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ZINC TOXICITY IN THE MALLARD DUCK[1]

WILLIAM C. GASAWAY, Department of Zoology, Washington State University, Pullman and Institute of Arctic Biology, Fairbanks, Alaska[2]

IRVEN O. BUSS, Department of Zoology, Washington State University, Pullman

Abstract: Levels of zinc from 3,000 to 12,000 ppm were fed to domestic mallard ducks (*Anas platyrhynchos*) in their diet. Food intake and body weight both showed closely parallel decreases as the level of zinc in the diet increased. In ducks fed zinc, the pancreas and gonads underwent reductions in relation to body weight, whereas ratios of adrenals and kidneys to body weights increased significantly. No significant change occurred in liver:body weight ratios in ducks fed zinc-supplemented diets. The gonads were so reduced in size that the reproductive function was probably lost. In birds fed zinc-supplemented diets, partial paralysis of the legs, diarrhea, and weight loss were noted within 10 days. Severe paralysis, with some ducks unable to walk, was noted after 20 days. Only slight anemia was present in some ducks after 30 days, but by day 45, extreme anemia (5–13 grams Hb per 100 cc of blood) was evident in most ducks. Normal hemoglobin concentrations averaged 18.6 grams per 100 cc of blood. Significant increases in concentrations of zinc were found in the liver, kidneys, pancreas, gonads, and breast muscle. High mortality was observed in all groups treated with zinc-supplemented diets; all but 2 of 24 ducks died within 60 days. Paralysis of the legs, high concentrations of zinc in the pancreas and kidneys, and yellowish-red kidneys may be used for diagnosing zinc toxicosis in mallards.

Heavy metals such as lead, mercury, copper, and zinc have long been known for their toxicity to animals. Copper and zinc are required as trace elements in the diets of animals, but when they occur in relatively high levels, they become toxic. In this particular set of experiments, toxicity of zinc was studied in domestic mallard ducks to determine the hazard of zinc as an environmental pollutant to waterfowl. This study stems from 70 years of observations, summarized by Chupp and Dalke (1964), of dead and dying ducks, geese, and swans in the Coeur d'Alene Lake area in northern Idaho where zinc is found in high concentrations.

The Coeur d'Alene area has been actively mined for at least 70 years and effluents from mines and smelters have been poured into the river systems of the area. These wastes then flow downstream, settling in the rivers and lakes, where the accumulation of metals becomes high.

Lewis and Denecke (1923) found 2.86 percent lead in a sediment sample collected in the Coeur d'Alene valley. Ellis (1940) found a range of 5 to 12 percent zinc in soil encrustations. Chupp and Dalke (1964) analyzed 12 soil samples (type not stated) and obtained from 150 to 9,600 ppm lead, 240 to 7,700 ppm zinc, and 31 to 320 ppm copper. Three samples of emergent aquatic plants (species not stated) were analyzed from which an average of 2,530 ppm lead, 1,930 ppm zinc, and 115 ppm copper were found. As a result, waterfowl that feed in these areas consume larger amounts of heavy metals than waterfowl feeding in areas of naturally low (normal) concentrations. Die-offs of ducks, geese, and whistling swans (*Olor columbianus*) have been noted in this area since the early 1900's (Chupp and Dalke 1964); however, these die-offs have been variable, occurring only in some years. Chupp and Dalke indicated that their tissue analyses for heavy metals and the early studies by the Bureau

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[2] Present address: Institute of Arctic Biology, Fairbanks, Alaska.

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of Biological Survey led them to believe that the waterfowl were dying from poisoning by heavy metals.

Growing white leghorn and white rock chickens can tolerate 1,000 ppm zinc in their feed, but 1,500 ppm and above depresses the growth rate (Roberson and Schaible 1960). These workers compared the oxide, sulfate, and carbonate forms of zinc and concluded that at higher levels of zinc in the diet, the carbonate resulted in the highest mortality and the greatest retardation of growth. The effect of the sulfate was less than the effect of the carbonate. Oxide was next in order. Roberson and Schaible (1960) suggested that as a result of the lower solubility of the compound, the oxide could be tolerated to a greater extent than the other forms. Johnson et al. (1962) reported similar findings for growing crossbred broiler chicks whose diets were supplemented with zinc oxide. They found low mortality after 10 weeks on levels up to 5,000 ppm zinc and reported only 2.5 and 15 percent mortality at 3,000 and 5,000 ppm zinc, respectively.

The findings of Brink et al. (1959) showed tolerance of swine for zinc to be similar to that of the chicken. They also found food intake and gain in weight to be depressed when on diets supplemented with greater than 1,000 ppm zinc. Death occurred frequently within 21 days at the level of 8,000 ppm zinc in the food.

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MATERIALS AND METHODS

Fifty mallard ducks 7 weeks of age were purchased from a commercial duck breeder. Four levels of reagent grade zinc carbonate (in ppm air-dried weight) were incorporated into the food of the ducks: 3,000, 6,000, 9,000, and 12,000, and a control diet with no zinc added. All ducks were fed the same basal food: a chicken developer–turkey finisher (Table 1). The grains were ground, the zinc mixed in, and this mixture was compressed into 3/16-inch pellets, insuring that the zinc carbonate would not separate from the food.

Table 1. Composition of basal food.[a]

Ingredient	Proportion
Corn	32.9
Wheat	32.9
Peas	21.5
Soybean oil meal (50 percent protein)	5.0
Fish meal, herring (70 percent protein)	2.5
Dehydrated alfalfa (18 percent protein)	2.5
Dicalcium phosphate	1.0
Ground limestone ($CaCO_3$)	1.0
Iodized salt (NaCl)	0.4
Chick developer premix[b]	0.25
Trace minerals[c]	0.25
Total	100.00

[a] Data from feed plant, Washington State University.
[b] Provides per pound of feed: vitamin A, 2,000 IU; vitamin D_3, 500 IU; riboflavin, 1 mg; Santoquin, 29 mg.
[c] Contains not less than: 10 percent Mn, 10 percent Fe, 1 percent Cu, 10 percent Zn, 0.3 percent Co, and 14 percent Ca.

Using a table of random numbers, we chose 30 ducks at random, 15 of each sex, and then 3 ducks of each sex were randomly selected to make five groups of 6 ducks each. After the level of zinc concentration was assigned at random to the groups, the ducks were randomly assigned one to a cage. Two laying batteries designed for chickens, each containing three layers of six individual cages, were used to hold the birds. The photoperiod was held constant at 10.5 hours per day throughout the experiment.

Food was made available ad libitum in individual containers so that consumption by each duck could be measured. All measurements were taken between 12:00 noon and 1:00 PM in an attempt to reduce sampling variability.

The weight of each duck was recorded

on day zero and at 10-day intervals thereafter.

To study the physiological effects on blood, hemoglobin levels and hematocrits were determined. Blood samples were taken from the brachial vein in the wing. Hematocrits were determined to the nearest percent in 75 mm heparinized microhematocrit tubes and read on a Clay–Adams microhematocrit reader.

The hemoglobin concentration was determined spectrographically after the method of Lucas and Jamroz (1961:231–232). A standard curve prepared by E. M. Denington and Lucas (personal communication) was used to convert the percentage transmission into grams of hemoglobin per 100 cc of blood.

Paralysis of the legs was noted in some ducks shortly after they were placed on the zinc diet. The degree of paralysis was observed at 10-day intervals by noting the duck's ability to walk, then subjectively rating the duck in one of four groups: only stand, barely walk, walks fairly well, or walks normally.

On the 60th day of the experiment, all surviving ducks were killed. Ducks that had died previously were preserved by freezing. Weights of the following organs were recorded for each duck: pancreas, liver, left kidney, adrenals, and gonads. Tissue samples were taken from each of the above organs plus breast muscle. Entire organs were used as the tissue sample if the organ weighed less than 3 grams. Organs that weighed more than 3 grams were blended and an aliquot taken. The breast muscle was sampled from the origin of the left pectoralis major near the keel of the sternum.

The concentration of zinc in various organs and tissues was determined by atomic absorption spectrophotometry. The method of preparation consisted of placing weighed fresh tissue into a test tube, adding approximately 10 ml of concentrated nitric acid, and heating the tube over a hot plate until the tissue dissolved and the solution became clear. The sample was then diluted to a known volume, and the absorption value of the solution was read on a Perkin Elmer, Model 310, atomic absorption spectrophotometer. A standard curve was prepared from six solutions of known concentration.

The statistical design was completely randomized. Analysis of variance was calculated for organ:body weight ratios and concentrations of zinc in tissues. Duncan's new multiple-range test was used to test the significance between mean values (Steel and Torrie 1960:107–109). A statistically significant difference at the 0.05 level will be referred to as significant and the 0.01 level will be referred to as highly significant.

RESULTS

Food Consumption

Ducks reduced their food intake immediately after being placed on their new diet containing zinc. The drop in food intake was related to the amount of zinc in the diet. Birds that were fed the lowest zinc supplements, on the average, ate the most; birds that were fed the highest concentrations ate the least amount of food (Table 2). All groups, including the controls, showed a reduction in food consumption after the initial 10-day period. The general trend of reduced food consumption seen in the controls may have resulted in part from a general increase in room temperature from about 65 F to 75 F during the experimental period. In several cases, ducks on diets containing 9,000 ppm zinc and 12,000 ppm zinc virtually stopped eating. Some ducks ate only 1 to 3 ounces of food

Table 2. Average total food consumed by each mallard fed zinc-supplemented diets.[a]

Content of Zinc Supplemented in Diet (ppm)	Sex	Average Total Food Consumed (ounces) by Each Duck During Each 10-Day Period					
		0–10	11–20	21–30	31–40	41–50	51–60
Control (0)	M	26.0 ± 6.9[b]	23.0 ± 6.1	22.0 ± 6.0	18.3 ± 2.9	12.7 ± 0.6	15.0 ± 2.6
	F	23.0 ± 3.0	18.0 ± 4.0	20.0 ± 3.6	17.0 ± 2.6	13.0 ± 2.0	17.0 ± 3.0
Mean		24.5 ± 5.0	20.1 ± 5.3	21.0 ± 4.6	17.7 ± 2.6	12.8 ± 1.3	16.0 ± 2.8
3,000	M	18.0 ± 3.6	11.0 ± 3.5	12.7 ± 4.1	12.0 ± 2.6	10.0 ± 1.7	12.5 ± 2.1(2)
	F	17.7 ± 10.8	9.0 ± 1.0	12.7 ± 6.6	14.0 ± 8.0	12.0 ± 0.0(2)	—
Mean		17.8 ± 7.2	10.0 ± 2.5	12.7 ± 5.0	13.0 ± 5.4	10.8 ± 1.6(5)	12.5 ± 2.1(2)
6,000	M	5.0 ± 2.6	5.7 ± 0.6	6.0 ± 4.6	11.0(1)	—	—
	F	8.7 ± 3.5	5.0 ± 3.6	6.3 ± 3.2	7.0 ± 1.4(2)	—	—
Mean		6.8 ± 3.4	5.4 ± 2.3	6.2 ± 3.5	8.3 ± 2.5(3)	—	—
9,000	M	4.3 ± 3.2	4.3 ± 2.5	5.0 ± 4.6	9.0(1)	—	—
	F	3.0 ± 2.6	2.0 ± 1.0	1.3 ± 1.5	3.7 ± 2.1	—	—
Mean		3.7 ± 2.7	3.2 ± 2.1	3.2 ± 3.7	5.0 ± 3.2(4)	—	—
12,000	M	3.7 ± 0.6	1.7 ± 0.6	1.3 ± 0.6	4.3 ± 2.3	5.0(1)	—
	F	4.7 ± 4.7	1.3 ± 0.6	2.0 ± 2.0	1.0(1)	—	—
Mean		4.2 ± 3.1	1.5 ± 0.5	1.7 ± 0.5	3.5 ± 2.5(4)	—	—

[a] Three birds of each sex were averaged except where the sample size is indicated in parentheses.
[b] Standard deviation.

per 10 days, whereas the controls consumed between 1.3 and 3.0 ounces of food per day. Both males and females followed similar patterns in reduction of food intake, and both consumed nearly the same amount of food for each respective level of zinc. In general, food consumption was consistently and inversely related to the concentration of zinc in the diet.

Changes in Body Weight

Body weight declined for all ducks fed a zinc-supplemented diet, as would be expected with the reduced caloric intake. The controls gained weight during the first 10 days and then fluctuated in weight during the next 60 days (Table 3). No major change in weight was expected in the controls, since they were at their adult weight at the beginning of the experiment. During the first 30 days, ducks fed 9,000 ppm zinc lost weight at the fastest rate, and ducks fed 3,000 ppm lost weight at the slowest rate. The greatest rate of weight loss occurred during the first 10 days and thereafter the rate diminished. By 30 days, birds fed the two highest concentrations of zinc had lost an average of 40 percent of their original body weight, whereas ducks fed 3,000 ppm had only lost an average of 18 percent of their initial weight. Females lost weight at a faster rate than males, with the exception of the ducks fed 3,000 ppm zinc. The average percentage of original body weight at the time of death or sacrifice ranged from 47 to 54 percent for groups supplemented with zinc (Table 3). In all cases, females lost, on the average, a greater percentage of their body weight than males.

In order to observe the physical effects of extreme reduction in food intake, two hen mallards were placed in separate pens without food. Weight loss in these two birds was greater than in any birds fed the zinc diet. After 29 days of starvation, they

ble 3. Average percentage of original weight of mallard ducks fed zinc-supplemented diets.[a]

CONTENT OF ZINC SUPPLEMENTED IN DIET (ppm)	SEX	AVERAGE PERCENT OF ORIGINAL WEIGHT ON DAY						
		10	20	30	40	50	60	At Death
ontrol (0)	M	101 ± 1[b]	99 ± 1	112 ± 4	104 ± 3	98 ± 3	97 ± 3	
	F	107 ± 3	101 ± 5	100 ± 6	100 ± 4	95 ± 12	96 ± 5	
Mean		104 ± 4	100 ± 4	101 ± 4	102 ± 3	97 ± 8	96 ± 3	
3,000	M	98 ± 8	89 ± 13	83 ± 17	82 ± 16	78 ± 18	81 ± 18(2)	47(1)
	F	95 ± 9	87 ± 6	81 ± 4	70 ± 10	64 ± 4(2)	—	48 ± 1
Mean		97 ± 8	88 ± 9	82 ± 11	76 ± 14	72 ± 15(5)	81 ± 18(2)	48 ± 1(4)
3,000	M	88 ± 10	75 ± 10	74 ± 13	74(1)	—	—	58 ± 9
	F	79 ± 6	67 ± 4	61 ± 9	62 ± 12(2)	—	—	50 ± 3
Mean		83 ± 9	71 ± 8	67 ± 12	66 ± 11(3)	—	—	54 ± 7
9,000	M	80 ± 3	68 ± 3	63 ± 2	58(1)	—	—	48 ± 5
	F	76 ± 3	62 ± 4	56 ± 8	50 ± 7	—	—	47 ± 9
Mean		78 ± 3	65 ± 5	60 ± 6	52 ± 7	—	—	48 ± 6
2,000	M	83 ± 5	68 ± 4	63 ± 6	55 ± 9	63(1)	—	50 ± 4
	F	81 ± 7	68 ± 6	57 ± 6	66(1)	—	—	44 ± 5
Mean		82 ± 5	68 ± 4	60 ± 6	58 ± 9	63(1)	—	47 ± 5

[a] Three birds of each sex were averaged except where sample size is indicated in parentheses.
[b] Standard deviation.

'eighed only 42 percent of their original ody weight. One of the hens died on day 9, the other died on day 31, and neither ıowed the signs exhibited by ducks fed inc-supplemented diets.

)rgan Weights

Highly significant reductions were found ı the wet weights of pancreases, livers, and onads of ducks fed zinc-supplemented diets. Weights of kidneys and adrenals were very near those of the controls (Table 4). Changes in organ weights did not parallel reductions in body weight of the test birds. Organ:body weight ratios (Table 5) demonstrate this more clearly. Only the pancreas and gonads showed weight reductions in relation to body weight, whereas weights of adrenals and kidneys increased significantly (Table 5).

ıble 4. Average fresh weights of organs from mallard ducks at time of death or when killed on day 60.

CONTENT OF ZINC SUPPLEMENTED IN DIET (ppm)	SEX	FRESH WEIGHT OF ORGANS (grams)					
		Pancreas	Liver	Adrenals	Left Kidney	Testes	Ovary
Control (0)	M	1.98 ± 0.51[a]	21.92 ± 3.87	0.11 ± 0.03	2.77 ± 0.77	4.72 ± 3.03	
	F	2.07 ± 0.31	16.00 ± 1.00	0.09 ± 0.02	2.31 ± 0.33		0.77 ± 0.68
3,000	M	0.62 ± 0.32	14.97 ± 5.24	0.13 ± 0.07	3.25 ± 0.82	0.16 ± 0.08	
	F	0.49 ± 0.96	10.49 ± 3.73	0.10 ± 0.01	2.23 ± 0.90		0.09 ± 0.02
6,000	M	0.61 ± 0.03	11.51 ± 1.76	0.12 ± 0.04	2.91 ± 1.25	0.20 ± 0.10	
	F	0.58 ± 0.07	9.17 ± 1.49	0.10 ± 0.05	2.02 ± 0.50		0.07 ± 0.07
9,000	M	0.90 ± 0.07	9.38 ± 3.63	0.11 ± 0.03	1.86 ± 0.38	0.09 ± 0.04	
	F	0.70 ± 0.09	10.78 ± 3.13	0.07 ± 0.01	1.60 ± 0.36		0.41 ± 0.60
2,000	M	0.72 ± 0.03	12.38 ± 3.28	0.13 ± 0.03	2.33 ± 0.41	0.10 ± 0.06	
	F	0.73 ± 0.10	8.29 ± 1.33	0.11 ± 0.03	1.85 ± 0.16		0.12 ± 0.06

[a] Standard deviation.

Table 5. Average organ:body weight ratios from mallard ducks fed zinc-supplemented diets. Six birds were used on each treatment, three of each sex.

Treatment and Sex	Organ:Body Weight Ratio × 100 (percent of body weight)				
	Pancreas	Liver	Adrenal	Kidney	Gonads[a]
Males and Females					
Control	0.22 ± 0.05[b]*a*[c]	1.97 ± 0.17*a*	0.01 ± 0.00*a*	0.27 ± 0.04*a*	0.46 ± 0.36*a* 0.10 ± 0.10*c*
3,000	0.09 ± 0.03*b*	2.10 ± 0.59*a*	0.02 ± 0.01*b*	0.45 ± 0.12*b*	0.23 ± 0.01*a* 0.02 ± 0.01*c*
6,000	0.12 ± 0.01*b*	2.01 ± 0.17*a*	0.02 ± 0.01*b*	0.47 ± 0.13*b*	0.03 ± 0.02*b* 0.01 ± 0.01*c*
9,000	0.17 ± 0.08*b*	2.07 ± 0.44*a*	0.02 ± 0.01*b*	0.36 ± 0.04*b*	0.02 ± 0.01*b* 0.02 ± 0.01*c*
12,000	0.15 ± 0.04*b*	1.98 ± 0.20*a*	0.02 ± 0.01*b*	0.42 ± 0.11*b*	0.02 ± 0.01*b* 0.03 ± 0.02*c*
Males					
All zinc supplemented	0.12 ± 1.07	2.00 ± 0.39	0.02 ± 0.01	0.43 ± 0.12	
Females					
All zinc supplemented	0.14 ± 0.04	2.08 ± 0.36	0.01 ± 0.01	0.42 ± 0.10	

[a] Males are listed first in this column.
[b] Standard deviation.
[c] Values within a column followed by a common letter are not significantly different at the 95 percent level. Comparisons are made within each sex in the column for gonads.

Liver:body weight did not fluctuate from normal even though there was a significant reduction in liver weight. Organ:body weight ratios for all organs did not differ significantly between males and females. Testicular and ovarian growth was significantly retarded at a time when gonadal growth of controls was high, and testicular wet weight of the controls was 24 to 52 times that of birds fed zinc (Table 4). This great reduction in gonad development implied that the impact on breeding success could be significant if certain zinc concentrations were present.

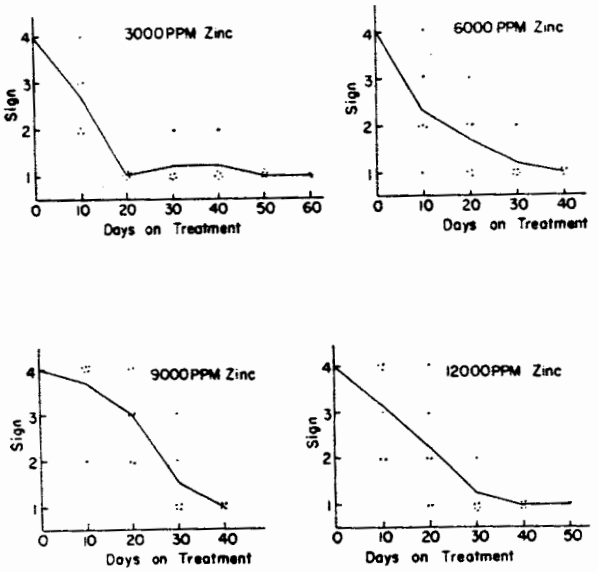

Fig. 1. Average degree of paralysis of legs observed in mallard ducks treated with zinc-supplemented diets—stand only, barely walk, walk fairly well, and walk normally. Dots are individual ducks.

Signs of Toxicity

Two conspicuous signs of zinc toxicosis were paralysis of the legs and diarrhea. The rate at which paralysis of the legs appeared in relation to the level of zinc in the diet was assessed by assigning a numerical rating to the duck's ability to use its legs. The mean values for these sign ratings were plotted against days (Fig. 1). Paralysis appeared at the fastest rate in the ducks fed 3,000 ppm zinc, the lowest level. Ducks fed more than 3,000 ppm zinc demonstrated little difference in the rate that paralysis

Table 6. Average hematocrit values of mallard ducks fed zinc-supplemented diets.

Content of Zinc Supplemented in Diet (ppm)	Sex	Average Hematocrit on Day 0	15	30	45	60
Control (0)	M	50 ± 3[a]	48 ± 2	49 ± 3	47 ± 1	47 ± 1
	F	45 ± 4	50 ± 4	50 ± 3	50 ± 5	48 ± 6
Mean		48 ± 4	49 ± 3	50 ± 3	49 ± 3	48 ± 4
3,000	M	44 ± 3	45 ± 4	47 ± 1	42 ± 4	43 ± 4(2)
	F	47 ± 4	46 ± 3	47 ± 3	42 ± 3(2)[b]	—
Mean		47 ± 4	46 ± 3	47 ± 2	42 ± 3(5)	—
6,000	M	47 ± 3	47 ± 3	37 ± 8	—	—
	F	49 ± 2	45 ± 3	43 ± 11	18(1)	—
Mean		48 ± 3	46 ± 3	40 ± 9	—	—
9,000	M	48 ± 3	47 ± 0	43 ± 4	27(1)	—
	F	46 ± 5	49 ± 5	47 ± 4	21(1)	—
Mean		47 ± 4	48 ± 3	45 ± 4	24 ± 4(2)	—
12,000	M	48 ± 1	48 ± 2	49 ± 5	36 ± 1(2)	—
	F	48 ± 2	50 ± 1	51 ± 1	34(1)	—
Mean		48 ± 1	49 ± 2	50 ± 3	35 ± 2	—

[a] Standard deviation.
[b] Three birds of each sex were averaged except where sample size is indicated in parentheses.

set in, as shown by the slope of the lines and the length of time necessary for all ducks to attain a rating of one. The controls remained normal throughout the entire 60-day experimental period.

There was a direct relationship between the amount of zinc and the amount of food consumed per bird, since zinc was an integral part of the food pellets. Zinc consumption showed an inverse relationship to concentration of zinc in the food. The reduction in food consumption and consequent reduction in zinc consumption was not anticipated to this extent.

Diarrhea was present in most birds after 15 days. Their excreta was a moderately viscous liquid.

Postmortem Observation

Ducks on zinc-supplemented diets had yellowish or reddish-yellow kidneys, whereas all the controls had deep red kidneys. No necrosis, lesions, or hemorrhaging was observed in any tissues.

Hematocrit and Hemoglobin Levels

Hematocrit values for the six controls throughout the experiment were 48.4—males were 48.2 and females were 48.6 (Table 6). No marked reduction in hematocrit value was noted until day 30 when two ducks that were being fed 6,000 ppm zinc showed hematocrit readings of 28 and 33. The remaining birds appeared normal, fluctuating slightly above or below their previous values. By day 45 there were only 12 living ducks on zinc-supplemented diets; 8 showed acute reductions in hematocrit values. Two ducks that were fed 3,000 ppm zinc and that were alive at day 60 showed only slight reductions in hematocrit values.

A reduction in hemoglobin occurred by day 30 in the group of ducks fed 3,000 ppm, 6,000 ppm, and 9,000 ppm zinc (Fig. 2).

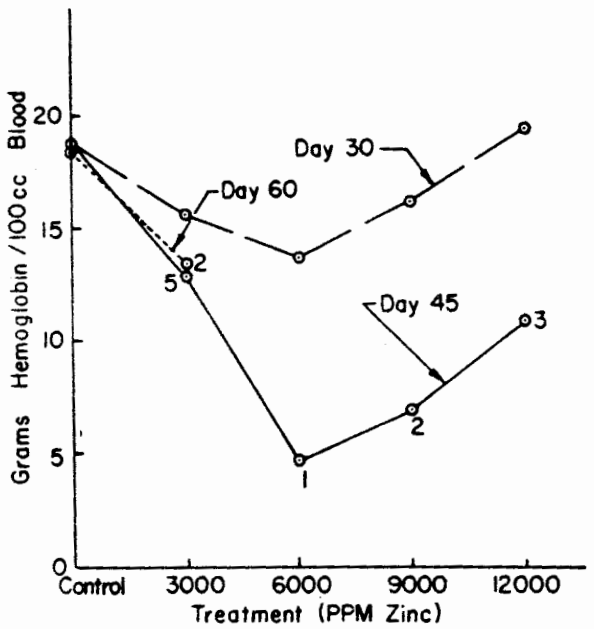

Fig. 2. Average grams hemoglobin per 100 cc of blood from mallard ducks treated with zinc-supplemented diets. Six ducks were averaged on each treatment unless noted otherwise.

Average grams of hemoglobin per 100 cc of blood in control ducks was 18.8 ± 0.6 (mean ± SD). On day 45, all birds fed zinc-supplemented diets showed marked reductions in the amount of hemoglobin. Two ducks that lived the full 60 days on a diet with 3,000 ppm zinc averaged 13.4 grams of hemoglobin. Although these two birds demonstrated a marked reduction in the amount of hemoglobin, they retained a normal hematocrit. The average amount of hemoglobin in the controls remained nearly constant throughout the experiment. A highly significant linear correlation was found between hematocrit and hemoglobin values on days 30 ($r = 0.89$) and 45 ($r = 0.94$). This correlation indicated that a reduction in the volume of red blood cells was correlated with a reduction in the amount of hemoglobin.

Mortality

Severe mortality occurred after 30 days in all groups of ducks fed zinc-supplemented diets. The survival rate was highest among ducks supplemented with 3,000 ppm zinc; two were living at day 60.

Zinc Concentration

Organisms fed high levels of zinc may encounter problems in maintaining homeostasis of tissue concentration of this particular element. Zinc regulatory problems were encountered by the ducks we studied, and tissue levels increased above the normal amounts found in the controls.

Marked increases in average zinc concentrations were noted in the pancreas, liver, testes, and kidneys, and smaller increases were found in the adrenals and ovaries (Table 7). Only in the kidneys was there a significant difference in zinc concentration among groups of ducks supplemented with zinc.

Concentrations in the pancreas averaged 89 ppm zinc in the controls, and groups fed zinc supplements averaged 15 to 30 times this level. Zinc concentrations in the pancreas seemed to show a direct relationship to the concentration of zinc in the diet, and only in the pancreas was this progression noted.

Zinc concentrations in livers of control birds averaged 54 ppm, and ducks fed zinc-supplemented diets averaged between 340 and 483 ppm zinc in their livers. These zinc concentrations represent a six- to nine-fold increase above the control level. Zinc concentration in livers of males and females did not differ significantly.

Zinc concentrations in the kidneys of controls averaged 27 ppm, and ducks with zinc-supplemented diets averaged between 311 ppm and 519 ppm. Zinc concentrations in male and female kidneys showed highly significant differences; females were consistently lower except for the group fed 6,000 ppm zinc.

Table 7. Average ppm zinc in fresh tissues of mallard ducks at time of death or sacrifice. Six birds were used on each treatment, three of each sex.

Content of Zinc Supplemented in Diet (ppm)	Average Zinc in Fresh Tissue (ppm)						
	Pancreas	Liver	Adrenals	Kidney	Breast Muscle	Testes	Ovary
Control	89 ± 36[a]*a*[b]	54 ± 17*a*	45 ± 21*a*	27 ± 5*a*	14 ± 8*a*	17 ± 3*a*	31 ± 33*a*
3,000	1,252 ± 472*ab*	401 ± 122*b*	88 ± 34*a*	413 ± 101*bc*	32 ± 12*b*	78 ± 49*a*	71 ± 22*a*
6,000	2,061 ± 507*ab*	483 ± 271*b*	100 ± 40*a*	453 ± 239*bc*	30 ± 5*b*	61 ± 21*a*	97 ± 10*a*
9,000	2,282 ± 456*b*	461 ± 162*b*	91 ± 31*a*	311 ± 69*b*	34 ± 7*b*	95 ± 55*a*	67 ± 22*a*
12,000	2,672 ± 745*b*	340 ± 141*b*	86 ± 49*a*	519 ± 359*c*	31 ± 5*b*	103 ± 7*a*	64 ± 33*a*

[a] Standard deviation.
[b] Values within a column followed by a common letter are not significantly different at the 95 percent level.

Samples of breast muscle averaged about twice as much zinc as the controls (14 ppm). The breast muscle had the lowest concentration of any tissues tested. Adrenal glands increased in zinc concentration to twice that of the controls. Controls averaged 45 ppm zinc in the adrenals, and zinc-treated ducks ranged between 86 and 100 ppm zinc. Testes also showed higher concentrations of zinc in birds fed zinc-supplemented diets than in the controls. The average level of zinc in testes of the controls was 17 ppm, and zinc-supplemented birds ranged from three to six times greater.

Ovarian zinc concentration showed the same trend evident in other organs. The average concentration in the ovaries was 31 ppm in controls, and averages for the four groups of ducks fed zinc were two to three times higher. One duck showed an extremely high concentration of zinc in the ovary, nearly 12 times higher than the next highest value recorded, thus the value was discarded.

DISCUSSION

One of the earliest signs of toxicity detected in this study was a loss of weight that appeared to correspond closely to food intake. However, ducks fed normal untreated food in the quantity consumed by zinc-supplemented ducks would probably have shown similar weight changes. Food intake was reduced by ducks fed zinc from the first day of treatment. This immediate reduction in food intake suggests unpalatability as the main causal factor rather than the toxic effect of zinc. Grant–Frost and Underwood (1958), Sadasivan (1951), and Ott et al. (1964) found similar responses to zinc-supplemented diets in rats and calves.

The ducks apparently detected the relative concentration of zinc carbonate in food, since their food intake was reduced in relation to the concentration of zinc in the diet. A reduction in food intake and a corresponding loss of weight in wild ducks during a portion of the migration could have profound effects on the distance of migration and reproductive potential. The question could be posed: Would ducks occupying an area containing high levels of zinc in plants and soil remain long in that area? The high mobility of ducks suggests that they could move to another area if the food were unpalatable. The fact that waterfowl appear to die from heavy metal poisoning in zinc-ladened areas supports the idea that wild ducks will tolerate heavy metals in their diet long enough to succumb to them.

In the Coeur d'Alene region, concentrations of zinc in plants are less than those used in the present study. Still, the level of zinc found in polluted areas could be

important considering the potential toxic effect on ducks. Based on the effects observed at 3,000 ppm zinc in the diet, mallard ducks feeding in an area that has about 2,000 ppm zinc in plants could soon suffer from toxicosis.

Closely related to total weight loss are weight changes in specific areas of the body, namely, in the visceral organs. The most spectacular weight loss and organ regression occurred in the gonads. With the regression in the size of the gonads, a nonfunctional status may be assumed, although it was not demonstrated. The reproductive capacity may be lost for a breeding season or longer in wild ducks with nonlethal exposures to high zinc in the diet. The implication of lowering the fecundity in birds migrating through or nesting in a zinc-ladened area is clear.

Ducks fed a basal chicken developer–turkey finisher in the present studies might have demonstrated zinc tolerances or signs of toxicity of different magnitudes on other diets. Sadasivan (1951) and McCall et al. (1961) showed changes in zinc tolerance in rats when the protein and fat levels were varied. Zinc was less available to chickens and pigs fed plant protein than to those fed animal protein diets (O'Dell and Savage 1957, Smith et al. 1959). Phytate, found in plant proteins but not in animal proteins, decreased zinc availability (O'Dell and Savage 1960). Wild mallards have an extremely diverse diet, which is determined by availability as well as by preference and leads to variability in the toxic response to zinc.

Paralysis common to all ducks on zinc-supplemented diets soon immobilized the birds. Similarly, the chance of survival for wild ducks ingesting moderate concentrations of zinc would be lessened in many ways, probably through impairment of the birds' ability to feed. Chupp and Dalke (1964) described no paralysis of the legs in sick whistling swans examined from the Coeur d'Alene region. Paralysis of the legs is rarely reported by investigators of zinc toxicity, although Vallee's (1959) account of paralysis in a dog may indicate that zinc poisoning does not affect all species similarly.

Although the diarrhea and accompanying reddish-yellow kidneys may have some value as a field technique in pointing to zinc poisoning, birds affected by lead toxicosis also usually have diarrhea. Yellowish kidneys, however, may be more indicative of zinc poisoning, since, to our knowledge, such coloring has not been reported by investigators of lead poisoning.

All ducks on zinc-supplemented diets were anemic but this sign occurred late. By day 30, only a small number of the ducks showed a reduced hematocrit value, or a reduction in the hemoglobin from the control value. As suggested by the corresponding decreases in hematocrit and hemoglobin values found in the present study, the observed anemia may result in part from the reduction in the red cell volume or in the number of red cells. Smith and Larson (1946) found that excessive zinc caused a microcytic and hypochromic anemia, which suggests that reduction in hematocrit values results partially from a decrease in the size of red blood cells. In the present study, a reduction in the size of the red blood cells as a cause of anemia did not appear to be the case with all ducks, since several showed reductions in hemoglobin with normal hematocrits.

Impairments in the physiological capacity of the blood to transport oxygen to body tissues could reduce mobility and activities of wild birds. If zinc toxicosis occurred under natural conditions, nearly 4 weeks would elapse before this effect would influence the ducks or could be detected.

A significant increase was found in the average zinc concentration in all tissues tested from the zinc-supplemented groups of ducks, but this increase was without a correlation between tissue concentrations and the level of zinc in the diet, or zinc consumption, or length of time on the diet. The organs concentrating the largest amount of zinc appear to be related to organ function—either zinc excretion or metabolism. The pancreas contained concentrations several times higher than the other tissues tested. A main excretion route of zinc from the dog's body is by the pancreas and into the intestine, where it passes in the feces (Montgomery et al. 1943, Birnstingl et al. 1956); the excretion of zinc by the pancreas may account for the higher concentration in the pancreas.

When determining whether wild ducks have been feeding on plants containing abnormally high concentrations of zinc and when testing to see if zinc toxicity has resulted, the levels of zinc in the pancreas, liver, and kidneys may be used as diagnostic supporting evidence. The concentration in the pancreas may be the best single criterion for diagnosis of zinc toxicity.

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