Biochemical parameters and regurgitation of implanted lead sinkers in largemouth bass, *Micropterus salmoides*

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Abstract

The original objective of this study was to investigate lead toxicity in largemouth bass (*Micropterus salmoides*) focusing on the possible implications of ingested lead sinkers (split shots). We hypothesized that lead in the gastrointestinal tract of a largemouth bass would result in detectable blood lead levels. This study was designed to determine the level and speed at which lead would be absorbed. Commercial lead sinkers were gastrically implanted with the aid of an endoscope and blunt forceps. Within 1 hour, one of the fish regurgitated the lead sinker and the sinker was immediately re-implanted. Within 3 days of the initial procedure, all fish had regurgitated the lead sinkers. The lead toxicity component of this study could not be completed given that all fish regurgitated the lead shots. Pre-implantation blood biochemical parameters for largemouth bass are presented.

Keywords: Ballast, Blood chemistry, Lead shots, Lead weights, Plasma, Split shots

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Introduction

This study began as a lead toxicity investigation in largemouth bass (*Micropterus salmoides*) (LMB) aimed at studying the implications of fish ingesting lead sinkers (split shots). The idea was generated from a clinical case involving a captive moribund electric eel (*Electrophorus electricus*) that had consumed lead aquatic plant sinkers. The lead levels in the eel were measured but there were no base line data to compare the post-ingestion lead levels (Minter *et al*. 2012). Furthermore, there were no publications on lead toxicity from lead sinkers consumption in fishes. It has been documented that waterfowl and other birds consuming lead fishing sinkers experience detectable blood lead levels and potential lead toxicity (Pokras *et al*., 1992; De Francisco *et al*., 2003; Franson *et al*., 2003; Wood and Newth, 2024). Lead toxicosis due to ingestion of lead sinkers has been reported in other wildlife including a harbor seal (Borkowski, 1997), a snapping turtle (Zabka *et al*. 2006), various European wild mammals (Chiverton et al., 2022) and a human (Gupta et al., 2021). Data on the consumption of lead sinkers and its effect in game fish were mostly anecdotal.

The LMB was chosen because it is a popular recreational game fish with a perceived risk from ingesting lead sinkers (Allen *et al*., 2008). Although the fish have a large and heavy premaxilla that enables grasping of large prey at any angle, they have the ability to use suction to inhale small foods and/or small objects (Werner, 1977). The largemouth bass' foraging behavior, ability to consume variable prey, and being a popular game fish, made it a likely candidate to swallow lead sinkers.

The initial objectives of the study were to establish blood biochemistry baseline values, determine if fish with lead in their stomach absorb lead into their bloodstream, and the time and level of lead absorption.

Materials and methods

All procedures performed on the largemouth bass were approved by North Carolina State University Institutional Animal Care and Use Committee (IACUC).

Largemouth bass purchased from a commercial vendor were placed in individual 57 L aquariums on a re-circulating system with weekly 15% water changes. The water temperature was maintained at approximately 15.5^0C with a salinity of 1 ppt. The fish were fed an omnivore gel food three times a week and fasted 1 day before and the day of the experimental procedures. An initial health assessment was performed while the fish were anesthetized with 200 ppm buffered (pH approx. 7.0) tricaine methanesulfonate (MS-222, Sigma Chemical Co.). A gill, fin, and skin biopsies were collected from three random fish. Each sample was viewed as a wet mount with a compound microscope.

Approximately 1 month after arrival, the fish were carefully netted, anesthetized, and weighed. Blood was collected from the caudal vein using a 1 ml sodium heparinized syringe and 25 g

needle to establish an initial healthy plasma biochemical base line. After the blood collection, one fish died. All fish were deemed too underweight for proceeding with the experiment that would involve multiple blood samples. A new fish was purchased from a commercial vendor to replace the deceased fish. To ensure that a sufficient volume of blood for analysis could be safely collected, all the animals were given additional acclimation time to reach an appropriate weight.

On the day of the experiment, approximately 5 months after the initial blood collection, the fish were carefully netted and anesthetized similar to above. Between 500 and 700 uL of blood was drawn from eight experimental fish and two control fish. After each fish was weighed, a lubricated lead sinker (Water Gremlin Removable Split Shot, White Bear Lake, MN, USA) of 10 g for every kg of fish was implanted in the stomach using blunt forceps. The identical procedure was performed on control fish #3 and #11 but no lead sinker was placed in the stomach. A rigid endoscope, Hopkins Telescope (2.7 mm X 18 cm, 30°), was used to verify the gastric implantation of the split shots of three of the experimental fish, #8, 9, and 10 (Table 1).

Table 1: The weight in grams of each largemouth bass (*Micropterus salmoides*) and the weight in grams of lead sinkers (lead dosing) and the Water Gremlin size lead sinkers placed into each largemouth bass. The dosage of lead implanted given by the percent of body weight is also given.

Fish	Weight of fish (g)	Water Gremlin Lead weight (g)-lead dosing	Water Gremlin size	% body weight
1	133	1.04	#3/0	0.78
2	145	1.04	#3/0	0.72
3	116	Control	Control	Control
4	191	1.83	#7	0.96
5	122	Dead	Dead	Dead
6	219	1.83	#7	0.84
7	159	1.83	#7	1.15
8	145	1.83	#7	1.26
9	241	2.54	#5	1.05
10	261	2.54	#5	0.97
11	86	Control	Control	Control

The fish were then returned to their individual tanks for recovery. At this time fish #7 expelled the lead shot in its tank, and the lead shot was immediately replaced and verified with the endoscope. The fish were monitored daily for any possible physical distress due to the lead implantation. The heparinized blood samples were placed in 1.5 ml with attached cap Greiner Bio-One natural graduated reaction tubes, initially stored at -80 ^{0}C for 10 days, and then centrifuged to harvest plasma. The plasma was isolated and analyzed using

a VetScan analyzer (Abaxis VetScan (Classic) Chemistry Analyzer, Avian Reptilian Profile Plus rotor, software version 5.118.2) designed to automatically determine albumin, bile acid, aspartate aminotransferase, creatine kinase, total protein, globulin, glucose, uric acid, calcium, phosphorus, potassium, sodium, hemolysis, lipase, hemolysis, lipemia, and icterus. The plasma from the deceased fish was stored at -80 ^{0}C for approximately 5 months and included in the results.

Statistical analyses were performed and basic statistics obtained (MINITAB version 14 software). Data for the biochemical parameters were not normally distributed and were reported in median, quartiles 1 (25%), quartiles 3 (75%), and minimum to maximum (Table II). Categorical data reported by the VetScan Analyzer (Abaxis VetScan (Classic) Chemistry Analyzer, Avian Reptilian Profile Plus rotor, software version 5.118.2) for calcium, potassium, bile acid, hemolysis, and lipemia were presented as percent of the 11 fish in the category (Table 3).

Results

The initial health assessment of the fish indicated they were too thin and small for adequate blood collection. A large number of the fish were hyperemic and gill biopsies revealed mild monogenean infection and one ciliated protozoan. This level of parasitism was not considered clinically significant.

The amount of lead placed into each subject was proportional to the animal's weight (Table 1). The two controls, fish #3 and 11, and the fish that died prior to experiment (#5), had no lead placed but blood was collected. Within one hour, a fish regurgitated the split shot and the last fish regurgitated its lead shot three days after the procedure.

Although the lead sinkers were regurgitated, data from the blood were analyzed to establish baseline blood biochemical parameters for LMB. The plasma values results included the minimum to maximum (Tables 2 and 3).

Table 2: Largemouth (*Micropterus salmoides*) bass plasma chemistry values (n = 11).

Parameter	Median	25th% Percentile	75th% Percentile	Minimum-Maximum
Albumin (g/dL)	2.0	1.9	2.3	1.5-2.4
Aspartate aminotransferase (μ /L)	57.0	22.0	82.0	21-115
Creatine kinase, (μ/L)	1401.0	618.0	2755.0	300-6493
Globulins (g/dL)	2.3	2.1	2.5	2.0-2.8
Glucose (mg/dL)	69.0	63.0	81.0	55-106
Icterus	0.0	0.0	0.0	0.0
Phosphorus (mg/dL)	10.8	9.4	13.1	7.5-14.2
Sodium (mmol/L)	154.0	151.0	157.0	147-161
Total protein (g/dL)	4.4	3.9	4.8	3.5-5.0

Table 3: Largemouth bass (*Micropterus salmoides*) calcium, potassium, bile acid, hemolysis, and lipemia reported percent per category.

Range of calcium (mg/dL)	≥16	11-13.6	14-15.9
Percent of subjects	27.3%	45.4%	27.3%
Range of bile acid (µmol/L)	<35	91	-
Percent of subjects	90.90%	9.10%	-
Amount of hemolysis	0	+1	+2
Percent of subjects	54.5%	27.3%	18.2%
Amount of lipemia	0	+1	+3
Percent of subjects	63.6%	27.3%	9.1%
Amount of potassium (mmol/L)	<1.5	1.7	-
Percent of subjects			

The values for calcium, potassium, bile acid, hemolysis, and lipemia are presented in categories in Table 3. The second row for each parameter represents the percent of fish in that category. Of the 11 largemouth bass, 45.5% had a calcium value between 11-13.6 mg/dL, 81.8% had a potassium level of <1.5 mmol/L, 90.9 % had a bile acid level of <35 µmol/L, 54.5% had no hemolysis detected, and 63.6% had no detectable lipemia.

Discussion

The lead toxicity component of this study could not be completed given that all fish regurgitated the lead shots. We speculate that regurgitation was due to the rapid digestion rate of LMB and the inability to digest the lead shots. In one study, LMB digested most of their meal within 2 hours and digested a 2.81% of body weight meal in 17.5 hours. The time of gastric emptying varied between 16 and 19 hours (Hunt, 1960). In our study, each fish was implanted with lead that ranged from 0.84% to 1.26% body weight (Table 1). It was thought that within the first 2 hours, gastric enzymes would have started acting on the lead. Although other factors may have contributed to gastric evacuation (temperature of water, starvation time, and size of meal), we believe the presence of a metallic gastric foreign body in the form of a lead shot precipitated expulsion.

The literature on plasma chemistry values in LMB is scant. The values reported in previous studies were limited to electrolytes and glucose (Dean and Goodnight, 1964; Hunn and Robinson, 1966; Clark *et al*., 1979; Carmichael, 1983; Stoskopf, 1993). Glucose concentrations and electrolytes can be affected by a number of factors, including fasting, temperature, and water quality parameters. In comparison, the sodium levels in our study were consistent with the expected values for LMB serum (159-166 mM/L) (Hazen *et al* 1978; Stoskopf, 1993).

Other factors that could have influenced changes in blood parameters

include age, sex, fish handling, MS-222 sedation, sample handling, and hemolysis. In has been noted that MS-222 can increase PCV, blood glucose, and potassium concentrations in teleosts (Hattingh, 1977; Soivio *et al*., 1977; Stoskopf, 1993; Sladky *et al*., 2001; Popovic *et al*., 2012). The whole blood samples were frozen before processing and the effect of freezing on LMB plasma have not been established. The table top analyzer used in this study reports that physical interference (hemolysis, icterus, and lipemia) may cause changes in the reported values.

The blood parameters distribution were not normal, therefore we reported findings in percentiles (median, 25th percentile, and 75th percentile) and range. A more comprehensive database is needed to characterize potential indicators for environmental monitoring. The most interesting finding from our study was that all LMB regurgitated the lead shots. If this is a common occurrence, lead intoxication in this fish may not be common.

References

Allen, M.S., Walters, C.J. and Myers, R., 2008. Temporal trends in largemouth bass, mortality, with fishery implications. *North American Journal of Fisheries Management*, 28, 2, 418-427. https://doi.org/10.1577/M06-264.1

Borkowski, R., 1997. Lead poisoning and intestinal perforations in a snapping turtle (*Chelydra serpentina*) due to fishing gear ingestion. *Journal of Zoo and Wildlife Medicine*, 28(**1**), 109-113. https://www.jstor.org/stable/20079497

Carmichael, G.J., 1983. Plasma characteristics of largemouth bass (*Micropoetrus salmoides*) exposed to aquaculture related stressors [dissertation]. Memphis, TN: Memphis State University.

Chiverton, L., Cromie, R. and Kock, R., 2022. European mammal exposure to lead from ammunition and fishing weight sources. *Heliyon*, 8(**8**). DOI:10.1016/j.heliyon.2022.e10014

Clark, S., Whitmore JR, D.H. and McMahon, R.F., 1979. Considerations of blood parameters of largemouth bass, *Micropterus salmoides*. *Journal of Fish Biology*, 14, 147-158. https://doi.org/10.1111/j.1095-8649.1979.tb03504.xDigital Object Identifier (DOI)

Dean, J.M. and Goodnight, C.J., 1964. A comparative study of carbohydrate metabolism in fish as affected by temperature and exercise. *Physiological zoology*, 37(**3**), 280-299.

De Francisco, N., Troya, J.D. and Aguera, E.I., 2003. Lead and lead toxicity in domestic and free living birds. *Avian Pathology* 32(**1**), 3-13. https://doi.org/10.1080/0307945021000070660

Franson, J.C., Hansen, S.P., Creekmore, T.E., Brand, C.J., Evers, D.C., Duerr, A.E. and DeStefano, S., 2003. Lead Fishing Weights and Other Fishing Tackle in Selected Waterbirds. *Waterbirds*, 26(**3**), 345–352. doi.org/10.1675/1524-4695(2003)026[0345:LFWAOF]2.0.CO;2

Gupta, S.R., Mezoff, E. and Dienhart, M., 2021. Lead toxicity from a swallowed fishing sinker: a case report. *JPGN reports*, 2(**3**), p.e084.

https://doi.org/10.1097/PG9.0000000000000084

Hattingh, J., 1977. The effect of tricaine methanesulphonate (MS-222) on the microhaematocrit of fish blood. *Journal Fish Biology*, 10, 453–455. https://doi.org/10.1111/j.1095-8649.1977.tb04077.x

Hazen, T.C., Esch, G.W., Glassman, A.B. and Gibbons, J.W., 1978. Relationship of season, thermal loading and Red-Sore disease with various haematological parameters in *Micropterus salmoides*. *Journal of Fish Biology*, 12, 491-498. https://doi.org/10.1111/j.1095-8649.1978.tb04192.x

Hunn, J. and Robinson, P.F., 1966. Some blood chemistry values for five Chesapeake Bay area fishes. *Chesapeake Science*, 7, 173-175. https://www.jstor.org/stable/1351166

Hunt, B., 1960. Digestion rate and food consumption of Florida gar, warmouth, and largemouth bass. *Transactions of the American Fisheries Society*, 89, 206-211. https://doi.org/10.1577/1548-8659(1960)89[206:DRAFCO]2.0.CO;2

Minter, L., Stoskopf, M.K., Serrano, M.E., Burrus, O. and Lewbart, G.A., 2012. Suspected lead toxicosis in an electric eel, *Electrophorus electricus* (Linnaeus). *Journal of Fish Diseases*, 35, 8. 10.1111/j.1365-2761.2012.01386.x

Pokras, M.A. and Chafel, R., 1992. Lead Toxicosis from Ingested Fishing Sinkers in Adult Common Loons (*Gavia immer*) in New England. *American Association of Zoo Veterinarians*, 23(**1**), 92–97. https://www.jstor.org/stable/20460274

Popovic, N., Strunjak-Perovic, I., Coz-Rakovac, R., Barisic, J., Jadan, M., Persin Berakovic, A. and Sauerborn Klobucar, R., 2012. Tricaine methane-sulfonate (MS-222) application in fish anaesthesia. *Journal Applied Ichthyology*, 28, 553–564. https://doi.org/10.1111/j.1439-0426.2012.01950.x

Sladky, K.K., Swanson, C.R., Stoskopf, M.K., Loomis, M.R. and Lewbart, G.A., 2001. Comparative efficacy of tricaine methanesulfonate and clove oil for use as anesthetics in red pacu (*Piaractus brachypomus*). *American Journal Veterinary Research*, 62(**3**), 337–342. https://doi.org/10.2460/ajvr.2001.62.337

Soivio, A., Nyholm, K. and Huhti, M., 1977. Effects of anaesthesia with MS 222, neutralized MS 222 and benzocaine on the blood constituents of rainbow trout, *Salmo gairdneri*. *Journal Fish Biology*, 10, 91–100. https://doi.org/10.1111/j.1095-8649.1977.tb04045.x

Stoskopf, M.K., 1993. *Fish Medicine*. Philadelphia, PA: W.B. Saunders Co.

Werner, E.E., 1977. Species packing and niche complementarity in three sunfishes. *American Naturalist*, 111, 553–578. https://doi.org/10.1086/283184

Wood, K.A. and Newth, J.L., 2024. Swans and lead fishing weights: a systematic review of deposition, impacts and regulations in Europe. *Wildfowl*, pp. 27-56.

Zabka, Tanja S., Haulena, M., Puschner, B., Gulland, F.M.D. and Conrad, P.A., 2006. Acute lead toxicosis in a harbor seal (*Phoca vitulina richardsi*) consequent to ingestion of a lead fishing sinker. *Journal of Wildlife Diseases*, 42, 3, 651-657. DOI:10.7589/0090-3558-42.3.651