = 60.90 percent Pb and 39.10 percent Fe  
 65-35 = 64.60 percent Pb and 35.40 percent Fe

<sup>b</sup> As if all expelled shot were expelled on day of death

<sup>c</sup> Although the experiment was terminated after 30 days, several ducks were not killed until the 31st day. These values should not be used for calculating average survival time or rate of loss of body weight

<sup>d</sup> One duck expelled all five shot and was eliminated from the analyses

<sup>e</sup> One duck escaped

may encourage greater consumption of food (Jordan & Bellrose 1951:21). During the present experiment, the weather was mild, and there were indications that the rate of food consumption, although not measured, was low. The rate of erosion for one lead pellet per duck in the present experiment was 1.8 percent per day compared with 3.6 percent per day in an earlier experiment (Sanderson unpublished) in which the pens and diet were the same as in the present study, but the time was November and December, when the weather was colder and food consumption was presumably higher. Average survival time for 10 ducks in that experiment was 17.2 days, the range being 10-25 days. In the present experiment, only four of nine ducks dosed with one lead pellet each had died by 30 days after dosing, and the average time the lead pellets were in the gizzard was 26.7 days (Table 1). This finding differs from that of Jordan & Bellrose (1951:21), who reported that "the proportion of dosed birds surviving in winter was greater than that in milder seasons."

Cook & Trainer (1966) reported that the rate of erosion is not related to the number of shot in the gizzard. We found (Table 3) that the number of pellets had no significant ( $P > 0.30$ ) effect on the rate of erosion per shot per day. Also, the total amount of lead administered had no significant effect ( $P > 0.05$ ) on the percentage of each shot eroded each day, but the heavier the pellet, the higher the percentage eroded per day ( $P < 0.001$ ). Lead percentage showed a positive but not significant ( $P > 0.05$ ) effect on the percentage of total weight eroded each

Table 2.—Mean amounts of lead and iron administered, mean amounts eroded daily, mean daily changes in body weight, and mean daily ratios of lead and iron eroded.

Dose <sup>a</sup>	Mean Amount in Grams Administered			Lead Eroded in Grams per Duck		Iron Eroded in Grams per Duck		Mean Ratio of Fe: Pb Eroded Daily per Duck
	All Shot	Lead	Iron	Total	Per Day	Total	Per Day	
0-0-0	0.000	0.000	0.000	0.000	0.000	0.000	0.000	. . .
1-100-0	0.213	0.213	0.000	0.100	0.004	0.000	0.000	0:1.00
5-0-100	0.751	0.000	0.751	0.000	0.000	0.275	0.090	1:0.00
1-37-63	0.181	0.066	0.115	0.038	0.001	0.065	0.002	1:0.58
3-37-63	0.541	0.198	0.343	0.080	0.003	0.138	0.005	1:0.58
5-37-63	0.905	0.332	0.573	0.150	0.005	0.260	0.009	1:0.58
1-61-39	0.220	0.134	0.086	0.083	0.003	0.053	0.002	1:1.56
3-61-39	0.651	0.396	0.255	0.161	0.006	0.104	0.004	1:1.56
5-61-39	1.070	0.652	0.418	0.268	0.009	0.172	0.006	1:1.56
1-65-35	0.189	0.122	0.067	0.059	0.002	0.032	0.001	1:1.82
3-65-35	0.551	0.356	0.195	0.137	0.005	0.075	0.003	1:1.82
5-65-35	0.957	0.618	0.339	0.128	0.006	0.070	0.003	1:1.82
<i>Grand mean</i>	<i>0.566</i>	<i>0.309<sup>b</sup></i>	<i>0.314<sup>c</sup></i>	<i>0.120<sup>b</sup></i>	<i>0.004<sup>b</sup></i>	<i>0.124<sup>c</sup></i>	<i>0.012<sup>c</sup></i>	<i>1:1.17</i>

<sup>a</sup> See Table 1, footnote a.<sup>b</sup> Includes only those ducks receiving lead.<sup>c</sup> Includes only those ducks receiving iron.

day (Table 3). Although we did not measure the hardness of the pellets, we had expected that pellets with a higher percentage of lead would erode faster than pellets with a higher percentage of iron. Sanderson (unpublished) found a daily rate of erosion for lead pellets (one per gizzard) of 3.62 percent compared with 0.23 percent for copper pellets (1.32 pellets per gizzard). The effect of the number of pellets on the rate of erosion was not considered in that study, but there appeared to be a slightly higher rate of erosion when more copper pellets were present.

AMOUNT OF LEAD AND IRON ERODED.—Because no duck in this experiment expelled more than one shot and because the time of expulsion was not determined, expelled shot are ignored in this analysis. However, every shot administered was included in calculating the dosage of lead and iron for each duck (Table 2).

In ducks dosed with shot containing lead, the average amount of lead eroded per duck varied from 0.0378 g to 0.2679 g (Table 2). As expected, there was a significant ( $P < 0.001$ ) positive relationship between weight of lead administered and the amount eroded each day. In

Table 3.—Mean weight of shot eroded per day.

Dose <sup>a</sup>	Mean Weight in Grams of Each Shot Given	Mean Weight in Grams of Each Shot Recovered	Percent of Total Weight Eroded	Percent of Weight Eroded Daily per Shot
0-0-0	0.000	0.000	0	0.0
1-100-0	0.213	0.113	47	1.8
5-0-100	0.150	0.095	37	1.2
1-37-63	0.181	0.078	57	1.9
3-37-63	0.180	0.105	42	1.5
5-37-63	0.181	0.099	45	1.6
1-61-39	0.220	0.084	62	2.0
3-61-39	0.217	0.112	48	1.7
5-61-39	0.214	0.122	43	1.4
1-65-35	0.190	0.098	48	1.6
3-65-35	0.184	0.113	39	1.4
5-65-35	0.191	0.151	21	1.0
<i>Grand mean<sup>b</sup></i>	<i>0.193</i>	<i>0.106</i>	<i>44</i>	<i>1.6</i>

<sup>a</sup> See Table 1, footnote a.<sup>b</sup> Controls are not included.

general, the daily amount and total amount of lead eroded per duck were in relative agreement, but there were some differences (Table 2). The largest daily amount (0.0087 g) and the largest total (0.2679 g) were eroded in ducks in the 5-61-39 group. However, ducks in the 5-65-35 group eroded the second highest *daily* amount (0.0063 g), but ducks in the 3-61-39 group eroded the second highest *total* amount (0.1614 g). A higher daily erosion rate may result in an earlier death and thus in a lower total amount eroded. Ducks in the 1-100-0 group ranked sixth in both the daily amount (0.0037 g) and the total (0.0996 g) of lead eroded.

Ducks dosed with five steel shot eroded the highest daily amount (0.009 g) and the highest total amount (0.275 g) of iron per duck (Table 2). Total iron eroded ranged from 0.032 to 0.275 g per duck and from 0.001 to 0.009 g per duck per day.

### Mortality Rate

Mortality rates for the 12 groups of ducks ranged from no mortality in three groups to 67 percent mortality in the 5-65-35 group (Table 4). Mild weather during the present experiment may have permitted relatively long survival, but Jordan & Bellrose (1951:21) reported that ducks dosed with lead survived better in winter than in milder seasons because of the higher intake of food in winter. As mentioned earlier, Sanderson (unpublished) found 100 percent mortality in a study conducted in November 1972.

Table 4.—Mortality and average survival time.

Dose <sup>a</sup>	Number of Ducks in Group	Ducks Dead Before End of Experiment		Survival in Days <sup>b</sup>	
		Number	Percent	Mean	Range
0-0-0	9	1	11	29	23-30
1-100-0	9	4	44	26	15-30
5-0-100	9	0	0	30	30
1-37-63	9	1	11	29	20-30
3-37-63	9	1	11	27	6-30
5-37-63	7	1	14	29	20-30
1-61-39	8	0	0	30	30
3-61-39	9	5	56	28	16-30
5-61-39	9	0	0	30	30
1-65-35	9	1	11	30	28-30
3-65-35	9	4	44	27	17-30
5-65-35	9	6	67	20	7-30
<i>Total or mean</i>	<i>105</i>	<i>24</i>	<i>22</i>	<i>28</i>	<i>6-30</i>

<sup>a</sup> See Table 1, footnote a

<sup>b</sup> Calculated as if surviving ducks were all killed on the 30th day (some were actually killed on the 31st day).

Mean survival time (Table 4) includes all ducks that were killed at the end of the experiment but has little meaning in the present experiment, because many ducks were still alive after 30 days. However, these values were used only to calculate average daily loss of body weight and average daily change in PCV and Hb.

### Body Weight

Body weight is an indicator of general condition. All surviving ducks progressively lost weight as the experiment continued (Table 5). All groups except the controls and 5-0-100 group weighed less at the end of the experiment than at the midpoint. Weights of controls were essentially the same and weights of the 5-0-100 group were the same at the midpoint and at the end. The controls, 5-0-100 group, and 1-37-63 group weighed 5.0, 5.3, and 6.8 percent, respectively, less at the end of the experiment than at the start. In contrast, groups 3-37-63, 5-37-63, 1-61-39, and 1-65-35 lost from 11.9 to 20.9 percent of their initial body weights. The remaining groups lost from 27.3 to 40.5 percent of their body weight. Ducks dosed with one lead pellet lost the greatest percentage.

Some surviving ducks had lost more than 50 percent of their body weights. Jordan & Bellrose (1951:5) discussed weight loss in lead-poisoned captive wild mallards and stated that "loss of body weight . . . at the time of death may average about 40 percent of the original weight."

The 1-100-0 group was sixth in total lead eroded and in lead eroded per duck per day (Table 2). Thus, the presence of iron in the shot resulted in a reduction of weight loss. The group of ducks (5-65-35) with the second highest (36.7 percent) average weight loss among surviving ducks (Table 5) was second in lead eroded per day and sixth in iron eroded per duck per day (Table 2).

All ducks were included in the calculations of the mean loss of body weight per duck per day. Again, the two groups having the lowest (1.86 and 2.59 g) daily weight losses were those dosed only with steel shot and the controls. However, the 1-61-39 group was third (4.31 g) in rate of daily weight loss, slightly ahead of the 1-37-63 group (4.72 g). The group dosed with one lead pellet each in the present experiment had the highest daily weight loss (17.15 g).

Weight losses for ducks dosed with one lead pellet in a previous experiment (Sanderson unpublished) have been calculated from data in our files. The mean weight of those 10 ducks was 1.208 kg at the start of the experiment on 22 November. Their average weight at death (an average 17.2 days after being dosed) was 0.800 kg, a 33.8 percent loss at the rate of 23.68 g per duck per day, compared with 40.5 percent and 17.15 g per duck per day in our 1-100-0 group. These latter values break down to 41.7 percent (at an average 21.2 days after being dosed) and 21.46 g per duck per day for the four ducks that died, and 40.5 percent and 14.74 g per duck per day for the five survivors (Tables 5 and 6). These findings indicate that lead-poisoned ducks on a corn diet survive longer and lose more weight during mild weather than during cold weather.

The mean weight of ducks that died (24 ducks, 0.772 kg) was lower than that of ducks that survived for 30 days (81 ducks, 0.890 kg, Table 6). The mean daily weight loss of the ducks that died (24.63 g per duck) was higher than the daily loss (7.79 g) of surviving ducks.

BODY WEIGHT IN RELATION TO LEAD DOSAGE.—The mean loss in body weight per duck per day was directly related to the mean amount of lead administered per duck

Table 5.—Mean body weight at start, middle, and end of the experiment; daily change in body weight per duck; and percentage of body weight lost by surviving ducks.

Dose <sup>a</sup>	Mean Body Weight in kg			Mean Daily Weight Loss in Grams per Duck <sup>b</sup>	Percent of Body Weight Lost in 30 Days <sup>c</sup>
	13 May	28 May	12 June		
0-0-0	1.13	1.06	1.07 (8) <sup>d</sup>	2.6—29 <sup>e</sup>	5.0 (8)
1-100-0	1.09	0.86	0.65 (5)	17.2—26	40.5 (5)
5-0-100	1.06	1.01	1.01	1.9—30	5.3 (9)
1-37-63	1.20	1.18	1.12 (8)	4.7—29	6.8 (8)
3-37-63	1.14	1.05 (8)	0.98 (8)	7.0—27	13.5 (8)
5-37-63	1.18 (8)	1.09 (8)	0.96 (6)	6.4—27	15.9 (6)
1-61-39	1.09 (8)	1.06 (8)	0.96 (8)	4.3—30	11.9 (8)
3-61-39	1.08	0.87	0.71 (4)	14.3—28	30.9 (4)
5-61-39	1.20	0.99	0.80	13.4—30	33.3 (9)
1-65-35	1.16	1.03	0.93 (8)	8.6—30	20.9 (8)
3-65-35	1.13	0.96	0.82 (5)	14.0—27	27.3 (5)
5-65-35	1.05	0.84 (7)	0.66 (3)	14.2—20	36.7 (3)
<i>Grand mean</i>	<i>1.13 (106)</i>	<i>1.00 (103)</i>	<i>0.89 (81)</i>	<i>9.0—28</i>	<i>20.7 (81)</i>

<sup>a</sup> See Table 1, footnote a

<sup>b</sup> Includes ducks that died during the experiment

<sup>c</sup> Includes only ducks that survived to the end of the experiment (30 days)

<sup>d</sup> Numbers in parentheses refer to the number of ducks weighed, in all other groups there were nine ducks

<sup>e</sup> Mean number of days to death

( $P < 0.001$ ) and the mean amount of lead eroded per duck ( $P < 0.005$ ) per day (Tables 2 and 5). Mean weights of surviving ducks on 28 May and 12 June were inversely related ( $P < 0.001$ ) to lead administered and directly related ( $P < 0.001$  and  $< 0.005$ , respectively) to mean weight on 13 May. The two groups not dosed with lead (0-0-0 and 5-0-100) lost the least weight per duck per day

(2.6 and 1.9 g, respectively, Table 5). Ducks dosed with one lead pellet had the highest daily weight loss (17.2 g), even though this group ranked sixth in total lead administered per duck and mean lead eroded per duck per day. The 3-61-39, 3-65-35, and 5-65-35 groups had similar daily rates of weight loss and ranked third, fourth, and second, respectively, in total lead administered per

Table 6.—Mean body weight and mean loss of body weight per day for ducks that died and ducks that survived.

Dose <sup>a</sup>	Mean Body Weight in kg at Death		Mean Daily Loss of Body Weight in Grams per Duck		Mean Number of Days to Death <sup>b</sup>	Percent of Body Weight Lost by Ducks That Died
	Survived	Died	Survived	Died		
0-0-0	1.07 (8) <sup>c</sup>	1.00 (1)	0.77	21.7	23.0	35.5
1-100-0	0.65 (5)	0.64 (4)	14.74	21.5	21.2	41.7
5-0-100	1.01 (9)	...	1.86	...	...	...
1-37-63	1.12 (8)	0.64 (1)	3.67	17.0	20.0	34.9
3-37-63	0.98 (8)	0.74 (1)	5.13	81.6	6.0	40.0
5-37-63 <sup>d</sup>	0.96 (6)	1.13 (1)	6.03	11.3	20.0	16.7
1-61-39	0.96 (8)	...	4.31	...	...	...
3-61-39	0.71 (4)	0.67 (5)	12.25	18.0	25.8	41.0
5-61-39	0.80 (9)	...	13.38	...	...	...
1-65-35	0.93 (8)	0.70 (1)	8.21	12.2	28.0	32.6
3-65-35	0.82 (5)	0.64 (4)	10.30	19.8	24.2	42.8
5-65-35	0.66 (3)	0.81 (6)	12.84	18.6	15.7	23.2
<i>Grand mean</i>	<i>0.89 (81)<sup>d</sup></i>	<i>0.77 (24)</i>	<i>7.79</i>	<i>24.6</i>	<i>20.4</i>	<i>34.3</i>

<sup>a</sup> See Table 1, footnote a

<sup>b</sup> Ducks that died

<sup>c</sup> Numbers in parentheses are numbers of ducks in each category

<sup>d</sup> One duck was accidentally killed during the experiment

duck. The 5-61-39 group, which was dosed with the largest amount of lead and the third highest amount of iron (Table 2), ranked fifth in rate of daily weight loss. However, we do not conclude that one or more lead-iron shot containing 37-65 percent lead has no effect on daily weight loss.

Ducks dosed with one lead shot lost more weight than ducks dosed with lead-iron shot that eroded more lead (total or daily), and the 5-37-63 group lost less weight than ducks that eroded similar amounts of lead but less iron (Tables 2 and 5). These weight losses suggest that iron provides some protection against weight loss in lead-poisoned ducks on a diet of corn.

Seven groups had noticeably lower daily weight losses than the other five had (Tables 5 and 6). These seven groups lost from 1.9 to 8.6 g per day, compared with 13.4-17.2 g for the remaining five. The seven included both groups that received no lead, all groups that received only one lead-iron shot, and all three that received lead-iron shot containing 63 percent iron.

Thus, ducks dosed with only one lead-iron shot with up to 65 percent lead and 35 percent iron (by weight) seem to have received substantial protection from lead poisoning by some action of the iron. The same conclusion is indicated by the data for ducks dosed with up to five lead-iron shot containing up to 37 percent lead and 63 percent iron. When ducks were dosed with three or more lead-iron pellets containing  $\geq 61$  percent lead and  $\leq 39$  percent iron, important weight losses occurred ( $\geq 13.4$  g per day compared with  $\leq 8.6$  g per day for all other groups dosed with lead-iron shot). There was no substantial difference between the daily weight loss of controls (2.6 g per duck) and that of ducks dosed with five steel shot (1.9 g). However, the next group (1-61-39) lost 4.3 g per duck per day. The slight difference in daily weight loss

between the controls and the group dosed with iron only favored the group dosed with iron (Table 5).

### Packed-Cell Volume

Packed-cell volume (PCV) is believed to reflect lead toxicity in ducks. One difficulty is that blood samples can be collected only from live ducks. We observed (Table 6) a greater daily loss of body weight in ducks that died than in those that survived. There may be similar differences in PCV (and Hb), but they cannot be measured.

At the start of the experiment, the mean PCV ranged from 46.2 to 52.2 percent (Table 7). Two weeks later the PCV ranged from 31.6 and 34.1 percent for the 3-61-39 and 1-100-0 groups, respectively, to 52.5 and 52.7 percent for the 5-37-63 and 0-0-0 groups, respectively. At the end of the experiment, values ranged from 31.2 and 34.8 percent for the 1-100-0 and 5-61-39 groups, respectively, to 54.6 and 55.6 percent for the 5-0-100 and 0-0-0 groups, respectively.

The larger the dose of lead, the greater the decline ( $P < 0.001$ ) in PCV during the experiment (Tables 2 and 7). Also, the greater the amount of lead eroded per day, the lower the average PCV at the end of the experiment for surviving ducks (Tables 2 and 7). As with body weight, the PCVs were lower in birds in the 1-100-0 group and higher in birds in the 5-37-63 group than in ducks that were dosed with or eroded similar amounts of lead.

The same seven groups that had relatively low weight losses (Table 5) also had high PCVs at the end of the experiment (Table 7), values that were not appreciably lower (in three groups they were higher) at the end of the experiment than at the beginning. In the remaining five groups (1-100-0, 3-61-39, 5-61-39, 3-65-35, and 5-65-35), the PCVs were markedly lower at the end of the experi-

Table 7.—Mean packed-cell volume (PCV) and hemoglobin (Hb) values at the start, middle, and end of the experiment and daily changes in PCV and Hb values per duck.

Dose <sup>a</sup>	Mean PCV (Percent)			Mean Daily Change in PCV per Duck (Percent) <sup>b</sup>	Mean Hb Value (g/100 ml)			Mean Daily Change in Hb per Duck (Percent) <sup>b</sup>
	13 May	28 May	12 June		13 May	28 May	12 June	
0-0-0	48.3	52.7	55.6 (8) <sup>c</sup>	+ 0.275	14.12	15.48	15.25 (8)	+ 0.053
1-100-0	46.2	34.1	31.2 (5)	- 0.547	14.40 (5)	7.88	9.72 (5)	- 0.071
5-0-100	48.8	50.3	54.6	+ 0.192	14.68 (5)	16.01	17.28	+ 0.091
1-37-63	47.6	49.6	51.9 (8)	+ 0.042	14.70 (7)	12.58	14.65 (8)	0.000
3-37-63	50.2	50.2 (8)	48.6 (8)	+ 0.033	13.39	12.55 (8)	14.94 (8)	+ 0.034
5-37-63	50.8 (8)	52.5 (8)	45.5 (6)	- 0.144	14.58 (8)	14.31 (8)	12.03 (6)	- 0.095
1-61-39	52.2	50.8 (8)	49.9 (8)	- 0.021	14.19	13.90 (8)	14.29 (8)	+ 0.005
3-61-39	46.9	31.6	36.2 (4)	- 0.225	14.48 (4)	7.94	10.70 (4)	- 0.037
5-61-39	51.0	41.1	34.8	- 0.541	15.10 (7)	9.62	8.54	- 0.231
1-65-35	46.2	46.7	46.0 (8)	+ 0.033	13.51	11.42	12.38 (8)	- 0.025
3-65-35	49.9	41.4	35.4 (5)	- 0.440	14.75 (6)	10.21	8.38 (5)	- 0.164
5-65-35	49.8	41.8 (7)	40.0 (3)	- 0.311	14.14 (7)	10.71 (7)	10.13 (3)	- 0.096
Grand mean	49.0	45.2	44.1	- 0.138	14.34	11.88	12.36	- 0.045

<sup>a</sup> See Table 1, footnote a

<sup>b</sup> Includes only ducks surviving on 12 June

<sup>c</sup> Numbers in parentheses are numbers of ducks in groups. All other groups numbered nine ducks

ment than at the beginning. Thus, the PCVs also indicate that ducks receive substantial protection against lead poisoning from one lead-iron shot with up to 65 percent lead and 35 percent iron and from up to five lead-iron shot containing 37 percent lead and 63 percent iron, but ducks dosed with three or more lead-iron pellets containing 61 percent or more lead and 39 percent or less iron had reduced PCVs. Compared with the controls and ducks dosed exclusively with steel shot, all groups had a modest reduction in PCVs. Again, we do not conclude that even one lead-iron pellet containing as little as 37 percent lead has no effect.

PCVs at the end of the experiment and the mean change in PCV during the experiment were not appreciably different in the controls and in ducks dosed with five steel pellets (Table 7). PCVs increased slightly in both groups but increased more in the controls.

PCV IN RELATION TO LEAD DOSAGE.—The PCVs of the ducks that survived to the end of the experiment were higher in five groups at the end of the experiment than they were at the start, perhaps indicating that these ducks were better conditioned to captivity at the end of the experiment (Table 7). Only in controls and in ducks dosed only with steel pellets were the daily increases in PCVs substantial. Changes in PCVs in five other groups were a slight daily increase in groups 1-37-63 (0.042 percent), 3-37-63 (0.033 percent), and 1-65-35 (0.033 percent); a slight daily decrease in group 1-61-39 (-0.021 percent); and an intermediate daily decrease in group 5-37-63 (-0.144 percent).

The remaining six groups showed larger daily decreases in PCVs ranging from -0.225 to -0.547 percent (Table 7). The group dosed with one lead shot showed the greatest daily decrease. The same seven groups of ducks

that showed the lowest daily weight losses (Table 5) and the highest PCVs (Table 7) at the end of the experiment also showed the smallest decreases (five showed increases) in PCVs. With slight deviations, group rank in weight loss, actual PCV, and change in PCV were the same (Table 8).

These data further indicate the protective value of iron against lead toxicity in ducks, one lead-iron pellet of up to 65 percent lead and 35 percent iron and three pellets of 37 percent lead and 63 percent iron having only slight effects. Although the 5-37-63 and 3-61-39 groups eroded similar amounts of lead daily (0.0054 and 0.0057 g, respectively), the first also eroded 0.009 g of iron daily compared with 0.004 g of iron for the second (Table 2). These two groups had daily decreases in PCVs of -0.144 and -0.225 percent, respectively (Table 7). We conclude that, as with total weight change and total change in PCV, all levels of lead administered caused detectable differences in daily changes in PCV, even in the presence of iron.

### Hemoglobin

As with PCV, a reduction in hemoglobin (Hb) is believed to reflect lead toxicity. At the start of the experiment, mean Hb for the various groups ranged from 13.4 (g per 100 ml) to 15.1, and the grand mean was 14.2. Two weeks later Hb ranged from 7.9 for the 1-100-0 and 3-61-39 groups to 15.5 and 16.0 for the controls and ducks dosed only with steel shot, respectively (Table 7). At the end, Hb ranged from 8.4 and 8.5 for the 3-65-35 and 5-61-39 groups, respectively, to 15.2 and 17.3 for the controls and ducks dosed only with steel shot, respectively. Hb for the ducks dosed with one lead pellet (9.7) was slightly higher at the end of the experiment than it had been 2 weeks earlier.

Table 8.—Rankings of treatment groups for 18 characters (most favorable condition received the lowest score).

Dose <sup>a</sup>	Character <sup>b</sup>																		Total	Order of Total
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18		
0-0-0	4	3	2	2	1	1	1	1	2	2	2	3	9	1	2	3	2	6	47	1
1-100-0	9	11	12	12	12	11	12	12	12	10	8	12	12	12	11	12	3	9	192	12
5-0-100	1	7	3	1	2	4	2	2	1	1	1	4	1	3	5	10	1	1	50	2
1-37-63	4	1	1	4	3	7	3	3	5	4	5	2	2	4	8	1	6	2	65	3
3-37-63	4	5	4	6	5	6	5	4	6	3	3	6	6	2	1	2	8	3	79	4
5-37-63	8	2	6	5	6	2	7	7	3	7	9	1	7	5	3	5	11	8	102	6
1-61-39	1	4	5	3	4	3	4	6	4	5	4	5	3	10	7	6	7	4	85	5
3-61-39	11	10	10	11	10	12	9	8	11	8	7	7	4	8	9	9	10	163	9	
5-61-39	1	8	9	8	9	10	11	11	10	11	12	8	5	9	4	7	10	7	150	8
1-65-35	4	6	7	7	7	4	6	4	7	6	6	11	8	6	5	8	5	5	112	7
3-65-35	9	9	8	9	8	9	10	10	9	12	11	10	11	11	12	11	4	11	174	11
5-65-35	12	12	11	10	11	8	8	9	8	9	10	9	10	7	10	4	12	12	172	10

<sup>a</sup> See Table 1, footnote a

<sup>b</sup> 1. Percentage of mortality within 30 days  
 2. Mean body weight on 28 May  
 3. Mean body weight on 12 June  
 4. Mean daily loss of body weight per duck  
 5. Percentage of body weight lost in 30 days (surviving ducks only)  
 6. Percentage of PCV on 28 May  
 7. Percentage of PCV on 12 June  
 8. Mean daily change in PCV per duck  
 9. Mean Hb on 28 May

10. Mean Hb on 12 June  
 11. Mean daily change in Hb per duck  
 12. Testes weight  
 13. Thyroid weight  
 14. Spleen weight  
 15. Pancreas weight  
 16. Liver weight  
 17. Kidney weight  
 18. Adrenal weight  
 Note. Ties are given equal numbers. Low weights for kidneys and adrenals are considered desirable

The more lead administered per duck, the lower the average Hb ( $P < 0.001$ ) in surviving ducks at the end of the experiment. Also, the higher the Hb at the start (13 May), the larger the decline ( $P < 0.001$ ) in Hb during the experiment, but there was a significant positive correlation ( $P < 0.05$ ) between the amount of Hb at the beginning and at the end of the experiment (Table 7).

The same seven groups of ducks that had relatively low weight losses (Table 5) and high PCVs (Table 7) also had high Hb at the end of the experiment, values that were not appreciably lower (four groups were higher) at the end of the experiment than at the beginning. In the remaining five groups (3-65-35, 5-61-39, 1-100-0, 5-65-35, and 3-61-39), Hb was lower at the end of the experiment than at the beginning. Thus, Hb also supports the conclusions and statements indicated by weight loss and PCV.

**Hb IN RELATION TO LEAD DOSAGE.**—In four groups, Hb was higher at the end of the experiment than at the start in ducks surviving to the end of the experiment, perhaps indicating that these ducks were better conditioned to captivity at the end of the experiment (Table 7). However, the only substantial increases were in the 5-0-100, 0-0-0, and 3-37-63 groups. The first two of these groups also showed increases in PCVs. In calculating the mean daily change in Hb per duck from the start of the experiment to the end, only ducks alive at the end of the experiment were included. However, in calculating the mean Hb for the start, middle, and end of the experiment, all ducks alive at the respective dates and for which we had suitable blood samples were included. Several blood samples collected on 13 May hemolyzed and were not suitable for Hb determinations. Hb increased slightly in group 1-61-39, was unchanged in group 1-37-63, and decreased slightly in groups 1-65-35 and 3-61-39.

Table 9.—Mean organ weights.

Dose <sup>a</sup>	Testes (g)	Thyroid (mg)	Spleen (mg)	Pancreas (g)	Liver (g)	Kidneys (g)	Adrenals (mg)
0-0-0	1.40	44	928	1.60	19.2	4.6	149
1-100-0	0.37	36 <sup>b</sup>	223	1.13	10.7	4.8	173
5-0-100	1.33	101	687	1.48	14.9	3.8	100
1-37-63	1.68	84	601 <sup>c</sup>	1.36	23.8	5.0	118
3-37-63	0.78	59 <sup>b</sup>	868	1.63	20.7	5.0	131
5-37-63	0.93 <sup>d</sup>	57 <sup>c</sup>	542 <sup>b</sup>	1.54 <sup>b</sup>	17.5 <sup>b</sup>	5.6 <sup>b</sup>	161 <sup>b</sup>
1-61-39 <sup>b</sup>	0.94	79	410	1.47	16.1	5.0	131
3-61-39	0.59	46	429	1.34	15.5	5.2	178
5-61-39	0.45	60	420	1.51	15.8	5.5	149
1-65-35	0.38	52	535 <sup>b</sup>	1.48	15.8	4.9	143
3-65-35	0.39	38	311	1.07	12.3	4.7	180
5-65-35	0.43	42 <sup>c</sup>	485	1.32	18.9	6.0	220
<i>Grand mean</i>	<i>0.80</i>	<i>58</i>	<i>536</i>	<i>1.41</i>	<i>16.8</i>	<i>5.0</i>	<i>153</i>

<sup>a</sup> See Table 1, footnote a

<sup>b</sup> Eight organ weights, unless indicated otherwise, nine organs were weighed in each group

<sup>c</sup> One spleen in this group weighed 9.891 mg, more than the spleens from the other eight ducks in this group combined. If this large spleen were included, the mean weight would be 1.6333 mg

<sup>d</sup> The testes of one duck in this group weighed 12.83 g, more than the testes of the other seven ducks in this group combined. If these large testes were included, the mean weight for this group would be 2.42 g

<sup>e</sup> Seven thyroids

The remaining five groups showed daily percentage decreases in Hb ranging from  $-0.071$  to  $-0.231$ . The group dosed with five pellets containing 61 percent lead showed the greatest daily decrease. This group also eroded the most lead per duck per day (Table 2). The same seven groups of ducks showing the lowest daily weight losses (Table 5) and the highest PCVs at the end of the experiment (Table 7) also had the highest Hb values at the end. Except for the 5-37-63 group, the same seven groups also showed the slightest decreases (four groups showed increases) in Hb. As with changes in weight, PCV, and total Hb, lead caused daily changes in Hb even in the presence of iron.

The 5-37-63 group showed Hb values superior to those of the 5-61-39 group, but not to those of the 5-65-35 group, in the mean daily change in Hb per duck. The Hb of the six surviving ducks in the 5-37-63 group was higher than the Hb of the surviving ducks in the 3-61-39, 5-65-35, 1-100-0, 5-61-39, and 3-65-35 groups (Table 7). Hb in the surviving ducks dosed with five pellets containing 37 percent lead and 63 percent iron declined relatively more than change in weight and PCV. However, the decrease in Hb in the 5-37-63 group was caused by exceptionally low values in two of the six surviving ducks. Thus, Hb in most ducks declined only slightly when five pellets containing 37 percent lead and 63 percent iron were ingested. Hb at the end of the experiment was slightly higher and the daily increase in Hb was greater in ducks dosed with five soft steel pellets than in the controls.

#### Weight of Testes

The weights of both testes for the 12 groups of ducks ranged from a low of 0.368 g for the group dosed with one lead pellet to 1.333, 1.401, 1.679, and 2.420 g for groups 5-0-100, 0-0-0, 1-37-63, and 5-37-63, respectively (Table

9). The 5-37-63 group had the highest mean weight, perhaps because the testes from one duck in the group weighed 12.83 g, more than three times as much as the testes of any other duck in the same group. If these testes are omitted, the mean weight of both testes is only 0.933 g.

The normal seasonal cycle of gonad weights in wild male mallards has apparently not been studied. The mean weight of both testes of two mallards collected during February and March in one of the royal parks in London was 13.10 g (Höhn 1947:288), and the mean weight of both testes of three mallards collected during May at the same location was 15.88 g. Sanderson (unpublished) found that the total weight of the testes of one captive wild mallard in a control group was 2.0 g on 2 March; the mean weight was 10.28 g for the two testes from each of 12 ducks dosed with copper shot and sacrificed 160 days later on 1 May 1973, the greatest weight being 19.1 g. Two factors, which may have inhibited the normal growth of testes in ducks used in that experiment, were the dosage of copper shot and a corn diet for the first 100 days (to 2 March). On day 100 the ducks were placed on an improved diet.

In the present experiment, it appears that either the effects of captivity or diet, or both, inhibited the normal growth of the testes. There was a negative, but not significant ( $P > 0.05$ ), linear relationship between the amount of lead administered and weight of the testes at the end of the experiment. The heaviest testes were the pair in group 5-37-63, mentioned previously, that weighed 12.83 g on 28 May. The second heaviest pair were from a duck in group 1-37-63 and weighed 7.05 g on 12 June. No other pair weighed as much as 5.0 g, and only two pairs exceeded 4.0 g.

Apparently the effects of diet or captivity, or both, which inhibited normal growth of the testes, prevented the amount of lead administered from showing a significant effect on testis weight. However, the correlation was negative; histology indicated that lead had an adverse effect on spermatogenesis; and the ducks in the group dosed with one lead pellet had the lightest testes, a finding similar to the findings about several other parameters measured. The seven groups with the highest mean weights of testes included the usual groups (0-0-0, 5-0-100, and all ducks dosed with one lead-iron shot) except that the 1-65-35 group had next to the smallest testes (0.381 g) and was replaced in the top seven groups by the 3-61-39 group. It appears that the mean testis weight of wild mallards on a diet of corn in captivity is not a sensitive indicator of lead toxicity.

### Weight of Thyroids

Weights of thyroids ranged from 36.1 mg for ducks dosed with one lead pellet up to 101.1 mg for ducks dosed with five steel pellets (Table 9). The usual seven groups had the heaviest thyroids except that the control group was replaced by the 5-61-39 group. The contrast between the thyroid weights of the two groups not dosed with lead (the controls and the 5-0-100 group) is striking—44.26 mg for the controls compared with 101.13 mg for the group dosed with steel pellets. There was no significant

( $P > 0.05$ ) correlation between thyroid size and amount of lead administered or between thyroid size and any other of our measurements. We have no explanation for the large size of the thyroids of ducks dosed with steel shot. In a later study with game-farm mallards, Sanderson found no significant effect of ingested steel shot on the size of thyroids (Sanderson unpublished).

### Weight of Spleen

Mean weights of spleens ranged from 222.9 mg for ducks dosed with one lead pellet to 928.4 mg for controls. The usual seven groups had the heaviest spleens except that the 5-65-35 group replaced the 1-61-39 group. Although the difference in weights of the spleens of the two groups not dosed with lead was appreciable, on a relative basis it was not as striking as the difference in thyroid weights—thyroids of the 5-0-100 ducks weighed 130 percent more than thyroids of controls, but spleens of the 5-0-100 ducks weighed 26 percent less than spleens of controls.

In most groups the spleen weights were highly variable, but there were few significant differences among groups. There was a significant ( $P < 0.005$ ) negative correlation between the amount of lead administered and the weight of the spleen.

### Weight of Pancreas

Mean weights of the pancreas varied from 1.07 g for the 3-65-35 group to 1.63 g for the 3-37-63 group (Table 9). The usual seven groups had the heaviest pancreases except that the 5-61-39 group ranked fourth (1.51 g) ahead of ducks dosed only with steel shot (1.48 g) and ducks in the 1-65-35 group (1.48 g). The pancreases of the group dosed with one lead pellet (1.13 g) averaged only slightly heavier than those of the 3-65-35 group (1.07 g). Also, there was little difference in the mean weights of the pancreases in the controls (1.60 g) and the 5-0-100 group (1.48 g). There was a significant ( $P < 0.005$ ) negative correlation between the amount of lead administered and the weight of the pancreas. However, the 5-61-39 group ranked fourth in weight of the pancreas but had the most total lead eroded and the most lead eroded daily per duck (Table 2). The same group had the third highest total amount and daily amount of iron eroded per duck. Thus, simultaneous high levels of lead and iron appeared to have less effect on the pancreas than lower levels of lead had when smaller amounts of iron were administered.

### Weight of Liver

Mean weights of the liver varied from 10.7 for the 1-100-0 group to 23.8 g for the 1-37-63 group. Livers of groups dosed with lead shot only (10.7 g) and with steel shot only (14.9 g) both averaged less than the mean liver weight (16.8 g) of all ducks (Table 9). Livers of ducks in the 1-37-63 group (23.8 g) and in the 3-37-63 group (20.7 g) had higher mean weights than livers of control ducks (19.2 g). Thus, it appears that dosing with lead shot or with steel shot caused a greater reduction in liver weight than did most lead-iron pellet doses. There was a significant ( $P < 0.001$ ) negative correlation between the amount of lead administered and the weight of the liver. Livers of ducks dosed with five steel pellets weighed less than livers of controls.

## Weight of Kidneys

The combined mean weight of both kidneys ranged from 3.8 g (5-0-100) to 6.0 g (5-65-35). Although the weights of kidneys of controls were little different from those of six other groups (1-100-0, 3-65-35, 1-65-35, 1-37-63, 1-61-39, and 3-37-63) or from the mean weight of all kidneys (Table 9), there was a significant ( $P < 0.02$ ) positive relationship between the amount of lead administered and kidney weight. All groups dosed with five lead-iron pellets and the 3-61-39 group had kidneys that weighed more than the kidneys of controls and more than the mean weight of all kidneys. Kidneys of the group dosed only with steel pellets weighed less than the kidneys of any other group.

## Weight of Adrenals

The mean weight of the adrenals ranged from 100.1 mg (5-0-100) to 220.1 mg (5-65-35). As with the kidneys, the adrenals of ducks dosed only with steel pellets weighed less than the adrenals of any other group (Table 9). There was a significant ( $P < 0.001$ ) positive linear relationship between the amount of lead administered and the mean weight of the adrenals. Adrenals of five groups of ducks weighed substantially more and of four groups substantially less than the mean weight of adrenals of controls (Table 9). Except for ducks dosed with steel shot only, all ducks whose adrenals weighed substantially less than adrenals of controls were dosed with low levels of both lead and iron.

## Ratio of Iron to Lead Eroded

The ratio of iron to lead in the pellets (Table 2) appears to be as important as the amount of lead eroded per day as measured by the mean daily rate of weight loss. For example, ducks in groups 5-37-63 and 3-61-39 eroded similar amounts of lead per day, but the ratio of iron to lead eroded daily was nearly three times as high in the

5-37-63 group as in the 3-61-39 group. The daily weight loss in the 5-37-63 group was only 44.8 percent of the rate in the 3-61-39 group (Table 5). Also, daily amounts of lead eroded were similar in the 1-37-63 group (0.0013 g per day) and in 1-65-35 (0.0019 g per day), but the ratio of iron to lead eroded was more than three times higher in the former group. Daily weight losses in the 1-37-63 group were only 54.7 percent of the losses in the 1-65-35 group.

## Lead and Iron Residues in the Liver

The lead in livers of two ducks from each group (Table 10) averaged from 1.4 and 2.2 ppm (wet weight), respectively, for the controls and the group dosed only with steel shot to a high of 54.9 ppm for the 5-61-39 group. Ducks in the 5-61-39 group by far eroded the most lead. There was a significant ( $P < 0.001$ ) positive correlation between the amount of lead eroded and ppm of lead in the liver. Iron eroded tended to be negatively associated with amount of lead in the liver, but the association was not significant ( $P > 0.40$ ). It may be that iron helps to mitigate the effects of lead poisoning in ducks by decreasing the absorption of lead or increasing its excretion, or both.

Livers of two ducks from each group averaged from 775 to 4,000 ppm of iron (wet weight), respectively, for the controls and the 3-65-35 group. The latter group eroded less than the average amount of iron. Ducks dosed only with steel shot eroded the most iron but had only 2,140 ppm of iron in their livers compared with a mean of 2,657 ppm of iron for all livers. There were negative but not significant correlations between the amount of iron eroded and the concentration of iron ( $P > 0.25$ ) and total iron in the liver ( $P > 0.50$ ). However, the reason that these relationships were not significant is probably that ducks dosed with high levels of lead had high levels of iron (both ppm and total amount of iron) in their livers. For

Table 10.—Lead and iron in the liver and the mean amount of lead and iron eroded daily per duck; two ducks were in each group.

Dose <sup>a</sup>	Mean ppm of Lead in Liver (Wet Weight)	Mean Lead Eroded in Grams	Mean ppm of Iron in Liver (Wet Weight)	Mean Iron Eroded in Grams	Mean Total Iron in Liver in mg
0-0-0	1.4	0.000	775	0.000	13.8
1-100-0	41.6	0.108	3,875	0.000	46.3
5-0-100	2.2	0.000	2,140	0.255	23.3
1-37-63	11.3	0.044	1,380	0.076	21.7
3-37-63	36.3	0.097	2,550	0.167	35.5
5-37-63	20.8	0.128	1,515	0.221	26.8
1-61-39	29.6	0.069	2,500	0.044	35.1
3-61-39	33.2	0.144	3,925	0.092	36.1
5-61-39	54.9	0.383	2,695	0.246	45.7
1-65-35	16.4	0.064	3,346	0.035	49.4
3-65-35	32.2	0.140	4,000	0.077	44.6
5-65-35	29.9	0.118	3,180	0.065	34.8
<i>Grand mean</i>	<i>25.8</i>	<i>0.108</i>	<i>2,657</i>	<i>0.106</i>	<i>34.4</i>

<sup>a</sup> See Table 1, footnote a

Table 11.—Lead in the blood; two ducks were in each group.

Dose <sup>a</sup>	Mean ppm of Lead in Blood (Wet Weight)		
	13 May	28 May	12 June
0-0-0	0.35	0.67	0.83
1-100-0	0.26	4.50	7.50
5-0-100	0.14	0.50	0.96
1-37-63	0.52	3.92	3.98
3-37-63	0.42	10.24	10.64
5-37-63	0.51	7.75	10.30
1-61-39	0.54	11.41	7.42
3-61-39	0.32	6.28	5.66
5-61-39	0.40	22.28	15.95
1-65-35	0.62	3.90	3.22
3-65-35	0.34	13.15	9.83
5-65-35	0.33	31.24	5.99
<i>Grand mean</i>	<i>0.40</i>	<i>9.66</i>	<i>6.86</i>

<sup>a</sup> See Table 1, footnote a

example, the 1-100-0 ducks had almost as high a concentration (3,875 ppm) of iron in their livers as the two groups with the highest concentrations of iron—3-65-35 (4,000 ppm) and 3-61-39 (3,925 ppm). The 1-100-0 ducks had the second highest total iron (46.3 mg) in their livers after the 1-65-35 ducks (49.4 mg). There was a positive, but not significant, relationship between the amount of iron administered and the concentration and total iron in the livers of controls, the 5-0-100 group, and the three groups of ducks dosed with shot that were 37 percent iron.

### Lead in the Blood

Lead in the blood of two ducks from each group was determined on the day when pellets were administered and 2 and 4 weeks later. It was higher in all groups 2 weeks after dosing than at dosing (Table 11). However, lead residues in the 5-0-100 and 0-0-0 groups increased only slightly compared with increases in groups dosed with shot containing lead. Lead in the blood of all but one group was essentially the same 4 weeks after dosing as it was after 2 weeks. One duck in the 5-65-35 group showed a sharp decline in lead in the blood from 2 to 4 weeks after dosing, which we cannot explain. Both samples were analyzed twice with similar results. There was a positive relationship ( $P < 0.001$ ) between mean weight of lead eroded and lead in the blood 4 weeks after dosing.

Iron eroded (Table 2) appeared to have little relation to the lead concentration in the blood (Table 11). In contrast, lead, administered either in lead shot or in lead-iron shot, caused substantial increases in concentration and total iron in the liver (Table 10).

### Histopathology of Organs

Abnormal changes occurred in liver, spleen, kidneys, and testes regardless of the concentration of lead, but the

changes related to the amounts of lead. Hemosiderosis was found in the spleens and livers of all ducks except the controls. Intranuclear inclusion bodies were found in the kidney tubule cells of all birds dosed with lead. Bone structure was normal in all groups. Gonad, spleen, liver, and kidneys showed histological variation from group to group.

**TESTES.**—Testes of controls and birds dosed only with steel shot showed active spermatogenesis. Sperm were present in all microscopic fields, and interstitial tissue was well developed. No duck in any other group showed evidence of spermatogenesis. The seminiferous tubules were, in most cases, poorly differentiated by the interstitium.

**SPLEEN.**—Spleens of the two control ducks appeared normal. Birds in the 5-0-100 group had spleens containing a granular pigment, possibly a siederin (iron-containing complex), deposited in both the red and white pulp. All ducks dosed with lead had a moderate amount of granular pigment deposited throughout the spleen. Large amounts of pigment were found in the spleens of the ducks in the 3-61-39 group.

**LIVER.**—Liver sections from controls were not typical of normal liver. Both livers showed vacuolation of hepatocytes, possibly as a result of glycogen storage or as part of a degenerative process, probably both. Livers from birds in the 5-0-100 group showed vacuolation of hepatocytes as well as deposition of a granular pigment similar in appearance to that found in the spleen. Both livers in the 1-37-63 group showed vacuolation of hepatocytes but no pigment deposition. Neither liver from the ducks in the 1-61-39 group showed vacuolation, but one (No. 154) had moderate amounts of pigment deposition. One duck of the 1-65-35 group had vacuolated hepatocytes, and both had pigment deposition in moderate amounts.

One bird from the 3-37-63 group had a normal liver. The other showed pigment deposition, but no vacuolation, and slight necrosis of hepatocytes. Necrosis may have occurred postmortem due to improper fixation of the tissues, but it is known to be an antemortem sign of lead intoxication.

In the other experimental groups, most ducks showed moderate pigment deposition and necrosis of hepatocytes. The necrosis obscured any vacuolation that might have been present by obliterating cellular boundaries. Two exceptions were the ducks of the 5-65-35 group, which, although showing heavy pigment deposition, showed no necrosis or vacuolation of hepatocytes. One duck in the 3-65-35 group had extensive cirrhosis of the liver in addition to pigment deposits and necrosis.

**KIDNEYS**—Intranuclear inclusion bodies were present in the proximal tubule cells of the kidneys of most of the groups of mallards dosed with lead (Table 12). The controls and ducks in the 5-0-100 and 1-61-39 groups had no inclusion bodies, and the kidneys appeared morphologically normal. One duck in the 5-65-35 group also had no inclusion bodies in the nuclei, but there was some degenerative change in the tubule cells.

The significance of the inclusion bodies is not known. Chronic nephritis is a common finding in mammalian

Table 12.—Number of intranuclear inclusion bodies in proximal tubule cells of the kidneys.

Dose <sup>a</sup>	Number of Ducks	Average Number of Inclusions per Oil Immersion Field
0-0-0	2	0
1-100-0	3	30
5-0-100	2	0
1-37-63	2	3
3-37-63	2	10
5-37-63	3	9
1-61-39	2	0
3-61-39	3	23
5-61-39	2	20
1-65-35	2	4
3-65-35	3	32
5-65-35	4	7

<sup>a</sup> See Table 1, footnote a

chronic lead poisoning but has not been related to the presence of inclusion bodies in the proximal tubule cells (National Academy of Sciences 1972).

### Hematology

The controls and birds in the 5-0-100 group showed a relatively constant blood picture throughout the experiment. There were few reticulocytes, and most red and white cells were normal in shape, size, and color. Platelet numbers appeared to be normal, judging from the predosage values.

Because changes over the 4-week period were similar among the groups dosed with varying amounts of lead and differed only in degree (which is difficult to evaluate), these groups are discussed in terms of the two extremes—1-100-0 and 1-37-63. Ducks in the 1-100-0 group showed the greatest changes over the 4-week period. Reticulocytes increased from baseline levels of approximately 5-6 percent of the red cell population to, in some cases, full fields of reticulocytes, poikilocytes, and anisocytes. White cells appeared normal, but differential counts were not performed. Platelets varied erratically in number. Some ducks had very few, others far more than the usual number.

The ducks in the 1-37-63 group showed few changes. The second and third samples contained more reticulocytes than the first. Approximately 15 percent of the red blood cells in the third sample were immature, as compared with 5 or 6 percent in the predosage samples and in control ducks. White blood cells and platelets appeared normal. Blood from ducks dosed with lead is typical of that of ducks suffering varying degrees of anemia.

### Ordering of Groups

We ranked the 12 groups of ducks in numerical order from 18 characters (Table 8). The condition considered

most desirable (usually the highest value, but in some cases the lowest value) was designated by 1. Ties were given equal values. The total scores increased relatively evenly from the first group (controls) to the seventh and from the eighth to the twelfth, but a large increase occurred between the seventh group and the eighth.

### SUGGESTIONS FOR FUTURE RESEARCH

Body weight, PCV, and Hb were all significantly correlated with the amount of lead administered. PCV and Hb gave similar results, but PCV is easier to determine. Only limited information regarding the effects of lead was gained from the weights of organs (most were variable) and from the histopathology studies.

Adequate data about the effects of lead-iron shot in captive mallards may be the mortality rate after 6 weeks and body weight and PCV at 2, 4, and 6 weeks. Such a regime would reduce handling of the ducks, which could be banded when randomly assigned to dosage groups. It would be necessary only to dose the ducks and, at 2, 4, and 6 weeks, to weigh them and take blood samples for PCV determinations.

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