Penobscot River Mercury Study

With comparisons to previous years

Report to:
Judge John Woodcock
U.S. District Court (District of Maine)
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By

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ABSTRACT

During the winter of 2013-2014, mercury (Hg) concentrations were determined in American black ducks wintering along the lower Penobscot River and in a reference area on the Downeast coast of Maine. Average Hg concentrations in duck breast muscle at the end of the fall hunting season in Mendall Marsh (0.43 ± 0.04 µg/g ww, mean ± SD) were over two times greater than the Maine Hg action level. In the previous two years when measurements were made (2010-11 and 2011-12), concentrations were close to 4 times the action level. In duck muscle, 99% of the mercury found is methyl Hg. Mercury concentrations in blood samples from ducks live-trapped in January and February 2014 were also used to compare Hg exposures among sites and over time. Total Hg in the blood from ducks trapped at Mendall Marsh (0.26 µg/g ww, adjusted for bill length) was significantly greater than found in ducks from ES13-South Verona (0.09) and from the reference site in Frenchman Bay (0.07). Compared to earlier years, Hg concentrations in blood were significantly lower in 2014 at Mendall Marsh and at ES13-South Verona. Blood Hg concentrations remained unchanged in ducks wintering in Frenchman Bay. The reason why Hg concentrations in 2014 were lower at sites along the lower Penobscot is not known. While it may indicate a reduction in Hg concentrations in the food web a more likely explanation is random variation in a range of variables. One possible explanation for the lower Hg concentrations in 2014 is the unusually cold weather in the region in the first half of the winter. Unlike previous years, extensive icing of the marsh platform and large portions of the mudflats in Mendall Marsh may have limited the foraging areas used by the ducks and reduced their exposure to Hg.

INTRODUCTION

Monitoring of mercury (Hg) concentrations in American black ducks (Anas rubripes) began in the winter of 2010-2011 as part of the Penobscot River Mercury Study, a larger assessment of Hg contamination in the lower Penobscot River. The monitoring work on black ducks began at the suggestion of Kelsey Sullivan, Biologist with the Maine Department of Inland Fisheries and Wildlife, who leads the black duck banding program for the State of Maine, and the DIF&W continues to support the monitoring work. The mercury in most ducks is analyzed as total mercury for this study, but it is known to be composed of about 99% MeHg, which is the most toxic form of mercury to animals and human consumers (Kopec and Bodaly 2013). Ducks were sampled in marshes along the lower Penobscot River and at a reference site in Frenchman Bay. Monitoring was repeated the following winter, and again two years later in the winter of 2013-2014. The findings of the 2013-2014 monitoring season are reported here, along with an analysis of temporal trends in Hg concentrations using the full 2010 through 2014 dataset.

Black ducks are a useful indicator of Hg exposure during the winter months in the tidal marshes along the lower Penobscot River and at nearby coastal marshes. The ducks arrive from breeding sites in the Canadian Maritimes in late September or early October, and stay at a given wetland through the fall, moving to coastal marshes only if solid ice prevents foraging at more inland sites. They leave Maine for northern breeding grounds in mid-March through early April (Longcore et al. 2000).

Black ducks forage on invertebrate prey gleaned from exposed mudflats during low tide and from marsh sediments and plants when high tides flood the marsh platform. The periwinkle snails (Littorina), amphipods (Gammarus), blue mussels (Mytilus), soft-shelled clams (Mya) and worms (Nereis) eaten by black ducks (Jorde and Owen 1990) have been previously found to have elevated Hg concentrations in the marshes and intertidal areas along the lower Penobscot River (Bodaly et al. 2009; Kopec and Bodaly 2013).

In the first two winters of duck monitoring, elevated Hg concentrations were found in duck breast muscle sampled in Mendall Marsh in December, after ducks were resident for several months along the lower Penobscot. This finding led to a human health advisory issued by the State of Maine recommending limited or no consumption of duck meat from the contaminated area.
When the ducks were at their summer breeding ground, they were exposed to low Hg concentrations, as shown by Hg in their primary feathers. Long-term banding studies indicate that ducks, which overwinter in Maine, breed in Maritime Canada. Given the timing of feather molt in black ducks, Hg analyses of the first primary feather (P1) reflects Hg exposure during the summer breeding season. Evidence from the first year of duck monitoring found uniformly low Hg concentrations in the P1 feathers collected from ducks over-wintering in the study area, whether they were collected along the lower Penobscot or at the coastal reference site in Frenchman Bay (Kopec and Bodaly 2013).

The black duck monitoring makes a unique contribution to the Penobscot River Mercury Study as it documents Hg exposure during the winter months. All other biota examined in the study, along with the sediment and water samples, were collected during the summer and early fall. This long term study also provides needed information for the health advisory on human consumption of black ducks, which has been instituted by the state of Maine.

METHODS

During the winter of 2013-2014 muscle samples were collected from black ducks sampled in Mendall Marsh and blood samples were collected from ducks trapped in Mendall Marsh, the southern tip of Verona Island, and from reference sites in northern Frenchman Bay. The winter weather created challenging conditions not experienced during the two previous winters when ducks were sampled.

Muscle samples (n = 3) were collected December 16 – 23, 2013 at the end of the second half of the fall hunting season, from ducks shot at Mendall Marsh by hunters and personnel from the Maine Department of Inland Fisheries and Wildlife. Samples were collected from the scapular region of the breast muscle and frozen prior to analysis.

Most of the samples collected were blood samples because this could be done with less injury to the ducks, and because mercury concentrations in the duck muscle can be accurately inferred from blood mercury concentrations (Kopec and Bodaly 2013). Blood samples (n = 51) were collected from three study areas in late January and early February 2014. Baiting all sites with whole corn began January 6, after the coastal hunting season was closed. As in past years, collection sites were baited to accustom the ducks to finding food at the site. Toward the end of the baiting period open wire traps were constructed on the bait site, allowing free access into and out of the baited trap. After ducks willingly entered the open trap to eat the bait, the trap was closed, leaving only narrow access chutes which allowed the ducks to enter, but not exit the trap. At Mendall Marsh, numerous capture methods were attempted, including several net gun arrays and wire traps with different configurations.

The initial two-week collection window for blood samples (January 27 to February 9) was extended due to difficulties capturing ducks at Mendall Marsh. Ducks were successfully trapped and sampled at sites in the reference area of Frenchman Bay and at ES13-South Verona between January 27 and 30. Ongoing efforts to trap ducks at Mendall Marsh were complicated by limited trapping areas, winds, tides, predators (bald eagles and coyotes) and competition from non-target species (Canada geese). On February 12th additional ducks were sampled at ES13-South Verona and in Frenchman Bay to confirm whether extended residence time at those sites increased the blood Hg concentration in the ducks. Ducks were sampled at Mendall Marsh on February 12 and 13. Trapping attempts were continued at Mendall Marsh through February 19, but no additional samples were collected.

Blood was collected from the inner brachial vein at the base of the wing, or from the femoral vein in the leg using a 25 gauge butterfly needle. Blood was collected into a trace element Vacutainer (royal blue) containing EDTA to prevent clotting. Up to 1.5 ml of blood was collected, gently mixed, and placed on ice for transport to the lab where samples were frozen prior to analysis. The right P1 primary feather was collected from each duck and archived for future Hg analysis.
Relevant biological data were collected from each bird, including weight, sex, age class (determined by appearance of primary feathers), and lengths of wing chord, tarsus, and bill.

Blood and muscle samples were analyzed for total Hg and a subset of muscle samples were analyzed for methyl Hg at the Battelle Marine Sciences Laboratory in Sequim, Washington. Breast tissue samples were freeze-dried and ball-milled at the lab. Blood samples were analyzed as whole blood homogenized with EDTA. All samples were analyzed for total Hg by EPA Method 1631e. Two breast muscle samples were analyzed for methyl Hg by EPA Method 1630 modified for tissues (by digestion for 25% KOH in methanol). QA/QC tests, including blanks, matrix spikes, replicates and SRMs were all within accepted criteria limits.

Power analyses were used to estimate the minimum sample sizes needed to detect significant temporal changes in Hg concentrations in the black ducks. The sampling goals were 5 breast muscle samples from ducks taken from Mendall Marsh in December, and 20 blood samples from ducks sampled in late January - early February from each of the three black duck collection sites.

**RESULTS AND DISCUSSION**

Muscle samples were collected from three ducks from Mendall Marsh in late December, two females and one male. A total of 51 blood samples were collected from three sampling areas in January and February. The majority of ducks sampled were males (75%) and adults two years of age and older (77%). In Frenchman Bay most blood collections were from ducks captured near the mouth of the Jordan River (n = 11), the reference area used in 2012, and at additional Frenchman Bay collection sites at Haynes Pt. (n = 4) and near the mouth of the Union River (n = 4 in January, n = 3 in February). At ES13-South Verona, blood was sampled from 18 ducks in January and 3 in February, and at Mendall Marsh blood was sampled from 8 ducks in February (Appendix 1).

Mercury concentrations in biota have been found to increase with residence time at contaminated sites (Eagles-Smith et al. 2009; Kopec and Bodaly 2013). However, our study design ensured that geographic comparisons of Hg concentrations among different sites were not influenced by residence time. In the initial design, all collections were planned to occur within a two-week period (January 27 – February 9). While adequate datasets of blood samples were collected in Frenchman Bay and at ES13-South Verona, we were unable to trap ducks at Mendall Marsh within the planned collection period. On February 12th an additional three blood samples were collected from both Frenchman Bay and from ES13-South Verona to test whether Hg concentrations had increased at either site since their original sample dates in January. No significant difference was found between Hg concentrations in blood samples collected in January and February at either ES13-South Verona (geometric mean total Hg, Jan = 0.10 µg/g ww; mean, Feb = 0.10 µg/g ww) or at the Union River site in Frenchman Bay (geometric mean total Hg, Jan = 0.11 µg/g ww; mean, Feb = 0.04 µg/g ww; 2-sample t-test, log-normalized total Hg, pooled variance, P > 0.05). As a result, samples were pooled within each site regardless of date collected, and geographic comparisons of Hg concentrations in duck blood were determined to be valid despite the extended collection period. Further, no significant difference was found in Hg concentrations from the three sites sampled in the Frenchman Bay reference area (ANOVA log total Hg vs site, P = 0.95); all data from Frenchman Bay were pooled for comparison with the other sites.

The percent of total Hg that was methyl Hg was determined in a subset (2) of the muscle samples. Similar to past years, 99% of the Hg in duck breast muscle was methyl Hg.

**Geographic Trends in Hg Concentrations in Black Ducks:** In 2014 the mean total Hg concentration in blood from ducks sampled in Mendall Marsh (0.26 µg/g ww; antilog of LSM, adjusted for bill length; Figure 1) was significantly greater than at ES13-South Verona (0.09 µg/g ww) or from the reference site in Frenchman Bay (0.07 µg/g ww). Hg in duck blood from ES13-South Verona was not significantly different from the reference site.
Of all the size variables tested, bill length was the only significant co-variate in the model (ANCOVA, log total Hg, adjusted for bill length, P < 0.05; Tukey HSD, α < 0.05; MM > ES13-SV = FB). Bill length increases with duck size, and was found to be significantly greater in male ducks (mean, 54.5 mm) compared to female ducks (mean 51.8 mm; 2-sample t-test, pooled variance, P = 0.001), reflecting the greater mass in male ducks (Longcore et al. 2000).

Figure 1. Map showing the location of American black duck (ABDU) study sites with graphs illustrating the relative Hg concentrations in blood and muscle (µg/g w.w. for both muscle and blood) for the period 2010-2014. The mean blood total Hg concentrations are antilogs of LSMs adjusted for duck bill length. Muscle total Hg values at Mendall Marsh are geometric means. Significant declines in mean blood Hg concentrations were found at Mendall Marsh and ES13-South Verona.
Temporal Trends in Hg Concentrations in Black Ducks: Monitoring in the winter of 2013-2014 created the third dataset for black ducks and the first opportunity to assess temporal trends in Hg concentrations for this species. As expected with only three years of data collection, there was no significant trend in Hg concentrations in duck breast muscle from Mendall Marsh (ANOVA, log total Hg, \( P = 0.50 \)). The power to detect a significant change was low due to the limited sampling period. The mean concentration in muscle in December of 2013 was notably lower than found in the previous two years (Figure 2).

Mercury concentrations in whole blood sampled from black ducks were significantly lower in 2014 at both of the sampling sites along the lower Penobscot River (independent ANOVA, adjusted for bill length, with pooled variance, \( P < 0.001 \); Figure 3). No significant change was found in blood Hg concentrations at the Frenchman Bay reference area. The significant drop in blood Hg concentrations found at Mendall Marsh and ES13-South Verona indicates reduced Hg exposure during this winter season. We do not interpret the observed decrease in 2014 in mercury concentrations at Mendall Marsh and ES13-South Verona to be evidence of abrupt changes in THg concentrations in the Penobscot marshes. Instead this is most likely due to year to year variance in MeHg bioaccumulation by the ducks. MeHg production and bioaccumulation rates are known to vary widely year to year (Kelly et al. 1997).
Figure 3. A temporal comparison of mean Hg concentrations in whole blood sampled from black ducks in January and February, 2011-2014. Significantly lower Hg concentrations were found in ducks sampled in Mendall Marsh and at ES13-South Verona. No changes were found at the Frenchman Bay reference site. Sample sizes are given at the base of each column. Error bars indicate the 95% confidence intervals.
One factor that may have contributed to the drop in Hg exposure during the 2013-2014 winter season is the weather. In November and December of 2010 and 2011, the first two seasons in which black ducks were sampled along the lower Penobscot, coastal Maine experienced unusually mild weather, with mean temperatures above freezing (34-37 °F) during the two month period (Figure 4). The marsh platform was generally free of ice, especially during high spring tides, and black ducks were observed foraging on the vegetated platform when it was flooded. Mudflats remained ice-free through late December along the Marsh River for over 2 miles south of the river’s mouth, providing further duck foraging area.

In 2013 mean temperatures along the Maine coast in November and December dropped below 30°F, the lowest in almost 20 years. By December the marsh platform was encased in solid layers of ice, preventing ducks from foraging on that habitat. Open water was limited to the northern section of Mendall Marsh between Treat Point and the boat launch. Upstream areas were generally filled with pack ice that covered the mudflats at low tide, further limiting access to invertebrate prey. By mid-January the only reliably open water and exposed mudflats in Mendall Marsh were found at Treat Point, maintained by strong tidal currents. During the winter of 2013-2014, the foraging black ducks were observed daily on the exposed mudflats at Treat Point but they had limited opportunities to forage.
in other areas of Mendall Marsh. This change in foraging area may have reduced the ducks' exposure to Hg relative to recent years.

**Return of Wintering Black Ducks to the Lower Penobscot Area:** The return of previously banded black ducks to the lower Penobscot indicates notable inter-annual site fidelity to this region. Mercury monitoring in black ducks along the lower Penobscot and Downeast coast was built on a larger study of black duck population dynamics. In the larger study up to 300 ducks are banded each year in lower Penobscot area and at coastal Downeast sites. Of the 13 previously banded black ducks that were re-captured for Hg sampling in the lower Penobscot in the winters of 2011-2012 and 2013-2014, all were re-captured on or near the site they were originally banded. This includes eight ducks re-captured at ES13-South Verona and five ducks re-captured within Mendall Marsh. The ducks were re-captured one to four years after their original banding date. Firm conclusions are not possible using this subset of the larger banding dataset, but the results suggest solid inter-annual site fidelity to these wintering grounds.

**CONCLUSIONS**

The primary objectives of the black duck monitoring work are: 1) to accumulate a multi-year data set of mercury concentrations in black ducks that will be capable of demonstrating the anticipated slow rate of mercury decline in the Penobscot over the coming years, and 2) to monitor the high mercury concentrations of mercury in duck muscle that exceed State of Maine guidelines for human consumption, and are data needed to make ongoing discussions about continuance of the health advisory for the consumption of black ducks. This report concludes that concentrations of mercury in the ducks of Mendall Marsh continue to be well above the advisory limit for human consumption set by the State of Maine. Further, that the apparent drop in Hg concentrations along the lower Penobscot during 2014 likely reflects interannual variation, given the currently short time period (3yrs) of monitoring. For these reasons we have proposed an ongoing, multiyear study of mercury in the ducks, in order to examine the anticipated slow changes over time.

**REFERENCES**


ACKNOWLEDGEMENTS

We thank K. Sullivan and H. Cady, Maine Department of Inland Fisheries and Wildlife, Bangor, ME, for cooperation in planning and executing this study.
Appendix 1. Black duck summary statistics for 2013-2014, using raw data for weight, bill length, total Hg concentrations in blood and muscle, and methyl Hg concentrations in muscle.

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<th>Site</th>
<th>Month</th>
<th>Sex</th>
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<th>Mean Blood Total Hg (µg/g ww)</th>
<th>SD Blood Total Hg</th>
<th>Minimum Blood Total Hg</th>
<th>Maximum Blood Total Hg</th>
<th>n of Muscle Total Hg</th>
<th>Mean Muscle Total Hg (µg/g ww)</th>
<th>Minimum Muscle Total Hg</th>
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<th>SD Weight</th>
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<th>Maximum Weight</th>
<th>Mean Bill Length (mm)</th>
<th>SD Bill Length</th>
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