Mercury deposition in nine designated feather regions of eight captive raised, dose response Common Loons (*Gavia immer*)

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ABSTRACT

The presence of methylmercury in natural environments is on the rise due to an increase in atmospheric mercury deposition rates. Piscivorous wildlife is at high risk due to the toxic effects of methylmercury bioaccumulation. Plumage development in birds has proven to be an elimination pathway for mercury, and may actually reduce total body mercury burdens. In this study, the elimination of methylmercury into nine designated feather regions was examined in eight captive raised, dose response Common Loons. These birds were divided into four dose treatment groups, were euthanized after 105 days, and their feathers were removed and analyzed for mercury content. Total mercury burdens were found to increase with an increased mercury dose, although variation in mercury content seen between feather regions could not be correlated with literature based molt sequence.

INTRODUCTION

Atmospheric mercury deposition poses ecological risks that are of primary concern for federal and state regulatory agencies as well as industry (1). The rate of mercury deposition has increased in the north central United States two to four times since the pre-industrial era (1). Naturally occurring inorganic mercury is transformed into methylmercury by anaerobic microorganisms inhabiting water sediments (2). This methylated form of the pollutant is highly toxic and capable of bioaccumulation, which places piscivorous wildlife at high risk (1-5).

A bird's plumage has proven to be a major elimination pathway for mercury (2-4, 6-11). During periods of molting, mercury concentrations in target tissues may actually decrease, lowering the total body burden, as a result of the mobilization of mercury into growing feathers (2, 6, 10).

The Upper Midwest Environmental Sciences Center (UMESC), La Crosse, Wisconsin, and the Wisconsin DNR are currently conducting a study to assess the ecological risk of mercury exposure in Common Loons (*Gavia immer*). Chicks from twenty-four Common Loon eggs, collected from a four-county region of northern Wisconsin, were reared in captivity at UMESC. The chicks were equally divided into six experimental blocks and were subjected to daily doses of methyl mercury. The birds were assigned to one of four dose treatment groups: control, 0.1μ g CH₃HgCl/g wet fish (treatment 1), $0.5^{-}\mu$ g CH₃HgCl/g wet fish (treatment 2), and 1.5μ g CH₃HgCl/g wet fish (treatment 3). Each of the experimental blocks contained a

representative of each dose group. The growth, food intake, and molting patterns of these dose response loons were closely monitored throughout the study. A variety of behavioral observations were also made. All dose-response loons were euthanized at an age of 105 days to allow for mercury analysis sampling.

Through cooperative efforts with UMESC, this project has been conducted to analyze the mercury content in the feathers of dose response Common Loons. The feathers of birds from two experimental blocks were analyzed to compare feather mercury levels among selected body regions, and among dose response treatment groups. Total feather mercury burden was also estimated for each bird.

METHODS

The carcasses of eight Common Loons were stored at < -20°C and required partial thawing before plucking could begin. Each bird was kept at room temperature for approximately 4-6 hours to thaw. The feathers of these birds were individually plucked and sorted into the following nine categories: head/neck, ventral body, apertium, tail, legs, primaries (right wing only), secondaries, remaining wing feathers, and dorsal body. Seven by nine inch whirl pak bags, labeled with the study number, sample number, initials and date, were weighed on a calibrated balance with 0.001g accuracy. Empty bag weights were recorded on a specialized data form.

A metal tray was covered with aluminum foil to avoid contamination. Each carcass was placed on the tray and the designated body regions were plucked one by one. Rubber gloves and a surgical mask were worn as safety precautions. Pliers were used to remove feathers in dense regions, such as the ventral body, and a feather collection vacuum aided in the collection of feathers. This device is simply a funnel (inside diameter = 20.5 cm) attached to the hose of a Metro Datavac Pro-3 or Royal Dirt Devil Plus vacuum. A screen and lose filter (plastic mesh size = < 2mm) were placed in the funnel to trap the feathers for easier collection. The feathers were placed near the funnel as they were plucked from the bird.

Feathers were transferred to the pre-weighed, pre-labeled whirl pak bags. Blood soaked feathers were rinsed with deionized water and were allowed to dry on paper towels before being transferred to the whirl pak bags. The plucked feathers were stored at \leq -20°C to avoid mercury loss until they were washed and weighed.

Care was taken to prevent cross-contamination among feather body regions within birds and among birds. Loose down and feathers were removed from the feather plucking area and feather collection vacuum screen and filter once a feather region had been completed. The pliers were decontaminated using a hexane/acetone rinse and the foil on the tray was changed between birds.

The whirl pak bags containing each feather group were weighed to the nearest 0.001g after thawing, and the weights were recorded on the specialized data form. The feathers were cleaned to remove external atmospheric mercury contamination and blood residue that may have altered the analysis results. This was done as follows. Deionized water was added to the whirl pak bags so all the feathers were submersed. This excess of water was used to slosh the feathers around the bag. The outside of the bag was gently squeezed for approximately 2-3 minutes, the water was drained, and the washing process was repeated two more times. This same procedure was then repeated twice with acetone to remove as much moisture as possible. A fine mesh net was used to capture down and feathers while draining the water and acetone. The net was washed with deionized water between feather groups. The feathers were stored at \leq -20°C until they were submitted for analysis.

The feather samples were sent to EnChem, Madison, Wisconsin, for the analysis of total mercury. Here, the feather samples were allowed to air-dry at room temperature for 24 hours before they were homogenized. A subsample of feathers from the homogenate was analyzed for total mercury by cold vapor atomic absorption for each body region (refer to procedures outlined in Evers et al. 1998 and Braune and Gaskin 1987), and another feather subsample was analyzed for moisture content.

RESULTS

Feather weights varied significantly among body regions. The ventral body feathers weighed the most with a mean weight of 46.63g for the eight loons. The remaining wing region had a mean feather weight of 28.66g and was the second heaviest feather region. The apertium and legs had mean weights of 3.37g and 2.08g respectively, and were the lightest feather regions.

A difference in mercury feather concentration was apparent among the nine body regions for each of the four treatment groups. The highest mean mercury concentration for the two control birds was found in the head/neck region (Table 1). The highest mercury concentration was found in the apertium for treatments 1, 2, and 3. The remaining wing feathers had the lowest mercury concentration in the control, treatment 1, and treatment 2 birds, while mercury was least concentrated in the secondaries for the treatment 3 birds (Table 1).

Total mercury levels were the highest in the ventral body feathers for all treatment groups. The mean amount of mercury in the ventral body was 45.45μ g for the control group, and 275.53μ g, 2079.97μ g, and 6052.06μ g for treatments 1, 2, and 3 respectively. The leg feathers accounted for the least amount of total mercury for the control, treatment 1 and treatment 2 birds. The mean amount of mercury present in the leg feathers was 1.73μ g for the control group, 14.03 µg for treatment 1, and 90.11 µg for treatment 2. The mean total mercury deposition for treatment 3 was the lowest in the secondaries, where 262.34μ g of mercury was found.

Region	Control	Treatment 1	Treatment 2	Treatment 3
Apertium	1.30	9.20	55.50	200.00
Head/Neck	1.55	8.35	44.50	165.00
Tail	1.04	7.15	41.50	160.00
Legs	1.25	7.40	42.50	140.00
Dorsal Body	0.67	5.90	37.00	120.00
Ventral Body	1.07	6.05	44.00	117.00
Right Primaries	0.77	6.15	35.00	110.00
Remaining Wing	0.28	3.10	23.00	93.50
Secondaries	0.57	4.00	26.50	53.00
All Feathers	0.94	6.37	38.83	128.72

Table 1. Mean mercury concentration (µ	µg/g) for each body region	among treatment groups
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The total feather mercury burden increased with an increased mercury dose. The feathers of the control birds contained the least amount of mercury with totals of 60.88 μ g and 95.99 μ g. The two treatment 1 birds had 582.86 μ g and 609.22 μ g of mercury in their feathers, while the feathers of the treatment 2 birds contained 3495.56 μ g and 4620.23 μ g of mercury. The feathers of the treatment 3 birds accounted for the most mercury, with amounts of 13186.99 μ g and 15988.25 μ g.

DISCUSSION

Molting sequence may account for the variation of mercury concentrations seen between feather regions. The concentration of mercury in a bird's feathers reflects blood mercury levels at the time of feather development (3, 7, 10, 11). As mercury is excreted into growing feathers, the body mercury pool is reduced, which therefore decreases the amount of mercury isolated in the feathers as plumage development continues (3, 7, 11).

The low mercury concentrations found in the remaining wing feathers and secondaries suggest that molting of these regions occur later in the molt sequence, while the high mercury content in the apertium suggests it is one of the first regions to undergo new growth (Figure 1). However, observations of prejuvenal molt sequences for two sets of hand-reared loons (n = 7) raised in different years in Ontario do not support this interpretation (12). In the Ontario study the molt sequence began with the eruption of the remiges after approximately three and a half weeks, soon followed by the appearance of the tail feathers near week four. By the fifth week, white ventral body feathers had replaced the down found on the belly, and the remaining wing feathers were in place. Head feathers were present by week six, and the prejuvenal molt was near completion after eight weeks, including the dorsal body, scapulars and legs. Flight feather growth continued until week fifteen (12).

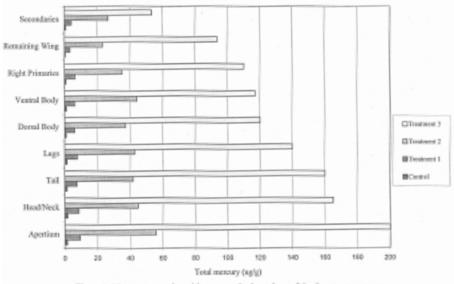


Figure 1. Mean mercury deposition among body regions of the four treatment groups.

Other explanations for the lack of correlation between feather region mercury content and molt sequence were also considered. It is possible for later developing feathers to contain higher mercury concentrations than those feathers that develop early in the molt sequence if the amount of ingested mercury exceeds the amount of mercury excreted (7). There is also less variation in mercury concentrations among feather regions in birds receiving smaller amounts of mercury since less mercury is eliminated with a decreased body burden of mercury ry (7).

Total feather mercury burden increased with an increased mercury dose as expected (Figure 1). Mercury elimination rates were higher for the more contaminated birds than those receiving smaller amounts of mercury. These results correspond with a study conducted by Monteiro et al.which also found a positive correlation between dietary mercury and mercury levels in seabirds (9). Becker et al. also came to this conclusion while studying Herring gulls, Black-headed gulls and Common terns (7). In Addition, Lewis and Furness found that 49 % of mercury administered to Black-headed gull chicks was eliminated into growing feathers, and that the total plumage accounted for 65 % of the total body burden of mercury after the completed molt (3).

The difference in mercury concentrations among body regions is relatively unclear. The primaries may prove to be one of the best choices for feather sampling based on the average mercury concentration found in this region along with the ease of their removal. However, without performing a statistical analysis of the data, it can not be determined whether or not the sampling of a representative feather region would be sufficient for mercury feather analysis. Statistical tests could be incorporated after the feather regions of the remaining sixteen birds involved in the UMESC study have been analyzed for mercury content.

ACKNOWLEDGEMENTS

This research could not have been performed without the support of the Upper Midwest Environmental Sciences Center, La Crosse, Wisconsin and the Wisconsin DNR. I would especially like to thank Kevin Kenow and Randy Hines for their direction and guidance. Thanks also to Sam Troxell and Jenny Fiedler for their plucking assistance, and Melissa Jenco for providing a loon body region diagram. In addition, I would like to thank Jean Ruhser, UW-La Crosse Biology Department, for her supervision and manuscript review.

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Appendix 1 Mean feather region weights by body region

Region	Mean Weight (g)
Apertium	3.37
Dorsal Body	6.27
Head/Neck	4.28
Legs	2.08
Right Primaries	10.44
Secondaries	4.41
Tail	4.21
Ventral Body	46.63
Remaining Wing	28.66

Appendix 2. Mean mercury deposition for each body region among treatment groups (µg)

Region	Control	Treatment 1	Treatment 2	Treatment 3
Apertium	3.33	32.25	161.19	904.00
Dorsal Body	3.20	36.56	235.97	912.95
Head/Neck	4.97	40.18	190.60	765.85
Legs	1.73	14.03	90.11	402.50
Right Primaries	7.18	62.46	357.97	1315.05
Secondaries	2.20	17.36	116.60	262.34
Tail	3.66	26.67	180.26	825.60
Ventral Body	45.45	275.53	2079.97	6052.06
Remaining Wing	6.71	91.01	645.21	3147.28
All Feathers	8.71	66.23	450.88	1620.85

Treatment Group	Loon ID	Total Mercury (µg)
Control	1416	60.88
Control	1426	95.99
Treatment 1	1414	582.86
Treatment 1	1427	609.22
Treatment 2	1428	3495.56
Treatment 2	1415	4620.23
Treatment 3	1417	13186.99
Treatment 3	1424	15988.25

Appendix 3. Total feather mercury burden for each loon