A comparison of mercury exposure and risk between artificial impoundments and natural lakes measured in Common Loons and their prey, 1996-97

1997 Field Season Report

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Executive Summary

In 1996, Central Maine Power (CMP) and Union Water Power Company (UWP) initiated a study of mercury (Hg) availability on their hydroelectric and storage reservoirs as part of the Federal Energy Regulatory Commission (FERC) relicensing process. Several regulatory agencies, such as the Maine Department of Environmental Protection (MDEP) and U.S. Fish and Wildlife Service (USFWS), and non-governmental organizations (e.g., Loon Preservation Committee (LPC) and Maine Audubon Society (MAS) expressed interest in determining Hg availability in reservoirs. BioDiversity Research Institute (BRI) designed a study to compare Hg concentrations in environmental biotic compartments between artificial impoundments (reservoirs) and natural lakes. The purpose of this study as outlined in the 1996 proposal was "to determine the exposure of mercury, other heavy metals, and organochlorines (including total PCBs) to Common Loons *Gavia immer* in the study area and identify potential effects on individual survival, behavior, and reproductive success." In 1997, CMP continued their funding and participation in the collaborative study. The primary focus of the 1997 study was to "increase the sample size for better statistical validation and investigate further whether there is a relationship between fluctuating water levels and Hg exposure."

U.S. Environmental Protection Agency (U.S. EPA) models have predicted mercury deposition in the northeastern Unites States to be at the highest levels in the country (U.S. EPA 1996). Since 1991, researchers with BRI have collected loon blood and feather tissue for contaminant analyses from over 1,600 breeding loons in five regions across North America from Alaska to Nova Scotia. BRI's results agree with the U.S. EPA Hg deposition models indicating a west-east geographic pattern of increasing exposure to Hg (Evers et al. 1998). Results from the 1994-95 Regional Environmental Monitoring Assessment Program (REMAP) study also show fish Hg concentrations at or above levels considered safe for human and wildlife consumption across Maine (Stafford and Haines 1996).

In 1998, representatives of New England and eastern Canada air, water, waste, and public health divisions compiled a technical and policy information document (NESCAUM 1998). This broad based workgroup recommended that, "Additional research is needed to assess the ecosystem impacts of mercury in the Northeast, particularly with respect to fish-eating wildlife." Similarly, Maine's Land and Water Resources Council (1997) recommended "additional research should be undertaken" in "determining whether adverse health effects are occurring in Maine fish and wildlife."

Although the REMAP fish results indicate widespread and elevated environmental Hg exposure, they provide limited information toward an understanding of the availability of Hg, particularly methylmercury (MeHg). In general, lower trophic level, short-lived biota are less sensitive to MeHg than birds that are higher on the food chain and longer lived. The subtle neurological effects of MeHg may not impact an individual until concentrations attain levels that are reached through bioaccumulation and biomagnification. For example, the U.S. EPA (1996) predicts a five-fold increase in MeHg exposure between forage fish (trophic level 3) and predatory fish (trophic level 4). Therefore, we evaluated MeHg availability and potential impacts on piscivorous birds.

By using these high trophic level organisms and BRI's broad geographic database we were better able to understand and interpret Hg concentrations in Common Loons. The relationship of MeHg production and availability with widely fluctuating water levels is an issue being addressed by BRI in association with the Northeast Loon Study Working Group (NELSWG). Studies funded by the USFWS from 1994-95 at Lake Umbagog National Wildlife Refuge found Common Loon blood and feather Hg levels to be elevated. These findings spurred the formation of NELSWG. This group consists of professionals from private industry, state and federal agencies, non-profit organizations, and universities. NELSWG has generated thousands of volunteer hours, including tremendous assistance by the USFWS and CMP with personnel (over 2,000 hours in 1996-97), equipment, logistics and lab analysis support.

There are many factors that affect the production and bioavailability of MeHg in waterbodies. Therefore, the interpretation of Hg exposure in loons and fish is complicated. The grouping of waterbodies for comparative purposes was placed in context with (1) hydrological factors such annual and summer water level fluctuations and flushing rates, (2) physical features such as watershed and shoreline acreage, surrounding terrain, and habitat types, (3) geochemical factors such as water chemistry and temperature, soil and parent substrate types, and (4) other factors such as age of trophic structure, water body, peat pressing, episodic events, and temporal trends. In addition, Hg levels in birds are affected by age, size, gender, and trophic forage level. The effects of Hg may be subtle and complicated to quantify; for example, elevated Hg may increase hyperactivity that alters loon incubation behavior and results in decreased hatching success. Parallel high resolution monitoring is required to distinguish contributions by variables, such as predation and weather, from actual Hg effects.

In 1996-97, data on Hg distribution and MeHg availability in reservoirs and natural lakes were collected. The prey sampling strategy was based on a Common Loon feeding selection study conducted by Barr (1996). In 1996, we targeted four size classes of Yellow Perch (*Perca flavescens*) and three size classes of White Sucker (*Catostomus commersoni*), both species preferentially chosen by loons. In 1997, Yellow Perch were emphasized because we found a strong relationship between perch Hg and adult loon blood Hg (Evers and Reaman 1997). On lakes where Yellow Perch were not present we collected three size classes of minnows and other species that were available to adult and juvenile loons. Whole fish (all organs and muscle tissue) composites were analyzed for total Hg in 1996, and both total and MeHg in 1997.

Every study lake (reservoir and natural) with the exception of Indian Pond had at least one perch size class with Hg concentrations above the level determined by Barr to cause reproductive harm to Common Loons. Barr found that loons feeding on fish with Hg concentrations of 0.30 ppm or higher produced fewer chicks than loons feeding on less contaminated fish. Loons that fed on fish with Hg concentrations of 0.40 ppm or higher did not produce young.

Flagstaff Lake had the highest level of Hg in all size classes of Yellow Perch and they were significantly higher than the other 7 lakes sampled. White Sucker composites had significantly lower Hg concentrations than similar-sized Yellow Perch. White Sucker composites from three reservoirs (Flagstaff, Aziscohos, and Richardson) and two natural lakes (Little Beaver and East Richardson Pond) had Hg levels > 0.30 ppm. White Sucker Hg levels did correlate with Yellow Perch Hg levels. Several species of minnows had high levels of Hg (>0.30 ppm) particularly Blacknose Dace (*Rhinichthys atratulus*) and Northern Redbelly Dace

(*Phoxinus eos*) in the small class, Golden Shiner (*Notemigonus crysoleucas*) in the medium class, and Fallfish (*Semotilus corporalis*) in the large class.

We found Yellow Perch Hg levels to correlate strongly with adult loon blood Hg concentrations, particularly medium sized fish composites ($p<0.001$). This suggests that Yellow Perch may be a significant source of the adult's diet in our study area and the loons are likely to assimilate much of their Hg from perch. Juvenile loon blood Hg did not correlate with Yellow Perch Hg ($p>0.05$), and adult and juvenile loon blood Hg did not correlate with White Sucker Hg ($p>0.05$). Therefore a mix of prey items probably accounts for juvenile Hg levels and White Sucker is a less important contributor than perch to adult blood Hg concentrations.

We found mean Hg concentrations in Yellow Perch and White Sucker to increase with fish size. In all our study regions, adult male loons had higher Hg concentrations than females in blood and feathers. Males in New England average 30% heavier than females and may be foraging on larger prey items. Juveniles feed on smaller forage items than adults but may receive proportionally higher amounts of Hg, particularly if they are feeding on minnow species with elevated Hg concentrations.

Common Loon blood, feather, and egg Hg concentrations varied considerably among all waterbodies. We designated each of the 9 reservoirs and 46 natural lakes into the following 5 categories based on hydrology:

- RSR - regulated storage or hydroelectric reservoirs with annual fluctuations > 1.5 m (5 feet),
- RPR - regulated "peak" hydroelectric reservoirs with weekly water fluctuation levels < 1 m (2-3 feet),
- RFP - regulated "full pond" reservoirs and raised lakes managed for minimal water level fluctuations < 1.5 m (5 feet),
- FNL - "flashy" natural lakes with annual water fluctuations > 1.5 m (5 feet)
- SNL - stable natural lakes with annual water fluctuations < 1.5 m (5 feet)

Analysis of adult and juvenile blood Hg levels within these categories did show patterns of Hg exposure. Variability in blood Hg concentrations was greater in adults (due to the age factor) and therefore only RSRs significantly differed from our SNL benchmark. The juvenile blood Hg index was less variable and showed that RSRs and FNLs are significantly greater than both FRPs and SNLs. These categorical data indicate water bodies that have water level changes greater than 1.5 meters are more likely to have higher MeHg availability than waterbodies with fluctuations less than 1.5 meters.

A Pearson Correlation Coefficient matrix was then developed with 14 lake morphometric and water chemistry variables to determine the factors most significant to predicting loon blood Hg levels. Nine factors were then selected and placed in a stepwise multiple regression analysis to explain the variability of adult and juvenile blood Hg. Simple regression analysis was then used for the factors best explaining the variability to show the strength of the relationship. We found that annual and summer water level fluctuations were the most important factor measured in this analysis and were significantly correlated with adult and juvenile blood Hg levels.

Limitations to these analyses are the number of lakes with complete morphometric and water chemistry data sets and lack of some parameters paramount to understanding Hg distribution and availability (e.g., watershed inputs and trophic structure). Nevertheless, lakes

with high water level fluctuations held a common pattern for enhanced MeHg availability whether they were FNLs (e.g., Chain of Ponds and Little Lobster) or RSRs. Not all RSRs have similar origins and we further showed that those originating from river valleys (e.g., Aziscohos and Flagstaff) had higher loon blood Hg levels than those originating from existing lakes (e.g., Mooselookmeguntic, Richardson, and Umbagog).

The risk Hg poses to loons (and other wildlife) was rated according to the number of matrices exceeding designated thresholds. FNLs were rated the highest (33%) followed by RSRs (29%), RPRs (25%), SNLs (23%), and RFPs (14%). RFPs exhibit the least water level change of all lake types while FNLs have the greatest. Risk within RSRs had the greatest variability but when it was broken down into reservoir origin, those from river valleys had an 80% risk rating and those from existing lakes rated 20%. We therefore recommend that when examining the risk of Hg enhancement in a waterbody that its hydrology and origin be considered.

A growing component of this study includes the investigation of potential Hg effects on loons and other wildlife. Preliminary information on Hg risk levels and the toxic impact on the loon's well being, productivity, and individual performance indicates problems. From a sample of 50 Maine lakes, we found 50% having enough MeHg to place individual loons at risk. Seven of the nine target reservoirs have elevated Hg levels that place them as a risk to aquatic wildlife. Whether this risk effects the breeding populations of loons and other wildlife is not yet known.

Background

The element mercury (Hg) has become an issue of concern in Maine in recent years, leading to a statewide fish consumption advisory. Hg is classified as a heavy metal and has been demonstrated to affect reproduction, behavior, and survival of humans and wildlife. Environmental Hg is a naturally occurring element in our landscape, especially in aquatic ecosystems. However, analyses of lake sediment cores indicate that the current rate of regional Hg deposition are 2-5x greater than historical levels (pre-1940s) (Swain et al. 1992, Haines pers. com.). Scientists have traced this Hg increase to dry and wet atmospheric particulate fallout. Atmospheric Hg primarily originates from coal burning and incinerator emissions. Studies comparing fish Hg concentrations with rates of atmospheric deposition have found that these anthropogenic sources account for much of the aquatic system load (Rada et al. 1989, Fitzgerald 1995, Rudd 1995). Inorganic Hg is eventually converted by bacteria into a more toxic organic form — methylmercury (MeHg). MeHg is then assimilated by phytoplankton and zooplankton. It accumulates over time and biomagnifies through the food chain in larger trophic level animals, from zooplankton and invertebrates to fish. Loons, osprey and eagles top this mercurial pyramid. Loons consume approximately two pounds of fish a day. Mercury can be toxic, but determining its effects on wildlife populations is difficult. A challenge to researchers is identifying the threshold at which Hg impairs the function of high trophic level foragers such as loons and humans.

Increases in Hg are of concern owing to the potentially small margin of safety between background levels of exposure and concentrations that can harm humans and other organisms (Michigan Environmental Science Board 1993). Mercury is second only to dioxin as an environmental hazard (per unit emitted to the atmosphere) (Pratt et al. 1993). Additionally, most

of the anthropogenic Hg deposited in a watershed is retained in the soil, water, and biota for over a decade (Lindqvist and Rodhoe 1991) and therefore has the potential to persist as a toxin. Although global inputs and natural removal mechanisms are not fully understood, annual increases of global atmospheric Hg concentrations may be 1% (Slemr and Langer 1992).

Numerous studies have investigated negative effects of environmental Hg on fish (e.g., Hoffman and Moore 1979, Eisler 1987, Wiener and Spry 1996). Recently, the U.S. EPA (1996) reported "a plausible link between methylmercury contamination in freshwater fish and anthropogenic mercury emissions" and "concentrations of mercury in the tissues of wildlife species have been reported at levels associated with adverse health effects in laboratory studies in the same species." Few studies, however, have measured Hg concentrations in a high trophic level wildlife species.

We identified the Common Loon to be one of the best indicators for investigating Hg exposure because it (1) can be reliably captured and recaptured, (2) is easily monitored and observed throughout the breeding season, (3) exhibits high fidelity to a breeding area (4) can tolerate moderate levels of human disturbance, and (5) is highly sensitive to the bioaccumulation of MeHg (6% each year - Evers et al. 1996). Using loons to assess risk of environmental Hg exposure and effects is supported by their (1) habitat preference — lentic systems that serve as endpoint repositories of atmospheric Hg for a watershed, (2) forage base — upper trophic levels organisms such as piscivorous fish, (3) top trophic structure position, (4) elaborate social, reproductive, and behavioral demands that can be possibly altered by neurological imbalances induced by contaminant interferences, (5) lifespan —possibly over 30 years, thereby providing ample bioaccumulation opportunities, and (6) prior reported reproductive impairment associated with fish Hg levels (Barr 1986).

Few studies have been conducted on the five target species (Common Loon, Bald Eagle, Osprey, Belted Kingfisher, and Common Merganser) to determine when Hg effects their survival, reproductive success and overall well being. Several studies have provided some avian references for no adverse effect level (NOAEL) and lowest observed adverse effect levels (LOAEL). Laboratory or captive experiments with wild birds provide ample information on NOAEL and LOAEL (e.g.Fimreite 1971, Heinz 1974, 1979, Finley and Stendall 1978, Finley et al. 1979, Scheuhammer 1988). While care is essential when applying these effect levels to free-ranging wildlife and for making interspecies comparisons, laboratory experiments provide toxicosis benchmarks and insights for measurable endpoints. Elevated MeHg levels are known to be related to neurological, immunological, and genetic toxicosis (Wolfe et al. 1998) as well as disrupting the biochemical functions with cortisol (Friedmann et al. 1996) in fish, cholinesterase in quail (Dieter 1974), and glutathione in mallards (Hoffman and Heinz 1998). In-situ wildlife studies complement many of these laboratory experiments.

Scheuhammer (1991) considered that the dietary concentrations of MeHg needed for adult birds to exhibit overt toxicity were five times those needed to produce significant reproductive impairments. One of the most important reference studies for piscivorous birds is a study of the effects of Hg on loons in a lake system highly contaminated from an upstream chlor-alkali plant in Ontario (Barr 1986). Barr found reproductive impairment with loons feeding on fish with 0.30 ppm (e.g., reduced egg laying and territorial fidelity) and no reproduction from loons feeding on fish with 0.40 ppm or more. Highly exposed adults (i.e., mean brain MeHg levels of 0.76 +/- 0.50 ppm) did not show overt signs of Hg toxicosis. Mercury levels without

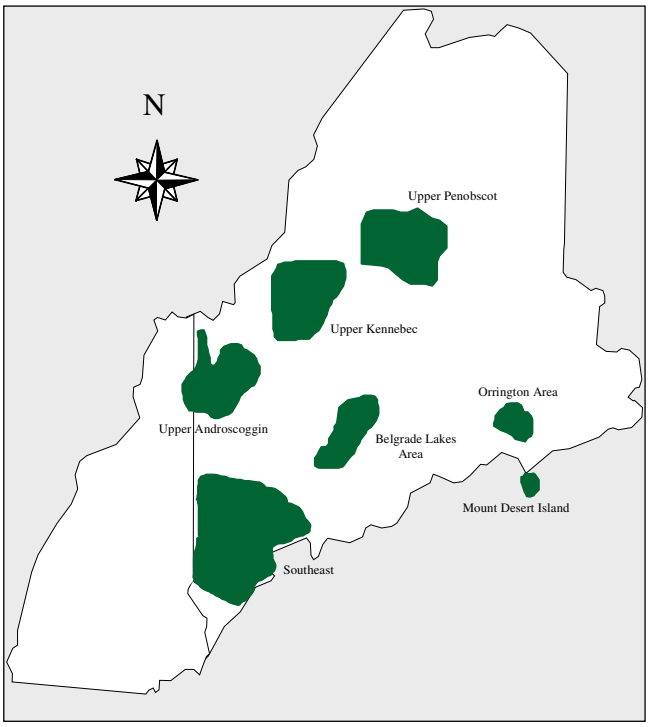
overt signs can cause a 35-50% decrease in reproductive success (Heinz 1974, Scheuhammer 1987). In Maine, Evers and Reaman (1997) found yellow perch Hg concentrations to exceed the 0.30 ppm threshold on 7 of 8 lakes studied in northern Maine. The highest adult and juvenile loon blood levels found during this study came from 2 adult and a juvenile loon on Little Lobster, a small natural lake. Loons on these lakes do produce chicks annually, but long-term productivity data is needed. We also documented the Hg levels in 18 species of fish in Maine for three size classes based on Barr's (1996) study of loon prey preferences. Between 62-67% of the fish had mean Hg levels of 0.30 ppm or more. Over 2/3 of the fish in the small size class (5-20g) have Hg levels that are reported to cause reproductive impairment in loons and are the size class most frequently used by the Common Merganser and Belted Kingfisher.

Maine's fish Hg levels are elevated across the state (Stafford and Haines 1996) and according to comparable studies, wildlife may be affected. How these prey Hg levels relate to exposure in piscivorous bird species varies. Although prey Hg levels are effective indications of MeHg availability to piscivorous birds, diet composition, species toxicokinetics, and biotransformation rates are needed to effectively model MeHg loads for piscivorous birds.

Study Area & Design

Field sampling was conducted in two watersheds (Upper Kennebec and Upper Andoscoggin) in 1996 and additional sites in southern and eastern Maine were sampled in 1997 as part of the Maine Outdoor Heritage Fund (MOHF) project "Determining Mercury Exposure in Maine's Fish-Eating Birds" (Figure 1).

Figure 1. 1997 MOHF Mercury Study Areas, 1997.



To investigate differences in Hg availability, sampling efforts were distributed among the following water body types:

- regulated storage or hydroelectric reservoirs (RSR) with annual fluctuations greater than 1.5 m (5 feet),
- "flashy" natural lakes with annual water fluctuations greater than 1.5 m (5 feet) (FNL)
- regulated "peak" hydroelectric reservoirs (RPR) with weekly water fluctuation levels of 2-3 feet,
- regulated "full pond" reservoirs and raised lakes managed for minimal water level fluctuations of less than 1.5 m (5 feet) (RFP),
- stable natural lakes with annual water fluctuations less than 1.5 m (5 feet) (SNL)

These classifications have been revised from last year to better reflect the types of waterbody with their corresponding water fluctuations.

For both years, sampling was concentrated in two major watersheds in Maine and New Hampshire: the Kennebec (3 reservoirs, 6 natural lakes) and the Androscoggin (6 reservoirs, 5 natural lakes). In 1997, 46 lakes from outside these two watersheds were sampled to increase the sample size of natural lakes. Sampling efforts on natural lakes were opportunistic based upon loon productivity. Lake morphometrics, water chemistry, water quality, and hydrology were compiled for many of the study lakes (Appendix I & II).

Methods

Field Capture and Collection

From June through October, 1996-97, 2-10 people assisted with the capture of adult and juvenile loons at night on their breeding lakes. Crews used boats powered with 10-150 hp outboard motors, 400,000 to 1.5 million candlepower spotlights, tape-recordings and mimicked vocalizations of loon calls to attract family groups. Loons were netted with large landing nets, restrained, then transported to shore.

The second secondary flight feathers were removed by cutting the calamus (i.e., below the base of the vane). Blood samples were taken by venipuncture from either the cutaneous ulnar vein or the medial metatarsal vein. We used 20-25 gauge straight needles with 1-12 cc syringes, or direct collection with 21-25 gauge needles and 7 inch tubing with multiple sample Luer adapter into 2-10 cc Vacutainers,, or combinations of the above. All loons were marked with a USFWS aluminum and 1-3 plastic color leg bands glued with an acetone-based adhesive cement.

The feathers were placed in polyethylene bags, labeled, and refrigerated within 12 hours of collection. The initial selection of remigials was based on sufficient mass and removal that would not negatively impact loon behavior (e.g., flight). Loons exhibit simultaneous remigial molt therefore selection does not bias analysis.

Blood was collected in 2-10 cc green top Vacutainers, (with powdered sodium heparin) for subsequent Hg analysis. Due to logistical and field limitations 10% formalin was added to preserve the blood following U.S. Fish and Wildlife Service protocol (0.05 cc of formalin / one cc of whole blood) (Stafford and Stickel 1981, Wiemeyer et al. 1984).

Egg materials have been collected from abandoned nests since 1993 by the Loon Preservation Committee and since 1996 by members of NELSWG. Eggs, eggshell membranes, and eggshells were placed in polyethylene bags and frozen. Volume, weight, length, and width were measured for each whole egg and the contents placed in I-Chem jars. Eggs before 1996 were archived in freezers for up to 3 1/2 years. Eggs were scored for 6 developmental stages. Some eggs could not be scored because they were autolyzed (18%). Shells and outer egg membranes were dried. Inner egg membranes were sampled independently.

Tissue sample analyses

All feather and blood samples in 1996-97 and eggs from all years were analyzed at the University of Pennsylvania under the supervision of Dr. Bob Poppenga. Samples from 1994-95 from Lake Umbagog, Aziscohos, and Rangeley Lake were analyzed at the Animal Health Diagnostics Laboratory at Michigan State University under the supervision of Dr. Emmett Braselton. Dr. Bob Taylor from the Trace Element Lab at Texas A&M analyzed the fish and sediment sample in 1996 and Dr. Bob Poppenga analyzed fish samples for total and methylmercury in 1997. Blood-Hg was measured in 1 ml heparinized whole blood by continuous flow cold vapor atomic absorption spectrophotometry using a Perkin-Elmer 3030b spectrophotometer (Scheuhammer and Bond 1991). The Hg residues measured in feather (fresh weight) and blood (wet weight) represent total Hg and are shown in ppm. Reagents were periodically tested to ensure low acceptable background levels of Hg contamination. Tests for residual Hg in the heparin and formalin were negative.

Feathers were analyzed for trace metal and mineral content using inductively coupled argon plasma (ICP) emission spectroscopy (Thermo Jarrell Ash Polyscan 61E). Feathers were cut, calamus discarded, and washed 3 times in acetone (chromatography grade, Burdick and Jackson, Muskegon, MI), 3 times in ultra pure water (4 bowl MilliQ System, Millipore Corp, Bedford, MA) one additional time in acetone, then allowed to dry overnight in a fume hood. Feather Hg strongly binds to disulfide linkages of keratin (Crewther et al. 1965) and is not disturbed by washing episodes (Applequist et al. 1984) or previous environmental exposure (Goede and de Bruin 1984). An 0.25-0.5 g aliquot of feathers was digitally weighed and digested overnight with 2 ml conc. HNO3 (Instra-analyzed grade, J.T. Baker Inc., Phillipsburg, NJ) in a closed, 30 ml Teflon container (Savillex Corp., Minnetonka, MN) at 90°C. The samples were quantitatively transferred to a 10 ml volumetric flask, mixed with 100 mg yttrium (JMC Specpure ICP/DCP Analytical Standards, Johnson-Matthey/Aesar, Ward Hill, MA) as internal standard, and diluted to volume. Samples were initially analyzed by inductively coupled plasma-atomic emission spectroscopy (Polyscan 61E, Thermo Jarrell-Ash Corp, Franklin, MA) (Stowe et al. 1985). An aliquot of the sample was then taken from the 10 ml volumetric flask and diluted an additional 1000 fold for analysis of Hg by cold-vapor atomic absorption spectroscopy (LDC mercury Monitor 3200, Thermo Separation Products, Riviera Beach, FL). Accuracy was monitored by concurrent analysis of procedural blanks (in triplicate), NIST Oyster Tissue SRM 1566a with Hg certified at 0.0642 +/- 0.0067 ppm (National Institute of Standards and Technology, Gaithersburg, MD) and NRC Tort 2 Lobster Hepatopancreas with Hg certified at 0.27 +/- 0.06 ppm (National Research Council of Canada, Ottawa, Canada).

Whole blood was thoroughly mixed and homogenized. From the whole blood sample, a 100 mg aliquot was placed into a 15 ml Teflon container, mixed with 2 ml conc. HNO3, sealed, and digested overnight at 90°C. Samples were quantitatively transferred to 25 ml (juveniles) or 100 ml (adults) volumetric flasks and diluted to volume with ultra pure water. Samples were analyzed as above by cold-vapor atomic absorption spectroscopy using the same quality assurance protocol.

Statistical procedures

We performed two-sample student's t-tests to detect differences in means. We tested normality with residual plots and homoscedasticity (whether variances were equal) with the Bartlett's Test. If normality or equal variance assumptions were violated we used a non-parametric heteroscedastic t-test. When variances were unequal we used non-parametric tests. Mercury concentration values were $\log^{10}$ transformed to stabilize the variances when variances were not equal. Sum of squares were corrected for inequity of unbalanced data sets within the computer analysis. The probability of determining differences between means was divided into two categories: significant ($p<0.05$) and marginally significant ($p<0.10$). All probability values are given for two-tailed tests. We performed multiple and linear regression analysis to provide us the strength of a correlation and the probability of a relationship. Pearson's Correlation Coefficient was used to test relationships between lake chemistry/morphometric data and mercury endpoint measures (e.g. blood Hg). Stepwise multiple regression was used on a subsample of lakes where all the independent variables were measured. Data were statistically analyzed using SYSTAT 5.0 and Microsoft Excel Toolpak 5.0. Hg concentrations are given as "mean +/- standard deviation."

Summary of Field Sampling Efforts

(See Results & Discussion for Hg concentration description & interpretation)

Sediment, Water Chemistry and Morphometric Sampling Efforts

Lake sediment cores were collected from reservoirs and natural lakes to determine recent and historical levels of mercury deposition. Hg deposition among lake sediments were compared to identify potential relationships between sediment and concentrations of Hg measured in loon and fish tissues. Sediment samples were analyzed for total Hg and Total Organic Carbon (TOC).

Water chemistry data collected by BRI were determined with a portable Hydrolab®, Hach kits (apparent color), and titration (alkalinity). All probes were calibrated daily. Samples were collected at 1 m intervals throughout the water column in 1996 and 0.5 m in 1997 (i.e., grab samples).

Water chemistry and quality measurements were provided by the Maine Department of Environmental Protection for the majority of the remaining lakes. Lake morphometric data were collected by the Maine Department of Inland Fisheries and Wildlife for all the lakes in this study. Water level fluctuations were measured at control structures for a number of lakes and by private individuals residing on natural lakes. Most natural lakes and several reservoirs in this study did not have reliable water level fluctuation data and were omitted from analyses.

Fish Sampling Efforts

To determine the relationship between Hg concentrations in loons and fish, we: 1). collected a sample of forage fish suitable for adult and juvenile loons following a custom-designed protocol based on loon prey preferences (Appendix III) and 2) collected larger fish to determine the predictive power of game fish Hg levels for assessing risk to piscivorous birds.

Loon prey items were collected from 6 reservoirs, and from 7 natural control lakes in 1996 and were composites of 5-15 fish. Fish fillets from 8 lakes were analyzed in 1996. Fish fillet Hg levels determined by the Maine DEP REMAP study in 1993-94 were used for 6 lakes with known blood Hg levels for loons and other piscivorous birds.

Bird Capture and Sampling Efforts

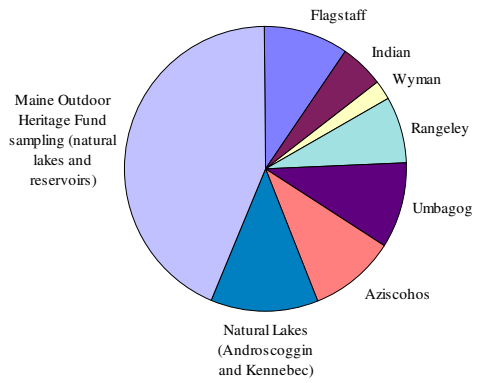
Common Loon blood, feathers, and eggs were collected from 49 natural lakes and 8 reservoirs in Maine and New Hampshire to determine Hg exposure. A total of 141 adult Common Loons were captured from 1993-97 in Maine and New Hampshire from the following waterbody types:

- Stable Natural Lake-SNL (12.6 %)
- Regulated Peaking Reservoir-RPR (5.9 %)
- Regulated Full Pond-RFP (11.9 %)
- Flashy Natural Lake-FNL (13.3 %)
- Regulated Storage/Hydroelectric Reservoir-RSR (56.3%)

Twenty two of these adults were recaptured for blood and feather Hg analyses, 1-3 years after their original capture. In addition, 64 loon chicks were captured in New England during this period. In 1997, capture efforts of loons were similar in reservoirs and natural lakes (Figure 2).

In 1997, 65 adult (8 recaptures) and 27 juvenile Common Loons were captured in the combined effort of the this study and the MOHF project, "Determining Mercury in Maine's Fish-Eating Birds" (Figure 3). Other piscivorous birds were also sampled. Species found on target reservoirs and included in this report are the Belted Kingfisher (*Ceryle alcyon*) and Common Merganser (*Mergus serrator*).

Figure 2. Percentage of capture nights distributed among selected natural lakes and reserviors in New Hampshire and Maine, 1997



Chick and adult blood samples were split for several analyses. Blood samples were collected from 139 adults and 56 juveniles and analyzed for total Hg. A subsample was tested for organochlorines and selenium. Feather samples from 137 adults were collected for Hg analysis. Loon eggs were analyzed for total Hg. The National Wildlife Health Research Center tested for lead concentrations as part of an EPA/USGS national lead study. Blood was also collected for genetic, blood physiology and hormone studies at Tufts University School of Veterinary Medicine. Common Loon eggs from abandoned nests were collected in Maine (n=45) and New Hampshire (n=72) as another measure of Hg exposure in loons.

Results & Discussion

The following discussion is in 8 parts. The first section describes factors that have an effect on the production and availability of MeHg in waterbodies. It provides context for the interpretation of the original data presented in later sections. This is followed by the factors that were measured for this study and their relationship to Hg concentrations in fish, loons, and other piscivores. We present results of our Hg testing for sediments, gamefish, and forage fish. The fish Hg levels are related with loon matrices. Following this is a summary of Hg concentrations

analyzed in: eggs, blood and feathers. We believe these matrices best reflect MeHg bioaccumulation and biomagnification in aquatic systems. Risk thresholds are discussed and fit into our findings of Hg concentrations in fish and loon matrices. Because the variability of MeHg production and availability are not fully explained by models, and patterns are difficult to describe, we summarize our findings for each reservoir and then group the natural lakes by known confounding factors. Because an understanding of water level fluctuations is crucial to this study, their relationship with other factors and various matrix Hg levels is emphasized.

1. Factors related to MeHg availability in waterbodies

There are many factors that affect the production and availability of MeHg in waterbodies. The production of MeHg is primarily by sulphur-reducing monobacteria that are most active in warm, acidic anaerobic environments. The availability of MeHg is dictated by many geochemical, hydrological, and physiognamy parameters, as well as biological factors such as uptake rates by plankton and efficiency of biotic transfer. Each factor has varying powers of influence and commonly have synergistic relationships which can be variable on different waterbodies. We have limited our data interpretation to a comparison of Hg exposure in biotic components between and among reservoirs and natural lakes. We have not fully evaluated the relative importance of each factor (i.e., through computer models), but recognize that each may influence Hg availability.

Water level fluctuations and related hydrological features are emphasized because they have direct management implications. Sorensen et al. (1989) were the first to link reservoir drawdowns to enhanced Hg concentrations. However, their work and others (e.g., Bloom et al. 1992) were not definitive, probably because their Minnesota study sites contained low Hg concentrations. Since then, a summary of the literature by Tetra Tech (1995) and recent findings in Maine reservoirs (e.g., Evers and Reaman 1997, Haines pers. com.) indicate that water level fluctuations are one of several variables for enhanced MeHg productivity on both natural lakes and reservoirs.

Hydrology

Much of the literature discussing water level fluctuations emphasizes reservoirs but omits references to potential Hg impacts from naturally fluctuating water levels. Studies that measure differences in MeHg bioavailability between reservoirs and natural lakes (Stein et al. 1996, Langlois et al. 1996) generally did not investigate natural water bodies with wide annual fluctuations. Several natural lakes within this study (e.g., Attean, Long, Pierce and Parmachenee) have regular annual water level fluctuations over 1.5 m (5 feet). Comparative studies also typically exclude high trophic level organisms (e.g., loons and other piscivorous birds) that have the opportunity to bioaccumulate and biomagnify MeHg. Interpretation of Hg exposure information must always be placed in context with the following parameters.

Reservoir Age: Newer reservoirs have higher fish Hg concentrations than older reservoirs. Tetra Tech, Inc. (1995) reviewed several studies that found Hg concentrations approach natural background levels 20 years after initial flooding. In piscivorous fish the process may take up to

21 years (Anderson et al. 1995) or 30 years (French et al. 1996). The initial flooding of areas releases Hg bound in terrestrial soils. However, older reservoirs can continue to be sources of Hg at levels higher than natural lakes because of inherent Hg inputs from large watersheds.

Reservoir Origin: The transport of mercury into aquatic biota is influenced by the surface area of the flooded acreage. Natural lake basins that were elevated several feet by the construction of a dam inundate significantly less land than flooded river valleys. Reservoirs constructed in relatively flat terrain will flood more acreage, thus releasing greater loads of Hg.

Watershed Acreage: The transport of mercury (inorganic and organic) from a watershed into a receiving lake can significantly influence Hg productivity. Lakes with large watersheds are more likely to have greater Hg contributions than smaller lakes. Studies have also shown that wetland acreage within a watershed is also important for estimating Hg input (Verta et al. 1986, McMurtry et al. 1989). The increased transport of organic material from heavily disturbed upland areas may increase total mercury input into a receiver lake.

Water Level Fluctuations: Fluctuating water levels from natural water level fluctuations and reservoir regulation have multiple effects on the methylation of Hg. Several factors are related to MeHg production during changing water levels (Verta et al. 1986, Bloom et al. 1992, Jargoe et al. 1994, Watson et al. 1994). These changes create a transitional zone that has a mixture of oxic and anoxic conditions that are known activity sites for microbial methylators (Wollast et al. 1975, Craig and Moreton 1986, Watras et al. 1994). Methylation is primarily an anaerobic process (Stein et al. 1996). Verta (1986) and Sorensen et al. (1989) found a strong relationship between fluctuation amplitude and fish Hg content — the wider the fluctuations the higher Hg levels in the fish. Flooding or the inundation of previously exposed sediments and particularly wetlands such as peat bogs increase MeHg availability (Dmytriw et al. 1995, Rudd 1995). However, the relationship between water level fluctuations and MeHg enhancement in high trophic wildlife species remains unclear.

Flushing rates: Flushing rate is the number of times water is exchanged in a lake during one year. The Hg methylation in a system may change seasonally and even more frequently based upon retention time. If the MeHg input (receiver lake production + watershed input) is less than the output (demethylation + riverine output) biota Hg uptake is less. MeHg production by microbes generally increases during the summer months and peaks in early fall. However, MeHg input varies throughout the year depending on climatic conditions. For example, “peat pressing” during the winter and the accumulated watershed snow pack can potentially flush a large amount of Hg into the receiver lake during the spring thaw.

Peat Pressing: Wetlands with sphagnum moss are ideal micro-habitats for methylating bacteria. The flushing of these areas during flooding or storm events can provide significant MeHg and inorganic Hg input to the lake. Additionally, Virtanen et al. (1994) has found that the pressure of ice and snow on the dewatered shoreline of a reservoir will force significant amounts of Hg rich humic matter into the waterbody. The freezing and thawing of these Hg-rich habitats also breaks down the organic matter and makes it more available to bind with MeHg and eventually

bioaccumulate.

Physiognamy

Shoreline Wetland Acreage: Wetlands, particularly bog mats containing sphagnum moss (*Sphagnum* spp.) harbor an abundance of sulfur-reducing bacteria that are effective methylators (Hurley et al. 1994, St. Louis et al. 1994, Krabbenhoft et al. 1995, Rudd 1995). MeHg produced in these areas exist in the pore waters and are then available for watershed transport during storm events or floodings. Inundation of shoreline wetlands, particularly peatlands, increases MeHg loading into the waterbody.

Geologic Source and Soil Types: Substrate materials and shoreline soil types can significantly influence background levels of Hg and impact the availability of new Hg inputs. Bedrock Hg concentrations can vary. For example, dry podosols have less impact than humic soils. Therefore floodplain soils such as are found in the flooded river valleys of Aziscohos and Flagstaff reservoirs provide higher Hg producing microhabitats than alluvial outwash sands common along lake shorelines. Frequent shoreline erosion of humic soils can increase MeHg production (Grondin et al. 1995).

Topography: Waterbodies surrounded by mountains having a large snowpac can receive high inputs of mercury during spring melt. Montane climates are often subject to higher rainfall and cloud precipitation events and may increase Hg inputs into the watershed as well (pers comm. Alan Van Arsdale). Mountainous topography also creates flashy runoff into waterbodies rather than retention by local soils.

Watershed Cover Type: Recent studies indicate throughfall from coniferous trees have 3 times higher Hg concentrations than deciduous trees (T. Haines, pers. com). Lakes surrounded by largely coniferous forests likely have a greater source of Hg availability.

Episodic Events and Temporal Trends: The amount of Hg input into a receiver lake varies over time and is related to seasonal variances. Spikes of MeHg input into a waterbody occur throughout the year. Spring thaw and storm events can produce short term inputs of MeHg while increasing microbial activity throughout the summer produces a steady flow of MeHg. Fall turnover of the water column (Gill and Bruland 1992, Slotton et al. 1995) and winter peat pressing are also associated with considerable release of MeHg into a water body. Determining the average annual input and cycling of Hg in an aquatic system is important.

Water Chemistry & Quality

Water Chemistry: Studies have shown low acid-neutralizing capacity (ANC) and high humic content of a lake to strongly correlate with increasing fish Hg concentrations (Wiener and Spry 1996). When compared to alkaline water bodies, low pH waters support microbial activity that has a greater capacity for MeHg production. Apparent color measurements indicate the amount of dissolved organic matter such as humic acid and possibly dissolved organic carbon (DOC). Total and MeHg bind to organic particles. This effect on subsequent uptake by biota is

inconclusive (Matilainen and Verta 1995). Chloride (Mason et al. 1995), and sulfate (Heyes et al. 1996) also relate to MeHg bioavailability. Driscoll et al. (1995) found that high concentrations of monomeric aluminum enhanced the bioavailability of MeHg. In addition, Tsalritzis et al. (1996) found that total Hg in water and zooplankton correlated with aluminum concentrations.

Water Temperature: Methylating microbes increase their activity with a rise in ambient water temperature (Fagerstrom and Jernelov 1972, Callister and Winfreys 1986). Therefore, increasing MeHg is positively correlated with the extent of a lake's photic and littoral zones. Waterbodies with extensive shallows are most at risk. The upper hypolimnetic layer (just below the thermocline) is also associated with elevated MeHg production (Slotton et al. 1995).

Biological

Trophic Structure: MeHg bioaccumulates (Evers et al.1998) and biomagnifies in biota (Kainz and Lucotte 1996). The longer the food chain the greater the potential for elevated Hg levels at the endpoint (i.e., loons).

2. Surficial Sediment Hg concentrations

The source of Hg in an aquatic system may be anthropogenically and naturally based. Although annual anthropogenic inputs may vary considerably at local levels, global emissions may account for upwards of half of the atmospheric load (EPRI 1994). Sediment Hg levels are a good indicator of atmospheric deposition loads (Lindqvist et al. 1991, Nater and Grigal 1992, Swain et al. 1992). A comparison of Hg concentrations between historic (samples taken in 20-30 cm deep) and recent (samples taken in 0-10cm deep) sediment cores (1-3 per lake) was used to provide a chronological indication of Hg deposition into a system. Total organic carbon (TOC) was also measured because organic matter in soils has a high affinity for Hg (Schuster 1991). Mean Hg levels were adjusted with percent TOC to allow equivalent comparisons.

Mean Hg concentrations in surficial sediments ranged from 0.10 to 0.36 ppm (dry weight, 0-10cm) and 0.10 to 0.34 ppm (dry weight, 20-30cm). Over 70% of the natural lakes and 83% of the reservoirs had top layer sediment Hg levels that exceeded Ontario's 0.20 ppm benchmark of lowest effect level (Persaud et al. 1993). Only sediment Hg concentrations from the natural lakes, West Carry (0.10 ppm) and Parlin (0.17 ppm), and from one reservoir, Umbagog (0.19 ppm) were below this benchmark.

We also divided sediment core Hg by %TOC when we compared waterbodies because MeHg binds with organic material. Whether this indicates more or less MeHg available to biota is debatable. The mean Hg index for surficial sediments ranged from 0.01 to 0.11 in natural lakes and 0.02 to 0.09 in reservoirs. The percent TOC concentrations varied from 4.2 (in Aziscohos) to 22.4 (Little Beaver). Percent TOC concentrations were highest in natural lakes like Little Beaver, Little Indian, and Akers and lowest in reservoirs. Although the sandy substrates of our study reservoirs are characteristic for impoundments that are regularly drawn down and flushed, natural lakes may also have similar rates of sedimentation and transfer.

Although our 1996 sediment samples did not significantly correlate with fish ($p>0.05$) or bird ($p>0.05$) Hg concentrations, recent Hg deposition tends to be greater than historic (for adjusted and unadjusted Hg levels). Some differences between the two sampling layers were substantial. Adjusted sediment Hg levels for Richardson and Umbagog reservoirs were respectively 3 and 4 times higher than historic levels. Most natural lakes and Mooselookmeguntic reservoir exhibited no change between the top and bottom sediment layers. These chronological tendencies are significant in other Maine lakes although varying sedimentation rates in reservoirs and natural lakes demands the aging of sediment cores to interpret current Hg deposition rates (T. Haines, pers. com.).

From 8% (Mierle 1990) to 26% (Swain et al. 1992) of Hg deposited atmospherically in an aquatic system is from the watershed runoff, the remainder falls directly on the waterbody. Mercury contributions to a lake are a function of watershed size and physiognamy, such as type of vegetation, topography, soils. Upstream transfer of Hg from upstream waterbodies also impacts downstream levels. However, when comparing the 7 study reservoirs, it appears that the similar unadjusted mean Hg concentrations in the upper layer of reservoirs ($p>0.05$, range = 0.15-0.18 ppm) indicate similar atmospheric deposition of Hg. Sampling efforts in the reservoirs' deeper portions minimized potential confounding factors such as water level fluctuations, water chemistry and temperature, and wetland influences. Unadjusted sediment Hg levels in natural lakes were more variable than reservoirs. The mean Hg concentration of the upper sediment layer ranged from 0.03 ppm in West Carry to 0.32 ppm in Little Beaver. Sampling of the deeper portions of the natural lakes (that are much smaller than the study reservoirs) indicates confounding factors have more influence on the sampling sites than in the larger reservoirs.

3. Relationship between Hg concentrations in loon blood and fish fillets

Total Hg concentrations from fish axial muscle representing 8 lakes in 1996 (Evers and Reaman 1997) and 6 lakes in 1997 (Stafford and Haines 1997) were compared with mean adult and juvenile blood Hg levels. Yellow Perch fillets were collected on 2 natural lakes and 5 reservoirs. We compared the axial muscle of fish larger than 35 g and found no significant differences in Hg concentrations between reservoirs and natural lakes ($p>0.05$). We also stratified the data to measure the relationship of axial muscle Hg concentrations and size of fish (using weight or weight x length as the predictor variables) and found a positive correlation with larger fish and higher Hg concentrations (Salmon $R^2= 0.265$,$p<0.05$; Smallmouth Bass $R^2=0.63$, $p<0.05$, and Yellow Perch $R^2=0.51$, $p<0.05$) (Evers and Reaman 1997).

The relationship between axial muscle Hg concentrations of Yellow Perch and loon blood Hg was not significant for males, females and juveniles ($p>0.05$) (Appendix IV). Sample sizes of other species of fish were too small for statistical comparison with Common Loon Hg concentrations. We tested mean fillet Hg concentrations of all fish species sampled from each lake with loon blood Hg and found no linear relationship ($p>0.05$). Therefore, fish fillets may not be a suitable predictor of wildlife Hg exposure on a given lake.

4. Hg concentrations measured in loon prey and correlations with loon blood

We measured Hg in loon prey items to determine 1) if Hg concentrations are different between reservoirs and natural lakes, 2) whether Hg levels are high enough to inhibit loon reproductive activities 3) if concentrations correlate with Hg levels measured in loon matrices to confirm the Hg biomagnification pathway and 4) whether certain prey items have higher Hg concentrations and pose a greater risk to loons.

To determine prey Hg concentrations in species and size categories that loons select, we developed a collection protocol based on a study by Barr (1996). In this study, adult and juvenile loons selected small- to medium-sized fish. Large fish (over 8 inches) were preferred but their capture availability is lower than other size classes. In Ontario, adult loons consumed 432g (small fish), 365g (medium-sized fish), and 163g (large fish) daily or approximately 20% of their body weight per day. The percentage of individual fish consumed by size class was: 80% small, 18% medium, and 2% large. Male loons from Maine and New Hampshire loons are 26% larger than their counterparts in western Ontario (geographic differences in female body mass are less). We adjusted the amount of fish consumed by the heavier New England loons to be: 45%,38%,17% for females and 33%,40%,27%, for males.

Barr (1996) determined that because of their "zig zag" swimming behavior, Yellow Perch (*Perca flavescens*) were the preferred food of adult and juvenile loons. Yellow perch are widely distributed across the loon's breeding range. Median size perch consumed during Barr's trials for 12-15 week old loons (adult sized individuals) was 5.0cm (2 inches) and ranged from 1.0-9.5cm (1/2 to 3 3/4 inches). Adult loons were observed eating perch up to 12.0-16.0cm (4 3/4 to 6 1/4 inches). We divided perch into four size classes for this study (Appendix III). The first three size classes were similar to Barr's, and the fourth was an extra-large size class that contains individuals in the upper size limit that the larger New England loons could consume (8-12 inches).

Barr (1996) also found sucker to be preferentially chosen by loons (e.g., White Sucker, *Catostomus commersoni*). The average size of suckers selected was larger than perch because suckers are presumable easier to swallow. Fusiform (i.e., streamlined) shaped fish are preferred by loons. We separated sucker into three size classes for this study (Appendix III). Other fusiform shaped fish sampled in our study included Chain Pickerel (*Esox niger*), Rainbow Smelt (*Osmerus mordax*), and cyprinnids (minnows). These species were placed in an "opportunistic" category and divided into size classes similar to White Sucker. The swimming abilities and "burst speed" of salmonids make them less available to loons than other species and their collection was incidental.

Bloom (1992) and others have shown that nearly all (95 to 99%) of mercury in fish is MeHg. Few studies have investigated MeHg in fish within a size class available to loons. Scheuhammer et al. (1998) found MeHg constituted 94 +/- 12% of total Hg in fish suitable for adult loons (i.e., 20-50g Yellow Perch and centrarchids). Other studies reported small fish exhibited higher inorganic Hg load than larger fish (T. Haines, pers. com.). Evidence also indicates that 90% of the methylmercury in fish originates from the diet (Harrison et al. 1990).

Waterborne methylmercury that passes over the gills accounts for much of the remainder (Rodgers and Beamish 1981). Uptake of dietary methylmercury from forage fish to a predator is 65-80% or more (Rodgers 1994). Therefore, piscivorous fish are the most susceptible foraging guild to MeHg biomagnification.

Numerous potential confounding variables for interpreting the toxicity of Hg in fish body burdens are reported in the literature and listed by Sorensen (1991). Inter- and intra-species variation in the uptake of Hg may be attributed to differences in metabolism, size, gender, water chemistry, synergistic relationship with other pollutants, and hydrological changes. Therefore, the interpretation of forage fish Hg must be viewed as preliminary and not as an indicator of lake differences in fish populations.

A total of 150 fish composites or single individuals were collected in 1996 for whole body Hg concentrations This represents 67 composites from 6 reservoirs and 83 composites from 7 natural lakes. We collected composites of 27 perch, 28 White Sucker, and 66 of 14 other species. Fish were divided into three (sometimes 4) size categories (Appendix III). Full size complements for perch were gathered for 3 reservoirs (Flagstaff, Umbagog, Richardson) and 2 natural lakes (Akers and Long).

In 1997, we emphasized the collection of Yellow Perch to standardize comparisons and because perch and loon Hg concentrations were highly correlated. On lakes and reservoirs where Yellow Perch did not occur small and medium size cyprinnid species were collected. Perch and other small- and medium-sized forage fish were collected from 6 reservoirs and 6 natural lakes.

Yellow Perch

Based on 27 samples collected in 1996, we found mean Hg concentrations for perch to increase with size. There were no differences in Hg concentrations between small (5-20g, 5-10cm) and medium (20-35g, 10-15cm) size categories while large (35-60g, 15-20cm) and extra-large fish (60-80g, 20-25cm) had higher body Hg concentrations than the smaller size classes. The Hg levels of small to medium size perch were similar in reservoirs and natural lakes and tended to be higher in large and extra-large perch in reservoirs.

White Sucker

The relationship between mean Hg concentrations in three size classes of White Sucker was similar to Yellow Perch. Small (5-25g, 5-13cm) and medium (25-75g, 13-20cm) size classes had no significant differences in Hg concentrations. However, large sucker (75-200g, 20-30cm) had higher body burdens of Hg than smaller size classes. Similar to Yellow Perch, Hg levels in large size class sucker tended to be higher on Flagstaff Lake than the other four reservoirs and natural lakes. Unlike perch, White Sucker occur on Aziscohos Lake and they had Hg concentrations similar to the other study lakes. For natural lakes, Little Beaver and East Richardson Pond had White Sucker Hg concentrations above the 0.30 ppm effects threshold found by Barr (1986).

Opportunistic Species Sampling

Although Yellow Perch and White Sucker are generally ubiquitous species, they are either absent on some lakes or are difficult to catch in suitable numbers. The only study reservoir without Yellow Perch is Aziscohos. In addition, loons forage on fish other than perch and sucker. Therefore, we collected a variety of other fish species that were dependent on the size

class limits set for perch and sucker. Brown Bullhead *(Ictalurus melas)* and Pumpkinseed Sunfish *(Lepomis gibbosus)* individuals were fit to perch size classes. All other species were fit to sucker size classes. Most, but not all, fish mean Hg concentrations represent composites of five or more fish.

In the small size class three species exceeded the upper impact threshold of 0.40 ppm documented by Barr (1986): Longnose Sucker *(Catostomus Catostomus)* (Aziscohos Lake), Black-nosed Dace *(Rhinichthys atratulus)* (Little Beaver and East Richardson), and Northern Redbelly Dace *(Chrosomus eos)* (Aziscohos Lake). In addition to perch, 4 other species exceeded the lower impact threshold (0.30 ppm) including a Common Shiner *(Notropis cornutus)* composite of 0.44 ppm from Aziscohos and a Golden Shiner *(Notemigonus crysoleucas)* composite of 0.63 ppm from Attean Lake. A composite from East Richardson Lake of two bullfrog *(Rana catesbeiana)* tadpoles had a Hg concentration of 0.18 ppm compared to a *Rana* species (i.e., mink, pickerel, leopard, or green frogs) composite of 0.08 ppm.

A total of 13 species of fish were collected in the medium size class. Only one species, the Golden Shiner, exceeded the upper impact threshold with a mean composite Hg concentration of 0.53 ppm. Seven species were above the lower impact threshold. Species (and their respective lakes) that were considerably higher included composites of Alewife *(Alosa pseudoharengus)* with 0.68 ppm (Mooselookmeguntic) and Rainbow Smelt with 0.45 ppm (Attean). Eight species of fish had composite Hg concentrations higher than Yellow Perch.

Nearly all of the large size class species of fish exceeded the lower impact threshold. Although only 1 in 45 fish generally eaten by adult loons per day fall within our large fish classification, their Hg concentrations tend to be higher and therefore proportionally contribute more Hg to the loon compared to a similar sized meal containing an equivalent number of smaller fish.

Relationship of forage fish and loon blood Hg concentrations

Barr (1986) found the lowest observed adverse effect level for adult loons feeding on fish with whole body Hg concentrations over 0.30 ppm (i.e., lower threshold, impaired reproduction) and 0.40 ppm (i.e., upper threshold, no reproduction). Barr did not specify daily dietary Hg ingestion thresholds in his study, but it is assumed that loons in this contaminated system were regularly foraging on fish at and above this level on a daily basis.

Loon prey Hg data from 1996 indicate that mean Yellow Perch Hg concentrations sampled from natural lakes and reservoirs from Maine were comparable with Barr's contaminated site and above the level he found to cause reproductive impairment. In addition, Yellow perch in every size class sampled in Maine had elevated mean Hg concentrations (> 0.30 ppm), notably higher than background levels found in Barr's study.

All size classes of perch and sucker from natural lakes and reservoirs showed at least some composites with Hg concentrations above the lower threshold of loon reproductive impact. A major source of the adult and juvenile loon's diet in our study area is likely Yellow Perch, because it is available on many of our study lakes. This hypothesis is strengthened by our finding of perch Hg levels to strongly correlate with adult loon blood Hg levels particularly medium-sized perch (R^2=0.85, $p<0.001$). Medium and large Yellow Perch Hg concentrations correlated more strongly with male than with female blood Hg concentration. Young loons appeared to forage to a lesser extent on perch since their indexed blood Hg levels were not significantly

correlated to perch Hg concentrations ($p>0.10$).

Mean perch Hg concentrations for small and medium sized individuals approached the lower threshold described by Barr (1986) while large and extra large perch exceeded the upper threshold. The Hg body burden of Flagstaff Lake perch tend to be more elevated than other lakes: all size classes of perch exceeded the upper impact threshold. Mercury concentrations for extra large perch exceeded the upper impact threshold for other reservoirs (Umbagog and Richardson) and natural lakes (Attean, Akers, and Long). Large perch Hg levels exceeded the upper impact threshold for Akers Pond (low pH natural lake) and the lower impact threshold for Attean and Long Pond (natural lakes with widely fluctuating water levels) and Richardson and Mooselookmeguntic Reservoirs. Indian Pond perch Hg concentrations were below the lower impact threshold.

White Sucker Hg levels for small, medium and large size classes did not correlate with adult or juvenile blood Hg concentrations. White Sucker Hg levels exceeded the upper impact threshold for large individuals on one natural lake (East Richardson) and one reservoir (Flagstaff) and the lower impact threshold for large individuals on the reservoir, Aziscohos, and the natural lake, Long Pond. Small and medium sized White Sucker exceeded the lower impact threshold on the natural lake, Little Beaver and the reservoir, Richardson Lake (only small size). Generally the omnivorous, bottom-feeding White Sucker contained less Hg than Yellow Perch and were less of a potential Hg source for loons. Because Yellow Perch Hg levels strongly correlated with adult loon blood Hg, we believe this fish species is a major component of the loons' diet.

Cyprinnids, large fish fry, crayfish, tadpoles, and other prey are also important food sources for adult and juvenile loons. We collected 7 species of the family Cyprinnidae. They included Golden and Common Shiners, Blacknose and Northern Redbelly Dace, Lake and Creek Chub, and Fallfish. The mean composite Hg concentrations on our study lakes were over the upper impact threshold for successful loon production for small Blacknose and Northern Redbelly Dace and medium-sized Golden Shiner. Small Golden and Common Shiners and medium sized Creek and Lake Chubs exceeded the lower impact threshold. Therefore, although many of these cyprinnids are small and have shorter life spans than larger fish, their Hg accumulation rates provided potentially dangerous levels to loons. Juvenile loons are particularly impacted since they feed more regularly on cyprinnids than on other fish.

It is important to collect a variety of loon prey items because loons in different lakes have specific feeding habits that can change seasonally and annually. Although loons forage opportunistically they are guided by prey availability. Genders may vary in prey selection. New England males average 30% larger than their mates and therefore tend to select larger size classes. Therefore, although Yellow Perch may potentially predict loon Hg body burdens, their availability may not be consistent across various waterbodies. Due to sexual dimorphism and consequently higher Hg exposure rates, adult male loons are more at risk than adult females.

5. Hg Concentrations measured in the piscivorous bird matrices

We measured total Hg concentrations in three matrices: bird eggs, blood, and feathers. Our efforts concentrated on the Common Loon, although we did captured and sampled other piscivorous

birds, particularly Common Mergansers and Belted Kingfishers. Interspecies relationships of the impacts of Hg exposure are difficult to interpret due to differing species pharmacokinetics. However, Hg uptake can be predicted with the following formula: body mass (g) x (fish Hg concentrations by fish size category x % of fish eaten per size category) x ingestion rate (g). This allows an interspecies comparison of potential exposure to Hg (Table 1). Because kingfishers have a high metabolic rate their MeHg uptake is double that of loons, mergansers, and osprey, while eagles are lowest because of their lesser daily food requirements. However, because loons and eagles are longer lived than most piscivorous birds their risk to bioaccumulation is greater. Our calculated daily exposure rate for the Common Loon in Maine is 69 and 63 ug MeHg per kg of body weight for males and females, respectively (Table 1). Maine loons are 4.3 (male) and 3.5 (female) times more exposed than the national average and are generally more exposed than other species. This agrees with Evers et al. (1998) study that shows significantly higher adult and juvenile blood Hg levels in the Northeast compared to the Upper Great Lakes, Pacific Northwest, and Alaska.

Table 1. Calculated MeHg exposure rate for five piscivorous birds in New England (ug/kg).

Sex	Body Weight(g)	Ingestion Rate (g/day)	Fish Size Class[a]	% fish diet by size class	Maine MeHg Exposure[b,c]	National MeHg Exposure[b,d]
Kingfisher	150	75	1	100%	150	40
Loon (male)	6,050	1300	1,2,3	33,40,27%	69	16
Loon (fem)	4,700	950	1,2,3	45,38,17%	63	18
Merganser	1,100	250	1,2	50,50%	68	18
Osprey	1,500	300	3	100%	60	16
Eagle	4,600	500	3,4	74,18%	32	14

a Fish size classes for New England are: small (5-20g), medium (20-35g), large (35-60g), and extra large (60-80g)

b Units are ug MeHg per kg of body weight per day.

c Based on fish Hg levels in Maine lakes (Evers and Reaman 1997).

d Based on national fish Hg levels (USEPA 1996). Trophic level 3 fish are 0.08 ppm and level 4 fish are 0.40 ppm.

The risk that these Hg concentrations pose to loons and other piscivores is not fully known, although other studies have developed thresholds that provide some context of impact. The following sections describe Hg concentrations for Common Loon eggs, blood, and feathers and provide context for these levels.

Abandoned Eggs Hg concentrations

Wolfe (1998) describes mercury as "an extremely potent embryo toxicant." Egg hatchability is potentially related to parental investment or high Hg loads. The LOAEL of Hg in eggs is accepted at 0.50 ppm (Scheuhammer 1991, Thompson 1996, Wolfe et al. 1998) and is partly based on Fimreite's (1971) pheasant study but has also been shown in the wild with Bald Eagles (Wiemeyer et al. 1993). Significant effect threshold levels vary considerably but are thought to range from 1-3.6 ppm (Fimreite 1974). Barr (1986) documented egg Hg levels on a series of lakes with differentiating exposures. The highest exposed series of lakes had a mean Hg level of 1.39 ppm and the lowest exposed lakes in this system had a mean Hg level of 0.59 ppm

and was similar to a control group of natural lakes independent of the contaminated system. The lowest reproductive success for this loon population was recorded for pairs that laid eggs with > 1.00 ppm, although a causative relationship was not established in Barr's study.

Table 2. Total mercury concentrations measured in Common Loon egg contents from Maine, 1994-97

	Mean Egg Hg (µg/g)			
Lake	Hg	SD	n	range
Wyman {R}	2.49	-	1	-
Brewer	1.75	0.11	3	0.62 - 1.83
Aziscohos {R}	1.27	0.48	11	0.56 - 2.42
Seven Tree Pond	0.85	-	1	-
Mooselookmeguntic {R}	0.85	0.03	2	0.83 - 0.87
Hancock Pond	0.77	0.24	2	0.61 - 0.94
Pierce Pond	0.63	0.41	4	0.38 - 1.25
Long Pond (Belgrades)	0.59	0.09	2	0.83 -0.87
Lake Umbagog {R}	0.59	0.33	8	0.31 - 1.62
North Pond	0.53	-	1	-
Howard Pond	0.47	0.07	2	0.42 - 0.52
Cold Stream Pond	0.46	0.11	2	0.38 - 0.53
Kennebunk	0.45	-	1	-
Moose Pond	0.37	0.22	2	0.22 - 0.52
Flagstaff {R}	0.27	-	1	-
ALL SITES	**0.93**	**0.55**	**43**	**0.22 - 2.49**

{R} = reservoir

In Maine, 43 loon eggs had a mean of 0.915 ppm with a range of 0.215 to 2.49 ppm (Table 2). A breakdown by threshold category shows 23% of these eggs had less than 0.50 ppm, 45% between 0.5 and 1.00 ppm, and 32% greater than 1.00 ppm Hg. Two eggs from reservoirs (one from Aziscohos Lake and the other from Wyman Lake) were above 2.00 ppm. The collection of loon eggs in 1997 represented 15 lakes, 56% of which the mean egg Hg level was above 0.50 ppm and 33% of which levels were above 1.00 ppm. Lakes with loon egg Hg concentrations greater than 1.00 ppm are some of the most at-risk lakes we sampled and include the natural lakes, Brewer and Pierce Pond; and the reservoirs, Aziscohos, Umbagog and Wyman. Aziscohos, Pierce, and Umbagog have water level fluctuations greater than 1.5 meters while Wyman reservoir and Brewer, a natural lake, has lower water level changes. Brewer Lake is 4 miles southeast of a Hg air emission point source (the Holtra-Chem Manufacturing Plant in Orrington).

The distribution of egg samples prevented suitable statistical comparisons with hydrology or other parameters related to Hg production or availability. Interpretation of egg Hg concentrations needs to account for egg laying chronology. Generally the first egg laid has higher Hg levels than the second and therefore the second egg is more likely to hatch if Hg is affecting hatchability. The relationship of embryo toxicity or teratogenic effects and Hg concentrations is not fully known for loons. In this study we have found that waterbodies with loon territories that have high Hg eggs also hatch chicks. Loon territories that produced young also occur in areas that have forage fish Hg levels above the 0.40 ppm impact level designated by

Barr (1986).

Adult Blood Hg concentrations

Blood provides an indication of recent dietary Hg uptake. Nearly all Hg in the blood is MeHg bound to erythrocytes and because the half-life of MeHg in avian blood is 2-3 months (Scheuhammer 1987) it is one of the better matrices for determining current exposure at a breeding site. Residual MeHg bound in the body (i.e., muscle tissue) may influence concentrations, particularly during stressful events so it is important to sample healthy, breeding individuals. Wolfe and Norman (1998) showed a significant correlation between blood and brain Hg. Because measurable effects have been associated with brain Hg exposure, it is useful to measure blood Hg in free-ranging wildlife as a means to assess potential risk. In addition, blood provides a non-lethal method of assessing Hg exposure.

Threshold Hg blood levels in adult Common Loons have not been described in the literature. However, based upon our professional judgement and results of studies in other parts of the county (M. Meyer & N. Burgess, pers. com), adult loons with blood Hg levels of 3-4 ppm are considered to have a moderate risk and those over 4 ppm are high risk. Risk evaluation includes potential effects on the adult's well being (as measured through blood profiles) and effects on productivity (through behavioral or physiological changes).

The mean blood Hg concentration in New England for adult males and females was 2.46 +/- 1.11 ppm (n=71) and 1.72 +/- 1.00 ppm (n=67), respectively (Table 3, Figure 4-5). Mean male Hg levels were significantly higher (30%) than females ($p<0.05$). We also found the log of Hg and adult male weight were significantly correlated ($R^2=0.10$, $p=0.04$). Therefore, we consider male loons more at risk. Larger male loons, and piscivorous birds in general, likely eat larger fish with higher Hg concentrations. Consequently, female blood Hg concentrations are lower because they eat smaller fish and depurate Hg into their eggs.

We used several statistical methods to identify the relationships between adult male and female blood Hg concentrations and water level fluctuations. Stepwise multiple regression analysis indicated water chemistry (alkalinity, pH, secchi) as a better predictor of adult blood Hg levels than water level fluctuations, however the sample size of lakes used in this analysis was limited due to missing morphometric and water quality data. Simple linear regression analysis showed that the only independent variables that were significantly correlated with female blood were annual and summer water fluctuations ($p<0.05$); however, water level fluctuation data from high natural lakes were not available. A more thorough analysis is needed with additional water level fluctuation data from natural lakes. Male blood Hg levels did not correlate with water level fluctuations ($p>0.05$). Adult loon Hg concentrations were significantly higher on storage and hydroelectric reservoirs than stable natural lakes ($p<0.05$) (Figure 6, Table 4). Mean adult blood Hg concentrations from Aziscohos, Flagstaff, and Wyman Lakes were significantly higher than the other reservoirs ($p<0.001$) and stable and flashy natural lakes combined ($p<0.001$). Mean adult blood Hg levels from Umbagog, Pontook, Indian Pond, Mooselookmeguntic, and Richardson were not significantly different than the stable and flashy natural lakes combined ($p<0.05$). Adults on territories downstream from Aziscohos Lake (i.e., Magalloway River) and Flagstaff Lake (i.e., Wyman) had high Hg levels suggesting downstream transport of Hg may be a potential factor.

Juvenile blood Hg concentrations

Several recent studies have used juvenile blood to document exposure to Hg. Derr (1995) collected blood from a suite of piscivorous birds in Minnesota, including the common merganser. Welch (1994) and Matz (pers. com.) collected blood from juvenile Bald Eagles in Maine. Several studies have documented exposure levels in loons, including Meyer et al. (1995) and Meyer et al. (1998) in Wisconsin, Scheuhammer et al. (1998) in Ontario, and Evers et al. (1998) across North America. Of these, the only study providing information on threshold impacts is Meyer et al. (1998). Four to six week old loons with blood Hg levels of 0.30 ppm or more were associated with territories where fewer chicks hatched or survived to 8 weeks of age. Although this threshold level is not necessarily useful for other species (e.g., Welch 1994 considered 0.50 ppm as a relevant threshold blood level in Maine juvenile eagles) we feel it is an adequate benchmark for loons. In New England, 27% of the juvenile loons sampled had blood Hg levels that exceeded 0.30 ppm, representing 20% of the lakes sampled.

Juvenile blood Hg concentrations reflect exclusive ingestion from the sample lake of dietary Hg during the current breeding season. Young loons depurate their Hg body burdens in three distinct feather molts and can use this pathway on their natal lake through their first 12 weeks of life. We found that chick weight strongly correlated with blood Hg concentrations (R^2=0.48, $p<0.001$). With the assumption that juvenile loon growth rates are similar among lakes, loon chicks from lakes with lower prey Hg levels (e.g., <0.30 ppm) had a significant increase of Hg over time ($p<0.001$), but showed a weaker correlation with weight (R^2=0.37) and exhibited a slower Hg accumulation rate than high risk lakes (Figure 7).

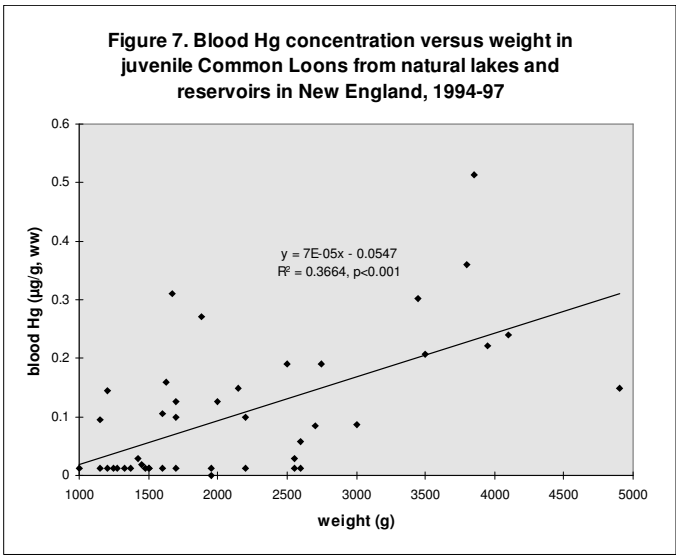
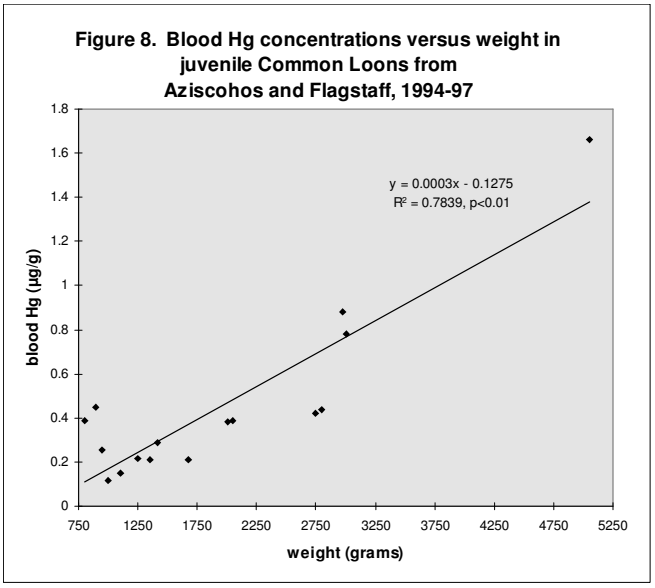


Figure 7. Blood Hg concentration versus weight in juvenile Common Loons from natural lakes and reservoirs in New England, 1994-97

A significant increase in blood and feather Hg levels during the loon's first 12 weeks was most apparent on lakes with small fish (5-25 g) that had Hg levels greater than 0.40 ppm (R^2=0.78, $p<0.05$) (Figure 8). Three of the 15 lakes sampled for juvenile loons had elevated MeHg levels that cause a significant accumulation over a 12 week period (i.e., Little Lobster lake and Flagstaff

and Aziscohos reservoirs).



Therefore, juvenile loons from lakes with elevated Hg levels are more likely to accumulate Hg and need to have their blood Hg levels adjusted by their weight (which correlates with age) to provide a standard index of Hg exposure. Other lakes with high Hg levels recorded in adults (eg. Green, Woodbury, Pemaquid, Branch, Sand and Chain of Ponds) have not produced chicks or chicks were not captured during this study and would likely accumulate mercury similarly. Therefore, because feathers depurate Hg (store excess blood Hg in an inert form) and larger chicks feed on larger prey items that tend to have higher Hg levels, we accounted for chick size with an index (ppm of MeHg/gram of loon).

The mean juvenile blood Hg level for New England was 0.22 +/- 0.28 (n=56) and was four times higher than concentrations found in similar-sized Common Loons breeding in Alaska (Evers et al. 1998). Corresponding mean blood Hg concentrations adjusted for weight were 1.10 + /- 1.24 ppm Hg/g x 10^4 (Table 5, Figure 9). The highest blood Hg index level in this study was 4.88 ppm Hg/g x 10^4 recorded on Little Lobster Lake in Maine. This lake also had the highest female (6.51 ppm) and the second highest male (5.86 ppm) blood Hg concentrations recorded in New England. Mean chick blood Hg index on natural lakes was 0.54 ppm Hg/g x 10^4 and 1.20 ppm Hg/g x 10^4 on reservoirs.

Table 5. Total mercury concentrations in juvenile Common Loon blood and blood Hg index at selected sites in Maine and New Hampshire, 1994-97

	Mean Blood Hg Values (ppm)				Mean Blood Hg Index†		
Location	Hg	SD	n	Range	Index	SD	n
Natural Lakes & Small Reservoirs	**0.15**	**0.22**	**17**	**0.01 - 0.79**	**0.54**	**1.00**	**17**

Little Lobster	*0.39*	-	*1*	-	*4.88*	-	*1*
Little Ossipee	*0.51*	-	*1*	-	*1.33*	-	*1*
Pierce	*0.10*	-	*1*	-	*0.83*	-	*1*
Northeast	*0.11*	-	*1*	-	*0.66*	-	*1*
Alamoosook	*0.24*	-	*1*	-	*0.59*	-	*1*
East Richardson	*0.08*	-	*1*	-	*0.31*	-	*1*
Moulton	*0.15*	-	*1*	-	*0.30*	-	*1*
Akers	*0.09*	-	*1*	-	*0.29*	-	*1*
Craig	*0.06*	-	*1*	-	*0.22*	-	*1*
Parlin	*0.03*	-	*1*	-	*0.20*	-	*1*
Echo	*0.01*	-	*1*	-	*0.10*	-	*1*
Hancock	*0.01*	-	*1*	-	*0.09*	-	*1*
Long (Belgrades)	*0.01*	-	*1*	-	*0.08*	-	*1*
Toddy	*0.01*	-	*1*	-	*0.06*	-	*1*
Lower Range	*0.01*	-	*1*	-	*0.06*	-	*1*
Pennesseewasse	*0.01*	-	*1*	-	*0.05*	-	*1*
Little Indian	*0.01*	-	*1*	-	*0.05*	-	*1*
Reservoirs	**0.25**	**0.31**	**39**	**0.01 - 1.66**	**1.20**	**1.10**	**39**
Flagstaff	*0.51*	*0.53*	*8*	*0.12 - 1.66*	*2.27*	*1.37*	*8*
Aziscohos	*0.39*	*0.19*	*7*	*0.21 - 0.78*	*2.06*	*0.43*	*7*
Lake Umbagog	*0.16*	*0.13*	*11*	*0.01 - 0.40*	*0.85*	*0.76*	*11*
Indian Pond	*0.19*	-	*1*	-	*0.76*	-	*1*
Pontook	*0.14*	*0.02*	*2*	*0.13 - 0.15*	*0.72*	*0.03*	*2*
Rangeley	*0.16*	*0.09*	*2*	*0.10 - 0.22*	*0.51*	*0.07*	*2*
Mooselookmeguntic	*0.06*	*0.12*	*6*	*0.01 - 0.30*	*0.23*	*0.32*	*6*
Richardson	*0.02*	*0.01*	*3*	*0.01 - 0.03*	*0.12*	*0.01*	*3*
ALL SITES	**0.22**	**0.28**	**56**	**0.01 -1.66**	**1.10**	**1.24**	**56**

†Index calculated as follows: {Individual blood Hg (ppm)/body weight (g)} X 10,000 = (ppmHg/g) x 10^4

Regression analyses using Pearson Correlation was performed for 12 lake chemistry, morphometric, and hydrology factors. Juvenile Common Loon blood index (weight adjusted) correlates with annual water level fluctuations (R^2=0.58, p<0.001). Analysis was limited however, to lakes with reliable water level fluctuation information. We predict that the addition of water level fluctuation data from several high Hg natural lakes to this analysis will show no significant relationship with water level fluctuations. We detected a marginally significant correlation between water color and juvenile loon blood Hg index (R^2=0.15, p=0.08). We found no relationship between juvenile blood Hg index and any other morphometric and water quality parameter.

Weight adjusted juvenile blood Hg levels were best predicted by annual water level fluctuations through a stepwise multiple regression analysis (R^2=0.82, p<0.001). Flushing rates were also significantly related to blood Hg levels (p=0.04). Stepwise multiple regressions were limited, however, to the 15 lakes where both water level fluctuations and juvenile blood Hg were measured. Common Loon tissue was collected from 54 lakes. These tests could be performed again after additional water level fluctuation data and exposed lake substrate area is collected.

We also compared mercury exposure on 25 study lakes (8 reservoirs and 17 natural lakes) divided into five categories sorted by annual water level fluctuations and lake type (Figure 10, Table 6). Mean juvenile blood Hg and a weight-adjusted index were highest for flashy natural lakes and regulated storage reservoirs and lowest for stable natural lakes and regulated full

ponds. Sample size was limiting for regulated peaking reservoirs. We detected no significant differences among lake types ($p>0.05$), possibly due to low sample size in some lake categories. However, when we stratified the data from categorical to continuous we found a significant correlation between juvenile blood Hg concentrations (weight adjusted) and greater water level fluctuations for three time periods: annual ($R^2=0.58$, $p<0.001$), summer ($R^2=0.39$, $p=0.02$), and nesting seasons' ($R^2=0.31$, $p=0.05$).

A lake by lake comparison shows that flashy natural lakes and some regulated storage reservoirs may have the capacity to contain high levels of MeHg. Stable natural lakes and regulated peaking reservoirs have mean juvenile blood Hg indices of 0.30 or lower. The natural lake Little Lobster was higher than any other waterbody sampled. While Aziscohos and Flagstaff reservoirs were significantly higher ($p<0.05$) than the other reservoirs (regulated storage and peaking) and the remaining reservoirs were not significantly different from each other ($p>0.05$).

Adult feather Hg concentrations

Feather tissue collection provides a simple and non-invasive method for detecting body burdens of Hg concentrations in wild birds. Feathers are widely recognized as major excretory pathways for Hg allowing adult birds to sequester 70-93% of their Hg body burden (Honda et al. 1985, 1986, Braune and Gaskin 1987, Burger 1993). Mean feather Hg concentration of adult loons was 7-10x greater in feathers than blood, illustrating the importance of feather Hg sequestering. Feathers are also indicators of chronic Hg body burdens. Evers et al. (1998) found recaptured loons showed significant Hg accumulation in their feathers over time, particularly for individuals with elevated blood Hg levels during the breeding season. Mercury ingested in prey during the loon's 6-7 months on Maine's breeding lakes is deposited in organs (e.g., liver and kidney) and muscle tissue. Although bile excretion contains some MeHg, most of the MeHg available for remobilization is found in muscle tissue. This Hg probably reenters the blood stream during stressful times such as full remigial molts. The correlation between flight feathers and breeding season blood Hg levels is strongest for individuals that are more exposed to Hg during the summer (e.g., Alaska $R^2=0.03$, New England $R^2=0.24$). Therefore, the feather Hg levels in breeding adult loons is related to their uptake of Hg in Maine lakes.

Feather Hg threshold levels vary according to feather type and bird species. Eisler (1987) considered 5 ppm as a LOAEL while 9 ppm was suggested by Heinz (1979). Scheuhammer (1991) and Thompson (1997) considered a higher risk threshold of 20 ppm and the co-authors have observed abnormal behavior in loons above 30 ppm. Adult Hg levels in loons, kingfishers, and mergansers were normally distributed using these various threshold levels (Table 7). Over 20% of the birds sampled in the past 4 years exceeded 20 ppm and 31% of the male loon breeding population have Hg levels over this threshold level indicating they are one of the most at risk cohorts of the New England piscivore community.

Table 7. Adult flight feather Hg levels for three target piscivorous birds in Maine, 1994-97.

Species/Sex	*<5 ppm*	*5-9 ppm*	*9-20 ppm*	*20-30 ppm*	*>30 ppm*	*Total*
Common Loon						
Male	0	7	39	16	5	67

Female	2	18	40	6	0	66
Belted Kingfisher	4	1	0	2	0	7
Common Merganser	0	0	2	0	0	2
Total	**6**	**26**	**81**	**24**	**5**	**142**

Mean feather Hg concentrations were 12.7 +/- 5.3 ppm for females and 17.2 +/- 7.3 ppm for males (Table 8, Figure 11-12). Compared to Alaskan levels, New England counterparts have 2.4x (females) and 3.1x (males) more feather Hg (Evers et al. 1998). Although age and individual stress levels were related to feather Hg levels, patterns of toxicity were similar to the blood Hg levels. Males from 6 natural lakes and 6 reservoirs had feather Hg levels above 20 ppm (29% of the lakes sampled), while high risk females were found on fewer lakes (12%). Simple and stepwise multiple regression indicated no relationship with feather Hg levels and water level fluctuations ($p>0.05$). Loons from higher exposed lakes tended to have the highest Hg levels.

6. Relationships of factors with biotic Hg concentrations from this study

We collected data for 14 of the parameters listed in section one and then related them to the Hg exposure levels found in fish and piscivorous birds. We selected more influential parameters including measures of acidity, apparent color (a predictor of DOC), shoreline and watershed vegetation and type, and magnitude and timing of water level changes (R. Harris, pers. com.). The Maine DEP and Central Maine Power Company contributed to databases created for lake morphometric, water chemistry, and water quality parameters (Appendix I & II). A stepwise multiple regression using 9 parameters as independent variables and loon blood Hg concentrations as the dependent variable indicates that annual and summer water level fluctuations may explain much of the variability of juvenile loon blood Hg concentrations. Lake acidity, measured by pH or total alkalinity, explained much of the variation in adult blood Hg concentrations although when we conducted simple linear regression analysis, annual and summer water level fluctuations were significantly correlated with higher adult female blood Hg concentrations and lake acidity did not.

The findings from these continuous data were supplemented with categorical data. Since annual, summer (May 1 to October 1), and nesting season (May 15 to July 15) water level tend to correlate we used annual fluctuations as the independent factor to separate lakes into 5 categories: (1) stable natural lakes, (2) regulated full ponds, (3) regulated peaking reservoirs, (4) flashy natural lakes, and (5) regulated storage reservoirs. Analysis of categorical data using ANOVA indicated that there was a significant difference among all categories ($p<0.05$), except regulated peaking reservoirs that were omitted from the analysis due to small sample size. Tukey's HSD post-hoc test indicated that storage and hydroelectric reservoirs with annual water level fluctuations greater than 1.5m had significantly higher Hg levels in adult and juvenile loon blood ($p<0.05$) than stable natural lakes. Adult and juvenile loon blood Hg levels were not significantly different among the other lake classifications.

Acidity is also a known contributor to enhanced Hg productivity. In a Wisconsin study, acidic lakes were associated with higher loon blood Hg levels and significantly lower loon

reproductive success (Meyer et al. 1995, 1998). Lakes sampled in this study ranged from 5.5 to 7.8. Effects of lake acidity were not statistically apparent ($p>0.05$ for pH and total alkalinity) although our range of lake acidity was relatively narrow and only 5 lakes were below the 6.3 pH threshold level that was found to have effects on juvenile blood Hg levels in Wisconsin.

Although we only elaborate on these two factors, we feel that the unaccounted variability in our statistical tests may be largely related to physiognamy. New data indicates that topography and vegetation types can be major contributors to a lake system. Preliminary data from upstream sources of Flagstaff Lake such as Chain of Ponds indicate high Hg availability to fish and loons.

Water Level Fluctuations

Annual water level fluctuations significantly correlated with adult female ($R^2= 0.40$, $p<0.001$) and juvenile ($R^2= 0.37$, $p<0.001$) blood Hg concentrations, but did not correlate with adult male blood Hg concentrations. The extent of summer fluctuations and loon blood Hg concentrations were also correlated for adult females ($R^2=0.37$, $p<0.05$) and juveniles ($R^2=0.31$, $p<0.05$) indicating a relationship with increased MeHg production and availability in the summer months. Around 82% of the variability in juvenile blood Hg concentrations could be explained by annual water level fluctuations ($p<0.01$). Flushing rates and lake surface area were also useful but not significant co-predictors of Hg ($p>0.05$). These 3 parameters explained 95% of the variability in the juvenile blood Hg concentrations. Adult male blood Hg concentrations were not correlated with annual and summer water level fluctuations. This may be because males have a significantly higher body burden of Hg than females (Evers et. al. 1998). Therefore their body burden may tend to reflect a chronic uptake of Hg rather than current prey Hg uptake.

A total of 8 reservoirs and 14 flashy natural lakes had annual water level fluctuations over 1.5 m. Some reservoirs have drawdowns over 1.5 m by early fall and some natural lakes such as Attean, Long, and Pierce also have substantial water level fluctuations. Aziscohos and Flagstaff Lakes represent situations where summer water level fluctuations were greater than 1.5m and their bathymetry created large areas of recently inundated substrate. These ephemeral microhabitats are known optimal areas for anaerobic sulphur-reducing bacteria that are responsible for increased methylation rates (Watras et al. 1994, Tetra Tech 1995).

Further evidence for this relationship is the Hg exposure patterns found on Aziscohos and Flagstaff lakes. The northern end of Aziscohos has more exposed shoreline microhabitat than the southern end. Similarly, the west side of Flagstaff Lake has more exposed shoreline during drawdowns than the east side. During the normal 2-3m summer drawdowns from June through September, both northern Aziscohos and western Flagstaff lakes have extensive areas of sandy shoals and widening shorelines that eventually create large mudflats and leave only the river channel with flowing water. Conversely, southern Aziscohos and eastern Flagstaff have far less exposed sandy or mud islands created by the drawdown and their shorelines are much more narrow and are more likely to have cobble and rock substrates (particularly on Aziscohos). When we compared the Hg levels of the various biotic matrices, northern Aziscohos and western Flagstaff were higher in each category (Table 9).

Enhanced methylation may not be the only explanation for spatial differences of Hg exposure in biota. Mercury inputs to northern Aziscohos and western Flagstaff have not been quantified, but evidence indicates that mercury exposure in upstream waterbodies is high. For example, Chain of Ponds, a group of lakes upstream of Flagstaff, has the highest lake trout Hg

level recorded in Maine. Parmacheenee, a lake upstream of Aziscohos has extremely high levels of Hg in loon prey (unpubl. data). These upstream Hg sources may be significant.

MeHg production is most likely tempered on Richardson and Mooselookmeguntic reservoirs because of substrates that have less organic soils than the richer floodplain soils in Aziscohos and Flagstaff valleys. Rangeley, Wyman, and Indian reservoirs do not have summer drawdowns greater than one meter. Rangeley and Indian reservoirs have low Hg levels in fish and loon blood for adults (<2.0 ppm) and juveniles (<0.30 ppm). A small sample of loon matrices from Wyman indicates exposure to high Hg concentrations.

Table 9. Mean Hg levels (ppm) and sample size in loon and fish by reservoir area

Reservoir compartment	Forage Fish (10-60g)	Adult Blood *Male*	Adult Blood *Female*	Juv. Blood	Eggs
Flagstaff - west	0.50	4.88	3.78	0.56	none
Flagstaff - east	0.46	2.83	3.16	0.42	0.27
Aziscohos - north	none	3.85	2.36	0.39	1.31
Aziscohos - south	0.28	2.85	1.65	0.29	0.94

Natural lakes that were found with annual water level fluctuations greater than 1.5 m were characterized as "flashy natural lakes (FNL)." There is evidence that these lakes have more of a tendency to have elevated Hg levels in the biota than stable natural lakes and regulated full ponds (Figure 11). Some of these FNLs indicated very high exposure to Hg including Little Lobster, Chain of Ponds, Pierce and Attean lakes.

Lake Acidity

The acidity of a water body was determined with two measurements: pH and total alkalinity. Mercury productivity increases with water acidity. Winfrey and Rudd (1990) found that MeHg production is highest in water with pH below 7. Demethylation or volatilization of dimethylmercury (methyl Hg is converted into dimethyl Hg which is a more volatile form of organic Hg) takes place in high pH lakes (>7). Over 77% of the lakes used in the study have pH below 7, therefore determining water acidity is crucial. Mercury that is not demethylized accumulates in the lake and may concentrate in high trophic level organisms.

MeHg production and chemistry in acidified lakes may differ from more alkaline lakes due to synergistic factors. For example in acidified drainage lakes elevated concentrations of DOC positively correlated with MeHg concentrations in water and fish (McMurtry et al. 1989). Conversely, in seepage lakes where precipitation supplies most of hydrologic input, Hg concentrations in fish negatively correlated with DOC in water (Grieb et al. 1990). From other studies (Tetra Tech Inc. 1995) it appears that lakes with low pH and high DOC have higher Hg levels in fish than lakes with high pH and low or high DOC.

The surface water pH values in natural lakes of this study ranged from 5.9 at Little Beaver to 7.6 at Pennesseewassee. Reservoir pH levels were generally circumneutral. Total alkalinity correlates with pH (Lane et al. 1997). It is considered a more precise measurement of acidity

although it is more time intensive to determine. Total alkalinity ranged from 2-45 ug/L in natural lakes and 6-16 ug/L in reservoirs. Lake acidity did not significantly differ between natural lakes and reservoirs ($p>0.05$ for pH and total alkalinity).

Since lake acidity correlates negatively with mercury concentrations in loons (Lane et al. 1997) and fewer loon chicks fledge from lakes with pH values of 6.3 or less (Meyer et al. 1995), we removed loon matrices from these acidic lakes when determining Hg relationships with specific factors (e.g., water level fluctuations). This reduced the ability of acidity to confound interpretations of the effect of other factors on Hg availability. Only 5% of the lakes measured from 1993-97 have pH levels below 6.3.

Unlike our Wisconsin study, simple linear regression analysis of lakes with annual water level fluctuations less than 1.5 meters indicated no significant relationship with lake acidity in our study ($p>0.05$) probably partly because of a limited pH and alkalinity range.

7. Study lake characterization for MeHg bioavailability

To determine if water level fluctuations affect MeHg productivity and availability, we sampled sediments, forage fish, gamefish, and bird blood, feathers, and eggs. Patterns of Hg concentrations in abiotic and biotic matrices were difficult to determine for the waterbodies except for extent of annual drawdown (the greater the drawdown the higher the Hg concentrations). Therefore, we treat each target reservoir separately and describe the factors that contributed the most toward MeHg productivity and availability and their relationship with our measured biotic matrices. Because annual and summer water level fluctuations are significantly correlated with adult female and juvenile loon blood Hg concentrations (see section 2) we emphasize this parameter for each lake characterization. Benchmarks for assessing risk are provided for context.

Comparisons are made among 9 reservoirs and 35 natural lakes. Factors related to MeHg bioavailability are discussed for each reservoir and two groups of natural lakes and are then compared with the 5 water fluctuation categories. Loon and fish Hg concentrations are given by waterbody (natural lakes grouped) and relevant comparisons follow. Individual natural lake characterizations are not given. Instead lakes are grouped according to hydrology (i.e., annual and summer water level fluctuations) or water chemistry (pH or total alkalinity). Some natural lakes have extensive water level changes and are characterized by a large watershed-small surface area ratio. Several of the storage reservoirs generally exhibit over a 1.5 m drawdown from May to October (except regulated full ponds) (Table 10). All of the reservoirs undergo the largest drawdowns during a period of slower mercury methylization in late winter. The two peaking reservoirs regularly fluctuate 0.5-1 m weekly but do not exhibit more than 1.5 m summer drawdowns.

There are several benchmarks that we use in the following lake characterizations. Top layer sediment Hg concentrations (unadjusted by %TOC) exceeding 0.20 ppm (dw) are considered elevated by Maine DEP standards (Persaud et al. 1993). Barr (1986) documented lowered productivity by adult loons eating fish with 0.30 ppm Hg (lower impact threshold - LIT) and no productivity when foraging on fish with over 0.40 ppm Hg (upper impact threshold - UIT). Adult blood Hg levels exceeding 3.00 ppm are considered elevated. We have yet to

establish a benchmark for juvenile blood Hg concentrations that are adjusted for weight, however, young loons over 1.00 ppm Hg/g x 10^4 are in waterbodies with elevated Hg levels. Unadjusted juvenile blood Hg levels over 0.30 ppm are considered elevated (Meyer et al. 1998). Feather Hg levels over 20 ppm indicate impact thresholds (Scheuhammer 1991). Average adult loon survivorship is 80% (Evers et al. 1996) and the percentage of loon territories producing fledged young in natural lakes in New Hampshire is 35% or (0.52 fledged young / territory) (K. Taylor, pers. com.).

Table 10. Mean Hg levels in Common Loon matrices sampled in 9 reservoirs and their summarized risk rating.

% Matrices	Sed	Fish			Whole	Adult -Blood		Adult Feather		Juv Blood		Rating
		Game	Forage	Perch	Egg	Male	Fem	Male	Fem	blood	Index	Exceeded
Threshold Levels	*0.20*	*0.2,0.7*	*0.3, 0.4*		*0.5,1*	*3.0*		*20.0*		*0.3*	-	
Wyman	-	-	-	-	2.49*	2.9	-	25.2	-	-	-	75%
Flagstaff	0.22*	0.71*	0.63*	0.42*	0.27	4.5*	3.6*	21.7*	15.3	0.51*	2.27	71%
Aziscohos	0.25*	-	0.28	-	1.27*	3.3*	2.3	21.8*	15.6	0.39*	2.06	70%
Mooselook	0.20*	0.92*	0.29	0.23	0.85*	1.8	1.3	14.0	8.5	0.07	0.23	29%
Richardson	0.24*	0.43*	0.26	0.16	-	1.6	1.6	18.9	20.6	0.02	0.12	25%
Indian	0.24*	0.28*	0.29	0.25	-	1.8	1.4	12.6	11.4	0.19	0.76	17%
Umbagog	0.19	-	0.28	0.18	0.90*	2.3	1.0	16.1	9.8	0.16	0.85	8%
Rangeley	-	-	-	-	-	1.4	1.0	10.3	9.3	0.16	0.51	0%

* Exceeds designated threshold level

Sampled matrices were compared with our designated threshold levels to determine their capacity to produce enough MeHg to threatens wildlife. If we summarize the various abiotic and biotic matrices by reservoir (Table 10) and lake (Table 11) category a more comprehensive and quantifiable evaluation can be made. A reservoir by reservoir analysis indicates tremendous differences in Hg risk to wildlife. Some reservoirs such as Rangeley Lake indicate low overall lake and individual loon risks. While Lake Umbagog rates low, a closer look does show differing Hg availability. The Magalloway River loon pairs have higher Hg exposure than those on the lake which have higher levels that those on the Androscoggin River. Indian, Mooselookmeguntic, and Richardson were moderately low while Wyman, Aziscohos, and Flagstaff were high. Although sample size and relevant threshold levels are crucial for reliable interpretation, flooded river valleys with or without water level fluctuations over 1.5 m have higher Hg exposure than other reservoir types.

When we categorized reservoirs and natural lakes according to their hydrology we found the percentage of matrices exceeding the designated thresholds ranged from 14-29 percent (Table 11). Regulated full ponds were the lowest and flashy natural lakes were highest. Five of the 9 target reservoirs fell into our category “regulated storage reservoirs (RSR)” even though they have different origins that have significant impacts on Hg production and availability. Therefore we divided reservoirs that originated from river valleys (i.e., Aziscohos and Flagstaff) from those that were historically natural lakes (i.e., Mooselookmeguntic, Richardson, and Umbagog) (Figure

13). That sub-designation indicates that not all RSRs are inherently highly exposed.

Table 11. Mean Hg levels in Common Loon matrices by five lake categories with summarized risk rating.

% Matrices	Sed	Fish			Whole	Adult -Blood		Adult Feather		Juv Blood		Rating
		Game	Forage	Perch	Egg	Male	Fem	Male	Fem	blood	Index	Exceeded
Threshold Levels	*0.20*	*0.2,0.7*	*0.3, 0.4*		*0.5,1*	*3.0*		*20.0*		*0.3*	-	
LAKE CATEGORY (sample size)												
Flashy Natural (14)	0.27*	0.39*	0.35*	0.26	0.74*	2.7	1.9	15.2	13.5	0.32*	1.39	33%
Reg. Storage Res (5)	0.22*	0.69*	0.35*	0.25	0.82*	2.7	2.0	18.5	14.0	0.23	1.11	29%
*(river valleys**)*	*0.24**	*0.71**	*0.46**	*0.42**	*0.77**	*3.9**	*3.0**	*21.8**	*15.5*	*0.30**	*2.17*	*80%*
*(old lakes **)*	*0.21**	*0.68**	*0.28*	*0.19*	*0.88**	*1.9*	*1.1*	*16.3*	*9.2*	*0.08*	*0.40*	*20%*
Reg. Peaking (2)	0.24*	0.28*	0.29*	0.25	2.49*	2.4	1.4	18.9	11.4	0.19	0.76	25%
Stable Natural (17)	0.30*	0.31*	0.37*	0.31*	0.46	1.7	1.1	18.6	11.3	0.05	0.19	23%
Regulated Full (17)	0.25*	-	-	-	0.91*	2.2	1.1	12.8	10.7	0.09	0.24	14%

* Exceeds designated threshold level

** RSRs were separated based on origin. Aziscohos and Flagstaff represent river valley origins while Mooselookmeguntic, Richardson, and Umbagog were old lakes.

We ordered the descriptions of the following 9 target reservoirs according to their Hg risk.

1. Wyman Lake (RPR): This peaking reservoir originally was a river valley that was flooded 70 years ago. Annual and summer water level fluctuations average less than 3 feet. The exposed shoreline is primarily bedrock and glacial till with few wetland areas. Water retention times are low. Upstream impacts are potentially high from Flagstaff Lake and Chain of Ponds because loon Hg samples from these lakes were well above Maine's average.

No sediment or fish samples have been collected from this reservoir. One egg and blood and feathers from one adult male collected for the Maine Outdoor Heritage Fund study indicate high exposure. The egg Hg level was the highest recorded in Maine (n=45 eggs) at 2.5 ppm. The males blood and feather Hg levels were above threshold levels. Hg risk rating was high - 75%.

2. Flagstaff Lake (RSR): This reservoir is characterized by numerous factors that relate to high MeHg productivity and bioavailability. It has extensive shoreline and watershed wetlands including peatlands. Water retention times are high and water temperatures are warm especially in the photic and littoral zones. The extensive shoreline wetlands, shallow water areas, and near

surface substrates on Flagstaff are conducive to MeHg production because of high inputs of organic matter and the extensive oxic-anoxic environments that are created during mid to late summer drawdowns. The drawdowns and influx of dissolved and suspended organic matter also contribute to enhanced methylation. Tributary influences are probably a significant component to Hg input to the lake (based on Maine DEP fish sampling (the highest Hg level for lake trout in ME) and a 5.5 ppm blood sample from an adult male loon on Chain of Ponds). Tributary Hg sources may be related to the topographical relief and land management practices in the watershed.

The top sediment layer was 0.22 ppm of Hg. Fish Hg levels were high in most all species and size classes sampled and Yellow Perch Hg levels were higher than all other reservoirs and lakes in all four size classes, exceeding the UIT for all sizes. Extra large perch were exceedingly high (1.38 ppm). Large White Sucker, large Chain Pickerel, medium Golden Shiner also exceeded the UIT while the LIT was surpassed by medium sized Brown Bullhead and Rainbow Smelt.

Mean adult loon blood Hg concentration (4.5 ppm in males and 3.6 ppm in females) was higher on Flagstaff than the other study reservoirs and mean juvenile blood-Hg index (adjusted for weight) was also the highest (2.27 ppm Hg / gram x 1,000). Feather Hg levels indicate a relationship with high summer blood Hg levels. One egg was collected from the Jerome Brook territory and had Hg concentrations below the threshold level. Western lake Hg exposure is apparently significantly higher than the east end and may be related to high Hg inputs from Cahin of Ponds and extensive exposed sediments in late summer. Hg risk rating was high - 71%.

3. Aziscohos Lake (RSR): This reservoir has the highest 30-year mean annual water fluctuations (20 feet). Riverine input of Hg from the Little Magalloway and Magalloway rivers is a potentially significant factor even though the total watershed area they drain is relatively small (214 sq. mi.). Mid-summer low water levels can expose extensive, formerly-inundated shoreline substrates. This transitional area of substrate and subsequent input of organic material (resuspension and erosion) creates optimal habitat for methylation by microbes. Wetland acreage (low peat shoreline acreage) and water temperatures are potentially moderately related to MeHg availability. Loon territories with potential increased MeHg inputs include most of those on the north and west end of the lake.

Sediment Hg levels exceeded the MDEP benchmark for total Hg (0.25 ppm). Yellow Perch are not present on Aziscohos. The LIT was exceeded by large white sucker, small Rainbow Smelt and small Fallfish. The UIT was exceeded by small Common Shiner (0.44 ppm), small Longnose Sucker (0.77 ppm) and small Northern Redbelly Dace (0.41 ppm). Loon adult mean blood Hg concentration were 3.3 ppm for males and 2.3 ppm for females and were the second highest level recorded for the target reservoirs. The juvenile blood Hg index was 2.06 and was considered high. Hg risk rating was high - 70%.

4. Mooselookmeguntic Lake (RSR): This reservoir's hydrology and geochemistry is similar to Richardson Lake with few variables related to enhanced MeHg bioavailability. This reservoir has the lowest pH (3 site grab sample mean=6.2) which may contribute to increased Hg methylation. Specific areas with potential increased dewatered substrates and shoreline wetland Hg input and pH effect include the north end of Cupsuptic Lake and the riverine territory, near

Upper Dam, and Bemis (particularly during late summer drawdowns). Exposed substrates on the shore tend to be alluvial sand beaches. Reservoir age, flushing rates, water temperature and soil types probably are minor contributors to lake MeHg production and bioavailability.

Sediment Hg levels were relatively low at the MDEP benchmark. Mean Yellow Perch Hg levels for all size classes was 0.23 ppm while all forage fish measured averaged 0.29 ppm. Yellow Perch fillet Hg concentrations averaged 0.92 ppm and were higher than the state average (0.27 ppm) and exceeded the MDEP upper limit. Although loon prey items were just below LIT, loon matrices indicate low levels of exposure to higher trophic web organisms. Adult blood-Hg concentrations were 1.8 ppm for males and 1.3 ppm for females. Low Hg concentrations were also found for indexed juvenile blood (0.23 ppm Hg / gram x 1,000). Two eggs collected from North Toothaker Island exhibited elevated levels of Hg (mean was 0.85 ppm). Hg risk rating was moderately low - 29%.

5. Richardson Lake (RSR): Richardson Lake had few factors related to enhanced MeHg bioavailability except for water level fluctuations (30 year mean is 10 feet). There are only select areas such as Cranberry Cove, Pine Island, and the mouth of Fish-Beaver Brook that have high potential for enhanced MeHg production because of shoreline wetland acreage, water temperature, water chemistry, or upstream inputs. Lowered water levels in these areas during late summer could contribute to enhanced Hg methylation. Most of the exposed substrates on Richardson Lake are also alluvial sand beaches with very little organic matter. Reservoir age, flushing rates, and soil types likely have minor contributions to lake MeHg production and bioavailability.

The upper sediment layer exceeded the MDEP benchmark. Mean Hg levels for Yellow Perch (0.16 ppm) and all forage fish (0.26 ppm) indicate low exposure for fish-eating wildlife. Landlocked Salmon fillet Hg concentrations were 0.514 ppm and exceeded MDEP lower limit acceptable levels. Results from Richardson indicate relatively low adult blood (1.6 ppm) and indexed juvenile blood (0.12 ppm Hg / gram x 1,000) Hg concentrations. Adult feather Hg concentrations were high on this reservoir (mean=19.8 ppm) and probably indicate a bias toward the sampling of older birds from the breeding population. Hg risk rating was low - 25%.

6. Indian Pond (RPR): Unlike the other study reservoirs, Indian has minimal seasonal water level fluctuations (3 feet) yet a high flushing rate. This former river gorge was flooded 45 years ago and has a minimal direct watershed. The inflow for Indian Pond comes from Moosehead Lake. Preliminary MDEP Hg data from Moosehead Lake indicates upstream Hg inputs are minimal. Shoreline wetland acreage is moderate and in some areas, such as the northwest and northeast corners, organic particulate and Hg input most likely enhances MeHg bioavailability.

Sediment Hg levels exceeded the MDEP benchmark. Unlike other reservoirs and natural lakes, Yellow Perch and White Sucker Hg levels did not exceed the LIT and UIT in any of the size classes. Smallmouth Bass fillet Hg concentrations were 0.28 +/- 0.11 ppm (n=17) and were lower than the statewide average of 0.77 +/- 0.51 ppm. Loon matrices indicate low adult blood (mean = 1.70 ppm) and indexed juvenile blood (0.19 ppm Hg / gram x 1,000) Hg concentrations. Hg risk rating was low - 17%.

7. Umbagog (RSR): Factors that contribute to MeHg production and bioavailability include the

large watershed, extensive shoreline and photic zone acreage, warm water temperature, and high amount of organic material. Reservoir age and flushing rates have minimal impact on MeHg bioavailability. Annual water level fluctuations are moderate (6 feet) and summer changes are minimal due to regulation restrictions. Upstream inputs of Hg may be substantial, but have not been quantified. Adult and juvenile loons from territories on the Magalloway River, one inlet of Umbagog and the outlet of Aziscohos lake, have higher levels of Hg than those birds on Lake Umbagog. Because Aziscohos has elevated Hg concentrations measured in loons and fish, downstream Hg transport is possibility. Another possibility is the extensive wetlands in this part of the Magalloway River. Hg exposure on the Androscoggin River (an outlet of Lake Umbagog) is lower than levels in the lake basin.

Sediment Hg levels were just under the MDEP benchmark. Of the four size classes of Yellow Perch sampled, the extra large class exceeded the UIT (as did extra large Bullhead). Two species exceeded the LIT including large and extra large Chain Pickerel and medium, large, and extra large Fallfish. Results of adult loon sampling since 1994 indicate a mean blood Hg concentration of 2.3 ppm for males and 1.0 ppm for females. Several males had blood Hg levels that exceeded our designated 3.0 ppm impact threshold. Indexed juvenile blood Hg concentrations were 0.16 ppm of Hg / g x 1,000. The mean egg Hg level for 22 samples was 0.90 ppm and was elevated. The Hg risk rating was low (8%) for all pairs associated with Lake Umbagog, however the rating increases for the 3 loon territories inhabiting the Magalloway River.

8. Rangeley Lake (RFP): A highly developed shoreline with a large density of homes and cottages characterizes this reservoir and sets it apart from the other study reservoirs. Water levels are regulated for stable full pond and changes are minimal during the summer. Winter drawdowns are up to 4 feet. Upstream inputs appear to be minimal and may be related to the relatively small drainage area (99 square miles). Flushing rates are low but the water chemistry is not conducive to high MeHg production. Much of the shoreline contains upland habitats except for a cove with extensive bog mats on the southwest corner.

The mean adult male blood levels was 1.4 ppm and the female was 1.0 ppm. The juvenile blood index was a low 0.51 ppm Hg / g x 1,000 but was higher than Mooselookmeguntic and Richardson. Adult feather Hg levels were low. Our sampling efforts so far show no risk of Hg to loons or other wildlife.

9. Pontook Lake (RFP): This small reservoir has water levels that are maintained for minimal changes. Inputs of Hg from the Androscoggin River are low as indicated by upstream loon territories (Harper's and Sweats Meadow). There are extensive areas of emergent wetlands that have the potential for increasing MeHg production however the flushing rate is also moderately high. Analysis of sediment and fish sampling is not complete, however adult and juvenile blood Hg levels are low. Sampling efforts so far indicate no risk of Hg to loons or other wildlife.

Natural Lakes: Control or reference natural lakes are being studied to compare potential MeHg exposure to loons and their prey. These lakes have been placed in two designations based on hydrology. Flashy natural lakes have annual water level changes over 1.5 meters and are usually characterized by large watersheds compared to the lake's surface area. Stable natural lakes have

annual water level changes less than 1.5 meters and many are spring fed. Because there is a demonstrated strong negative correlation with MeHg availability and acidity we have also designated 3 natural lakes as acidic and removed them from some of the comparative analysis to avoid additional confounding factors (i.e., Little Beaver, West Carry, and Kennebunk).

Lakes with Wide Water Level Fluctuations (FNL)

We collected loon blood, feather, or egg samples from 14 lakes with dynamic hydrologies fitting into this category. Some lakes such as Attean, Long, and Parmachenee fluctuated 8-11 feet but most hovered around 1.5 meters (5 feet). Sediment Hg samples from 3 lakes had a mean of 0.27 ppm and exceeded the MDEP benchmark for total Hg concentrations. Forage fish exceeded the LIT however perch did not for the 3 lakes sampled (Attean, Long, and Pierce). The mean egg Hg level was 0.74 ppm and exceeded the lower threshold level. Mean adult blood levels for males (2.7 ppm) and females (1.9 ppm) were similar to storage reservoirs and generally exceeded other lake categories (Table 11). Juvenile blood Hg indexed for weight was also higher than most lake situations indicating that these flashy natural lakes are at higher risk than those natural lakes with more stable water regimes. Little Lobster and Chain of Ponds had extremely high levels of blood Hg in adults and juveniles. The Hg risk rating was moderate for this category - 33%.

Lakes with minimal Water Level Fluctuations (SNL & RFP)

A total of 34 lakes had minimal water changes through the year. Three of these lakes were confounded by high acidity levels and 17 lakes had water levels regulated by check dams to dampen minor changes. These two lake categories, especially lakes categorized as regulated full ponds provide reference for interpretations and provide context for the other categories impacted by hydrological manipulations. Mean adult and juvenile blood Hg levels were the lowest in these two categories. The mean juvenile blood Hg index (ppm Hg / g x 1,000) for SNL was 0.19 and for RFP was 0.24. These levels are approximately 10x lower than the highest at risk category (RSR with a river valley origin). The Hg risk rating was low for SNL and RFP - 14 and 23%, respectively.

Conclusions

This study quantified MeHg distribution and availability in aquatic systems of northern Maine and New Hampshire. It was conducted in collaboration with a network of member agencies and organizations of the Northeast Loon Study Workgroup. Nearly 2,000 hours were contributed by 30-40 people in 1996 and 1997. Sampling by a concurrent study in 1997 funded by the Maine Outdoor Heritage Program and administered by the Maine Department of Environmental Protection greatly increased the samples from natural lakes (Evers and Reaman 1998).

This report summarizes Hg exposure data we collected in aquatic wildlife over a four year period. We have found a number of trends and tendencies verified by statistical tests. However, the biological significance of these findings is under investigation. We are currently determining the potential affects of Hg on reproduction and behavior. The following is a summary of our conclusions:

1. FACTORS RELATED TO MeHg AVAILABILITY IN WATERBODIES FOR THIS STUDY

- No single factor primarily accounts for MeHg exposure in waterbodies
- Lakes designated as flashy natural lakes generally have large watersheds that may be responsible for the highest Hg levels we recorded in this study.
- Water level fluctuations could be affecting Hg availability on both natural lakes and reservoirs, however, other factors such as watershed inputs, wetland inputs, tributaries, soils and precipitation runoff could all be equally or more important.
- Several high Hg waterbodies in this study have upstream lakes and ponds with high Hg exposure measured in biota. The contribution of these potential Hg flux inputs needs to be evaluated.
- Both natural and regulated water level fluctuations are at least partially related to MeHg enhancement. This factor is further discussed in sections 6 and 7.
- Flushing rates are important to reducing MeHg availability in reservoirs such as Indian Pond.
- The origin of reservoirs may be one of the most important contributing factors for MeHg enhancement. Reservoirs with river valley origins (i.e., Aziscohos, Flagstaff and Wyman) are more likely to have higher Hg concentrations in their biota than reservoirs that originated from raising the water levels of existing lakes (i.e., Mooselookmeguntic, Richardson, and Umbagog).
- Soil type is related to reservoir origin. Soils found in river floodplains are higher quality microhabitats for MeHg production versus glacial till soils found along lake shorelines.
- Mountainous terrain may be more likely to capture atmospheric Hg and recent studies indicate coniferous forests are 3 times more productive than deciduous forests for MeHg. Contributions from these potentially important sources is unknown.
- Reservoirs and most natural lakes were generally circumneutral in acidity and had similar dissolved organic carbon and apparent color measurements, thereby providing adequate comparisons between reservoirs and most natural lakes.
- Reservoir age is probably not a contributing factor toward MeHg enhancement.

2. SEDIMENTS

- Lake sediment total Hg (dw) of the top layer (0-10cm) exceeded the 0.20 ppm MEDEP benchmark of lowest effect levels in 5 of the 7 natural lakes and in 5 of the 6 reservoirs tested.
- Lake sediment total Hg adjusted for percent total organic carbon was not significantly different between natural lakes and reservoirs and suggests our study lakes are receiving similar levels of Hg deposition statewide.

3. GAMEFISH HG AND ITS RELATIONSHIP WITH LOON BLOOD

- The mean mercury concentrations for Yellow Perch, Land-locked Salmon, and Brook Trout fillets (natural lakes and reservoirs combined) exceed the MDEP Fish Consumption Advisory of 0.20 ppm for pregnant women, women of childbearing years and children of 8 years and under.
- The mean mercury concentrations for Yellow Perch, Land-locked Salmon, and Brook Trout

fillets (natural lakes and reservoirs combined) did not exceed the MDEP Fish Consumption Advisory of 0.70 ppm for the general population, however several individual fish fillet samples exceeded this limit.

- Samples of extra large (>80 grams) Yellow Perch fillets did not have Hg concentrations that significantly correlated with adult or juvenile loon blood Hg concentrations.
- Fish fillet Hg concentrations for a number of species did not significantly correlate with adult or juvenile loon blood Hg concentrations.

4. LOON PREY HG AND THEIR RELATIONSHIP WITH LOON BLOOD

- Larger size classes of fish have higher Hg concentrations than smaller size classes.
- Yellow Perch Hg have a significantly strong correlation with adult loon blood Hg indicating probable selection of this species by loons and possible primary exposure of Hg from perch.
- Hg concentrations in adult male loon blood Hg and medium-sized fish were highly correlated.
- Juvenile loon blood Hg index did not correlate with Yellow Perch or White Sucker, and juvenile Hg levels are probably more affected by a more varied forage base.
- The 3 natural lakes and 4 of the 5 reservoirs (excluding Indian Pond) had at least one size class of perch over the lower impact threshold (0.30) for loon productivity.
- Some minnow and other selected species of potential loon prey had Hg levels similar or higher than piscivorous species and larger omnivores.

5. LOON EGG HG CONCENTRATIONS

- Of 43 eggs analyzed from Maine lakes, 77% had Hg levels greater than 0.50 ppm (lowest observed adverse effects level) and 32% were greater than 1.00 ppm (lowest observed adverse effects level in loons based on a Hg study on loons at a contaminated site (Barr, 1986)).
- Mean egg Hg concentrations exceeded 1.00 ppm on Wyman reservoir, Brewer Lake, and Aziscohos reservoir. At least one egg from Wyman and Aziscohos reservoirs exceeded 2.00 ppm.
- Sample sizes of eggs from reservoirs and natural lakes with known water level fluctuations did not allow parametric statistical testing.

6. LOON BLOOD AND FEATHER HG CONCENTRATIONS

- Adult and juvenile blood Hg concentrations were elevated across Maine and New Hampshire and are significantly higher than Hg levels found in loons in the Upper Great Lakes, Pacific Northwest, and Alaska.
- Males had significantly higher blood and feather levels than females and are probably more at risk. Over 30% of the males had feather Hg levels above our designated 20 ppm threshold.
- Adult blood Hg levels from stable natural lakes with circumneutral acidity were significantly lower than regulated storage reservoirs (RSRs). When we separated RSRs according to origin, only those originating from river valleys had Hg levels that were significantly higher than those on stable natural lakes.
- Adult blood Hg levels from regulated storage reservoirs (RSR) were not significantly

different than flashy natural lakes (FNL) or regulated full ponds (RFP).

- Juvenile blood Hg levels were significantly correlated with age (indicated by weight) and were therefore adjusted as an index for comparative purposes.
- Juvenile blood Hg index levels were highest on Flashy Natural Lakes (FNLs) and RSRs and lowest on Regulated Full Ponds (RFPs) and Stable Natural Lakes (SNLs).
- Because FNLs and RSRs tended to have the highest adult and juvenile blood Hg concentrations further statistical analysis using continuous vs. categorical data for stepwise multiple and simple regressions were used to show significant positive correlations between adult and juvenile blood Hg concentrations and annual and summer water level fluctuations.

7. RELATIONSHIP OF FACTORS INFLUENCING HG AVAILABLITY

- Determining the importance of the single factor of water level fluctuations availability is extremely difficult to assess due to the interrelationship of other contributing factors.
- Water level fluctuations are enhancing MeHg availability on some reservoirs and natural lakes, however other factors such as soil types, topography, watershed cover types, and tributary inputs may greatly contribute to MeHg enhancement.
- Summer (May 1 - October 1) and annual water level fluctuations on all of the reservoirs and a sub-sample of natural lakes significantly correlated with female blood Hg and juvenile blood Hg index. It did not significantly correlate with adult male blood Hg or egg Hg concentrations
- The statistical testing of 14 lake morphometric and water chemistry factors with a Pearson's Correlation Coefficient Matrix showed significant relationships with the independent variable (adult or juvenile blood Hg) for annual and summer water level fluctuations.
- Statistical tests were limited to a handful of lakes with reliable water level fluctuation data and the biological significance of these findings needs to be evaluated.

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