August 2017


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Abstract

Mercury contamination within aquatic ecosystems is a concern in the Adirondack Park of New York State due to mercury deposition from global and regional atmospheric sources coupled with watershed characteristics that drive biophysical processes which mobilize and transform mercury, increasing its bioavailability. Short-term internal biological forces also impact mercury bioaccumulation as fish communities and populations change due to species introductions and lake management practices causing alterations in food web structure and energy transfer. Little Moose Lake, located in the southwestern region of the Adirondacks, provides an opportunity to study how shifts in food web dynamics may impact biological cycling of mercury. To promote the native Lake Trout fishery in Little Moose Lake, large-scale annual removal of non-native Smallmouth Bass has been utilized as a management strategy for over 15 years. Utilizing archived tissue and otolith samples and historical data, changes in total mercury concentrations, stable carbon ($\delta^{13}$C) and nitrogen ($\delta^{15}$N) isotopes, diet, age and growth were evaluated over time for Smallmouth Bass (*Micropterus dolomieu*) and Lake Trout (*Salvelinus namaycush*), the native top-predator in Little Moose Lake. Mercury concentrations in Lake Trout have increased over the 15-year period, while Smallmouth Bass mercury concentrations decreased over the same interval. Thus, changes in mercury deposition were likely not the main driver for these observations. Diets also changed over time, with Lake Trout consuming higher trophic level prey with higher concentrations of mercury and growth for both predator species increased. Changes in stable isotope signatures were also observed for both predator fish species and several lower trophic level organisms with a tendency for both $\delta^{13}$C and $\delta^{15}$N to be more depleted over time. The annual Smallmouth Bass removal resulted in shifts in trophic transfer mechanisms that influenced the temporal mercury trends in the two top-predator species. The knowledge gained
from this in-depth study will allow better understanding of spatial patterns and temporal trends in sportfish mercury concentrations in the context of food web changes.

By

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THESIS

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1. **Introduction**

Mercury (Hg) contamination within aquatic ecosystems is an important toxicological issue for environmental scientists, policy makers and the public who consume contaminated fish and are exposed to the neurotoxic pollutant. Toxic risks are also posed to fish and other wildlife due to elevated intake of Hg from their diets (Scheuhammer et al. 2007). For humans and wildlife, exposure to Hg occurs primarily via consumption of contaminated fish (Kamman et al. 2005; Driscoll et al. 2007a).

Global and regional atmospheric sources of Hg are linked to anthropogenic activities such as mining, coal combustion and waste incineration among others (Pirrone et al. 2010; UNEP 2013). Once Hg is emitted to the atmosphere, it can be deposited into areas remote from local sources (Fitzgerald et al. 1998) where it may be converted to methylmercury (MeHg) by sulfur and iron-reducing bacteria and archaea under reduced conditions (Benoit et al. 2003; Kerin et al. 2006). MeHg is strongly absorbed by lower trophic organisms and biomagnifies in aquatic food webs. This process results in particularly high concentrations of MeHg in piscivorous sportfish comprising >95% of the total mercury (THg) in their muscle tissue (Bloom 1992; Downs et al. 1998). Fractions of MeHg are usually lower and more variable in lower trophic level consumers, predominantly influenced by diet, trophic position and habitat (Chetelat et al. 2011; Yu et al. 2011).

Remote regions in the northeastern United States, including the Adirondack Park of New York State, are particularly sensitive to Hg contamination. Atmospheric Hg deposition in the Adirondacks is facilitated by high amounts of forest cover providing a large surface area for deposition (Xue et al. 2013). Chemical interactions mobilize, transform and enhance Hg bioavailability due to lake and watershed characteristics prevalent in the Adirondacks including
low productivity, high dissolved organic carbon (DOC) concentration, low acid neutralizing capacity (ANC) and low pH (Driscoll et al. 2007a). Additionally, the abundance of wetland cover enhances Hg transport through supply of dissolved organic matter and promotes the formation of MeHg due to reducing conditions thereby increasing the overall availability of Hg (Dennis et al. 2005). Due to levels of Hg above the EPA human health criterion of 0.3 μg•g⁻¹ observed in a variety of biota, the Adirondack region of New York State has been deemed a ‘biological mercury hotspot’ (Evers et al. 2007).

Decreases in Hg emissions have occurred regionally in northeastern North America since the 1990s when Clean Air Act amendments initiated policy changes with stronger regulations on anthropogenic emissions (Zhang et al. 2016). In contrast, global emissions, particularly those originating in Asian countries, are elevated and have continued to increase in recent years (UNEP 2013). Recent decreasing trends in Hg deposition in the Adirondack region have been attributed to controls on regional Hg emissions (Gerson & Driscoll 2016; Zhou et al. 2016). However, trends in Hg deposition have been spatially and temporally variable across other parts of the U.S. and increases in Hg deposition have been observed in the Rocky Mountain and Great Plains regions since the early 2000s (Weiss-Penzias et al. 2016). Associating decreases in Hg emissions and deposition with decreases in concentrations Hg in fish is extremely relevant to environmental policies and management strategies regarding mercury contamination in New York State and the northeastern United States. Recently, a study in eastern Massachusetts reported decreasing temporal trends in fish Hg concentrations associated with local emissions reductions indicating more pronounced trends in this local ‘mercury hotspot’ compared with the rest of the state (Hutcheson et al. 2014).
Although atmospheric Hg deposition is an apparent and important driver affecting concentrations of bioavailable Hg, other factors influence mercury concentrations observed in fish. Many physical, geographical and chemical characteristics that alter spatial and temporal mercury dynamics in Adirondack lakes and fish have been explored (e.g. Driscoll et al. 1994; Simonin et al. 2008; Dittman & Driscoll 2009). For example, temporal decreases in mercury concentrations of Yellow Perch (*Perca flavescens*) have been observed in several Adirondack lakes and was linked to changes in the acid-base chemistry of lakes mediated by decreases in acid deposition (Simonin et al. 2009). However, the impacts of food web alterations and other dynamic trophic interactions are also important to the bioaccumulation of Hg in fish, albeit less well-studied considerations. Changes in food web structure, energy transfer and mercury bioaccumulation have been documented for several regions following species introductions (Johnston et al. 2003; Eagles-Smith et al. 2008; Thomas et al. 2016), large scale species removal (Gothberg 1983; Verta 1990; Lepak et al. 2009), changes in prey assemblages (Johnson et al. 2015) and other fish-management related food web manipulations such as stocking practices (Lepak et al. 2012a; Lepak et al. 2012b). Additional research investigating how changes in aquatic food web structure influence bioenergetics and contaminant accumulation within specific, well-studied systems is needed to better understand and predict how ecosystem processes influence cycling and bioaccumulation of harmful pollutants such as mercury. As environmental policies to control mercury emissions result in decreases in Hg deposition in the U.S. and elsewhere (Driscoll et al. 2013), links between changes in atmospheric Hg deposition and changes in fish Hg levels have become increasingly important to understand. Internal biological forces at work must also be considered in this context as fish communities and
populations change due to species introductions, removals, stocking and other lake management practices.

Measurements of stable nitrogen (δ\(^{15}\)N) and carbon (δ\(^{13}\)C) isotopes are often used to determine energy sources and flows within ecosystems (Vander Zanden & Rasmussen 1999) and therefore provide information regarding trophic pathways for Hg within aquatic ecosystems. Signatures of δ\(^{13}\)C are distinct for pelagic phytoplankton compared to benthic algae which are typically enriched in \(^{13}\)C relative to phytoplankton (Post 2002). Additionally, the unique and characteristic δ\(^{13}\)C signatures of pelagic and littoral primary producers are transferred up the food chain, as little fractionation, or changes in δ\(^{13}\)C values, occur via consumption. Therefore, the differences in the δ\(^{13}\)C values for baseline primary producers will be accurately reflected in the δ\(^{13}\)C values of primary and secondary consumers obtaining energy from the same habitat. In contrast, signatures of δ\(^{15}\)N for consumers are typically enriched in \(^{15}\)N relative to their prey by +3-4‰ (Minagawa & Wada 1984) and δ\(^{15}\)N values are therefore indicative of relative trophic position (Vander Zanden & Rasmussen 1999). Because mercury biomagnification is strongly influenced by individual diets and consumption rates as well as the length of the food chain (Trudel & Rasmussen 2001), trophic positions estimated from measured δ\(^{15}\)N values are strong predictors of mercury concentrations in aquatic food webs (Cabana & Rasmussen 1994).

Predatory Smallmouth Bass (Micropterus dolomieu) have been introduced into many temperate North American lakes and resulted in changes in trophic structure that have been documented in several invaded ecosystems (Vander Zanden & Rasmussen 2002), including the site of the current study, Little Moose Lake in the Adirondack region of New York State (Weidel et al. 2000; Lepak et al. 2006). Annual Smallmouth Bass removal was initiated over a decade ago in Little Moose Lake to improve Lake Trout (Salvelinus namaycush) populations which are
the native top predator. In this study, archived tissue and otolith samples and historical data were utilized to evaluate changes in THg concentrations, stable $\delta^{13}C$ and $\delta^{15}N$ isotope ratios, diet, age and growth over time for Lake Trout and Smallmouth Bass from Little Moose Lake in the Adirondacks to:

1) Determine temporal trends in mercury concentrations for the two top-predator species over the >15-year interval of bass removal;

2) Evaluate internal trophic mechanisms resulting from annual bass removal that may have influenced temporal trends in mercury concentrations of the two top-predator species; and

3) Discuss how the knowledge gained from this in-depth study could be applied more broadly to better evaluate and monitor spatial patterns and temporal trends in sportfish mercury concentrations in the context of food web changes to inform policies that protect human health and the environment.
2. Methods

2.1 Study Site

Little Moose Lake is a 271-ha oligotrophic lake located within the Adirondack Park, New York, USA (43° 38’N, 74° 56’W). It has a mean depth of 15 m, a maximum depth of 39 m and is surrounded by a predominantly forested watershed with five first-order tributaries. Little Moose Lake supports a diverse fish community consisting of eleven native and four introduced species. Smallmouth Bass were introduced during the 1940s and quickly became established replacing native Lake Trout as the dominant top-predator in the lake. Surveys from spring and summer during the mid-1990s determined that Smallmouth Bass were the most abundant littoral fish species (Brown et al. 2000). Beginning in 2000, Smallmouth Bass were removed via electrofishing along the shoreline in spring and fall. This effort has resulted in the removal of nearly 100,000 individuals from the lake (Table 1). Observations indicate that the management efforts have been successful in increasing native littoral fish abundance. Additionally, a shift in the population size structure of the Smallmouth Bass was observed due to selective removal of larger fish by the gear used (Weidel et al. 2007). A diet shift was observed for Lake Trout in response to Smallmouth Bass removal from the consumption of primarily daphnids and benthic invertebrates to more prey fish (e.g., Rainbow Smelt (Osmerus mordax), Pumpkinseed Sunfish (Lepomis gibbosus)). The diet data were further supported by stable δ15N analysis indicating Lake Trout increased in trophic position (Lepak et al. 2006).

2.2 Fish and Water Sampling and Processing Methods

Lake Trout and Smallmouth Bass tissues, otoliths and stomach contents have been sampled and archived annually by Cornell University personnel at the Little Moose Field Station.
near Old Forge, NY. Smallmouth Bass samples were collected from 1999-2015 predominantly via boat electrofishing from May-June and September- November although some were captured during gill net and angling surveys from July-August. Lake Trout were collected from 2000-2015 using a combination of gill-netting, angling and boat electrofishing primarily during spring (May-June) surveys. Select forage fish species (i.e., Pumpkinseed Sunfish, Rainbow Smelt, Slimy Sculpin \((Cottus cognatus)\) collected via boat electrofishing in 2015 and burrowing mayfly larvae \((Hexagenia\) spp) obtained from Lake Trout gut samples were frozen for isotope and Hg analysis. Zooplankton samples were also collected during July and August of 2015 via a 163 µm mesh zooplankton net and were sorted for \(Daphnia\) spp. for comparison with data from previous sampling by Cornell in the early 2000s. Additionally, isotope and some THg data for these organisms from years 2001-2003 and for Lake Trout from years 2000-2007 were available from previous studies (Lepak et al. 2006 & 2009).

In the laboratory, total lengths (TL, mm) and wet weights (ww, g) were taken for individuals after sampling events. A small (~1 cm\(^3\)) portion of tissue was removed from below the dorsal fin, the skin was removed and the muscle tissue was placed into a 1.5 mL centrifuge tube. Samples were stored frozen at -5° C. Sagittal otoliths were also extracted and prepared by sectioning and mounting for interpretation of fish age for both predator species. A sub-sample of fish sampled each year of the study period were selected for THg analysis in the current study. The number of fish analyzed varied among years due to availability of samples (ranging from 4 to 28 individuals).

Diet analysis was completed for archived stomach contents obtained from fish after sampling events. The entire gut (buccal cavity to the pyloric caeca) was removed and stored in 80% ethanol for preservation. Contents were identified to the lowest possible taxon, counted, and
body lengths were measured for whole prey or estimated based on sizes of identifiable parts. For a limited number of samples, length data were not available so mean prey length for the organism in the diet dataset or median length for the length category of the diet item (in 50 mm increments) was used (e.g., 75 mm was used for a prey item in the 50 mm-100 mm length category). Stomach contents were converted into percent composition as dry biomass using length-mass regression relationships established in literature for specific organisms down to the species level when possible (i.e., Anderson & Neumann 1996; Benke et al. 1999; Methot et al. 2012; Sabo et al. 2002; Schneider et al. 2000). Diet compositions were sorted and categorized by year to observe changes in feeding habits over time.

Ancillary chemistry and in-situ measurements of water quality in Little Moose Lake have been completed annually since the early 2000s or earlier. Water samples were collected from the subsurface (approximately 0.5 m depth) above the deepest location of the lake during July and August. Additional water was collected for THg and MeHg determination in 2015 and 2016. The Hg samples were collected in 500-mL acid-washed polyethylene bottles after rinsing three times with sample water following USEPA trace-metal clean techniques method 1669 (USEPA 1995).

2.3 Analytical Methods

Stable isotope analysis was completed by Cornell University Stable Isotope Laboratory using a ThermoFinnigan Delta Plus mass spectrometer. Accuracy and precision of the stable isotope measurements (expressed in the standard per mil notation relative to V-PDB for δ\textsuperscript{13}C and atmospheric nitrogen for δ\textsuperscript{15}N) were verified by reference materials provided by the International Atomic Energy Association (IAEA). Daily precision of the instrument was verified by repeated analyses of internal laboratory standards during the sample runs. Stable isotope composition is
expressed in parts per thousand (‰ or ‘per mil’) as a deviation from a standard material denoted as ‘delta’ (δ): δ^{13}C or δ^{15}N = ([R_{sample}/ R_{standard}] - 1)·1000, where R = ^{13}C/^ {12}C or ^{15}N/^ {14}N.

Fish and invertebrate tissues were analyzed for THg using a Milestone DMA-80 (Direct Mercury Analyzer, Milestone, Monroe, CT, USA) in accordance with US EPA method 7473 (1998) at Syracuse University. Frozen archived samples were freeze-dried at -80°C and 0.080 mBar (FreeZone Type 6 plus freeze drier by Labconco) and homogenized by hand prior to analysis yielding THg concentrations on a dry tissue weight basis. Percent moisture of tissue samples were calculated from dry and wet mass measurements taken before and after freeze-drying. Samples were run in duplicate along with certified reference material Dorm 4 (dogfish muscle proteins; NRC Canada). Method and sample blanks and matrix spikes were analyzed in batches of 20 samples. Daily accuracy and precision of the instrument was also verified using certified reference materials Dolt 4 (dogfish liver; NRC Canada) and 2976 (mussel tissues; US NIST).

Concentrations of THg in unfiltered water samples were determined using oxidation, purge and trap, desorption and cold-vapor atomic fluorescence spectrometry following U.S. EPA method 1631, revision E (USEPA 2002) with a method detection limit of 0.2 ng·L^{-1}. MeHg in water samples were analyzed by distillation, ethylation, purge and trap, desorption and cold-vapor atomic fluorescence spectrometry per U.S. EPA Method 1630 (USEPA 2007) with a method detection limit of 0.02 ng·L^{-1}.

2.4 Data and Statistical Analyses

To evaluate temporal trends in fish Hg, THg concentrations were standardized for length due to the positive correlation often observed between fish length and Hg concentration (e.g., Scudder Eikenberry et al. 2015; Simonin et al. 2008). Linear regression analyses of the log of the Hg concentrations as a function of fish length were performed for each year of the study to
calculate the Hg concentration of a standard-length fish for that year. Residual differences between the calculated length-standardized Hg estimate and each individual fish Hg concentration for a given years’ data were calculated and added back into the overall annual standardized estimate. This approach allowed the spread of the data to be represented and incorporated unexplained variation back into the estimate (Eagles-Smith et al. 2016). For years with no meaningful length-Hg relationship (p>0.05), unadjusted Hg concentrations were used. Standard lengths of 480 mm for Lake Trout and 250 mm for Smallmouth Bass were selected to avoid extrapolating beyond observed values.

Estimates of trophic position were made for years where baseline information was available (2001-2002 from Lepak et al. 2006 and a recent survey in 2015) using the formula:

\[
\text{Trophic position}_{\text{consumer}} = \delta^{15}N_{\text{consumer}} - \delta^{15}N_{\text{baseline}} / 3.4 + \lambda
\]  
(1)

Where 3.4 ‰ represents the assumed trophic fractionation (Minagawa & Wada 1984) and \( \lambda \) represents the trophic position of the baseline organism (e.g., is 2 for primary consumers). It is important that \( \delta^{15}N_{\text{baseline}} \) values reflect the same habitat and time frame as the Trophic position\(_{\text{consumer}} \) value being estimated as variations in baseline stable isotope values are habitat and system-specific and potentially time dependent (Post 2002; Vander Zanden & Rasmussen 1999).

Data organization and statistical analyses were completed using Excel and R. THg concentrations are reported on a wet-weight basis and were calculated from dry weights using percent moisture contents determined for each sample when possible and mean moisture content for each species was used when individual values were not available (~30 samples of each species). Temporal trends in fish THg and water chemistry were assessed by calculating Kendall’s tau coefficients (ranging from -1 to 1) with a significance of \( \alpha=0.05 \).
Estimates of Hg mass associated with Smallmouth Bass tissues removed from Little Moose Lake were made for each year and cumulatively. Length standardized THg concentrations for a 250 mm fish were estimated each year. A log space length-weight regression equation was calculated using all Smallmouth Bass data to obtain a standard weight (in g) for a 250 mm fish which was used to estimate the mass of Hg removed per fish per year. Total mass of Hg removed per year and the total since 2000 could then be estimated based on the number of Smallmouth Bass removed each year.
3. **Results**

3.1 *Fish Mercury Trends*

A total of 179 Lake Trout (N=10-15 individuals per year) and 214 Smallmouth Bass (N=4-28 individuals per year) were analyzed for THg in this study. Samples included a range of fish lengths (360 mm to 640 mm with a mean of 481 mm ± 51 mm for Lake Trout; 46 mm to 412 mm with a mean of 207 mm ± 80 mm for Smallmouth Bass). Length-mercury relationships were significant for seven of the years for Lake Trout and for ten of the years for Smallmouth Bass. Lake Trout wet weight Hg concentrations ranged from 0.066 µg·g⁻¹ to 0.430 µg·g⁻¹, with a mean value of 0.219 µg·g⁻¹ ± 0.088 µg·g⁻¹ for the entire dataset. Smallmouth Bass wet weight Hg concentrations ranged from 0.021 µg·g⁻¹ to 0.604 µg·g⁻¹, with a mean value of 0.173 µg·g⁻¹ ± 0.110 µg·g⁻¹. Lake Trout THg concentrations standardized for length (480 mm) increased significantly over the period of this study (Figure 1 a.; tau= 0.419, p<0.001), while a decreasing trend in THg concentrations were observed for length-standardized Smallmouth Bass (250 mm; Figure 1 b.; tau= -0.172, p<0.001).
Figure 1. Wet-weight (ww) total mercury concentrations for length-standardized Lake Trout (a) and Smallmouth Bass (b) in Little Moose Lake, NY. Trend analysis identified a significant increase in 480 mm Lake Trout THg concentrations over time (tau= 0.419, p<0.001) and a significant decrease in 250 mm Smallmouth Bass THg concentrations over time (tau= -0.172, p<0.001). The dotted line represents the EPA human health criterion of 0.3 µg·g⁻¹ ww.

A total of 92,811 Smallmouth Bass were removed from Little Moose Lake from 2000 to 2015 with a minimum of 2,417 removed during 2008 and a maximum of 10,121 removed during 2001. A standard wet weight of 203 g was estimated for a 250 mm length bass using length-mass
regressions of the Smallmouth Bass size data in log space. Based on the annual estimates for a 250 mm length-standardized Smallmouth Bass THg concentration and the number of fish removed each year, mass of Hg associated with the Smallmouth Bass removed each year ranged from 0.05 g in 2010 to 0.56 g of Hg in 2001. An estimated total of 3.45 g of Hg associated with Smallmouth Bass has been removed from Little Moose Lake from 2000-2015. However, MeHg concentrates in white muscle tissues associated with the fillet (Bloom 1992) and the resulting values are likely overestimates as the edible fillets are approximately 33% of the fish’s mass (Ebert et al. 1993). Because correcting the values to reflect this would likely result in an underestimate of Hg removed, no adjustments to the values were made. Therefore, the calculations of Hg associated with the Smallmouth Bass removed are an estimation and values may be inflated (Table 1).

Table 1. Number of Smallmouth Bass and associated mass of THg removed each year from Little Moose Lake based on standardized THg concentrations and weights of 250 mm fish.

<table>
<thead>
<tr>
<th>Year</th>
<th>N SMB removed</th>
<th>Standardized [T-Hg (µg/g ww)] for 250 mm SMB</th>
<th>Standardized Weight of 250 mm SMB (g)</th>
<th>µg of Hg removed per fish per year</th>
<th>Total g of Hg removed per year</th>
</tr>
</thead>
<tbody>
<tr>
<td>2000</td>
<td>9490</td>
<td>0.213</td>
<td>203</td>
<td>43</td>
<td>0.41</td>
</tr>
<tr>
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<td>10121</td>
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<td>55</td>
<td>0.56</td>
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<tr>
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<td>203</td>
<td>40</td>
<td>0.35</td>
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<td>8917</td>
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<td>203</td>
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<td>203</td>
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<td>0.37</td>
</tr>
</tbody>
</table>

Total as of 2015: 92811 3.45
3.2 Growth and Diet

Growth for each species was evaluated using age data over two time intervals: pre-bass removal (year 2000 and before) and post-bass removal (2005 and after). Age data from years 2001-2004 were excluded from the growth comparisons to allow for a stabilization period. Von Bertalanffy growth curves (Ricker 1975) were constructed using lengths and ages at capture for each species during the pre- and post-removal intervals (Figure 2).

Figure 2. Lake Trout (a) and Smallmouth Bass (b) total length (mm) at age of capture pre- (red triangles and lines) and post- (blue circles and lines) Smallmouth Bass removal in Little Moose Lake. The lines represent Von Bertalanffy growth trajectories during the pre- and post-removal conditions.
Smallmouth bass diets analyzed included a total of 1399 individuals from years 2005-2014 (Figure 3b & 3d) with annual sample sizes ranging from 5 (2014) to 352 (2007). Of the Smallmouth Bass samples analyzed for diet, 297 individuals had empty stomachs. Overall, crayfish (*Cambarus* and *Orconectes* spp.) were the largest percent composition of smallmouth diets comprising an estimated 82.4% of the diet dry mass for all samples. Fish comprised 16.8% of all smallmouth diets during the study period. The other generalized categories represented small contributions by mass and included aquatic insects (0.74%), terrestrial invertebrates (0.057%; e.g. Hymenoptera, Formicidae) and other crustaceans (0.003%; e.g., Isopoda and Amphipoda). The percent composition of crayfish in Smallmouth Bass diets ranged from 33.2% in 2013 to 94.3% in 2006. The percent composition of fish in Smallmouth Bass diets ranged from 4.8% in 2006 to 64.2% in 2013. Slimy Sculpin and Rainbow Smelt dominated the fish components of Smallmouth Bass diets for all years with data.

Lake Trout diets were analyzed for a total of 127 individuals from years 2000-2010 (Figure 3a & 3c) with annual sample sizes ranging from 4 (2010) to 27 (2008). Of the Lake Trout samples analyzed for diet, 18 individuals had empty stomachs. Overall, fish were the largest component of Lake Trout diets comprising 92.7% of the diet mass over the sampling period, 63.1% of which were other Lake Trout. Other prey fish that were important components in Lake Trout diet samples were Centrachidae spp. (20.7%; i.e., Smallmouth Bass, Pumpkinseed Sunfish) and Rainbow Smelt (10.8%). Other prey fish included Salmonidae spp. (3.7%; i.e., Rainbow Trout (*Oncorhynchus mykiss*), Atlantic Salmon (*Salmo salar*)), Slimy Sculpin (1.2%) as well as unidentifiable fish (0.6%). Crayfish and other invertebrates were smaller components of Lake Trout diets overall comprising 5.7% and 1.7% of diets over the sampling period, respectively, with chironomids, *Hexagenia* spp. and cladoceran zooplankton the most abundant.
invertebrates after crayfish. During the year 2000, diet for Lake Trout consisted of 100% ‘other’ consisting primarily of *Hexagenia* spp. and chironomid larvae. A shift to 47.8% fish and 52.2% ‘other’ was observed in 2001. Fish dominated Lake Trout diet after 2001 with percent compositions as fish ranging from 66.3% in 2005 to 99.9% in 2010. Dominant fish species in Lake Trout diet varied from year to year with Slimy Sculpin (2001), Rainbow Smelt (2002 and 2010), centrachids (2006 and 2008) and other Lake Trout (2005 and 2007) comprising the majorities for each year with data.

**Figure 3.** Percent compositions of Lake Trout (a & c) and Smallmouth Bass (b & d) diets by year from 2000-2010 for Lake Trout (except for 2003-2004) and from 2005-2014 for Smallmouth Bass. Prey were categorized into large broad groups (a & b) in addition to percent compositions of the fish dietary components (c & d).
3.3 Prey Fish Mercury and Stable Isotope Results

In addition to the top predator species, select lower trophic level organisms that were sampled and analyzed in 2001-2002 (data from Lepak et al. 2006) were sampled in 2015 and analyzed for stable isotope ratios and THg concentrations for comparison (Table 2).

Table 2. Mean total lengths (TL), wet weight THg concentrations, stable nitrogen (δ15N) and carbon (δ13C) isotope ratios (±1 S.D.) and estimates of trophic position for select organisms within the Little Moose Lake food web sampled during 2001-2002 and 2015.

<table>
<thead>
<tr>
<th>Organism</th>
<th>2001-2002</th>
<th>2015</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>N</td>
<td>Mean TL (mm)</td>
</tr>
<tr>
<td>----------------------------</td>
<td>---</td>
<td>---------------</td>
</tr>
<tr>
<td>Lake Trout</td>
<td>30</td>
<td>455</td>
</tr>
<tr>
<td>Smallmouth Bass</td>
<td>15</td>
<td>277</td>
</tr>
<tr>
<td>Slimy Sculpin</td>
<td>3</td>
<td>75</td>
</tr>
<tr>
<td>Rainbow Stickle</td>
<td>5</td>
<td>105</td>
</tr>
<tr>
<td>Pumpkinseed Sunfish</td>
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</tr>
<tr>
<td>Hexagenia Spp.</td>
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<td>30</td>
</tr>
<tr>
<td>Daphnia Spp.</td>
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<td>NA</td>
</tr>
<tr>
<td></td>
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</tr>
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<td></td>
<td>10</td>
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</tr>
<tr>
<td></td>
<td>10</td>
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<tr>
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<td>7</td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>-</td>
</tr>
</tbody>
</table>

(*) reported THg values for different samples than those analyzed for stable isotope ratios
(-) no data available

Except for Daphnia spp., 2015 δ13C ratios were more depleted (more negative) than 2001-2002 δ13C ratios for all organisms. Decreases in δ15N ratios were also observed between the 2001-2002 and 2015 samples for all organisms except for Hexagenia spp. and Daphnia spp. (Figure 4).
Figure 4. Stable nitrogen ($\delta^{15}$N) and carbon ($\delta^{13}$C) isotope ratios (±1 S.D.) for select organisms within the Little Moose Lake food web sampled during 2001-2002 (triangles) and 2015 (circles).

During the 2001-2002 period, mean (±1 S.D.) Lake Trout trophic position was 3.3 ± 0.2 (where $\lambda=2$ for *Daphnia* with a mean 2000s $\delta^{15}$N$_{baseline}$= 7.5‰; Equation 1). Mean Smallmouth Bass trophic position was also estimated to be 3.3 ± 0.1 for the early 2000s period (where $\lambda=3$ for Pumpkinseed Sunfish with a mean 2000s $\delta^{15}$N$_{baseline}$= 9.7‰; Equation 1). During the 2015 period, mean Lake Trout trophic position increased to 5.0 ± 0.1 (where $\lambda=2$ for *Daphnia* with a mean 2015 $\delta^{15}$N$_{baseline}$= 1.0‰; Equation 1) and Smallmouth Bass trophic position remained at 3.3 ± 0.2 (where $\lambda=3$ for Pumpkinseed Sunfish with a mean 2015 $\delta^{15}$N$_{baseline}$= 7.8‰; Equation 1; Table 2).

Trend analysis using Kendall’s tau indicated declining trends in Lake Trout (tau= -0.305; p-value<0.001) and Smallmouth Bass (tau= -0.482; p-value<0.001) $\delta^{15}$N signatures occurred.
over time. Significant decreases in Smallmouth Bass $\delta^{13}$C signatures were also observed over time (tau= -0.306, p<0.001), while no trend was observed in Lake Trout $\delta^{13}$C over time (tau= -0.077, p=0.13; Figure 5).

**Figure 5.** Stable nitrogen (a & b) and carbon (c & d) isotope ratios for Lake Trout (a & c) and Smallmouth Bass (b & d) over time in Little Moose Lake, NY. Trend analysis identified significant decreases in $\delta^{15}$N for both species over time (tau= -0.305 for Lake Trout; tau= -0.482 for Smallmouth Bass; both p-values<0.001) and a significant decrease in Smallmouth Bass $\delta^{13}$C over time (tau= -0.306, p<0.001) while no trend was observed in Lake Trout $\delta^{13}$C.

### 3.4 Little Moose Lake Water Chemistry

The mean surface temperature of Little Moose Lake was 20.1 °C ± 3.2 °C from 1979-2012 and lake temperature profiles did not change significantly over time (p-value= 0.6). Total phosphorus (mean ±1 S.D. = 4.7 µg P·L$^{-1}$ ± 2.7 µg P·L$^{-1}$) and chlorophyll a (1.3 µg·L$^{-1}$ ± 0.9 µg·L$^{-1}$) measurements from 1998-2012 indicated that productivity in the lake also did not change
significantly (p-values= 0.8 and 0.6, respectively). Declining trends in sulfate (tau= -0.26; p-value= 0.01) and nitrate (tau= -0.186; p-value= 0.08) concentrations were observed from 2003-2012 and are consistent with declines in emissions and deposition of these two anions in the Adirondack region (Waller et al. 2012; Driscoll et al. 2016). However, no changes were observed in mean ANC (149 μeq·L⁻¹ ± 39 μeq·L⁻¹; p-value= 0.6) or pH (6.7 ± 0.5; p-value= 0.4) indicating that acid-base chemistry of the lake is not changing and impacts from acid precipitation are not as important in Little Moose compared to other Adirondack Lakes due to high ANC. Concentrations of dissolved organic carbon increased significantly from 2003-2012 (tau=0.31; p-value= 0.002), although concentrations remain relatively low (2.3 mg C·L⁻¹ ± 0.7 mg C·L⁻¹). Mean Secchi depth did not change over the same time interval (7.3 m ± 1.8 m; p-value=0.9).

The analyses of THg and MeHg in Little Moose Lake surface waters yielded values that were below the analytical detection limits (<0.02 ng·L⁻¹ MeHg and <0.2 ng·L⁻¹ THg) for repeated analytical and field duplicate and replicate analyses.
4. Discussion

4.1 Conceptual Model of Role of Smallmouth Bass Removal in Fish Mercury and Trophic Dynamics

Several internal and external factors have likely contributed to the decreasing trends in THg concentrations for Smallmouth Bass, and the simultaneous increase in Lake Trout THg concentrations in Little Moose Lake since the Smallmouth Bass removal was initiated in 2000. The key drivers of these changes are likely independent of changes in Hg and acid deposition and lake chemistry due to the contrasting trends for the two species within the same ecosystem, indicating food web dynamics are at play and altering Hg bioaccumulation in these species differently.

After a 5-year stabilization period following the initiation of the Smallmouth Bass removal program (after 2005), Lake Trout growth increased and larger individuals were observed at the same age classes compared with the smaller size of individuals prior to the bass removal. It has been hypothesized that increased fish growth can lead to “growth dilution” whereby increased growth rates due to consumption of higher quality prey result in decreases in Hg concentrations in fish (Verta 1990). The increases in Lake Trout growth and THg in this study are consistent with earlier patterns observed by Lepak et al. (2009) where increases in Lake Trout Hg was attributed to consumption of higher MeHg content of prey post-removal (prey fish), regardless of increased growth. The diet data from this study were also consistent with observations by Lepak et al. (2009). Lake Trout have continued to consume mostly fish post-removal, which made up the largest components of their diet after 2001. This pattern was partially due to increased consumption of littoral prey fish by Lake Trout likely resulting from an increase in littoral prey fish abundance after Smallmouth Bass removal (Weidel et al. 2007) as well as increased consumption of non-native Rainbow Smelt whose populations increased
between 2005-2007 in Little Moose Lake (D. Josephson, unpublished data). The impacts of smelt invasion on Hg bioaccumulation in native predators in other smelt-invaded lakes have been variable and thought to be mediated by increases in growth rates of piscivores due to switching to a smelt diet (Johnston et al. 2003).

Growth patterns of Smallmouth Bass also changed after the post-removal stabilization period, with a greater number of larger individuals within a given age class compared with pre-removal bass which were smaller at comparable age classes (i.e., growth increased). There is also evidence of compensatory recruitment consistent with observations by Weidel et al. (2007) and a population size structure shift with fewer fish older than age class four. This pattern could be a direct manifestation of the removal because electrofishing preferentially selects for larger individuals. In contrast to Lake Trout, increases in Smallmouth Bass growth coupled with decreases in THg concentrations post-removal may reflect the growth dilution phenomena. Diet compositions for Smallmouth Bass were primarily crayfish from 2005-2014. Increased Smallmouth Bass growth was observed with little corresponding changes in prey consumption which could result in decreased overall dietary intake of MeHg per unit of tissue growth thereby resulting in lower MeHg concentrations in the tissues of the faster growing, post-removal individuals. This conclusion is based on the premise that MeHg content of crayfish and other prey organisms did not change over the removal period and is therefore speculative as these data are not available.

Changes in δ¹³C and δ¹⁵N between the early 2000s and 2015 were consistent for most of the Little Moose Lake organisms sampled with both δ¹³C and δ¹⁵N becoming more depleted. The exceptions to these patterns are likely driven by small sample sizes for Hexagenia in the earlier survey (N=1) and Daphnia in the recent survey (N=2; Figure 4). Except for Smallmouth Bass
$\delta^{13}C$ signatures, decreasing temporal trends in stable isotope ratios were also observed over the entire sampling period of this study for the top predator fish (Figure 5). These observations could indicate an overall shift in baseline $\delta^{13}C$ and $\delta^{15}N$.

The baseline-adjusted estimates of trophic position for the two top predators indicated trophic position increased from the early 2000s to 2015 by nearly two trophic levels (from 3.3 to 5.0) for Lake Trout, but remained constant for Smallmouth Bass (3.3). These data are consistent with the fish THg concentration trends and diet data and support the notion that Lake Trout are consuming higher trophic level prey and therefore have increased MeHg intake and bioaccumulation regardless of increased growth as well as the possibility of growth dilution occurring for Smallmouth Bass. The 2015 estimate of trophic position for Lake Trout may be inflated due to occurrences of cannibalism resulting in a trophic position >4. However, the lack of lower trophic level organism stable isotope data between the two time periods make inferences about changes in trophic position over time difficult. Adjusting for trophic position changes are thought to be important to understanding rates of change in contaminant concentrations (Hebert & Weseloh 2006). These results are also consistent with earlier findings from Little Moose Lake by Lepak et al. (2006) who observed that Lake Trout trophic position increased from 2000-2002. Estimates of trophic position from 2005-2007 (Lepak et al. 2009) indicated the initial increase in trophic position did not continue through time, however changes in baseline signatures from 2000-2002 to 2005-2007 were not evaluated and may have resulted in underestimations of Lake Trout trophic position relative to decreases in baseline $\delta^{15}N$ signatures. The authors also reported increased (less depleted) Lake Trout $\delta^{13}C$ signatures indicating increased utilization of littoral prey after 2000. Data from 2005-2007 and 2015 support that this pattern did not continue, however. Changes in $\delta^{13}C$ signatures of lower trophic
level organisms were observed between the 2001-2002 and 2015 surveys and changes in Lake Trout $\delta^{13}$C may be occurring due to changing baseline signatures.

4.2 Management and Monitoring Program Implications

The results from this study have important implications for fish population and Hg management. Consistent with observations by Gothberg (1983), intensive electrofishing and Smallmouth Bass removal in Little Moose Lake has resulted in decreased THg concentrations in the Smallmouth Bass remaining in the lake. However, reduced competition with Smallmouth Bass resulted in consumption of prey at higher trophic levels and therefore higher MeHg concentrations in Lake Trout. Although growth for Lake Trout increased after the Smallmouth Bass removal began, the slow-growing and long-lived characteristics of Lake Trout have resulted in increased Hg biomagnification with current levels of Lake Trout Hg approaching or exceeding the EPA human health criterion of 0.3 µg·g$^{-1}$ ww.

These results also have important implications for spatial and temporal monitoring of fish Hg concentrations, especially for a region considered a biological Hg hotspot such as the Adirondacks where decisions based on monitoring may be more significant than other, less Hg impacted regions. It is important to understand how changes in food web structure on fish Hg biomagnification (and how these may affect species differently) differ from abiotic factors in their influence on fish Hg trends to provide sound interpretation of data for a given lake or region. Knowledge of food web structure may help explain specific cases when trends in fish Hg concentrations are not consistent with trends in Hg deposition or changes in lake physio-chemical variables that drive trends in fish Hg concentrations. Frequent collection of lower trophic level organisms is needed to provide good baseline information for interpreting stable $\delta^{13}$C and $\delta^{15}$N signatures of predators and is crucial to understanding food web dynamics in a
system being altered. Based on the results from this study, it appears baseline $\delta^{13}$C and $\delta^{15}$N signatures may be changing in Little Moose Lake. Sabo et al. (2016) observed decreases in $\delta^{15}$N in tree rings from a western Adirondack watershed. This may reflect changes in nitrogen and carbon cycling within the ecosystem as lake nitrate and DOC concentrations were both observed to decrease over the study period. Decreases in nitrate deposition and climate change have been hypothesized to cause tightening of the watershed nitrogen cycle (Duran et al. 2016; Sabo et al. 2016), although this notion needs to be further explored in aquatic ecosystems.
5. Conclusions

Temporal Hg trends in Little Moose Lake top-predators were strongly related to changes in food-web dynamics over the last 15 years resulting from the removal of thousands of Smallmouth Bass annually. Decreasing THg concentrations were observed for Smallmouth Bass while concentrations in Lake Trout increased. Growth rates for both Smallmouth Bass and Lake Trout increased and Lake Trout diet distinctly changed since the removal began in 2000. Therefore, it is likely that disparate factors are driving the observed trends for each species. Additionally, changes in stable nitrogen (δ\textsuperscript{15}N) and carbon (δ\textsuperscript{13}C) isotope signatures were observed in lower trophic level organisms from the early 2000s to 2015 and decreases in the isotope ratios were observed for the top-predators over the entire time series which has implications for shifting baseline signatures and changes in the availability of \textsuperscript{15}N and \textsuperscript{13}C within the ecosystem.

These observations have implications for fisheries management and fish consumption advisories as removal of the non-native bass resulted in reduced concentrations of Hg in the bass but indirectly contributed to increases in Lake Trout Hg concentrations over time. There is inherent variability in fish Hg concentrations that can depend on many factors. Therefore, understanding processes in freshwater ecosystems that affect the bioaccumulation of MeHg in top-predator fish species are essential to consider from a public policy perspective. The story of Little Moose Lake in the current study demonstrates how understanding management impacts on food web dynamics can provide relevant and valuable information regarding Hg bioaccumulation needed to provide comprehensive and meaningful monitoring strategies for freshwater ecosystems.
6. References


EDUCATION
SYRACUSE UNIVERSITY COLLEGE OF ENGINEERING AND COMPUTER
SCIENCE – Syracuse, NY
M.S. in Environmental Engineering Science August 2017; GPA-3.83

SUNY COLLEGE OF ENVIRONMENTAL SCIENCE AND FORESTRY (SUNY-ESF) –
Syracuse, NY
B.S. in Aquatic and Fisheries Science & minor in Water Resources received December 2012;
GPA- 3.52, Magna Cum Laude
Honors & Activities: Distinguished Biology Scholar Award for Aquatic and Fisheries Science,
2013; 1st Place for Undergraduate Poster at SUNY-ESF Spotlight on Student Research &
Outreach, 2013; Competitor for the SUNY-ESF Woodsman Team; Student member of the
American Fisheries Society; Volunteer at Carpenter’s Brook Fish Hatchery

WORK EXPERIENCE
Syracuse University, Dept. of Civil and Environmental Engineering Syracuse, NY
Laboratory Assistant and Project Coordinator March 2014 – June 2017
♦ Organized sampling and managed databases for NYS Energy and Resource Development
Authority (NYSERDA) funded project monitoring spatial patterns and temporal trends in
mercury concentrations of New York State sportfish.
♦ Coordinated and performed laboratory analyses for mercury-related research projects
including training of students and technicians, equipment scheduling, reviewing data and
quality control and updating standard operating procedures.
♦ Regularly interpreted, distilled and presented research findings in various professional
settings.

SUNY-ESF, Thousand Islands Biological Station Clayton & Syracuse, NY
Lead Research Technician April - October 2013
♦ Monitored fish populations of the Upper St. Lawrence River using trap nets, seine nets and
juvenile traps focusing on northern pike and muskellunge reproduction and recruitment.
♦ Completed vegetation surveys in coastal bays and wetlands to assess fish habitat and the
encroachment of the hybrid cattail Typha x glauca.
♦ Sampled and analyzed water chemistry in the laboratory and lower trophic level interactions
in wetlands.
♦ Lead, managed and instructed crews of students and technicians in the field and the lab
including boat operation/navigation, sampling techniques, and identification of fish,
invertebrates and vegetation.

Research Analyst January- March 2013
♦ Completed analysis of fish stomach contents using a dissecting microscope in the laboratory.
♦ Assisted with winter muskrat surveys in wetlands of the Upper St. Lawrence region
involving macrophyte identification, navigation using GPS and long hours outdoors.
Undergraduate Research Fellow  Summer 2012
  • Developed and conducted an independent research project studying the diet, distribution and isotopic signatures of the invasive round goby in various embayment and deep water habitats of the upper St. Lawrence River.
  • Presented results to the SUNY Board of Directors and at the SUNY-ESF Spotlight on Student Research & Outreach.

Hutton Junior Fisheries Biology Scholar (American Fisheries Society)  Summer 2008
  • Assisted with field and laboratory work completing limnological surveys, wetland vegetation surveys, submerged aquatic vegetation surveys and fish population surveys using various sampling techniques and specialized equipment including trap nets, seine nets, GPS and boat operation.

National Science Foundation  Arima, Trinidad & Tobago  Summer 2011
  • Assisted with field and lab work including bimonthly biomass surveys and mark/recapture procedures for Poecilia reticulata.
  • Participated in weekly meetings concerning micro-evolution and the factors involved in specific ecosystem responses.

PUBLICATIONS AND PRESENTATIONS

SPECIAL SKILLS AND INTERESTS
  • Microsoft Office; ArcMap GIS; R Studio; MatLab; NYS Boater & Hunter Safety Certified
  • Playing guitar/writing music; Cooking; Fishing; Boating/canoeing; Hiking