

Initial indications of polycyclic aromatic hydrocarbon exposure in Saskatchewan common loons

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Abstract

Mercury (Hg) and polycyclic aromatic hydrocarbons (PAHs) are global pollutants known for their toxicity to wildlife. Because of their trophic position, common loons (*Gavia immer* (Brünnich 1764)) are excellent indicators of environmental quality. In 2014 and 2015, tissue samples of ten adult common loons (plus one recapture) were obtained in Meadow Lake Provincial Park, Saskatchewan, and assessed for Hg and PAH exposure. Blood and feather levels of these contaminants are indicative of exposure during breeding and in wintering areas, respectively. Compared with an international Hg database, blood Hg levels were low (<1 µg/g). In most loons (90.5%, 10 out of 11), blood PAH concentrations were also low (<10 ng/g), but high (120 ng/g) for one individual (9.5% 1 out of 11). Feather PAH concentrations were high (95.9 ng/g and 250.6 ng/g) in two of the four loons (50%) caught in 2015. These data indicate that loons breeding in Meadow Lake Provincial Park were exposed to low levels of Hg; however, some individuals are being exposed to PAHs in both their breeding and wintering locations. The effect of these environmental pollutants on individual loon fitness is unclear, but because of their extreme toxicity in biological systems we suggest that future monitoring in the surrounding region is warranted.

Key words: common loons, *Gavia immer*, mercury, polycyclic aromatic hydrocarbons, Saskatchewan, contaminants, Meadow Lake Provincial Park



Citation: Paruk JD, Chickering M, Mager JN III, Wilkie SC, and Espie RHM. 2018. Initial indications of polycyclic aromatic hydrocarbon exposure in Saskatchewan common loons. FACETS 3: 849–857. doi:[10.1139/facets-2018-0009](https://doi.org/10.1139/facets-2018-0009)

Handling Editor: Mark Mallory

Received: March 29, 2018

Accepted: June 18, 2018

Published: September 6, 2018

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Published by: Canadian Science Publishing

Introduction

Mercury (Hg) toxicity to wildlife has been widely documented (Scheuhammer et al. 2007; Burgess and Meyer 2008). Common loons (*Gavia immer* (Brünnich 1764)) are good indicators of Hg contamination in aquatic systems (Evers et al. 2008). Loons exposed to Hg experience neurological and physiological impairments, which may contribute to diminished reproductive success (Nocera and Taylor 1998; Burgess and Meyer 2008). Reproductive success declines with increasing blood Hg concentrations, possibly due to aberrant incubation behavior in birds with high Hg burdens (Burgess and Meyer 2008; Evers et al. 2008). There is some evidence that western Canada is experiencing an increase in Hg contamination (Tozer et al. 2013). Although the origin of the atmospheric Hg deposition is unclear, the oil sands operations in Alberta (Cold Lake Oil Sands and Athabasca Oil Sands) or coal emissions from Asia are possible sources (Tozer et al. 2013).

Furthermore, the oil sands operations may be responsible for elevated polycyclic aromatic hydrocarbon (PAH) levels in the Athabasca watershed (Headley et al. 2001; Kelly et al. 2009). PAHs are global pollutants derived from fossil fuels and produced during oil production and combustion (Albers 2006). These compounds are transported long distances and can be found in air, soil, surface water, and groundwater. They can persist in sediment for decades and are toxic to wildlife at low concentrations (e.g., ng/g) (Wang et al. 2002; Albers 2006). PAHs cause a range of negative health effects, including liver damage, hemolytic anemia, weight loss, changes in salt gland weight, gut damage, and immunosuppression (Albers 2006). In addition, they are carcinogenic, mutagenic, and tumorigenic (Albers 2006).

Common loons are large, heavy-bodied, (3.5–7 kg), piscivorous migratory birds that breed on freshwater lakes in temperate regions across North America and winter predominately in marine environments. Common loons are long-lived (>25 years), and have high adult survival and low fecundity (Evers et al. 2010). They exhibit both breeding and wintering site fidelity (Paruk et al. 2015) and they provision their young primarily with fish from the natal lake. Breeding populations of common loons occur throughout the northern half of Saskatchewan, as southern lakes are not suitable because they are often shallow with reduced fish stocks (Smith 1996). Breeding loons in western Saskatchewan overwinter in both the Pacific Ocean and the Gulf of Mexico (Paruk et al. 2014a, 2014b).

Although common loons are not at formal risk across their breeding range in Canada, the Canadian Lakes Loon Survey documented a significant and nation-wide decline in overall productivity over the past two decades (Tozer et al. 2013). Although eastern populations have been more at risk from acid precipitation and elevated Hg levels than western populations (Evers et al. 2010), a recent survey documented a steeper decline in loon productivity in western provinces compared with eastern provinces (Tozer et al. 2013). Assessment of contaminants such as Hg and PAHs may help provide insight as to causes of these declines.

Our objective was to assess Hg and PAH exposure in breeding common loons in northern Saskatchewan.

Methods

Tissue sample collection

Common loons were captured from seven lakes within Meadow Lake Provincial Park in west-central Saskatchewan from 27 May to 5 June 2014 and 9–15 June 2015 (Fig. 1). Loons were caught using a suspended mist net with a loon decoy placed in the center (Uher-Koch et al. 2015). A playback of loon calls (yodel, tremolo, and wail) was used to lure individuals or pairs to the decoy. Captured loons were banded with a United States Geological Survey (USGS) aluminum band and a unique combination of plastic colored bands, enabling the individual identification of birds by field biologists during subsequent surveying. A suite of morphometric measurements was taken from captured individuals, including bill width, length, and depth, tarsal width and length, and mass. Blood samples were collected from the tibiotarsal vein to evaluate short-term Hg accumulation in individuals. Blood was drawn using a luer adapter directly into two 8 cc Vacutainers (Becton Dickinson, Franklin Lakes, New Jersey, USA) with sodium heparin. A total of 5–8 cc of blood was obtained from each individual. In addition, several capillary tubes were filled directly by placing them over the injection site. Upon filling, the ends were sealed (Critoseal, Fisher Scientific, Hampton, New Hampshire, USA).

To assess blood PAHs in 2014, several drops of blood were transferred to FTA cards (Fast Technology for Analysis of Nucleic Acids, GE Healthcare Life Sciences, Piscataway, New Jersey, USA) and placed in envelopes. To assess blood PAHs in 2015, 2–3 mL of blood were spun at 3000 rpm for 10 min and the plasma was decanted and frozen for analysis.

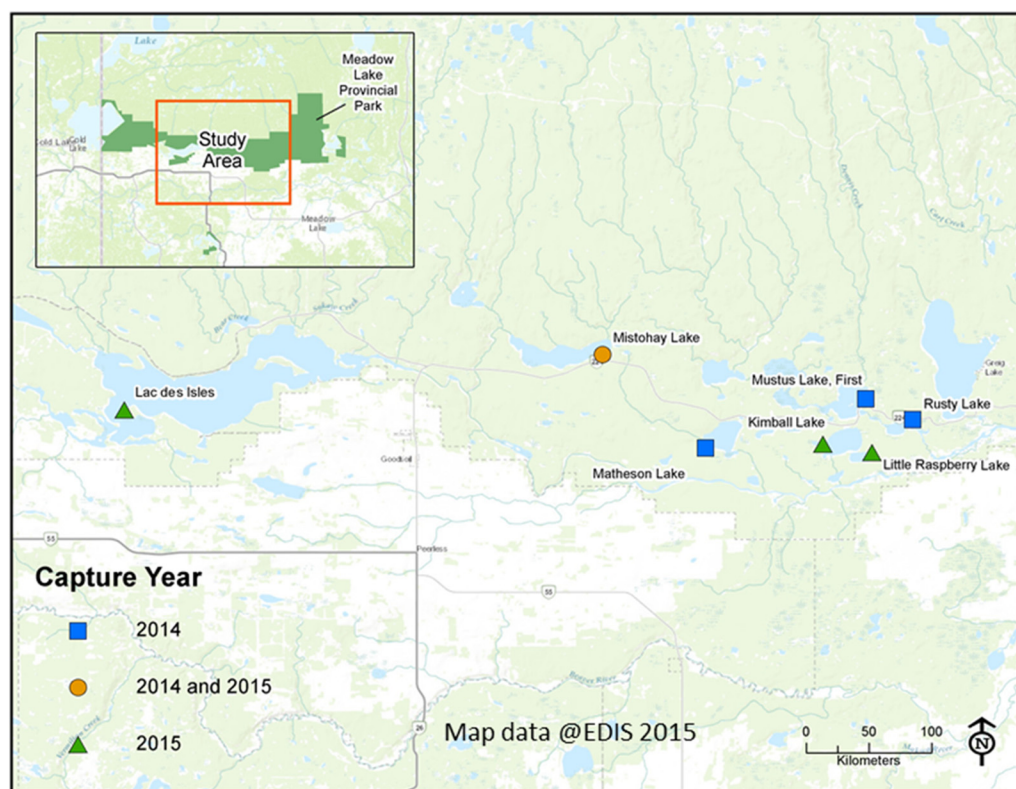


Fig. 1. Capture locations of common loons banded in Meadow Lake Provincial Park, Saskatchewan, in 2014 and 2015.

Feather samples, including one secondary feather from each wing, were collected to provide information on long-term Hg accumulation. To assess exposure to PAH during the winter period, four scapular feathers were collected in 2015 (scapular feathers were not obtained from loons in 2014). PAHs do not bioaccumulate, but their presence in feathers is indicative of circulating blood levels at the time of feather formation (C. Perkins, personal communication, 2014).

Laboratory analysis

Standard laboratory protocols for analyzing total Hg in common loon tissues for blood and feathers as described by Evers et al. (1998) were followed. Analyses of blood and feather tissue samples were conducted at the Biodiversity Research Institute Laboratory (Gorham, Maine, USA). Analyses of blood and feather PAHs were previously described by Paruk et al. (2016), and were conducted at the University of Connecticut's Center for Environmental Sciences and Engineering Laboratory. Sixteen parent PAHs were measured, as established by Keith and Telliard (1979) for the United States Environmental Protection Agency (phenanthrene, anthracene, naphthalene, acenaphthalene, fluorene, acenaphthene, fluoranthene, pyrene, chrysene, benzo(a)anthracene, benzo(b)fluoranthene, benzo(k)fluoranthene, benzo(a)pyrene, dibenz(a,h)anthracene, indeno(1,2,3-cd)pyrene, and benzo(g,h,i)perylene).

To assess loon health, hematocrit was determined by placing 0.1 cc of blood in a hematocrit tube and spinning it for 7 min at 12 000 rpm using a hematocrit rotor. The percentage of whole blood vs. plasma was obtained using a hematocrit reader (Haefele et al. 2005). In addition, a full screening of

Table 1. Summary of blood and feather mercury (Hg) and polycyclic aromatic hydrocarbon (PAH) concentrations from common loons banded in Meadow Lake Provincial Park from May to June 2014 and 2015.

Lake name	Year	USGS band No.	Sex	Mass (g)	Blood		Feather
					Hg concentration (µg/g)	PAH concentration (ng/g)	PAH concentration (ng/g)
Kimball	2015	0669-21952	Female	3400	<1	<5	250.6
Lac des Isles	2015	0669-21901	Female	3000	<1	<5	<19 ^a
Little Raspberry	2015	0669-21948	Female	3400	<1	<5	<19
Mistohay	2015	1118-15998	Male	4500	<1	<5	95.9
Matheson	2014	0938-03492	Male	4500	0.624	<3	DNT
Matheson	2014	0938-78822	Male	4300	0.619	<3	DNT
Matheson	2014	1118-15954	Male	4100	0.967	<3	DNT
Mistohay	2014	1118-15998	Male	4750	0.883	120	DNT
Rusty	2014	0968-87799	Female	3500	0.53	<3	DNT
Rusty	2014	1118-15953	Female	3100	0.419	<3	DNT
First Mustus	2014	1118-15955	Female	3350	0.295	<3	DNT

Note: Bold text indicates data from the same individual caught in consecutive years. USGS, United States Geological Survey; DNT, did not test.

^aLowest detectable concentration.

blood chemistry for key health parameters was conducted in 2015 at the University of Miami Avian and Wildlife Laboratory (UMAWL). The UMAWL does a full range of screening, but for this study we focused on several key enzymes that are indicative of organ or tissue damage (aspartate aminotransferase (AST), alanine aminotransferase (ALT), lactate dehydrogenase (LDH), gamma glutamyl-transferase (GGT), and creatine phosphate (CPK).

Bird capture and handling methods complied with the guidelines of the American Ornithological Council on animal care under Environment Canada permit number #22636 and Institutional Animal Care and Use Committee permit number 042214-104.

Data analysis

Blood and feather Hg levels obtained from breeding common loons in Saskatchewan were compared with the Biodiversity Research Institute's (briloon.org/) national loon mercury database. The database consists of blood and feather Hg levels obtained from over 5000 loons captured across North America. We used Paruk et al. (2016) to compare our blood PAH concentrations. This study examined blood PAHs obtained from common loons ($n = 93$) wintering off the Louisiana coast in an area that was heavily exposed to oil from the 2010 Deepwater Horizon oil spill. Blood chemistry data were compared with the only comparable data collected for 33 breeding loons from New York performed by the same lab (N. Schoch, personal communication, 2015).

Results and discussion

Seven and four adult common loons were caught in 2014 and 2015, respectively (Fig. 1). A male on Mistohay Lake was captured during both years; therefore, a total of 10 uniquely color-banded loons (4 males and 6 females) were included in this study. Males were significantly larger than females (4350 ± 95.7 g vs. 3292 ± 80.0 g; $p < 0.01$) (Table 1), which is consistent with findings by Gray et al. (2014).

Moreover, these loons, along with those captured in Manitoba, are the smallest loons in North America (Evers et al. 2010; Gray et al. 2014). Previous research has shown that common loons from this province are long-distance migrants (>1800 km) that winter in either the Pacific Ocean or the Gulf of Mexico (Paruk et al. 2014a, 2014b). Common loons have one of the highest wing-loading values of any North American bird (Poole 1938); thus, energetic costs associated with long-distance migration are likely high and constraining. Therefore, factors that influence loon health and body condition prior to migration may influence their survival and future reproductive success. Exposure to environmental contaminants is likely to affect loon body size and condition and may, therefore, significantly affect these loons' abilities to successfully move between breeding and wintering sites.

Contaminant exposure

Blood Hg values for all loons captured were low (<1.0 µg/g; Table 1), as established by Evers et al. (2008). Blood Hg concentrations reflect the near-term availability (i.e., days of exposure) of methylmercury in prey items. Loons are excellent indicators of Hg contamination (Evers et al. 2008); thus, the low Hg levels observed in the loons at Meadow Lake Provincial Park suggest that atmospheric fall-out from anthropogenic and natural sources was low at this time.

One male loon captured 30 May 2014 on Mistohay Lake had a blood PAH value of 120 ng/g (Table 1). Based on blood PAHs levels obtained from common loons wintering off the Louisiana coast in an area that was heavily exposed to oil from the 2010 Deepwater Horizon oil spill (range 0–270.2 ng/g; Paruk et al. 2016), this level is considered high. Most birds can rapidly metabolize PAHs (<2 weeks) because of a well-developed mixed-function hepatic oxygenase system (Albers 2006). As spring migration takes 6–8 weeks for breeding Saskatchewan loons (Paruk et al. 2014a, 2014b), any PAHs they may have received during time spent in their winter environment would have been metabolized by the time they reached the breeding area. Loons typically return to breeding lakes within a day or two after ice out (Evers et al. 2010). Because the 2014 winter was exceptionally mild and ice out was 3 weeks earlier than normal (R. Espie, personal observation, 2014), loons likely returned to the breeding lakes in early to mid-May. This early arrival would have given the male loon on Mistohay Lake two to three weeks to eliminate any PAHs it acquired during its migration. Therefore, we feel that this individual was likely exposed to PAHs on its breeding lake. Also, elevated concentrations of PAHs were found in the scapular feathers of two of the four loons (50%) sampled in 2015. This indicates that these loons were exposed to PAHs in their wintering areas. Anthracene was observed in loon blood and phenanthrene was observed in feather tissue (Table 2). Both are light-weight, airborne-dispersed PAHs that persist for a long time in the environment. Because they are global pollutants it is difficult to identify their source, but oil sand operations west of the park produce PAHs and prevailing winds could deposit them in this region.

Health assessments

Hematocrit, glucose, total protein, and white blood cells fell within the normal range (Haeefe et al. 2005) for loons (Table 3). All loons tested negative for avian influenza. Of the four loons assessed for blood chemistry, two (those breeding on Lac des Isles and Little Raspberry Lake) had no detectable feather PAH concentrations, and two (those breeding on Mistohay Lake and Mustus Lake) had high feather PAH concentrations, making an investigation of the physiological health of the birds with high PAH concentrations possible. One could posit that if oil exposure was high enough to cause tissue or organ damage then these loon individuals might also have elevated AST, ALT, LDH, and GGT levels compared with those individuals not exposed to PAHs. The results, however, were the reverse of what we expected (Table 3). Loons with no or low blood or feather PAH concentrations had high AST and ALA levels, whereas loons with high PAH concentrations had low AST and ALA levels. Given our small sample size, many other factors that we did not measure as part of

Table 2. Summary of polycyclic aromatic hydrocarbon (PAH) compounds identified in blood and feathers from common loons banded in Meadow Lake Provincial Park in 2014 and 2015.

Individual band No.	Source	Year	Total PAH (ng/g)	Phenanthrene (ng/g)	Anthracene (ng/g)
1118-15998	Feather	2015	95.9	95.9	None detected
	Blood	2015	<5	None detected	None detected
	Blood	2014	120.0	None detected	120.0
0669-21948	Feather	2015	0.0	0.0	None detected
	Blood	2015	<5	None detected	None detected
0669-21952	Feather	2015	250.6	250.6	None detected
	Blood	2015	<5	None detected	None detected
0669-21901	Feather	2015	0.0	0.0	None detected
	Blood	2015	<5	None detected	None detected
0669-21952	Blood	2014	0.0	None detected	None detected
0669-2194a8	Blood	2014	0.0	None detected	None detected
0669-21901	Blood	2014	0.0	None detected	None detected
1118-15998	Blood	2014	0.0	None detected	None detected
0669-21948	Blood	2014	0.0	None detected	None detected
0669-21901	Blood	2014	0.0	None detected	None detected

Table 3. Comparison of health assessment data (mean or mean \pm SE) for breeding loons in Meadow Lake Provincial Park, Saskatchewan, and Adirondack Park, New York.

Lake where individual caught	PAH (ng/g)	PCV (%)	TP (g/dL)	Gluc (mg/dL)	AST (U/L)	ALT (U/L)	LDH (U/L)	CPK (U/L)	GGT (U/L)
Kimball	120	40	4.4	238	196	88	1465	133	7
Mistohay	95	50	2.4	321	270	77	674	69	5
Lac des Isles	0	48	4.2	224	485	117	3656	416	9
Little Raspberry	0	49	3.2	283	639	205	4290	333	17
New York Lakes ($n = 28$)	DNT	48 \pm 0.6	3.5 \pm 0.7	261 \pm 6	290 \pm 17	82 \pm 3	856 \pm 113	581 \pm 98	DNT

Note: PAH, polycyclic aromatic hydrocarbon; PCV, packed cell volume; TP, total protein; Gluc, glucose; AST, aspartate aminotransferase; ALT, alanine aminotransferase; LDH, lactate dehydrogenase; CPK, creatine phosphate; GGT, gamma glutamyl-transferase.

our study could have brought about this result (e.g., stress, other contaminants, etc.). Baseline blood parameters are important references for assessing and monitoring population health and for understanding future population trends. As loons from this region of Saskatchewan winter in different ocean basins, there is the potential for the transmission of avian diseases from one basin to the other (Tracey et al. 2004). Thus, such baseline data are extremely valuable for monitoring the health of avian populations across the continent. More baseline data from loon populations across this region

would be useful to obtain a broader understanding of overall loon health and exposure to environmental contaminants.

Collectively, these data suggest that loons in Meadow Lake Provincial Park are not at risk for Hg contamination; however, some individuals are exposed to PAHs in both their wintering and breeding areas. What effect, if any, these PAH concentrations have on loon fitness remains unclear. However, elevated blood PAH concentrations lower both body mass (body condition) and hematocrit levels (aerobic capacity) in common loons (Paruk et al. 2016), both of which are key factors for their survival and future reproductive success because they are long-distance migrants (Kenow et al. 2002; Paruk et al. 2014a, 2014b). Chronic exposure to environmental contaminants such as PAHs decreases fitness and survival (Iverson and Esler 2010). It is unclear what demographic or population level effects PAH exposure have on breeding loons in Meadow Lake Provincial Park, but the preliminary data suggest that it may warrant further monitoring.

Acknowledgements

This project was funded by the Saskatchewan Ministry of the Environment and Earthwatch Institute. We are grateful for the following volunteers who assisted us in catching loons: Glenda Booth, Janice Spence, Bonnie Mabee, Dick Mabee, Elizabeth Johnson, Nathan Johnson, Beth Branthaver, and Lisa Waag. Finally, we wish to thank Jeffrey Tash for producing the figure and Kristin Kovach and Nina Schoch for helpful comments on an earlier draft.

Author contributions

JDP and RHME conceived and designed the study. JDP, MC, JNM, SCW, and RHME performed the experiments/collected the data. JDP and RHME analyzed and interpreted the data. JDP, MC, JNM, SCW, and RHME contributed resources. JDP, MC, JNM, SCW, and RHME drafted or revised the manuscript.

Competing interests

The authors have declared that no competing interests exist.

Data availability statement

All relevant data are within the paper.

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