Preliminary Assessment of Sources of Lead Contamination in Tundra Swans Frequenting the Weaver Bottoms

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ABSTRACT

Sources of lead contamination for tundra swans were studied in the Weaver Bottoms, a backwater area of the Mississippi River in southeastern Minnesota in 1985 and 1986. Densities of lead shot in the top 15-20 cm of sediment within 200 m of sites used by waterfowl hunters were 27.6 shot/square meter in an area heavily used for waterfowling and 13.1 shot/square meter in an area less heavily used. Sampling in two areas closed to hunting yielded no shot in any sample. Other lead objects, such as fishing sinkers, were not encountered in any sample. Tubers of the arrowhead plant, Sagittaria latifolia, which are used by tundra swans for food were analyzed for lead content. The mean level of contamination among four sampling areas was 0.35 ppm wet-weight (1.87 ppm dry-weight). No significant trends were noted between sites near or far from a major highway and those open or closed to hunting. Daily intake of arrowhead tubers by swans was estimated by determining the tuber population in an area where swans concentrate during the fall migration before and after the migration to determine total loss of tubers (185,034 kg) and dividing this by the total swan-days of use of the area (23,537). The total loss of tubers/swan-day of use was 7,860 g. This is regarded as an overestimate of swan intake, however, because other species steal tubers from the swans as they feed. Even if all of the tubers were taken by swans and all of the lead from the tubers were transferred to the swans' bloodstream, it would take 46 days of feeding to ingest the equivalent of one #6 lead shot from this area. Since most swans spend no more than three weeks in the area, it is unlikely that the lead from arrowhead tubers contributes significantly to lead poisoning in tundra swans in this area. Two swans found dead or moribund in the area were x-rayed for shot and the livers analyzed for lead. No shot were found and the lead levels were low. Death was attributed to unknown factors.

INTRODUCTION

This study was undertaken for several reasons. Lead poisoning has long been recognized as a major cause of waterfowl mortality (Sanderson and Bellrose, 1986). It has more recently been recognized as a major mortality source for bald eagles. Reichel et al. (1984) reported 17 of 293 bald eagles necropsied between 1978 and 1981 probably died of lead poisoning. The Weaver Bottoms area of the Mississippi River near Weaver, Minnesota is host each fall to tens of thousands of waterfowl including tundra swans (*Cygnus columbianus*) and canvasbacks (*Aythya valisineria*). It also hosts several hundred bald eagles which feed extensively on crippled waterfowl. Waterfowl exhibiting symptoms of lead poisoning are easy targets for bald eagle predation. Eagles also scavenge carcasses of waterfowl already dead. The Weaver Bottoms is a popular waterfowl hunting area and has been so for many years. As such, contamination of the area with spent lead shot is to be expected. Although use of lead shot has been prohibited in the Weaver Bottoms area for a decade, some hunters have persisted in its use. A portion of the Weaver Bottoms is completely closed to hunting and thus is not expected to have spent shot in its sediment. This is the area where swans congregate during the day in their fall migration. However, some of the swans do fly out into hunted areas during the night and even during the day late in the season and may be exposed to spent shot while feeding in these areas.

Another reason for the study comes from contamination of the plants growing within the Weaver Bottoms. In 1976, Anderson (1978) sampled various plants from the area. He found lead in arrowhead (*Sagittaria latifolia*) roots at an average concentration of 1.97 ug/g wet-weight (16.1 ug/g dry-weight). Another researcher found that arrowhead tubers were extensively utilized by tundra swans while at the Weaver Bottoms. By sampling tuber density before and after the swans' migration, and by counting swan numbers in the area throughout their migration, he estimated an intake of 2.8 kg/bird/day (6.1 pounds/bird/day) (Limpert, 1974). If the tubers held the same amount of lead as the roots, and if consumption were indeed 2.8 kg/bird/day, then an amount of lead equivalent to that in one #6 lead shot could be ingested in 25 days' feeding.

This study was initiated in 1985 with four objectives:

1. To determine the density of lead shot in Weaver Bottom sediments both in areas open to hunting and closed to hunting.
2. To determine residues of lead in samples of arrowhead tubers taken from four areas of the Weaver Bottoms.
3. To determine the amount of arrowhead tubers eaten each day by tundra swans using essentially the methods of Limpert (1974).
4. To determine tissue concentrations of lead in dead or moribund swans found in the Weaver Bottoms as well as to determine presence of shot in their bodies by x-ray.

METHODS

Lead Shot Distribution

Sediment sampling for lead shot was conducted in four areas of the Weaver Bottoms within beds of arrowhead (Figure 1). The four areas are designated hereafter as follows: CL-RI, an area closed to hunting and adjacent to the Mississippi River channel; CL-HWY, an area closed to hunting and adjacent to Highway 61, a four-lane highway carrying substantial traffic; OP-RI, an area open to hunting and adjacent to the Mississippi River channel; and OP-HWY, an area open to hunting and adjacent to Highway 61. These same areas were sampled for determination of lead in arrowhead tubers. Sampling was first attempted with a Ponar grab sampler. This failed because of the interfering vegetation and because some of the sediments were very hard to penetrate. The method finally adopted was to place a 61 cm (24 inch) diameter barrel with both ends removed in the sediment. It was pushed down into the sediment about 25 cm. Then the water was bailed out to permit easier access to the sediment. A scoop was then used to remove the top 15-20 cm. This material was placed in a screening box and rinsed until the fine sediment was removed. The screen was tested to ensure that even #8 shot would be easily retained. Shot encountered were returned to the lab for testing with a magnet to differentiate lead from steel shot. I found this method very effective and recommend it to anyone working in shallow water. Its disadvantages are that the depth of sampling cannot be strictly controlled and the exact depth of the shot encountered cannot be determined. Sixteen samples of 0.292 m (3.14 ft) each were taken in each of the four areas. It was originally intended that half of the samples would be taken from areas within 200 m of places where hunters stand in the areas open to hunting. However, virtually all areas open to hunting and with beds of arrowhead are within 200 m of hunters' blinds. The samples thus reflect shot densities near blinds but also reflect areas used for waterfowl feeding. Samples were taken along a randomly-placed transect within each bed.

Determination of Lead in Arrowhead Tubers

Sixteen tuber samples were taken in each of the four areas. Eight samples were taken in 1985 and another eight were taken in 1986. Tubers were dug from the sediment using a post-hole digger, rinsed of sediment in the marsh water and placed in polyethylene bags immediately. After returning from the field, the samples were frozen until analysis. Samples were taken along a randomly-placed transect within each area. Samples consisted of 2-5 tubers each.

For lead analysis, all glassware was thoroughly cleaned with detergent, rinsed with distilled water, and placed in a 50% nitric acid solution for at least 2 hrs. It was then rinsed twice with distilled water. For sample dissolution, I had intended to use muffle furnace combustion. However, tubers are very lightweight and not enough material could be fit into the crucibles for analysis. I was forced then to use nitric acid digestion, as is commonly done in many heavy metal analyses. In order to speed the digestion and minimize loss of sample, a pressure

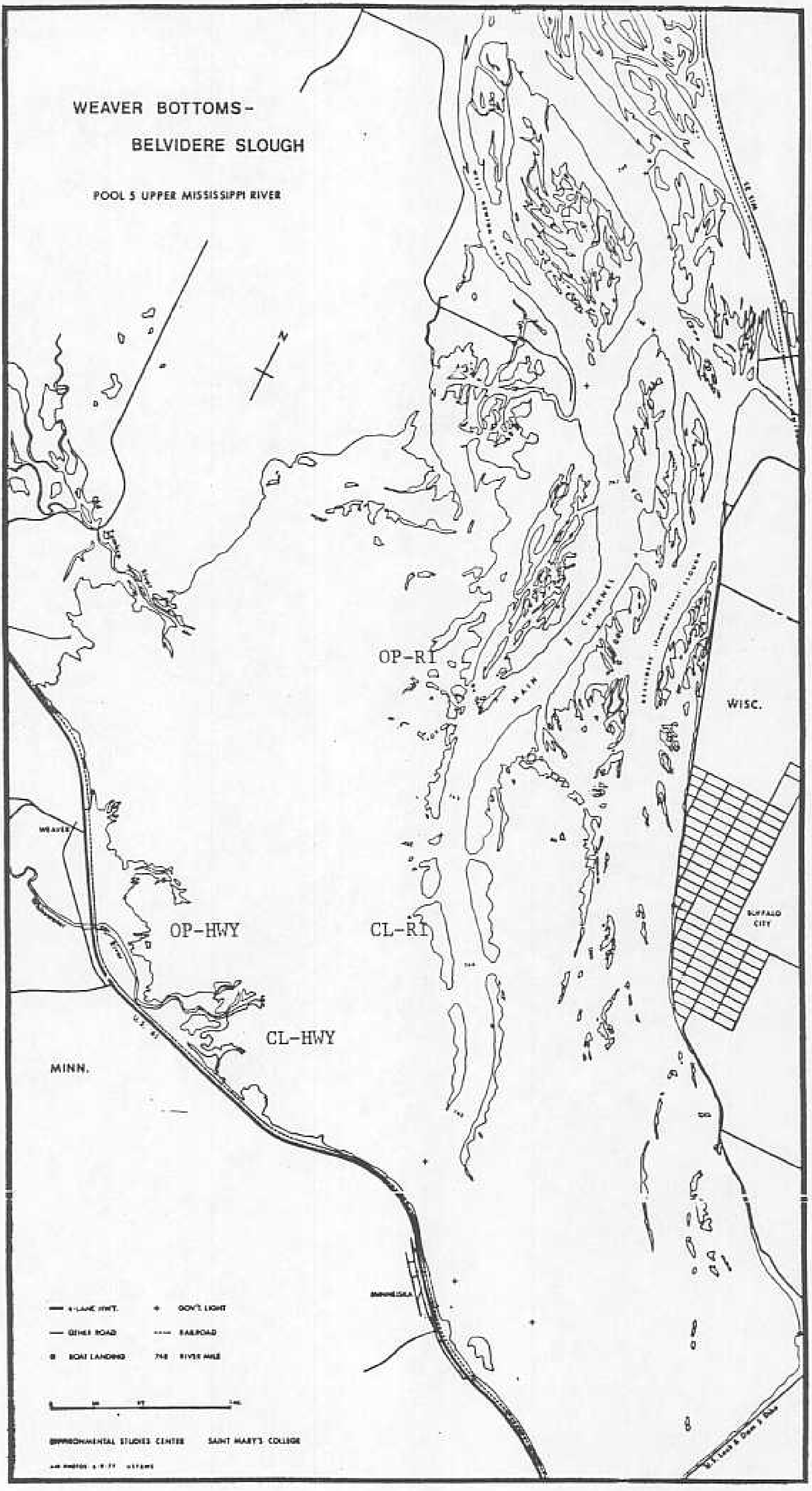

Figure 1. Weaver Bottoms study area. Collection sites are marked CL-RI, CL-HWY, OP-RI, and OP-HWY

cooker was used to heat and pressurize the tuber-acid mixture. Twenty grams of tubers were weighed into an erlenmeyer flask along with 20 ml of a 1:1 nitric acid:distilled water mixture. The flasks were covered and placed in the pressure cooker for 2 hrs at a pressure rating of 10. This liquified most of the sample but not all of it. Repeated pressure cooking and even addition of more acid failed to totally liquify the samples. Silicates are not digested by nitric acid and probably made up most of the sediment in these samples. These solids were removed by filtration through glass wool previously rinsed with nitric acid followed by distilled water. Resultant liquid digestates were then concentrated with gentle heat and made to a constant volume for analysis. Analysis by atomic absorption was attempted but discontinued because the lead concentrations were very near detection limits of the instrument. Instead a plasma emission spectrophotometer was used because it has a lower detection limit of 20 ppb. Both blanks and standards were run between each sample in order to insure linear response of the instrument.

Determination of Swan Tuber Intake

At the outset, it was assumed that almost all of the tubers that disappeared from the area during the fall season were taken by tundra swans. This seemed a valid assumption because the tubers tend to buried deep in the sediment and are reachable only by a species capable of digging in the sediment. Few tubers were encountered at depths less than 25 cm. Most were encountered between 25 and 40 cm depth.

This portion of the study was conducted in the area designated CL-HWY. It is closed to hunting and traditionally swans have congregated there in great numbers during November. The area was sampled for tuber density during August and September, 1985 before any swans had arrived. Swan numbers were recorded throughout October, November, and December. Unfortunately, the sampling after migration could not be completed because of two factors. First, the water froze quickly and deeply immediately after the swans left; by the time that we could get out onto the ice to begin sampling, freezing had extended deep into the sediment layer. This delayed sampling until the spring thaw. Second, by the time that thawing was completed, the river was in flood condition and it was impractical to sample in such deep water. The 1985 data on tuber density and swan numbers therefore were ignored in favor of a second attempt in 1986.

In 1986, pre-migration tuber density was determined by sampling along a randomly-placed transect through the arrowhead bed. A piece of stovepipe 25.4 cm (10 inches) in diameter was driven into the sediment a depth of 61 cm. The sediment containing the tubers was removed with a post-hole digger and strained through poulty wire (2.5 cm mesh). This technique was effective and relatively rapid. It permitted sampling deeply to collect all tubers yet in a defined area. Virtually all tubers were collected; those that escaped through the wire were still collected because they floated to the surface of the water. This technique was repeated along roughly the same transect in January after the migration had ended. In this case an ice auger was used to dig through the ice to the sediment below. The stovepipe was then inserted through the hole in the ice into the sediment. Many fewer tubers were found as might be expected. Those that were found were at depths greater than previously encountered. All of the tubers collected were returned to the lab and weighed to determine the weight of tubers in each sampling unit.

This pre- and post-migration sampling permitted calculation of tuber density. In order to calculate the total tuber population, the area covered by arrowhead plants had to be determined. This was done by planimetry of aerial photos of the area which had been taken by the U.S. Fish & Wildlife Service in August, 1986. I was able to ground-truth the aerial photos by measurement of an identifiable stretch of Highway 61 on the ground.

Finally, the numbers of swans using the area throughout the migration had to be determined. This was done simply by counting every bird with the help of a spotting scope and a tally counter. An elevated frontage road beside the area made counting of individual birds possible. Counts were relatively accurate, I feel, because repeat counts by myself and even duplicate counts by others were always within 5 % of the original count total. These counts were repeated at least three times per week, except when poor weather conditions made accurate counts impossible. In the calculation of total swan-days of use, it was assumed that the number of swans remained constant from one count to the next; i.e., if there were 500 swans on day 1 and 1000 swans on day 4, it was assumed that there were 500 on day 2 and day 3 as well. Thus, the count is perhaps slightly conservative. The total swan-days of area use was then calculated by adding the estimates for each day together from the first day swans appeared until the area froze over and swans could no longer dig for food within the arrowhead bed. The total amount of tubers that disappeared from the area during the migration was then divided by the total swan-days of area use to determine tuber intake per day.

Analysis of Dead/Moribund Swans

In spite of substantial publicity about the project and searches of the vegetation surrounding the area used most by swans, only two specimens were found in condition suitable for analysis. These were both found in 1985 in December. One was found dead, the other was extremely moribund and died on the way to the lab. Both were juvenile females in good flesh (not emaciated). Five other carcasses were found (two in 1985, three in 1986), but they had been scavenged severely and could not be analyzed. The two individuals not scavenged were taken to a veterinarian experienced in x-ray analysis of lead shot in animals. Both front and side-view x-rays were taken of the entire body. No shot, broken bones, or other abnormalities were found. The livers were removed and a 20 gram portion taken for analysis of lead. These tissues were analyzed in the same manner as done for the tubers described above. Livers were analyzed in preference to blood samples because of the greater concentrations of lead found in livers as opposed to blood and because it is a reliable indicator of lead toxicosis.

RESULTS AND DISCUSSION

Lead Shot Distribution

Numbers of lead shot encountered in the areas open to hunting (OP-RI and OP-HWY) were surprisingly high, especially since the samples were collected in the spring of 1987 several months after the hunting season had closed (Table 1). Those arguing for continued use of lead shot in this area often claim that the shot sink quickly and become unavailable to waterfowl. This is certainly untrue for swans and may be untrue for most waterfowl. Many of these shot were found within 8 cm of the surface. The sediment in which these shot were found was variable in character, but I would describe it subjectively as relatively loose. I found myself sinking deeply into it while doing the sampling. The concentration of 27.6 pellets/square meter found at the OP - HWY site is among the highest reported in the scientific literature (Bellrose, 1959; Mudge, 1984). This site is used intensively by duck and goose hunters throughout the entire waterfowl season, whereas the OP - RI site is used intensively early in the season, but receives less usage later and is little used by goose hunters.

As expected, the closed areas yielded no shot. This is not to suggest that no shot exist in the area, but rather that the sampling regime was not sensitive enough to detect the few that are there. Also, no fishing sinkers or other lead objects were found in any of the samples.

Of interest to me is the high incidence of lead shot relative to the number of steel shot found. This may be deceiving because steel shot certainly do rust and would be expected to disappear over time. The steel shot that were encountered were in relatively good condition with little corrosion. A major question to be answered is the age of those lead shot encountered. If they have been deposited recently, then the data indicate a major enforcement problem. Further studies are needed to determine sinking rates of shot in these sediments and to determine degree of compliance with the ban on use of lead shot in waterfowl hunting. Whether young or old, these shot exist in the sediments in high concentrations and may be causing a serious lead poisoning problem for species utilizing these arrowhead beds.

Table 1. Shot in upper 15-20 cm of Weaver Bottoms sediments.

Area	N	Mean shot per square meter	
		Lead	Steel*
CL - RI	16	0.0	0.0
CL - HWY	16	0.0	0.0
OP - RI	16	13.1	10.3
OP - HWY	16	27.6	10.5

* may be underrepresented relative to lead because of rapid corrosion

Residues of Lead in Arrowhead Tubers

Lead concentrations in arrowhead tubers from all sites were low compared with the findings of Anderson (1978) for arrowhead roots (Table 2). These four sites were chosen to permit comparisons of the influence of proximity to Highway 61, the Mississippi River flow, and hunting on lead concentrations in the tubers. No clear trends were evident from these analyses. Means for 1985 were not significantly different from 1986 and there was no net trend among the four sites over the two years. The highest level (0.46 ppm wet-weight) was from the OP - RI site, suggesting that lead shot might have an influence on tuber concentrations. Yet, the other site open to hunting, OP - HWY, had the lowest mean level of any site even though it had more shot in the sediment (Table 1). Likewise, comparison of river and highway sites showed no clear trend. The analysis of variance did show a significant difference among the means ($F = 4.04$, $p < 0.05$). The Newman-Keuls Multiple Range Test was applied and showed significant differences between the mean for OP - RI compared with both OP - HWY and CL - RI. The mean for OP - RI was highly influenced by one extraordinarily high value. Removing this datum from the analysis as a "flyer" removes the finding of significant differences. Without a biological or chemical explanation for this particular site having high values, I am inclined to ignore the results of the statistical analysis. In any case, none of the data show extraordinarily high levels of contamination in the tubers. Indeed, they are much lower than those found by Anderson (1978) in arrowhead roots. There are few data in the literature comparing lead or other heavy metals in tubers vs. roots. Lee *et al.* (1976) did such a comparison in a laboratory study of heavy metal uptake by marsh plants (not including arrowhead). They found that *Cyperus esculentus* concentrated far more lead in its roots than in its tubers. In terms of heavy metals, it appears then that tubers may be an excellent food source for swans in addition to their significant nutitive value.

Table 2. Lead concentration in arrowhead tubers from Weaver Bottoms.

Area	N	Mean ppm wet-weight	Mean ppm dry-weight
CL - RI	16	0.30	1.61
CL - HWY	16	0.36	1.93
OP - RI	16	0.46	2.49
OP - HWY	16	0.27	1.44
Combined	64	0.35	1.87

Swan Tuber Intake

Tuber density in area CL - HWY decreased significantly over the migration period from 1706 g/square meter before migration to 968 g/square meter after migration, indicating a net loss of 738 g/square meter (Table 3). Measurements of the arrowhead bed taken from aerial photos indicated a total area of 250,723 square meters. Thus, the total disappearance of tubers was 185,034 kg during the migration. Swan-days of area use totalled 23,537 (Table 4). Therefore, for every day of swan use of the area, 7860 g of tubers disappeared from the area. This is an incredibly high figure, especially since most tundra swans weigh roughly this amount. It is also much higher than the 3670 g loss found by Limpert (1974). Limpert utilized a slightly different sampling method than that of this study and may not have found all tubers in the sediment. Although these tubers are substantially water (81.5 %), it is unlikely that swans could actually eat this much. Limpert assumed that 25 % of the tubers dug were wasted based on visual observations. I believe that this "wastage" is actually greater because of behaviors that I observed. Very often swans are accompanied by waterfowl of various species, but especially mallards (Anas platyrhynchos) and canvas-backs. It is common to see these species eating parts of tubers or even whole tubers that have floated to the surface. But another behavior exists. When swans are using their feet to dig holes in the sediment to loosen tubers, these waterfowl dive alongside. I believe they are grabbing tubers loosened before the swans have a chance to get to them. Also, the holes leave some tubers that were formerly deep in the sediment now just below its surface. These tubers may then be dug by other species, including canvasbacks and even muskrats (Ondatra zibethica). Thus, it is possible that actual intake by the swans is much less than 7860 g/day. Unfortunately, it would be difficult if not impossible to calculate the actual intake given this kind of loss to other competitors. Any method which might exclude the competitors would also exclude the swans.

The major purpose of making this estimate of intake was to permit calculation of intake of lead from the tuber source. Let us assume that swans actually do eat 7860 g/day and that all of the lead from the tubers is absorbed into the bloodstream. The tubers contain an average of 0.35 mg/kg lead. Thus each swan takes in 2.75 mg lead each day. One #6 lead shot weighs 127 mg. For a swan to ingest the same amount of lead from tubers would then require 46 days of feeding. It is rare for swans to remain in the Weaver Bottoms for that long. The first birds arrive in mid-October and most are gone by early December. Most individual birds stay in the area for only one to three weeks before moving on. An additional factor of importance is the absorption of lead into the bloodstream. Stone et al. (1981) found that biologically incorporated lead in oyster meat was only 69-75% as available as lead acetate to Japanese quail (*Coturnix coturnix*). I conclude that it is unlikely that lead in tubers from the Weaver Bottoms contributes significantly to lead poisoning of tundra swans frequenting the area.

Table 3. Parameters of tuber utilization in the Weaver Bottoms.

Parameter	Quantity
Tuber density pre-migration	1706 g/square meter
Tuber density post-migration	968 g/square meter
Tuber loss during migration	738 g/square meter
Size of arrowhead bed utilized	250,723 square meters
Total amount of tubers lost	185,034 kg
Total swan-days of use of area	23,537 swan-days
Loss of tubers/swan-day of use	7,860 g/day

Table 4. Counts of tundra swans present at Weaver Bottoms, 1986.

Date	Number
October 22	3
23	7
24	7
25	7
26	7
27	22
28	29
29	42
30	52
31	[52]*
November 1	441
2	1161
3	891
4	1236
5	[1236]
6	1360
7	[1360]
8	[1360]
9	3566
10	[3566]
11	[3566]
12	[3566]
13	0 (area froze overnite; swans could not feed in bed henceforth)
Total	23,537

*brackets indicate estimated number based on previous count

<u>Analysis of dead/moribund swans</u>

Even though searches were conducted, only two dead/moribund swans were found in condition suitable for analysis. These were x-rayed and the livers analyzed for lead content. The x-rays were completely negative without shot or indication of broken bones. The birds also were in good flesh without signs of emaciation. The liver analysis showed levels of 0.20 and 0.28 ppm wet weight. This is much below the level needed to indicate lead toxicosis. These juvenile birds died of causes other than lead poisoning.

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APPENDIX-Raw Data

Lead shot sediment sampling

Each datum is the number of shot found in the top 15-20 cm of sediment from a circle 61 cm (2 feet) in diameter from arrowhead beds within 200 m of sites used for waterfowl hunting in the fall. Sixteen samples from each of the areas CL - RI and CL - HWY yielded no shot at all.

Sample Number	# Lead Shot	#Steel Shot
Site OP - RI:		
1	0	1
2	3	2
3	0	1
4	5	5
5	1	1
6	0	0
7	10	7
8	3	5
9	6	4
10	3	1
11	8	6
12	7	6
13	4	4
14	6	3
15	1	5
16	4	0
Site OP - HWY:		
1	2	0
2	2	2
3	1	3
4	4	1
5	0	0
6	14	7
7	22	7
8	3	7
9	29	4
10	3	0
11	17	3
12	20	4
13	6	3
14	0	1
15	3	5
16	3	2

Lead Concentrations in Arrowhead Tubers

Year-Site-#	Ppm wet-weight	Year-Site-#	Ppm wet-weight
85-CL-RI-01	0.32	86-CL-RI-01	0.41
02	0.35	02	0.41
03	0.32	03	0.22
04	0.30	04	0.39
05	0.27	05	0.27
06	0.25	06	0.28
07	0.29	07	0.22
08	0.25	08	0.22
85-CL-HY-01	0.26	86-CL-HY-01	0.39
02	0.30	02	0.32
03	0.34	03	0.32
04	0.30	04	0.25
05	0.55	05	0.28
06	0.58	06	0.28
07	0.21	07	0.37
08	0.46	08	0.50
85-OP-RI-01	0.28	86-OP-RI-01	0.43
02	0.22	02	0.33
03	0.27	03	0.44
04	0.27	04	0.68
05	1.54	05	0.44
06	0.50	06	0.38
07	0.34	07	0.41
08	0.47	08	0.39
85-OP-HY-01	0.20	86-OP-HY-01	0.23
02	0.25	02	0.41
03	0.36	03	0.30
04	0.27	04	0.28
05	0.20	05	0.21
06	0.22	06	0.21
07	0.25	07	0.21
08	0.32	08	0.35

Tuber Density Sampling

Data below are the weights in grams of tubers taken before and after the swan migration. They are listed in order of sampling along randomly-placed transects.

Sample Number	Pre-migration	Post-migration
01	57.0	48.7
02	138.6	0.0
03	42.6	10.6
04	15.1	27.9
05	83.5	0.0
06	50.0	40.9
07	68.3	33.3
08	62.5	0.0
09	166.3	38.3
10	63.9	156.4
11	103.6	55.6
12	203.3	0.0
13	223.2	7.9
14	25.8	39.5
15	126.8	61.2
16	66.8	145.4
17	90.6	164.0
18	67.5	35.4
19	27.1	141.9
20	107.2	0.0
21	118.2	
22	111.0	
23	21.0	
24	105.5	
25	121.3	
26	35.9	
27	88.4	
28	70.0	
29	112.4	