# AN EXPLORATORY STUDY OF METHYLMERCURY AVAILABILITY IN TERRESTRIAL WILDLIFE OF NEW YORK AND PENNSYLVANIA, 2005-2006

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NEW YORK STATE ENERGY RESEARCH AND DEVELOPMENT AUTHORITY





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Prepared for the

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#### **Overall Abstract**

Atmospheric deposition of sulfur, nitrogen, and mercury (Hg) has potential widespread and severe ramifications to ecosystem health and therefore it is critical that the impact of these pollutants and the ecological response be quantified. It is plausible that elevated methylmercury (MeHg) levels and depleted available calcium levels, particularly within acidic environments, are negatively contributing to the viability of wildlife populations. We sampled organic soils, Ca-rich invertebrates, herpetofauna, bats, and breeding songbirds from multiple sites in New York and Pennsylvania. We demonstrate three major outcomes from this exploratory study: (1) a better understanding of the dynamic relationship within terrestrial, acid-impacted ecosystems for Hg and Ca in wildlife, (2) added value to the identification of biological hotspots, and (3) improved information that will be used to identify long-term monitoring locations for a forthcoming national Hg monitoring program.

Keywords: Mercury, Songbird, Bats, Herpetofauna, Invertebrates

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#### SECTION I – BIRD MERCURY EXPOSURE PROFILE

#### **Abstract**

Atmospheric deposition of sulfur, nitrogen, and mercury (Hg) has potential widespread and severe ramifications to ecosystem health and therefore it is critical that the impact of these pollutants and the ecological response be quantified. It is plausible that elevated methylmercury (MeHg) levels and depleted available calcium levels, particularly within acidic environments, are negatively contributing to the viability of songbird populations. We build on existing work in northeastern North America that demonstrates the potential landscape-level effects of acidification on pools of Ca and on Ca-rich invertebrate prey populations, and trace the transfer and fate of MeHg in songbirds. We sampled organic soils, Ca-rich invertebrates, and breeding songbirds (e.g., thrushes, vireos, and blackbirds) from 20 stations in New York and Pennsylvania. We demonstrate four major outcomes from this exploratory study: (1) a better understanding of the dynamic relationship within terrestrial, acid-impacted ecosystems for Hg and Ca, (2) a spatial gradient for linking major Hg emission sources in the Ohio River Valley with rates of MeHg availability in New York and New England (3) added value to a forthcoming national Hg monitoring efforts and identification of biological hotspots, and (4) improved information that will be used to identify long-term monitoring locations for a forthcoming national Hg monitoring program.

#### **EXECUTIVE SUMMARY**

Atmospheric deposition of sulfur, nitrogen and mercury (Hg) has potential widespread and profound ramifications on the health of forest ecosystems and their inhabitants. It is critical that the impact of these pollutants and the ecological response be determined. Here a template is described to demonstrate the potential landscape-level effects of acidification on the transfer and fate of methylmercury (MeHg) in songbirds of northeastern North American. In time, these same effects will be linked with pools of available calcium (Ca). For this effort, there are scientific, conservation, and policy oriented goals. Scientifically, there are new and compelling reasons for studying MeHg availability in northern forests, particularly in areas at high elevation or with acidic conditions. For conservation purposes, the long-term decline of some neotropical migrant songbirds may be linked to environmental stressors within their breeding range. It is plausible that elevated MeHg levels and depleted available Ca levels, particularly within acidic environments, are negatively contributing to the viability of their populations. Lastly, this exploratory investigation contributes to the policy arena in three ways: (1) documenting a potential spatial gradient of Hg and Ca availability across an area that has major Hg emission sources to the West (i.e., the Ohio River Valley) and lower emissions in the East (i.e., most of New York and New England) (2) linking biotic responses with atmospheric Hg deposition stations and (3) providing improved information that will be used to identify long-term monitoring locations for a forthcoming national Hg monitoring program.

Songbirds that tended to exhibit elevated blood Hg levels were bog-obligate species (e.g., Palm Warbler), upper canopy foragers (e.g., Red-eyed Vireo) and species regularly associated with riparian areas (e.g., Louisiana Waterthrush). Larger songbirds exhibited higher risk of MeHg bioaccumulation than did smaller songbirds from the same foraging guild. Although blood Hg levels in upland forest songbirds are elevated, these levels remain below thresholds for likely negative reproductive impacts. What remains to be investigated is the influence of moderate Hg levels on songbird populations in areas that are deficient in Ca.

Although sampling efforts were conducted for all thrush species, Wood Thrushes were the target indicators for low to moderate elevations (63 Wood Thrushes from 12 sites) and the Bicknell's Thrush for high elevations (19 Bicknell's Thrushes from two sites). Thrush Hg levels provided the basis for standardizing the reporting of Hg body burdens among all thrush species relative to the levels found in these two particular species. Blood Hg levels in thrushes varied by geographic area, elevation, and likely trophic level. Based on preliminary results, thrush Hg levels converted into Wood Thrush blood Hg tended to be higher in the Catskill Mountains. High elevation thrush blood Hg levels tended to be higher than thrushes in low elevations from the same mountain slope.

Wood Thrushes are a primary bird indicator of the impacts of MeHg because (1) they had blood Hg levels higher than other thrushes and most other songbirds, (2) preliminary evidence indicates strong relationships with soil pH and available Ca levels and (3) they have shown major distribution and density declines

throughout New York and the Appalachian Mountains and are now nearly absent from the Adirondack Mountains.

To understand Hg and Ca pathways, three compartments were sampled: organic soils, Ca-rich and likely high Hg invertebrates, and breeding forest songbirds (e.g., thrushes, vireos, and warblers). Only songbird blood Hg levels are presented here. A total of 20 sampling stations were identified and sampled across New York and Pennsylvania. In 2006, 359 individuals of 47 species were sampled.

The three overarching objectives of this exploratory study are to: (1) better understand the dynamic relationship within terrestrial, acid-impacted ecosystems for Hg and Ca, (2) develop a spatial gradient for linking major Hg emission sources in the Ohio River Valley with rates of MeHg availability in New York and New England, and (3) integrate with a forthcoming national Hg monitoring network by identifying potential monitoring stations. This report represents a limited, exploratory effort to provide baseline information that can be furthered by other investigations. While compiled data may provide compelling evidence for further studies, limited sample sizes restrict statistical analysis.

#### 1.0 INTRODUCTION

Air pollution has been linked to adverse effects in wildlife (Lovett et al. 2009), including impairing reproductive success in songbirds (Saldiva and Bohm 1998, Llacuna et al. 1993, Janssens et al. 2003). Specifically, wet atmospheric deposition of acidifying emissions (i.e., nitrogen and sulfur oxides), has been linked to declines of bird species in Europe (Graveland 1990, 1998) and, recently, the United States (Hames et al. 2002). This phenomenon may be the result of depletion of soil pools of extractable calcium by leaching (Driscoll et al. 2001), leading to decreases in the abundance of Ca-rich invertebrate prey used by breeding female birds as necessary supplemental sources of Ca during egg production and when feeding nestlings (Graveland 1996, Graveland and Drent 1997). The study by Hames et al. (2002) is particularly relevant and compelling for follow-up efforts in this study. Based on logistic regression analysis that accounted for several habitat-related variables, the investigators found a particularly strong negative relationship between acid rain and the probability of detecting Wood Thrush breeding evidence. Findings also indicate further insult to breeding populations in areas that are at high elevation, with low pH soils, and exhibiting fragmentation of forest habitat.

Acid rain and related lowering of soil pH not only reduce calcium availability but add sulfates. The addition of sulfates is known to increase MeHg production in wetlands. In Minnesota, the estimated MeHg flux from an experimentally dosed wetland increased 2.4x (Jeremiason et al., forthcoming). Therefore, Hg increases in the environment are driven not only by the amount of Hg deposited and by acidifying deposition in general, but by sulfate deposition in particular.

The negative effects of Hg are well documented for aquatic ecosystems – through the biomagnification of biologically-active MeHg (Evers et al. 2003, 2005, 2008; Burgess and Meyer, 2008). Although studies of Hg cycling in terrestrial ecosystems are limited, upland soils have considerable capacity to store large quantities of atmospherically deposited Hg, particularly in the forest floor (Mason et al. 1994). Recently, new and compelling evidence connects MeHg availability in upland forests with acidic soils; regressions based on spatial models of atmospheric Hg deposition across terrestrial ecosystems in the Northeast predicted 50% of the variation in Bicknell's Thrush blood Hg levels (Rimmer et al. 2005). For the first time, scientists in the Northeast are realizing that the problem of MeHg availability in the environment is not restricted to aquatic habitats.

Other recent work suggests that accumulation in terrestrial ecosystems directly involves plants, through absorption of gaseous Hg stomatally, incorporation by foliar tissue, and subsequent release of Hg in litterfall (Ericksen et al. 2003). While litterfall may represent the bulk of Hg input to forested ecosystems, the wash-off of dry-deposited Hg species in throughfall, direct deposition in precipitation, and uptake of dissolved Hg by roots with translocation to foliar tissue may also play roles (Rea et al. 2002). Total Hg inputs to eastern forests may largely be incorporated in the leaf-litter and forest floor, where it is available

to invertebrates, such as gastropods (snails and slugs), isopods (woodlice), myriapods (millipedes), and to predators, such as centipedes (myriapods) and spiders (arachnids). Spiders can have a particularly influential impact on biomagnifying MeHg in invertivore food webs. Cristol et al. (2008) found some terrestrial songbird species to exceed aquatic-based songbirds; even piscivorous species such as the Belted Kingfisher had Hg body burdens that were lower than terrestrial songbird species that regularly prey on spiders. The abundances of many of the Ca-rich prey species decline with declines in soil pH. This pattern can have important ramifications on the health of songbird populations, particularly on females laying eggs and on the growth of hatchlings.

Based on these and other studies, the complex and potentially synergistic relationship of sulfur-driven acidification, associated soil Ca depletion, and MeHg enhancement may lead to potentially important anthropogenic, landscape-level impacts. The incorporation of Hg from the leaf litter by invertebrates feeding on leaf tissues and by predaceous invertebrate species (centipedes and spiders) feeding on these detritivores also leads to potentially elevated Hg levels in songbird species. Thus, breeding bird populations in eastern forests, in particular thrushes and songbirds found in acidified habitats, such as subalpine and bog habitats, may be at greatest risk.

#### 2.0 PROJECT GOALS AND SUPPORTING OBJECTIVES

- 1. Establish an exposure profile of Hg for songbirds foraging within the insectivorous pathway
- 2. Establish an exposure profile of calcium for monitoring stations
- 3. Assess the potential risk of elevated Hg and deficient calcium availability to songbirds
- 4. Determine habitats and areas that are highly sensitive to potential impacts
- 5. Link efforts with a broader network across the Appalachian Mountains

#### 3.0 STUDY AREA

In 2006, samples were taken of songbirds and soil at twenty locations, with an emphasis on New York and Pennsylvania (Figure 3-1). In New York, seven sites were sampled in the Adirondacks, which include Arbutus Lake, Elk Lake, Ferd's Bog, Lake George, Spring Pond Bog, Sunday Pond and Whiteface Mountain, a high elevation site. Five sites were sampled in the Catskill Mountains, which include Devil's Tombstone, Emmons Bog, Lake Capra, Neversink Reservoir, and Plateau Mountain, a high elevation site. In southeastern New York, three sampling stations were chosen at Mohonk Preserve, Sam's Point, and Black Rock Forest. In addition, samples were collected at Tug Hill and at two sites in Allegany State Park. Two sites sampled in western Pennsylvania included Powdermill Nature Preserve and Spruce Bog, which is near Powdermill Nature Preserve. Outside the scope of this project, BRI and collaborators sampled nine additional locations in Delaware, Maine, North Carolina, Tennessee, and Virginia as part of the Terrestrial Ecosystem ReseaRch and Assessment (TERRA) Mercury Network (Appendix 1).

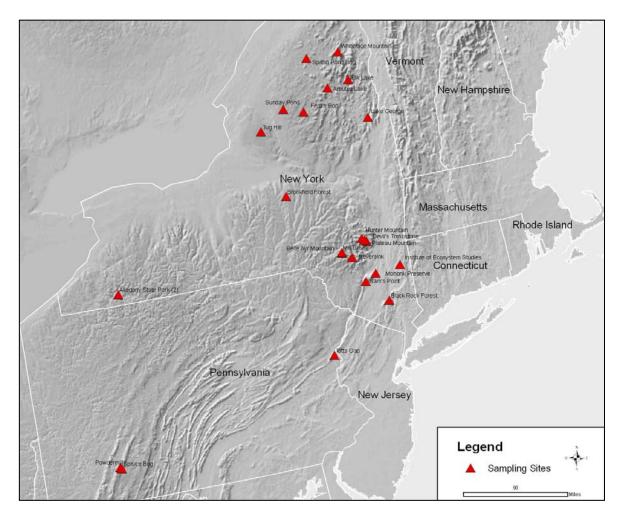


Figure 3-1. Distribution of sampling locations in New York and Pennsylvania, 2005-2006

#### 4.0 METHODS

#### 4.1 BIRD CAPTURE AND SAMPLE COLLECTION

An emphasis was placed on the sampling of Bicknell's Thrush at high elevation sites and the Wood Thrush and Red-eyed Vireo at low elevation sites. Other common forest invertivores, such as the Ovenbird, were opportunistically sampled. Thrushes and other songbirds were captured using mist nets in concert with decoys, playback of conspecific territorial vocalization, and playback of a flock of songbirds mobbing a small owl (Gunn et al. 2000). Both playbacks elicit a strong response from territorial breeding birds, allowing reliable capture. Sampling efforts were timed for June and July to allow time for depuration of Hg body burdens that could reflect winter and/or migratory MeHg uptake. It is well established that blood reflects recent dietary uptake of MeHg (Evers et al. 2005).

Typically, 8-10, 12m mist nets with a 36mm mesh size were used to harmlessly catch songbirds. Nets were placed on 6m bamboo and/or metal poles. The nets were checked every 20-40 minutes. Captured birds were removed and placed in cotton holding bags until processing. All birds were released unharmed 15-45 minutes after capture. Birds were captured during both dawn and dusk periods. All birds were measured using standard wing, tail, tarsi, bill, and mass measurements, and banded with USFWS bands. Information on age, sex, and body condition, (which is indicated by the external thickness and extent of fat), was also collected.

For all birds 28 gauge disposable needles were used to puncture a cutaneous ulnar vein in the wing to collect a small blood sample. Second secondary feathers from adults and selected juveniles were taken for Hg analysis. Each blood sample was collected in a 75 uL capillary tube, which was then sealed on both ends with Crito-seal or Critocaps ® and placed in a labeled plastic 7 cc vacutainer. Generally, 2-4 capillary tubes half-filled with blood were taken from each bird. The feathers were placed in a labeled plastic bag. All samples were stored in a field cooler with ice, and samples were later transferred for temporary storage (blood in the freezer, feathers in the refrigerator).

#### 4.2 INVERTEBRATE AND SOIL COLLECTION

Sampling efforts emphasized areas where songbirds were mist-netted, and focused on calcium-rich invertebrates found on or near the forest floor, such as isopods, predatory ground beetles, and myriapods. Calcium-rich prey items are necessary for egg laying and chick skeletal development (Graveland 1996; Graveland and Drent 1997). In addition, higher trophic invertebrates such as spiders were collected. Invertebrates were sampled near net lanes used for songbird capture to ensure that prey items were sampled within the songbird territory. Captured invertebrates were placed in plastic containers with snap lids and placed in the freezer. Using various keys, samples were identified to the lowest possible taxonomic level (usually, at least to the family level). Soil samples (2.5 cm) were opportunistically collected in the same

area as the collected invertebrates. All soil samples were double bagged in plastic bags and placed in a freezer.

#### 4.3 SAMPLE ANALYSIS

Laboratory analysis of songbird blood was conducted by Texas Agriculture and Mining University (Texas A&M) Trace Element Research Lab, College Station, Texas. Laboratory procedures follow standard U.S. Fish and Wildlife Service protocols. Blood samples were analyzed for total mercury using direct mercury analyzer DMA 80 by Milestone Inc. Mercury concentrations are presented on a wet weight (ww) basis. Instead of analyzing methylmercury levels (MeHg), total Hg was analyzed because it is less costly, and because > 95% of total Hg in bird blood is known to occur in MeHg form (Rimmer et al. 2005; Wolfe et al. 2007; BRI unpubl. data for 50 songbirds was 99% +/- 7%; D. Cristol unpubl. data for 30 songbirds was 95% +/-7%).

Mercury and methylmercury analysis of invertebrate samples was conducted by CEBAM Analytical Inc. Samples were analyzed using a direct mercury analyzer and by following the EPA Method 1630 series. Mercury concentrations were presented on a dry weight (dw) basis. Soil was analyzed for exchangeable calcium, available calcium and total mercury at Syracuse University. Mercury samples were analyzed using a direct mercury analyzer and calcium was analyzed using ICP-MS.

#### 5.0 RESULTS AND DISCUSSION

The 2006 sampling effort focused on multiple songbird species, with an emphasis on the Wood Thrush (*Hylocichla mustelina*). This neotropical migrant is experiencing significant negative population trends; postulated as being partly related to availability soil Ca deficiencies (Hames et al. 2002). Another species, the Bicknell's Thrush (*C. bicknelli*) is restricted to mountaintops where cloud and fog acidic and Hg deposition occur, in addition to increased precipitation due to orographic effects. This species serves as a previously identified indicator of the effects of high levels of both acidic and Hg deposition (Rimmer et al. 2005) and was therefore emphasized for high elevation areas. A primarily riparian species, the Louisiana Waterthrush (*Seiurus motacilla*), was chosen because samples collected in other states showed that this species' mean blood Hg levels are generally high in mercury. The Palm Warbler (*Dendroica palmarum*) is a bog-obligate species. Bogs may have the potential for increased Hg methylation and calcium depletion due to their acidity. Common species such as the Ovenbird (*Seiurus aurocapillus*) and the Red-eyed Vireo (*Vireo olivaceus*) were also emphasized. Their ubiquitous distribution provides a basis for comparisons across geographic areas.

Although neotropical migrant thrushes and other songbirds that are of high priority to environmental trustees were chosen, many songbird populations are exhibiting negative population trends that apparently are not wholly linked with neotropical wintering areas. For example, the Rusty Blackbird (*Euphagus carolinus*) has experienced continental declines of over 95% since the 1970s (Russ Greenberg pers. comm. 2008). Because a related species, the Red-winged Blackbird (*Aegalius phoenicius*) has recently been shown to have mean blood Hg levels ~10x higher than found in songbirds associated with the same habitats and 3x higher than in associated piscivores, wetland-associated blackbirds and other songbirds are of high conservation concern and will be emphasized in the future. How species react to air pollution within their breeding habitat may vary (Eeva et al. 1997); in some cases there can be toxicological impacts to the physiology, reproductive success, or survival of individuals within the study population; while in other cases normal food supply and nutrient rates can be disrupted and ultimately reduce fitness and breeding success.

The following description of findings for this exploratory study illustrates sampling efforts, blood Hg exposure for thrushes and other associated songbirds, ability to normalize thrush Hg levels for the Wood Thrush, a comparison with known Hg threshold levels in songbirds, and a description of the relationship between MeHg and exchangeable calcium in soil.

#### **5.1 SAMPLING EFFORT**

From June 5, 2006 to August 7, 2006, 20 sites were sampled in New York and Pennsylvania. With an average of 18 songbird samples per site, a total of 359 birds representing 47 species in 18 families were sampled. Cumulatively in New York and Pennsylvania, from 2005 to 2006, 515 birds were sampled (Appendix 3). Mean blood Hg levels for adults ranged from 0.01 ug/g, ww (Hairy Woodpecker) to 1.49 ug/g, ww (Palm Warbler) in 2006.

In addition, soil and invertebrates were sampled at 18 of the 20 sites. A total of 162 invertebrates were sampled, including 62 spiders, 36 beetles, 14 centipedes, 22 millipedes, 2 beetle larvae, 2 mealworms and 24 isopods (Appendix 2). Despite dedicated search efforts, beetles, centipedes, millipedes or isopods were not found at every site. In addition, spiders were not sampled at Black Rock Forest due to inclement weather, which forced spiders into hard-to-find, dry hiding spots. For Catskill Mountain area samples, the mean wholebody Hg levels ranged from 6.18 ng/g, dw (camel cricket) to 178.60 ng/g dry weight (spider). Soil total Hg levels ranged over an order of magnitude from 0.067 ug/g, dw (Neversink-Frost Valley, NY) to 0.70 ug/g, dw (Arbutus Lake, NY).

#### 5.2 MERCURY EXPOSURE IN TARGET BIRD SPECIES

Because blood Hg levels in adults are regularly significantly higher than in juveniles (Evers et al. 2005), sampling and analysis of adult individuals was targeted. Six songbirds were emphasized because of their conservation concern, predicted elevated Hg levels, or ubiquitous distributions.

#### Wood Thrush

This neotropical migrant is well known for being impacted by habitat fragmentation and associated increases in nest predation and parasitism (Robinson et al. 1995, Trine 1998), including in areas in New York (Driscoll and Donovan 2004). However, only recently have other threats, such as those related to air pollution, been identified. Hames et al. (2002) found a strong negative relationship between acid rain and occurrences of breeding Wood Thrushes. Acidification of forested landscapes and negative impacts on forest songbird breeding populations are known elsewhere as well, including in Europe (Graveland 1998).

The 2005-2006 capture effort resulted in 69 Wood Thrushes sampled for Hg analysis (63 adults and six juveniles) at twelve sites. Blood Hg levels in adults had a mean of  $0.08 \pm 0.04$  ug/g, ww and ranged from <0.01 to 0.18 ug/g, ww. Juveniles had a mean of  $0.03 \pm 0.02$  ug/g, ww and ranged from 0.01 to 0.08 ug/g, ww (Figure 5-1, Appendix 3).

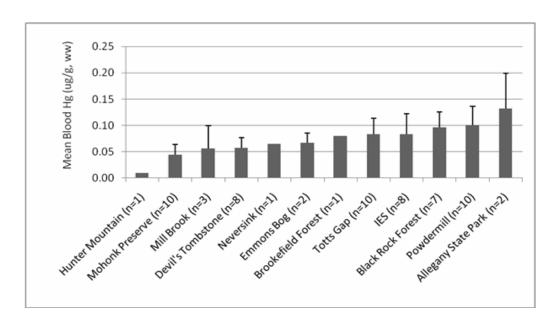


Figure 5-1. Comparison of mean blood Hg concentrations of Wood Thrushes in New York and Pennsylvania, 2005-2006.

Small sample sizes of adult Wood Thrushes from this exploratory effort makes spatial comparisons difficult for statistical robustness. To statistically characterize each location an estimated 26 individuals of one age class is needed (based on fixed precision analysis using the mean and variation from Totts Gap and a percentage of relative precision value [PRP] of 25%, which means that the upper and lower confidence limits will fall within 25% of the mean with a 95% certainty (Sutherland 1996). By normalizing blood Hg levels of various low-to-moderate elevation thrush species into a surrogate "Wood Thrush" measurement, a preliminary spatial analysis was conducted (see section 5.7).

When comparing limited Wood Thrush Hg results from a limited number of locations, with the geographic patterns of air pollution deposition and sensitive habitats (i.e., poorly buffered soils in montane areas), there is indication of a correlative relationship with changes in Wood Thrush presence and breeding intensity (Figure 5-2). Based on New York State's standard Breeding Bird Atlas efforts for the two time periods (1980-1985 and 2000-2005), Wood Thrush breeding populations indicate dramatic negative changes: (1) Wood Thrush presence in the Adirondack Mountains has declined in spatial extent as well as in the number of blocks with confirmed breeding (currently, there are very few areas with confirmed breeding in the Adirondack Mountains); and (2) the density of blocks with confirmed Wood Thrush breeding has declined substantially in the Catskill Mountains and several other areas of New York. A region-wide analysis of biological hotspots based on piscivorous birds and mammals identified parts of the Adirondack and Catskill

<sup>&</sup>lt;sup>1</sup> If we used 20% PRP, a sample size of 35 Wood Thrushes is needed for each location.

Mountains as falling within such hotspots (Evers et al. 2007). Although that analysis did not include potential MeHg availability and impacts on insectivores and northern forests, the overlap of biological hotspots of Hg for piscivores and concerns for insectivores in the same areas are compelling reasons to continue monitoring of species like the Wood Thrush.

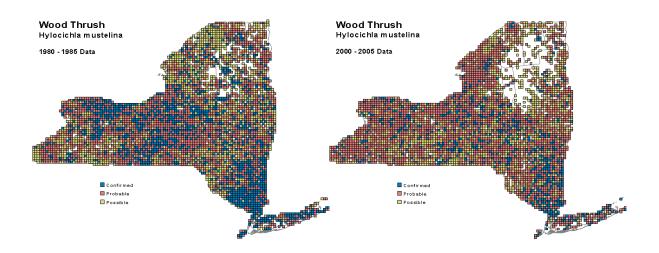


Figure 5-2. Breeding distribution of the Wood Thrush. Source: NY Breeding Bird Atlas (NYSDEC 2007).

#### Bicknell's Thrush

This neotropical migrant is a species of high conservation concern (Partners in Flight rank of 18 out of 20), is listed as a species of Special Concern in New York and appears on the Audubon Watchlist (Rich et al. 2004; 18,74). The Bicknell's Thrush breeds only in subalpine areas of conifer-dominated forests with elevation thresholds that are latitudinally controlled (Lambert et al. 2005); in the U.S., lowest elevations occupied are in northern Maine at 750m, while in the southernmost extent of its range in the Catskill Mountains, the Bicknell's Thrush generally breeds on mountains 1,100 m or higher (Rimmer et al. 2001). While the Wood Thrush serves as the indicator for MeHg availability in low to moderate level sites, the Bicknell's Thrush is an indicator of MeHg availability in high elevation levels.

Only two sites were included in the 2005-2006 sampling effort for Bicknell's Thrush. The mean blood Hg for adult Bicknell's Thrush was 0.08 +/- 0.02 ug/g (ww) with a range of 0.05 to 0.14 ug/g, ww (n=19) (Figure 5-3, Appendix 3). There was little difference in the mean blood Hg levels for Plateau Mountain in 2005 and 2006 (Figure 5-3). This site is lower in elevation than many sites in New England (Rimmer et al. 2005). To statistically characterize each location, an estimated 21 individuals of one age class is needed (based on fixed precision analysis using the mean and variation from Plateau Mountain and a percentage of

relative precision value [PRP] of 25%, which means that the upper and lower confidence limits will fall within 25% of the mean with a 95% certainty<sup>2</sup>) (Sutherland 1996).

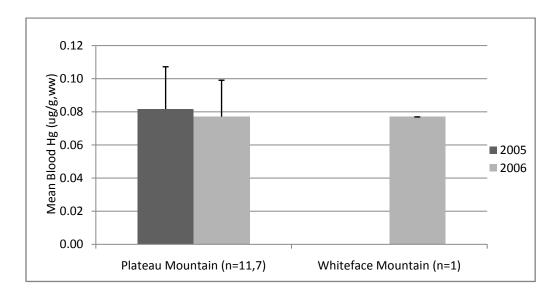


Figure 5-3. Comparison of mean blood Hg levels of Bicknell's Thrush at Plateau and Whiteface.

Unlike the Wood Thrush, the distribution and density of confirmed breeding by the Bicknell's Thrush in the Adirondack and Catskill Mountains is relatively similar between the two time periods of data collection by the New York Breeding Bird Atlas (Figure 5-4).

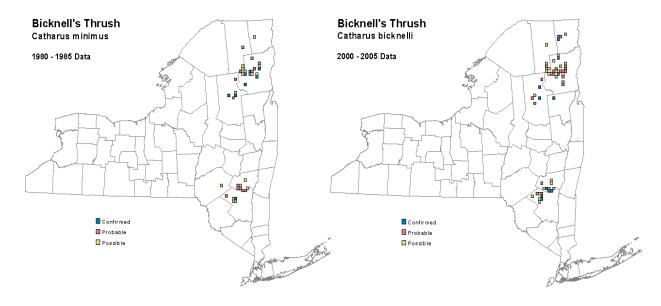


Figure 5-4. Breeding distribution of the Bicknell's Thrush. Source: NY Breeding Bird Atlas (NYSDEC 2007).

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<sup>&</sup>lt;sup>2</sup> If we used 20% PRP, a sample size of 27 Bicknell's Thrushes is needed for each location.

#### Palm Warbler

This neotropical migrant is not a species of concern, with a Partners in Flight (PIF) rank of 8 out of a possible 20 (Rich et al. 2004, 75). In addition, it appears that confirmed breeding blocks in New York have increased for the Palm Warbler (Figure 5-5). However, the Palm Warbler is a focal species for investigating MeHg bioaccumulation due to its preference for sphagnum habitats or open wet areas in coniferous forests, both naturally acidic environments. In addition, like Wood Thrushes, Palm Warblers tend to feed on or near the ground, and thus represent an indicator for MeHg in ground-feeding invertivores in bog areas (Wilson and Herbert 1996).

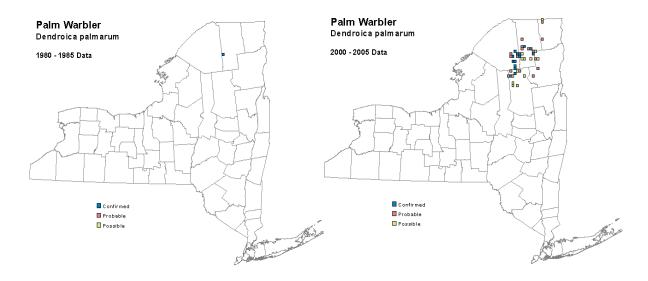


Figure 5-5. Breeding distribution of the Palm Warbler. Source: NY Breeding Bird Atlas (NYSDEC 2007).

Palm Warblers were sampled in 2006 at Spring Pond Bog in the Adirondack Mountains. The mean blood Hg for adult Palm Warblers was 0.92 ug/g,ww (n=2) (Figure 5-6). The Palm Warblers caught at Spring Pond Bog had higher mean blood Hg than any other songbirds within this study. Other songbirds sampled at Spring Pond Bog also tended to be higher in their Hg concentrations than observed at other monitoring stations.

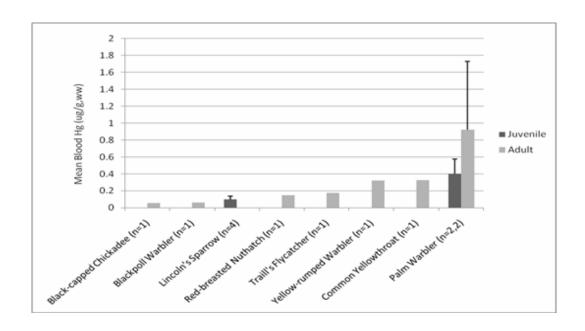


Figure 5-6. Comparison of blood Hg levels in songbirds from Spring Pond Bog, 2006.

#### Louisiana Waterthrush

The Louisiana Waterthrush has a PIF rank of 13 out of a possible 20 (Rich et al. 2004, 75), and is listed as a Species of Greatest Conservation Need in the New York Comprehensive Wildlife Conservation Strategy. The Louisiana Waterthrush mainly breeds and feeds in riparian forests and feeds primarily on aquatic invertebrates (Robinson 1995). This species is an indicator for MeHg availability in wet, forested habitats. Comparisons of NYS standard Breeding Bird Atlas efforts for the two time periods indicates a substantial loss of overall breeding range, density and confirmed breeding (Figure 5-7).

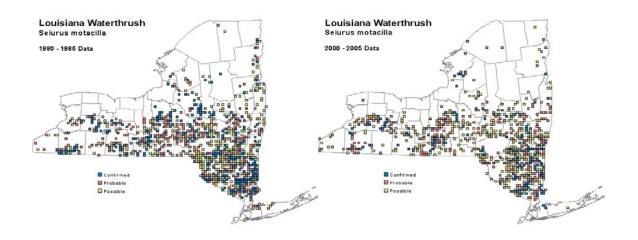


Figure 5-7. Breeding distribution of the Louisiana Waterthrush. Source: NY Breeding Bird Atlas (NYSDEC2007).

The 2005-2006 capture effort resulted in 9 Louisiana Waterthrush sampled for Hg analysis (5 adults and 4 juveniles) at three sites. Blood Hg levels in adults had a mean of  $0.13 \pm 0.04$  ug/g, ww, and ranged from 0.08 to 0.18 ug/g, ww. Juveniles had a mean of  $0.15 \pm 0.07$  ug/g, ww and ranged from 0.05 to 0.24 ug/g, ww (Appendix 3). Although a small sample size, Louisiana Waterthrush juvenile mean blood Hg from Allegany State Park tended to have higher levels than adults at Devil's Tombstone (Figure 5-8). Both New York sites, Devil's Tombstone and Allegany State Park, tended to have higher mean blood Hg levels than adults and juveniles in Powdermill, PA.

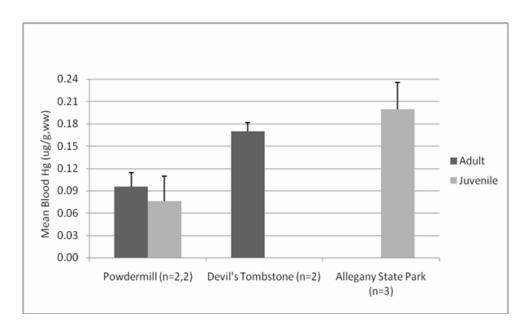


Figure 5-8. Comparison of mean blood Hg levels in Louisiana Waterthrush in NY and PA, 2005-2006.

#### Red-eyed Vireo

The Red-eyed Vireo has a PIF ranking of 7 out of 20 (Rich et al. 2004, 72). In comparing the Breeding Bird Atlas for the two time periods, Red-eyed Vireos show a decrease in confirmed breeding blocks for the Catskill region of New York (Figure 5-9). In general, however, confirmed breeding blocks have increased for most of the rest of the state. The Red-eyed Vireo provides an indicator of MeHg availability in canopyforaging birds.

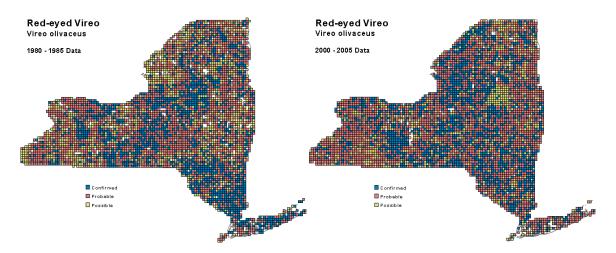


Figure 5-9. Breeding distribution of the Red-eyed Vireo. Source: NY Breeding Bird Atlas (NYSDEC).

The 2005-2006 capture effort resulted in 90 Red-eyed Vireos sampled for Hg analysis (85 adults and 5 juveniles) at 17 sites. Blood Hg levels in adults had a mean of  $0.09 \pm 0.07$  ug/g, ww, and ranged from 0.01

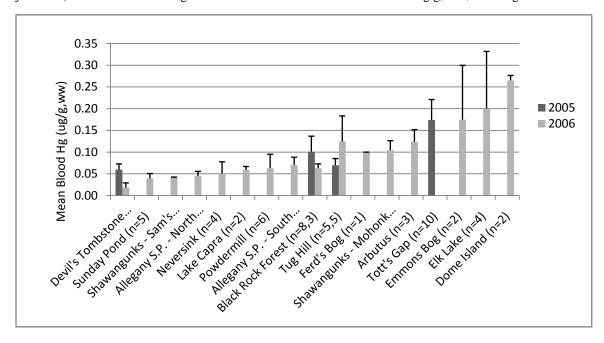


Figure 5-10. Comparison of mean blood Hg levels in Red-eyed Vireos from NY and PA, 2005-2006.

to 0.35 ug/g, ww. Juveniles had a mean of  $0.06 \pm 0.01$  ug/g, ww and ranged from 0.05 to 0.07 ug/g, ww (Appendix 3). The highest mean blood Hg occurred at Lake George's Dome Island in New York (Figure 11). Birds from sites such as Devil's Tombstone and Black Rock Forest located in the southern region of New York, averaged higher levels in 2005 than in 2006. However, birds from Tug Hill, located in northwestern New York had mean blood Hg levels higher in 2006 than in 2005.

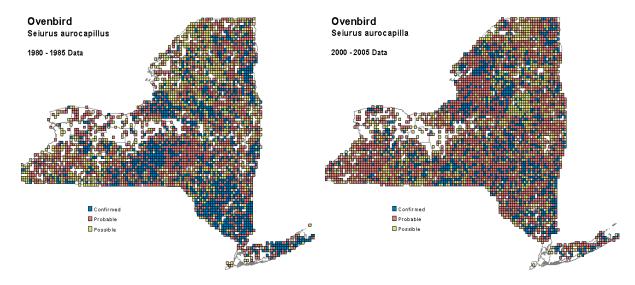


Figure 5-11. Breeding distribution of the Ovenbird. Source: NY Breeding Bird Atlas (NYSDEC 2007).

#### **Ovenbird**

The Ovenbird has a PIF ranking of 10 out of 20 (Rich et al. 2004, 75). While the rank does not indicate any special concern, a comparison of the two time periods of the Breeding Bird Atlas indicated an overall decline in confirmed breeding blocks most notably in southern New York (Figure 5-11). The Ovenbird shares the same foraging guild as the wood thrush. But unlike the Wood Thrush, which tends to feed within the leaf litter layers, the Ovenbird gleans prey from the leaf litter surface (Holmes and Robinson, 1988).

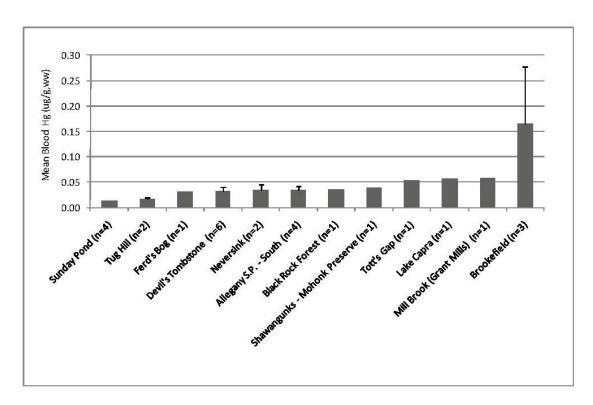


Figure 5-12. Comparison of Ovenbirds captured for all sites, 2005-2006.

A total of 27 adult Ovenbirds were sampled from 2005-2006 from 12 sites. Hg levels averaged  $0.05 \pm 0.05$   $\mu g/g$ , ww, with a range of 0.01 to 0.30  $\mu g/g$ , ww (Appendix 3). Preliminary comparisons show that Ovenbirds in Brookfield, NY have higher Hg levels than other sites in New York (Figure 5-12).

#### 5.3 REGIONAL COMPARISONS OF MERCURY LEVELS

The 2005-2006 sampling effort resulted in 28 soil samples for 22 sites. Mercury levels ranged from 0.07 ppm at Neversink-Frost Valley to 0.70 ppm at Arbutus Lake in the Adirondacks (Appendix 4). Exchangeable calcium ranged from 0.20 cmolc/Kg-soil at Black Rock Forest to 14.44 cmolc/Kg-soil at Arbutus Lake. Regional mean soil Hg was highest at Tug Hill and lowest in Eastern Pennsylvania (Figure 5-13). Regional exchangeable calcium levels were highest in the Adirondacks and lowest in the Allegheny Plateau region of New York (Figure 5-14).

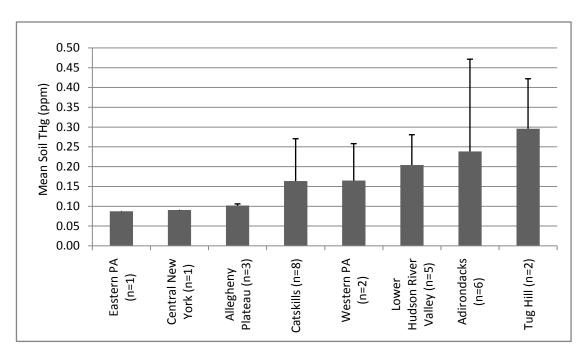


Figure 5-13. Regional soil Hg levels, 2005-2006.

Although the soil sample from eastern Pennsylvania had the lowest Hg concentration compared to samples from the other seven regions, Red-eyed Vireos sampled in eastern PA tended to have the highest mean blood Hg levels (Figure 5-15) of all regions. Adirondack Red-eyed Vireos had mean blood Hg levels higher than Catskill Red-eyed Vireos. However, the ground foragers, Hermit Thrush and Ovenbird, tended to have higher mean blood Hg levels in the Catskill region than in the Adirondack region.

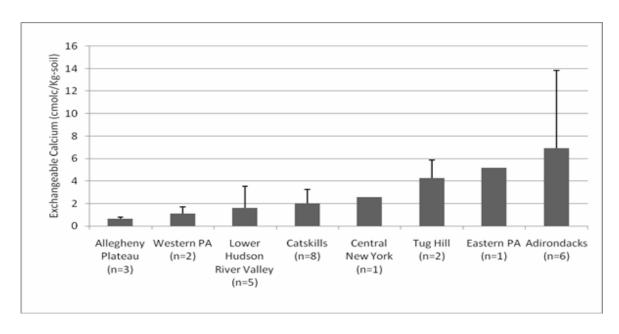


Figure 5-14. Mean exchangeable calcium for multiple regions in New York and Pennsylvania, 2005-2006.

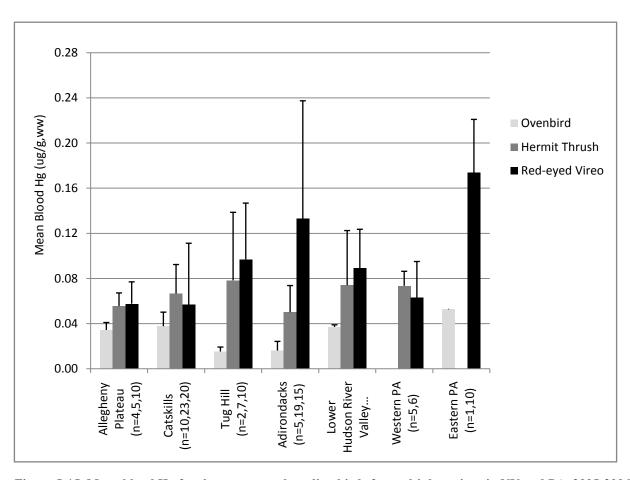


Figure 5-15. Mean blood Hg for three common breeding birds for multiple regions in NY and PA, 2005-2006

#### 5.4 MERCURY EXPOSURE IN INVERTEBRATES

Invertebrate data was provided by Jeff Loukmas of the NY State Department of Environmental Conservation (J. Loukmas pers. comm. 2008). Centipedes, spiders, and slugs from three sites in the Catskill region had the highest Hg levels (Figure 5-16). However, MeHg in slugs was not as high as in centipedes. For example, based on the limited 2006 data (Figure 5-17) and preliminary data from 2007, it appears that percentage of MeHg is related to taxa type and site.

Although a greater sample size is needed, preliminary comparisons of soil Hg and invertebrate MeHg show an emerging positive relationship between these two variables among all invertebrate types except beetles (Figure 5-17). Another emerging relationship occurs between thrush blood total Hg levels and both beetle and centipede MeHg levels (Figure 5-18). As centipede or beetle MeHg levels increase, so do thrush blood Hg levels. Isotope analysis and invertebrate species differentiation will enable us to further explore this preliminary relationship in the future.

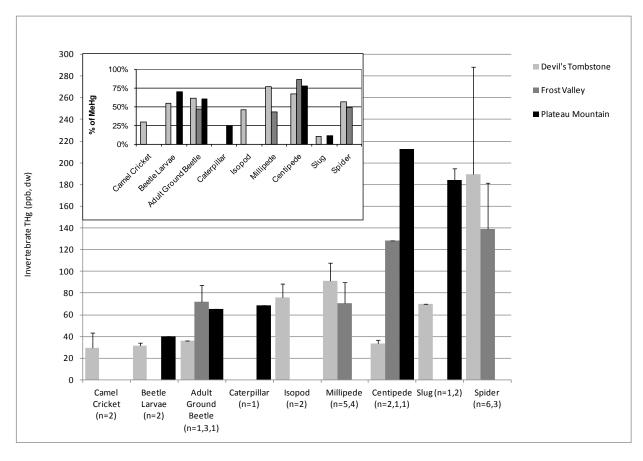


Figure 5-16. Comparison of mean invertebrate total Hg (ng/g, dw), 2005-2006 for the Catskills sites.

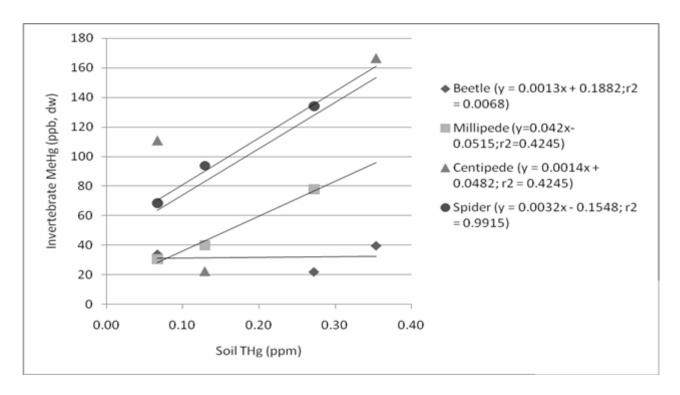


Figure 5-17. Invertebrate MeHg (ng/g,dw) vs. soil Hg (ug/g), 2005-2006.

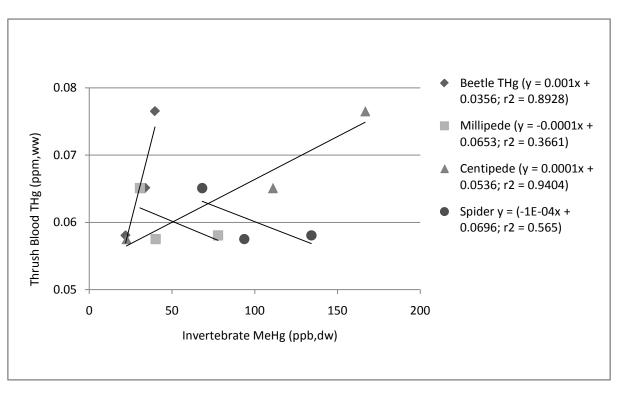


Figure 5-18. Thrush species blood Hg (ug/g,ww) vs. invertebrate MeHg levels (ng/g,dw), 2005-2006.

#### 5.5 MERCURY EXPOSURE IN SONGBIRDS BY FORAGING GUILD

Foraging guilds are likely important factors when assessing risk of Hg exposure. Songbirds foraging on seed and fruit had significantly lower Hg levels than other foraging guilds (Figure 5-19). In addition, bill volume (depth x length x width) may also be a factor that contributes to Hg risk. As bill volume increased, mean Hg levels also increased (Figure 5-20). Bill volume likely relates to food type and prey size.

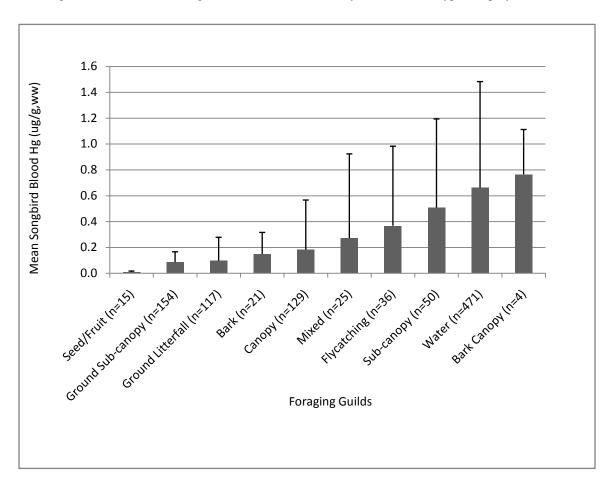


Figure 5-19. Comparison of mean songbird blood Hg levels by foraging guild.

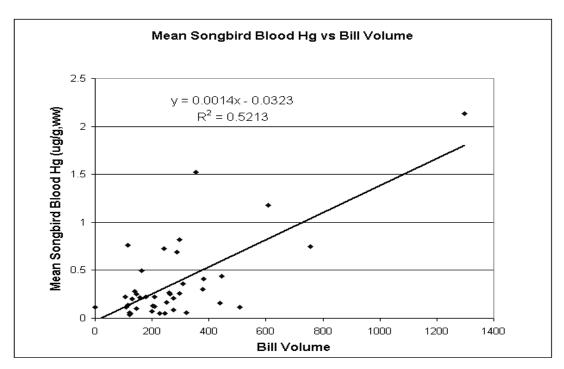


Figure 5-20. Mean songbird blood Hg vs. bill volume (length x width x depth mm<sup>3</sup>).

### 5.6 RELATIONSHIP OF MERCURY AND CALCIUM AVAILABILITY IN ACIDIFIED HABITATS

The relationship between soil total Hg (Figure 5-21) and soil exchangeable calcium (Figure 5-22) and songbird blood Hg concentrations is not clear and requires further field sampling.

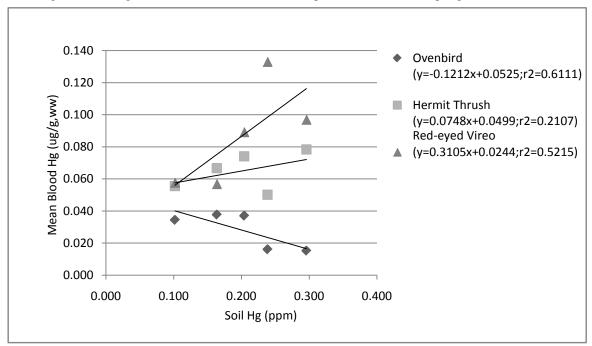


Figure 5-21. Mean blood total Hg (ug/g, ww) vs. soil total Hg (ug/g).

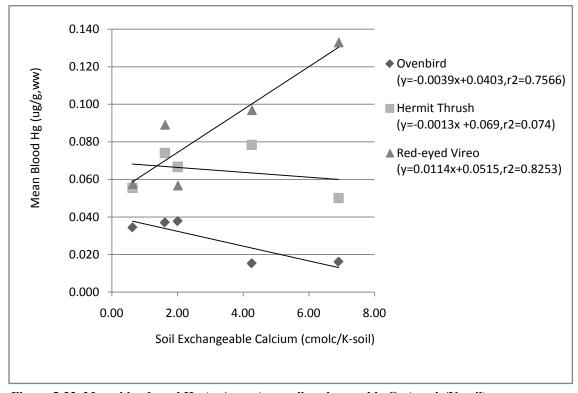


Figure 5-22. Mean blood total Hg (ug/g, ww) vs. soil exchangeable Ca (cmolc/K-soil).

#### 5.7 NORMALIZED SONGBIRD HG LEVELS: THRUSHES AS A CASE STUDY

Thrush Hg exposure is dictated by hydrogeology and biogeochemistry as well as (1) foraging technique, (2) prey selection, (3) body size, and (4) elevation of breeding habitat. These factors may be additive or antagonistic. For example, the American Robin, averaging 76g in body weight and the largest thrush, tends to exhibit blood Hg levels similar to those of the much smaller Veery (31g). Robins likely forage on prey items that are in lower trophic levels (e.g., earthworms) than do Veerys. The foraging technique and prey selection of Wood Thrushes are likely different than that of American Robins, emphasizing higher trophic level prey items (e.g., beetles and centipedes) (Holmes and Robinson 1988), because Wood Thrushes (adult average weight of 48g) are approximately 37% lighter than robins. There are also gradients of soil chemistry that tend to become more acidic and less buffered as elevation increases. High elevations are more prone to higher Hg deposition (Miller et al. 2005) and their geochemistry likely predisposes habitats to higher MeHg production and availability (as indicated by Rimmer et al. 2005). Therefore, high elevation species such as the Bicknell's Thrush (28g) and even the Swainson's Thrush (29g) have proportionally higher Hg levels than associated low-elevation mountainside neighbors. Lastly, moisture has an important role in MeHg availability to thrushes and other songbirds. Areas with more mesic soils are likely to have thrushes with higher Hg body burdens than the same species with territories on xeric soils. For example, at the Cary Institute of Ecosystem Studies site, the Veery was sampled in both mesic and xeric soils and those individuals in habitats with mesic soils had higher blood Hg levels (Cary Institute of Ecosystem Studies, unpubl. data). Although Hermit Thrushes average 30g body mass and are similar in size to the Bicknell's and Swainson's Thrushes, indications are that Hermit Thrushes have lower Hg body burdens. This may be because Hermit Thrushes generally inhabit drier forests tracts (Holmes and Robinson 1988).

Table 5-1. Within-site comparisons of thrush species for normalizing non-target thrushes to target thrush blood Hg levels.

Target Thrush	Non-target Thrush	Conversation factor to Target Thrush
Low Elevation		
Wood Thrush	Hermit Thrush	1.19
	American Robin	1.75
	Veery	2.21
High Elevation		
Bicknell's thrush	Swainson's Thrush	1.04
	Hermit Thrush	1.21

Although there are subtle differences in foraging strategy among the thrush species, blood Hg levels for thrushes were normalized within the same site to attain standard blood Hg levels for the Wood Thrush in low elevation areas and the Bicknell's Thrush in high elevation areas (Table 5-1). This process converts blood Hg levels in target species through same-site comparisons and provides a method for comparing

thrush Hg exposure with larger sample sizes and therefore is the basis for a preliminary attempt to develop a spatial gradient for New York.

Within-site comparisons are based at multiple sites; however, only the Plateau Mountain site provided the ability to relate the deposition of Hg and its subsequent availability at multiple elevations on a single mountainside gradient (Figure 5-23). At the base of Plateau Mountain at approximately 2,000 feet, lies the Devil's Tombstone State Park Campground site of sampling efforts for the Wood Thrush. Capture and sampling efforts at this site in 2005 resulted in the ability to compare the blood Hg levels from three species with overlapping breeding territories. The Wood Thrush had the highest Hg body burden of the three thrush species. Approximately 2,000 feet above this site was the sampling area for three other thrush species. Of these, the Bicknell's Thrush had the highest mean Hg body burden. All thrush species at Plateau Mountain tended to have higher Hg body burdens than those at Devil's Tombstone State Park, and providing preliminary evidence for higher MeHg availability as elevation increases.

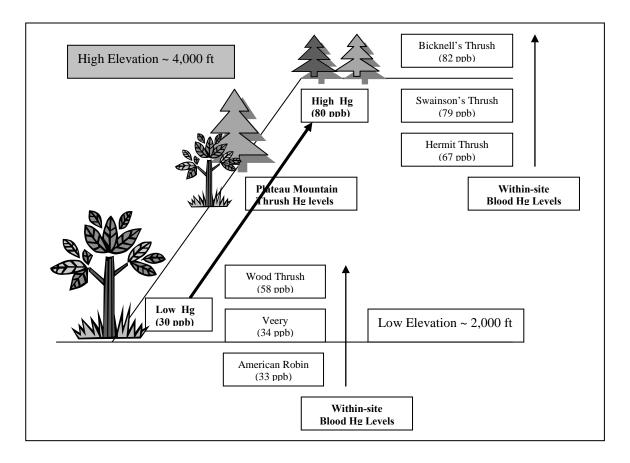


Figure 5-23. Comparison of thrush blood Hg levels on Plateau Mountain, New York.

To provide some geographic context to the New York and Pennsylvania thrush body burdens of Hg, comparisons are made with Maine, New Hampshire, and Vermont. Generally, blood Hg levels in thrush species from the 2005 New York and Pennsylvania sampling effort were similar to those measures at New England sites for the Veery, Hermit Thrush, Swainson's Thrush, and Wood Thrush. There appears to be significant differences between the two geographic areas for the American Robin and Bicknell's Thrush. Small samples limit statistical comparisons, but higher levels in Maine generally follow known geographic trends of MeHg availability in other birds, such as the Common Loon (*Gavia immer*; Evers et al. 2003). Such established geographic trends result from the interaction of many environmental variables including point source proximity, habitat sensitivity, and hydrological management and all may not necessarily apply to forest songbird geographic patterns of Hg levels.

#### 5.8 SPATIAL RELATIONSHIPS AMONG SITES FOR MERCURY

The spatial relationship of MeHg availability as measured through a standard thrush blood Hg unit could provide a basis for assessing habitat sensitivity and potential proximity to Hg emission sources. Because sample sizes within sites and the number of distinct sites and their juxtaposition are severely limiting, the concept requires further sampling efforts.

#### 5.9 MERCURY THRESHOLDS FOR SONGBIRDS

There are few datasets available that provide data on a level of concern (blood or feather concentration associated with adverse effects) for the biological effects of MeHg accumulation and exposure for avian insectivores. However, a risk assessment by Baron et al. (1999) estimated the lowest observed adverse effect level (LOAEL) for Northern Rough-winged Swallows (*Stelgidopteryx serripennis*) was a MeHg dose of 0.25 ug/g, ww per day. Recent additional work on swallows, specifically the Tree Swallow (*Tachycineta bicolor*), by scientists at the U.S. Geological Survey's Patuxent Wildlife Research Center provides more relevant information for this pilot assessment. Through formal Hg dosing experiments of Tree Swallow eggs based on protocols established for the CALFED-Bay Delta Mercury Project (Heinz 2003), Hg levels that posed a cause of concern were identified (Heinz et.al. 2009). Using an endpoint of embryo survival at 90% within the hatching date for the Tree Swallow, the median lethal dose (LC50) of MeHg in eggs was 0.32 ug/g (ww). (Heinz et al. 2009).

The use of eggs as a sampling tissue for our assessment was not feasible. Blood Hg levels were used instead. However, a regression model developed from an unpublished dataset for Tree Swallows from BioDiversity Research Institute, provides a relevant tool for predicting blood Hg levels from egg Hg levels. Based on 184 paired Hg levels from eggs and blood collected from the same female, a correlation where approximately 91% of the variability is accounted, a level of concern in the blood of 1.01 ug/g, ww was projected (Figure 5-24).

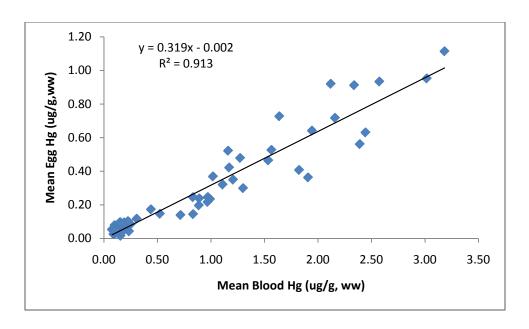


Figure 5-24. Model of the relationship of Hg in paired blood and egg samples.

## 6.0 POLICY IMPLICATIONS

The reduction of pollutant emissions is particularly important in northeastern North America. Industrial areas in the Ohio River valley are likely sources of sulfur and Hg deposition in New England and New York. Keeler et al. (2005) used back trajectories of air masses to directly link Hg deposition in western Vermont with sources in the Ohio River Valley and neighboring areas. This likely explains the dichotomy for why significant efforts in New England and New York to reduce Hg emissions have not yet achieved region-wide declines across these seven states. That is, current Hg deposition in many areas of the Northeast is still dictated by emissions from neighboring areas to the west.

Recent findings indicate a growing body of evidence that atmospheric deposition and biological Hg hotspots can be created by local emission sources. Five such studies include: (1) Steubenville, Ohio, where a U.S. Environmental Protection Agency funded project demonstrated that nearly 70% of the Hg collected at a monitoring site originated from a neighboring coal-burning facility (Landis et al. 2005); (2) northeastern Massachusetts, where the Massachusetts Department of Environmental Protection reported a 32% decline in yellow perch Hg during a seven year decline in nearby Hg emissions from municipal and hospital incinerators (Hutcheson 2003); (3) southern Florida, where fish and wading bird Hg concentrations declined 60-70% following research efforts directed by the state of Florida and University of Florida (Frederick et al. 2005); (4) Cresson, Pennsylvania where researchers from Pennsylvania State University and the Pennsylvania Department of Environmental Protection demonstrated that Hg levels were 47% higher in areas closer to power plants than in more distant areas such as in Wellsboro, Pennsylvania (Commonwealth of Pennsylvania Department of Environmental Protection, 2006); and (5) a study of nine southeastern New Hampshire lakes, where Hg levels declined over 50% in the blood of the Common Loon between 2001 and 2004 (Evers et al. 2007) correlated with the removal of 6,600 pounds of Hg from upwind incinerator emission sources located within 200 km of the study area.

If these examples are not exceptions, then local emission sources can cause significant local impacts. With its high density of Hg emission sources (e.g., coal-fired electric generator, incinerators, cement manufacturing facilities), the eastern U.S. would be expected to have high levels of atmospheric deposition and large numbers of local biological hotspots for Hg deposition. The recent identification and characterization of biological Hg hotspots in the Northeast by Evers et al. (2007) further demonstrates that they exist and are present for multiple reasons, which include proximity to local emission sources as well as landscape biogeochemical cycling. The contentious nature of both atmospheric and biological Hg hotspots was recently highlighted by a 15 May 2006 report by the Acting Inspector General for the U.S. Environmental Protection Agency (USEPA 2006). Within the report, the Inspector General questioned the U.S. Environmental Protection Agency's premise for its "Clean Air Mercury Rule" that emissions from power plants after emissions trading will not lead to local hotspots of Hg deposition. In response, the U.S.

Environmental Protection Agency Inspector General's office recommended the development and implementation of a mercury monitoring plan to address both emissions and deposition.

The influence of local, regional, and global atmospheric Hg emission sources on downwind ecosystems therefore needs careful consideration – particularly in the context of the recent U.S. Appeals' Court ruling that the U.S. Environmental Protection Agency's "Cap-and-Trade" Clean Air Mercury Rule (CAMR) is not valid [U.S. Court of Appeals for the District of Columbia Circuit, February 8, 2008, No. 05-1097, State of New Jersey, et al. v. Environmental Protection Agency]. A cost-benefit analysis with new information on the existence of biological Hg hotspots that are controlled by local and likely regional Hg sources may diverge from the federal analysis<sup>3</sup>. Until such an analysis is conducted, the development of Hg emission regulations should take into account (1) current Hg emission levels and (2) nearby habitat sensitivity.

Based partly on new evidence provided by the New York State Energy Research and Development Authority (NYSERDA 2006), New York State has approved one of the most stringent Hg emission reduction standards for coal-burning facilities in the country (on 18 December 2006). The rule will reduce Hg emissions from electricity-generating stations within the State by 50% by 2010, and would require a 90% reduction by 2015. Under the current federal rule, power plants need to reduce their Hg emissions by 50% by 2018 and, sometime after 2020, reduce emissions by a total of 70% (40 CFR Part 60, Appendix B; 70 Fed. Reg. 28,606). The New York standard increases the overall reduction of Hg and requires a much more aggressive timeline in accordance with Maximum Available Control Technologies.

The rapid identification of sensitive habitats and the species of greatest risk in New York and nearby states and provinces is paramount for avoiding increased impacts from regional Hg emission sources. Because Hg emissions in one part of the world have the ability to pollute distant parts, eventually the solution will need to be an international one. However, as long as biological Hg hotspots can be traced to local emission sources, local solutions are possible<sup>4</sup>.

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<sup>&</sup>lt;sup>3</sup> In addition, the USEPA (2005) did not include ecological impacts in its cost-benefit analysis, even with scientific precedent for the importance of ecological impacts on socioeconomic interests (Bockstael et al.

<sup>&</sup>lt;sup>4</sup> Technologies are rapidly being developed to reduce Hg emissions from coal-fired generation facilities in an efficient and economic way (Srivastava et al. 2006).

## 7.0 CONCLUSIONS

There are compelling reasons to be concerned about the impact from airborne pollutants to breeding songbirds in eastern forests. Much is already known about the effects of acidic deposition on northeastern landscapes and the depletion of available Ca in soil. Only recently has acidification been implicated in increased MeHg availability as well. The distribution of Hg and the availability of MeHg are now well documented in the Northeast. Detection of this pattern was accomplished through a four-year study funded by the USDA Forest Service. BRI and their collaborators compiled and synthesized most of the publicly available mercury data in the Northeast into a series of 21 papers in a special issue of Ecotoxicology (Evers and Clair 2005). From this comprehensive review on how Hg is distributed across the landscape, three findings emerged that partly serve as a basis for this current investigation: (1) new findings indicate MeHg availability is more prevalent in terrestrial birds than previously considered (Evers et al. 2005); (2) birds in montane terrestrial habitats may be at risk (Rimmer et al. 2005), likely as a consequence of a higher rate of atmospheric deposition of wet and dry Hg than in lower elevation habitats (VanArsdale et al. 2005); and (3) there is a significant relationship between wet and dry Hg deposition models based on Miller et al. (2005) and on Bicknell's Thrush blood Hg levels (Rimmer et al. 2005).

With this exploratory sampling effort of forest songbirds in 2005 and 2006, blood Hg levels were elevated and in some areas above levels of concern. Patterns of blood Hg levels indicate that body size, habitat type, elevation, and geographic location are important variables to measure. Because of the rapid and near-complete loss of breeding populations of the Wood Thrush in the Adirondack Mountains and prominent declines in other areas of eastern New York, including the Catskill Mountains, we hypothesize that elevated Hg levels and deficient Ca levels throughout the breeding range of the Wood Thrush may partly account for some of these declines. Other species, such as the Louisiana Waterthrush and Rusty Blackbird, exhibit similar population trends in New York, and appear to bioaccumulate greater amounts of MeHg than the Wood Thrush, and are therefore also potentially adversely impacted.

As electric utilities are the major sources of atmospheric Hg in the U.S., results from this investigation may provide important information to policy makers on the pervasiveness of Hg in the Northeast and how synergy with other stressors such as acidic deposition could have broad-scale impacts to bird populations and ecosystem health. If future efforts link emission sources from the Ohio River Valley with biological Hg hotspots in New York, the need for regulations by the U.S. Environmental Protection Agency is compelling. No individual point source in New England, New York, or New Jersey releases more than 500 pounds of Hg per year, while several sources in Pennsylvania and Ohio exceed this annual rate of release (Figure 7-1).

Because BRI and colleagues, including those from the Hubbard Brook Research Foundation, are actively linking Hg scientific findings with national policy through Hg briefings to Congress and other means, the jointly funded research by The Nature Conservancy (2005 and 2006) and NYSERDA (2006) could ultimately contribute to a framework for new national legislation to regulate Hg emissions and eventually standardized monitoring efforts. Should the decline of Wood Thrushes and other songbirds truly signal a widespread and major disruption in how forests function in New York, New England, and surrounding areas, then this effort becomes timely to better define potential sources of declines in songbird populations.

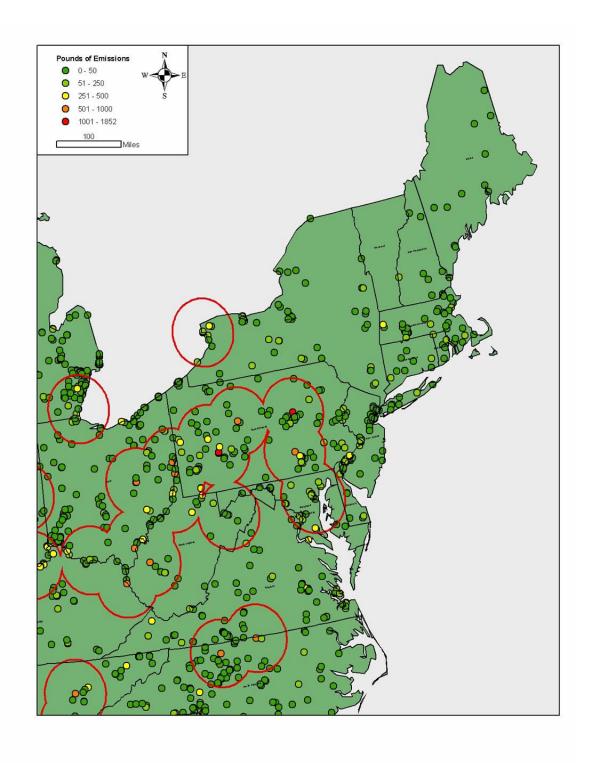


Figure 7-1. Mercury emission point sources in eastern U.S with sources of >500 pounds associated with a 50 mile radius gray circle.

#### 8.0 FUTURE RECOMMENDATIONS

- 1. Continue development of Hg exposure profile to (see Appendix I for map and listing of 2007 sampling stations):
  - a. Identify potential biological Hg hotspots;
  - b. Determine regional spatial gradient of MeHg availability;
  - c. Search for areas and habitats that might exceed the level of concern for Hg in the blood (i.e., bog areas).
- 2. Include an analysis of Hg and Ca in soil, prey items and birds to:
  - a. Quantify the relationship of Hg and Ca in birds for different geographic areas;
  - b. Quantify the relationship of Hg and Ca among soil, prey, and bird compartments;
  - c. Use relationships to develop a predictive model for identifying problem areas and extrapolating such findings across the region.
- 3. Add a field sampling component that measures bird response to pollutant stressors using nationally standardized protocols, such as:
  - a. Birds of Forested Landscapes to determine presence and density of singing males of target species;
  - b. Monitoring Avian Productivity and Survivorship (MAPS) to determine productivity and survivorship of target species.
- 4. Assess risk from Hg to neotropical migrants in the wintering areas.
- 5. Present findings at forums that include scientific and policy oriented outlets, including:
  - a. Scientific conferences and journals;
  - b. National policy arenas, such as the U.S. Senate's Environment and Public Works Committee.
- 6. Explore abilities to add this and similar Hg monitoring efforts to the national Hg monitoring network.

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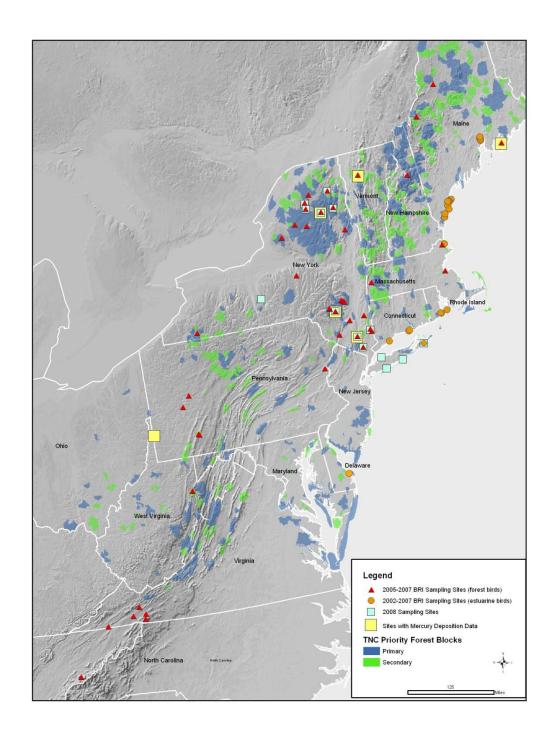
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**Appendix 1.** Current and future sampling stations.



**Appendix 2.** Sampling effort for soil, invertebrates and birds in New York and Pennsylvania, 2005-2006

Site	State	2005	2006	Songbirds	Invertebrates	Soil
Arbutus	NY		х	16	9	1
Allegany State Park	NY	Х	Х	53	24	4
Belle Ayr Mountain	NY	Х		1	6	4
Mill Brook	NY	Х		11	0	0
Black Rock Forest	NY	Х	х	37	17	
Brookfield Forest	NY	Х		7	3	
IES	NY	Х		9	0	0
Devil's Tombstone	NY	Х	Х	69	22	3
Plateau Mountain	NY	Х	х	43	14	4
Tug Hill	NY	Х	х	33	12	2
Shawangunks -Mohonk Preseve	NY		Х	19	15	1
Shawangunks-Sam's Point	NY	Х	Х	29	17	3
Whiteface	NY		х	13	0	0
Hunter Mountain West	NY	Х		11	4	1
Dome Island	NY		Х	10	10	1
Elk Lake	NY		Х	15	10	1
Emmons Bog	NY		х	18	5	1
Ferd's Bog	NY		х	12	6	1
Lake Capra	NY		Х	10	6	1
Neversink-Frost Valley	NY		Х	20	12	1
Spring Pond Bog	NY		Х	19	10	1
Sunday Pond	NY		Х	21	10	1
Totts Gap	PA	Х		23	3	2
Powdermill	PA		Х	36	9	1
Powdermill -Spruce Bog	PA		х	16	3	1
	-	Gran	nd Total	551	227	40

**Appendix 3.** Species, number, age, Hg levels and ancillary natural history and conservation information for avian species sampled.

Species	n	Age	Mean ± SD	Range	Family	Partners in Flight Conservation Rank	Forage Guild	Forage Habitat
Green Heron	1	A	1.31		ARDEIDAE		Water	
Sharp-shinned Hawk	1	A	0.74		ACCIPITRIDAE	8	Hawk	
Hairy Woodpecker	1	A	0.01		PICIDAE	6	Bark	
Yellow-bellied Flycatcher	1	A	0.19		TYRANNIDAE	10	Flycatching	
Traill's Flycatcher	2	A	0.14 ± 0.04	0.12 - 0.17	TYRANNIDAE	9 90	Flycatching	
Eastern Kingbird		Α	0.08		TYRANNIDAE	10	Flycatching	
White-eyed Vireo		A	$0.04 \pm 0.01$	0.03 - 0.05	VIREONIDAE	10	Foliage	Sub-canopy
Blue-headed Vireo		A			VIREONIDAE	8	Foliage	Canopy
Red-eyed Vireo		A			VIREONIDAE	7	Foliage	Canopy
		J	0.06 ± 0.01		***************************************	A.*	· silege	oupy
Black-capped Chickadee		A	0.10 ± 0.08	0.03 - 0.07	DADIDAE	6	Bark	
Diack-capped Chickagee		J	0.14 ± 0.09		FARIDAL		Dain	
Eastern Tufted Titmouse		A	0.06	0.07 - 0.20	PARIDAE	8	Bark	
Eastern Tuited Titmouse	_	J	0.04	0.05 - 0.04			Dark	
0-41	_		0.14	0.05 - 0.04		-	D. J.	-
Red-breasted Nuthatch		A		0.07.000	SITTIDAE	6	Bark	
White-breasted Nuthatch		A	0.07 ± 0.01	0.07 - 0.08		6	Bark	2
Brown Creeper		A	0.09		CERTHIIDAE	9	Bark	
House Wren		A	0.12		TROGLODYTIDAE	6	Foliage	Sub-canopy
Veery	49		$0.05 \pm 0.03$		TURDIDAE	11	Ground	Sub-canopy
		J	$0.01 \pm 0.01$		Secretaria de la composición del composición de la composición de la composición del composición de la composición de la composición del composición de la composición del com		0/7/00/	(3.24 (3.54 (3.55)
Bicknell's Thrush		A	$0.08 \pm 0.02$	0.05 - 0.14	TURDIDAE	18	Ground	Sub-canopy
Service and the service of the servi	1	J	0.06	A 10 1 100	CHEMICAL P.	7.65	- 2000-000	(20) 10 (20) (10) (10) (10) (10) (10) (10) (10) (1
Swainson's Thrush	41	A	$0.08 \pm 0.03$	0.03 - 0.19	TURDIDAE	10	Ground	Sub-canopy
	1	J	0.04	0.05 - 0.04	200000000000000000000000000000000000000	880	180000000	250000000000000000000000000000000000000
Hermit Thrush	64	A	0.06 ± 0.03	0.01 - 0.17	TURDIDAE	6	Mixed	Mixed
		J	0.04 ± 0.02			150	1200000	NEXT
Wood Thrush	63	100	0.08 ± 0.04		TURDIDAE	14	Ground	Litterfall
TTOOG TINGSIT		J	0.03 ± 0.02		TOTOTOTAL	177	Cidana	Littorian
American Robin		A	0.03 ± 0.02		TUDDIDAE	5	Ground	Litterfall
American Robin		J	0.03 ± 0.01		TORDIDAL	9	Ground	Litteriali
Gray Cathird		A	0.05 ± 0.02	0.02 - 0.04	MIMIDAE	9	Mixed	Mixed
Magnolia Warbler		A		0.04 0.20	PARULIDAE	8	Foliage	
Black-throated Blue Warbler		A			PARULIDAE	12		Canopy
							Foliage	Canopy
Yellow-rumped Warbler		A			PARULIDAE	6	Foliage	Canopy
Black-throated Green Warbler		A		0.04 - 0.11	PARULIDAE	11	Foliage	Canopy
Prairie Warbler		Α	0.04		PARULIDAE	14	Foliage	Sub-canopy
Palm Warbler	2	Α			PARULIDAE	8	Foliage	Sub-canopy
4-17-12-12-12-12-12-12-12-12-12-12-12-12-12-		J	$0.40 \pm 0.17$	0.28 - 0.52			2010	
Blackpoll Warbler		Α	0.06		PARULIDAE	12	Foliage	Canopy
Black-and-White Warbler		A			PARULIDAE	9	Bark	
American Redstart		A			PARULIDAE	8	Foliage	Mixed
Worm-eating Warbler		A			PARULIDAE	14	Foliage	Mixed
Ovenbird	27	A		0.01 - 0.30	PARULIDAE	10	Ground	Litterfall
00	1	J	0.01					
Louisiana Waterthrush	4	Α	$0.13 \pm 0.04$	0.08 - 0.18	PARULIDAE	13	Water	
		J	0.15 ± 0.07	0.05 - 0.24		. 0	5500	-
Mourning Warbler	2	A	0.02 ± 0.00	0.01 - 0.02	PARULIDAE	11	Foliage	Sub-canopy
Common Yellowthroat		A			PARULIDAE	8	Foliage	Sub-canopy
	1	J	0.06			10.00		
Hooded Warbler		A		0.03 - 0.18	PARULIDAE	13	Foliage	Sub-canopy
Scarlet Tanager		A	0.03	0,100	THRAUPIDAE	12	Foliage	Canopy
Eastern Towhee		A	0.08		EMBERIZIDAE	11	Ground	Litterfall
Chipping Sparrow		A	0.13		EMBERIZIDAE	7	Ground	Enterial
Savannah Sparrow		A	0.02 ± 0.00		EMBERIZIDAE	9	Ground	
				0.02 0.00				
Song Sparrow		A			EMBERIZIDAE	8	Water	
Lincoln's Sparrow	2	A			EMBERIZIDAE	7	Water	
		J	0.10 ± 0.04	0.06 - 0.14				
Swamp Sparrow		A	0.20		EMBERIZIDAE	7	Water	
Slate-colored Junco		A	0.03		EMBERIZIDAE	8	Ground	
Rose-breasted Grosbeak		Α	0.02		CARDINALIDAE	12	Foliage	Mixed
Indigo Bunting	1	A	0.02		CARDINALIDAE	11	Foliage	Sub-canopy
Red-winged Blackbird		Α	$0.07 \pm 0.01$	0.06 - 0.08	ICTERIDAE	8	Water	

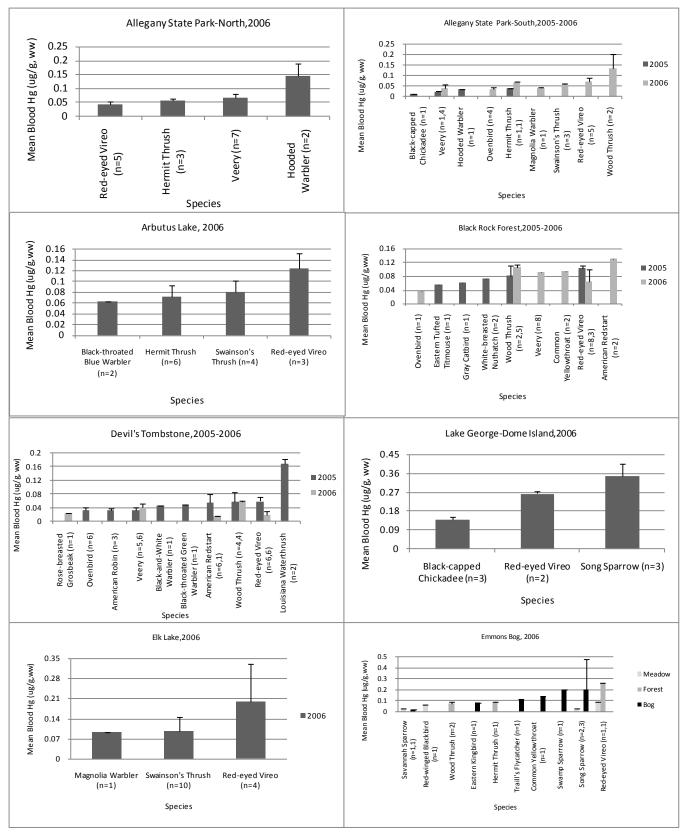
Appendix 4. Soil and exchangeable calcium for all sites, 2005-2006.

		Hg (ppm)		Exchangeable	Ca (cmolc/Kg-soil)	
Site	n	Mean ± SD	Range	Mean ± SD	Range	Region
Arbutus	1	0.70		14.44		Adirondacks
Dome Island	1	0.11		0.57		Adirondacks
Elk Lake	1	0.24		13.53		Adirondacks
Ferd's Bog	1	0.19		11.57		Adirondacks
Spring Pond Bog	~	0.10		0.75		Adirondacks
Sunday Lake	1	0.09		0.57		Adirondacks
Allegany State Park	ფ	$0.10 \pm 0.00$	0.10 - 0.11	$0.63 \pm 0.15$	0.49 - 0.79	Allegheny Plateau
Belle Ayr Mountain	1	0.07		2.86		Catskills
Devil's Tombstone	2	$0.20 \pm 0.10$	0.13 - 0.27	1.57 ± 1.41	0.58 - 2.57	Catskills
Emmons Bog	1	0.08		3.71		Catskills
Hunter Mountain West	1	0.12		1.75		Catskills
Lake Capra	1	0.22		1.16		Catskills
Neversink	1	0.07		0.28		Catskills
Plateau Mountain	1	0.35		3.14		Catskills
Brookefield	1	0.09		2.58		Central New York
Tott's Gap	1	0.09		5.16		Eastern PA
Black Rock Forest	2	$0.24 \pm 0.04$	0.21 - 0.27	$0.20 \pm 0.04$	0.17 - 0.23	Lower Hudson River Valley
Mohonk Preserve	1	0.09		1.08		Lower Hudson River Valley
Sam's Point	2	$0.23 \pm 0.07$	0.18 - 0.28	3.30 ± 2.16	1.77 - 4.83	Lower Hudson River Valley
Tug Hill	2	$0.30 \pm 0.13$	0.21 - 0.39	4.26 ± 1.61	3.12 - 5.40	Tug Hill
Powdermill	1	0.10		0.64		Western PA
Spruce Bog	1	0.23		1.54		Western PA

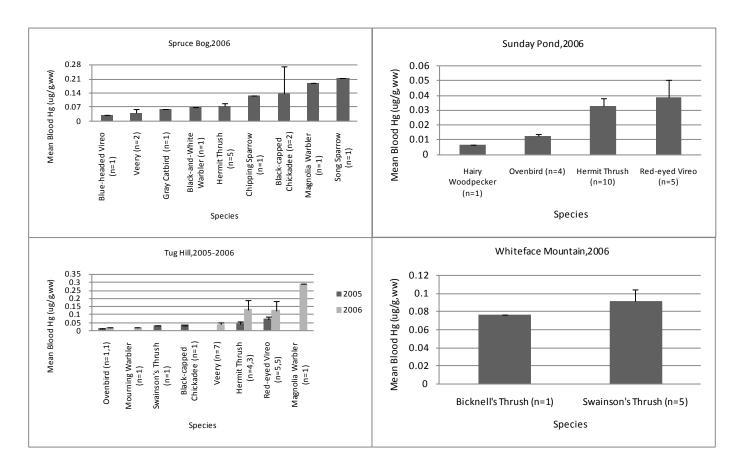
**Appendix 5.** List of species opportunistically observed on Dome Island, NY (Observed by Melissa Duron, Amy Sauer and Maria Neal from 7/24/2006 to 7/26/2006).

	Amphibians	
Red-backed Salamander	Plethodon cinereus	
Red Eft	Notophthalmus viridescens	
	Birds	
Cedar Waxwing	Bombycilla cedrorum	
American Crow	Corvus brachyrhynchos	
Song Sparrow	Melospiza melodia	
Black-capped Chickadee	Poecile atricapillus	
White-breasted Nuthatch	Sitta carolinensis	
Red-eyed Vireo	Vireo olivaceus	
Hairy Woodpecker	Picoides villosus	
	Invertebrates	
Funnel Web Spider*	Agelenopsis spp.	
Orb Weaver*	Kaira spp.	
Fishing Spider with egg sac		
Long-jawed Orb Weaver*	Glenognatha spp.	
Darkling Beetle*	Merinus spp.	
Pill bug*	Armadillidium vulgare	
Harvestmen		
Millipede*		
	Mammals	
Eastern Chipmunk	Tamias striatus	
	Plants	
Helleborine	Epipactis helleborine	
Canada Mayflower	Maianthemum canadense	
False Solomon Seal	Maianthemum racemosum	
Maple Leaf Viburnum	Vibumum acerifolium	
Indian Pipe	Monotropa uniflora	
Black Birch	Betula lenta	
Paper Birch	Betula papyrifera	
Ironwood	Carpinus caroliniana	
Amercian Beech	Fagus grandifolia	
Red Oak	Quercus rubra	
Beech drops	Epifagus americana	
Red Trillium	Trillium erectum	Protected-Exploitably Vulnerable
Sarsaparilla	Smilax regelii	
Quaking Aspen	Populus tremuloides	
American Basswood	Tilia americana	
White Cedar	Thuja occidentalis	
Balsam Fir	Abies balsamea	
Red Pine	Pinus resinosa	
White Pine	Pinus strobus	
Eastern Hemlock	Tsuga canadensis	
Rock Polypody	Polypodium virginianum L.	Protected-Exploitably Vulnerable
Bracken Fern	Pteridium spp.	
Striped Maple	Acer pensylvanicum	
Red Maple	Acer rubrum	
Sugar Maple	Acer saccharum	

Appendix 6. Average Hg levels by species at each sample site.



# Appendix 6, continued:



## SECTION II – BAT MERCURY EXPOSURE PROFILE

## **Abstract:**

Recent research indicates that the northeastern United States contains some of the highest levels of methylmercury (MeHg) in North America. Furthermore, levels in the Northeast exceed those associated with behavioral and reproductive impacts in aquatic wildlife. While most studies have emphasized MeHg exposure and effects in birds, few studies have included bats. In summer 2006, a pilot bat sampling effort was conducted in the Adirondack Mountain Region of New York to assess Hg exposure. Eight sites were chosen to represent broad spatial distribution of Hg exposure. Our findings indicate that some areas have environmental Hg loads above levels of concern when compared to dosing studies for mammals. While no data are available for bat-specific Hg levels of concern the multiple anthropogenic threats that New York bats currently face, including white-nose syndrome and wind power turbines, elevate the need to better understand potential additional stressors such as Hg.

## Figures and Table for Bats

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Table 13-3. Comparisons for all pairs using Tukev-Kramer HSD	14-7

## 10.0 EXECUTIVE SUMMARY

In the interest of assessing potential impacts and injury to invertivore mammals in New York, an exploratory study determined mercury (Hg) concentrations and potential effects on the local breeding bat community. Based on findings from the assessment for 2006, there is insufficient information to conclude if breeding bats from New York have potentially harmful body burdens of Hg.

Blood provides an evaluation of recent dietary uptake of MeHg (e.g., likely within the past 1-2 weeks), while fur provides a long-term evaluation and is likely related to individual age. Of the 96 individual bats sampled in New York, 5-16% contained fur Hg burdens that exceeded levels of concern identified for other mammals. The highest fur Hg concentration of 35 ug/g (fw) was detected in an adult male little brown bat (*Myotis lucifugus*) from Big Moose Lake.

Bats are increasingly of high concern to conservation agencies because of recent,large-scale mortality stressors such as fungal diseases (i.e., white nose syndrome) and the growing interest in industrial wind turbines. Therefore, high resolution investigations to determine spatially explicit effects from Hg on reproductive success, survival, and physiological effects are of even greater importance and urgency.

## 11.0 STUDY AREA

Bat capture and sampling occurred at eight different sites in June and July of 2006 across the eastern portion of New York (Figure 11-1)

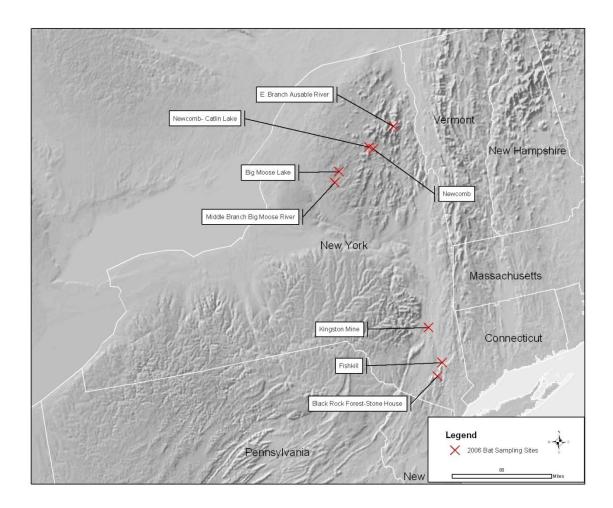


Figure 11-1. New York sampling locations, 2006.

## 12.0 METHODS

## 12.1 CAPTURE AND SAMPLE COLLECTION

Single, double, and triple high mist nets were strung directly in front of ledge outcroppings, between trees along small access roads, or in the middle of rivers to funnel bats into nets. Using the assumption that bats fly to water for drinking and feeding purposes after leaving daytime roosts, roads that led towards water were chosen.

Nets were set at dusk and monitored until at least 2300 hours; if bats were being captured, nets were left open until 0100 hours. All bats captured were identified to species, checked for reproductive status, sexed, and aged. Small blood samples were collected by puncturing the uropatagial vein (or wing vein) or the femoral vein in the leg with a 27.5 gauge needle. Blood was collected in heparinized capillary tubes. Fur samples were collected with clean stainless steel scissors. All bats were released unharmed at the site.

#### 12.2 SAMPLE ANALYSIS

Laboratory analysis was conducted by Texas A&M Trace Element Research Lab (TERL), College Station, Texas. All tissue samples were analyzed for total mercury using Cold Vapor Atomic Absorption (CVAA) methods. Mercury concentrations are presented as fresh weight (fw) for fur. Only fur samples were analyzed. Blood samples were archived. Instead of analyzing methylmercury (MeHg) concentrations we focused on total Hg. Total Hg is less costly and it is well established that fur Hg concentrations are >95% methyl (Wolfe et al. 2007).

## 13.0 RESULTS

## 13.1 SAMPLING EFFORT

In 2006, a total of 96 bats were sampled at six sites in New York. Collaborators provided fur samples from two additional sites, Fishkill and Kingston Mine. During 17 nights of trapping, the following seven species were captured: little brown bat (*Myotis lucifigus*, n=64), northern long-eared bat (*Myotis septentrionalis*, n=11), Indiana bat (*Myotis sodalis*, n=11) big brown bat (*Eptesicus fuscus*, n=10), eastern small-footed bat (*Myotis leibii*, n=2), eastern red bat (*Lasiurus borealis*, n=1), and the hoary bat (*Lasiurus cinereous*, n=1) (Table 1).

Table 13-1. Location and species captured during 2006 effort.

Species*	Black Rock Forest	E. Branch Ausable River	Newcomb	Newcomb- Catlin Lake	Fishkill	Big Moose Lake	Middle Branch Big Moose River	Kingston Mine	Total
<b>EPFU</b>	0	2	0	0	7	0	0	1	10
LABO	0	0	0	0	0	0	0	1	1
LACI	0	1	0	0	0	0	0	0	1
MYLE	0	0	0	0	0	0	0	2	2
MYLU	1	0	15	2	14	11	19	2	64
MYSE	6	0	0	1	0	0	0	0	7
MYSO	0	0	0	0	11	0	0	0	11
Total	7	3	15	3	32	11	19	6	96

\*EPFU=Big Brown bat, LABO=Eastern Red bat, LACI=Hoary bat, MYLE=Eastern Small-footed bat, MYLU=Little brown bat, MYSE=Northern Long-eared bat, MYSO=Indiana bat

#### 13.2 MERCURY EXPOSURE IN BATS

Total fur Hg concentrations were determined for 96 individuals. Site-specific mean fur Hg concentrations ranged from 3.43 ug/g (+/-2.72) at Newcomb-Catlin Lake to 14.90 ppm (+/-11.25) at Big Moose Lake. Fur Hg concentrations from all areas ranged between 0.94 ug/g and 35.00 ug/g (Table 2). The lowest Hg concentration was found in a juvenile male little brown bat at Middle Branch Big Moose River (0.94 ppm) while the highest concentration was detected in an adult male little brown bat from Big Moose Lake (35.00 ug/g) (Table 2).

Table 13-2. Mean fur Hg concentrations (ug/g, fw) by location.

Location	n	Mean	s.d. +/-	Min	Max
Black Rock Forest-Stone House	7	5.22	2.41	2.62	9.72
E. Branch Ausable River	3	3.47	1.86	1.63	5.35
Newcomb	15	6.61	3.53	1.16	13.80
Newcomb- Catlin Lake	3	3.43	2.72	1.48	6.54
Big Moose Lake	11	14.90	11.25	3.65	35.00
Middle Branch Big Moose River	19	4.26	2.66	0.94	10.70
Kingston Mine	6	8.73	5.79	2.11	16.00
Fishkill	32	8.59	6.72	1.23	30.20

We used a Tukey-Kramer HSD to compare fur Hg means among sites (Table 3). Big Moose Lake had significantly different means from Newcomb, Black Rock Forest and the Middle Branch of the Big Moose River (Table 3). All other areas did not have significantly different fur Hg means (Table 3).

Table 13-3. Comparisons for all pairs using Tukey-Kramer HSD\*.

Location	Big Moose Lake	Kingston Mine	Fishkill	Newcomb	Black Rock Forest- Stone House	Middle Branch Big Moose River	E. Branch Ausable River	Newcomb - Catlin Lake
Big Moose Lake	~	-3.31	22	0.87	0.63	3.55	-0.74	-0.70
Kingston Mine	-3.32	~	-8.17	-6.90	-6.89	-4.28	-7.96	-7.92
Fishkill	-0.23	-8.19	~	-3.86	-4.43	-1.08	-6.17	-6.13
Newcomb	0.87	-6.91	-3.86	~	-7.17	-4.11	-8.69	-8.65
Black Rock Forest- Stone House	0.63	-6.89	-4.43	-7.17	~	-7.30	-11.15	-11.11
Middle Branch Big Moose River	3.56	-4.28	-1.08	-4.11	-7.30	~	-10.83	-10.79
E. Branch Ausable River	-0.75	-7.96	-6.17	-8.69	-11.15	-10.83	~	-15.22
Newcomb- Catlin Lake	-0.71	-7.92	-6.13	-8.65	-11.11	-10.79	-15.22	~

\*Positive values show pairs of means that are significantly different.

#### 14.0 DISCUSSION

## 14.1 COMPARISONS WITH OTHER BAT HG CONCENTRATIONS

There have been very few investigations on Hg exposure in bats. Miura et al. (1978) examined various species of Chiroptera from areas in Japan sprayed with Hg fungicides. They measured total fur Hg levels and found 33.0 +/- 6.3 ug/g (fw) in 1965 and 33.7 +/- 4.2 ppm (fw) in 1966. While the mean fur Hg concentrations found in Chiroptera from New York did not exceed the Japanese study, two individual bats from Big Moose Lake did.

Massa and Grippo (2000) examined various Chiroptera species from rivers in Arkansas that were under fish consumption advisories and found fur Hg concentrations ranging from 1.0 to 30 ug/g (fw). They concluded that Hg accumulation had exceeded the hazard criteria set by the U.S. Fish & Wildlife Service and that Hg accumulation in the bats is a serious problem that warrants further investigation.

Hickey et al. (2001) examined fur Hg concentrations in various Chiroptera species from eastern Ontario and adjacent Quebec, Canada. In 1997, they pooled samples from five sites and found Hg concentrations ranging from 2.0 to 7.6 ug/g (fw) in fur. In 1998, they sampled the same sites and found fur Hg concentrations that approached or exceeded 10.0 ug/g (fw). Burton et al. (1977) found that mice with fur Hg concentrations of 7.8 ug/g (fw) and 10.8 ug/g (fw) showed behavioral deviations including decreased ambulatory activity and stress tolerance, and decreased swimming ability, respectively. The bats from New York had mean Hg concentrations higher at Big Moose Lake and 16% of bats sampled exceeded 10.8 ug/g (Figure 14-1). Based on the study by Burton et al. (1977), our findings suggest that free-ranging bats at some sites in New York may be exposed to levels of Hg sufficient to cause sublethal effects.

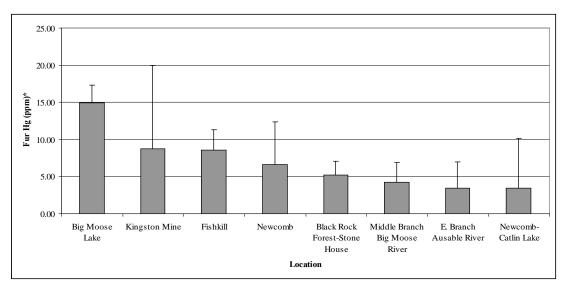


Figure 14-1. Mean fur Hg (ug/g, fw) by site.

#### 14.2 PREY CHOICE AND FIELD DOSE LEVELS

When bats emerge from their day roosting areas, they require drinking water. Bats use both aerial and gleaning techniques when foraging over river surfaces and floodplain edges. Carter et al. (2003) found northern long-eared bats (*Myotis septentrionalis*) main prey was Coleoptera and Lepidoptera followed by Diptera. This study showed diets of bats from West Virginia did not differ from diets of bats in other regions of the United States. Other studies (Whitaker and Hamilton 1998, Brack and Whitaker 2001) found northern long-eared and little brown bats typically preyed on moths and beetles, but overall had a varied diet including spiders. Spiders have been shown, in previous studies, to have elevated Hg concentrations (Adair et al. 2002, Cocking et al. 1991). Powell (1983) showed that aquatic nymphs of flying insects from a Virginia river polluted by a point source had elevated mercury. Insectivorous eastern pipistrelles (*Pipistrellus subflavus*) showed elevated Hg concentrations in livers and muscle tissue, but reference areas were not sampled.

Baron et al. (1999) completed a risk assessment for aerial insectivorous wildlife on the Clinch River, TN (Oak Ridge Reservation). Using a model, they determined the dose levels for the NOAEL and LOAEL for little brown bats are 0.11 and 0.56 ug/g, respectively. Bats experiencing exposure equal or greater than the LOAEL were found to display impaired growth, reproduction, and offspring viability (Verschuuren 1976). Although bat prey items were not sampled in our New York study, we recommend collections in future efforts.

## 15.0 CONCLUSIONS

Based on findings from the pilot assessment for bats in 2006, there is sufficient information to conclude that bats from in the Big Moose Lake, Fishkill and Kingston Mine area may have potentially harmful body burdens of Hg. Of the 96 individual bats sampled in New York, 16% were considered to contain fur Hg burdens that exceeded concentrations of concern in mice (i.e., 10.8 ug/g, fw). Five of the 96 individuals exceeded the 20.0 ug/g (fw) fur Hg concentrations of concern in mink and otter (Yates et al. 2004, 2005). Some bat fur Hg concentrations in Big Moose Lake and Fishkill area exceeded studies in Arkansas, Japan, and Ontario where researchers considered their bat populations at risk. Although the effects of methlymercury on bats are unknown, Hg effect levels from other mammalian species indicate that free-ranging bats in New York are likely at risk.

Bat species should be included in further assessments for determining injury to insectivorous mammals. Because some bats are endangered and are of high conservation concern, higher resolution investigations to determine spatially explicit effect levels on reproductive success, survival, and physiological effects should be based on these species.

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#### SECTION III – HERPETOFAUNA MERCURY EXPOSURE PROFILE

## **Abstract:**

Recent research indicates that the northeastern United States contains some of the highest levels of methylmercury (MeHg) in North America. Furthermore, levels in the Northeast exceed those associated with behavioral and reproductive impacts in aquatic wildlife. While most studies have emphasized MeHg exposure and effects in birds and mammals, few studies have included herpetofauna. In summer 2006, an exploratory herpetofauna sampling effort was conducted in the Adirondack Mountain region of New York to assess Hg exposure in amphibians and reptiles. Thirteen lakes were chosen to represent broad spatial and elevation distribution. Our findings indicate that amphibians from the Black River, Oswegatchie River, Raquette River, Hudson River, and St. Lawrence River watersheds have Hg levels below the malformation threshold. Depending on species, however, Hg concentrations in herpetofauna from the Adirondack Park were generally greater than those found in other established studies. While effect levels are relatively unknown, further insight into the relationship among species and between size and Hg levels was developed. Evidence from other studies indicates that further investigations in New York are recommended.

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## 17.0 EXECUTIVE SUMMARY

Recent research indicates that the northeastern United States contains some of the highest levels of methylmercury (MeHg) in North America. Furthermore, levels in the Northeast exceed those associated with behavioral and reproductive impacts in aquatic wildlife. While most studies have emphasized MeHg exposure and effects in bird and mammals, few studies have included herpetofauna. In the summer of 2006, an exploratory herpetofauna sampling effort was conducted in the Adirondack Mountain region of New York to assess Hg exposure in amphibians and reptiles. Thirteen lakes were chosen to represent broad spatial and elevation distribution. Samples were collected from a subset of lakes previously sampled in 2005 for Hg in Common Loons (Gavia immer) and other biota. Seven species, comprising of 76 individuals of amphibians and reptiles were collected during the 27 May to 24 September 2006 sampling period. Tissues were collected from the Black River, Oswegatchie River, Raquette River, Hudson River, and St. Lawrence River watersheds. Our findings show that amphibians from the Black River, Oswegatchie River, Raquette River, Hudson River, and St. Lawrence River watersheds have Hg levels below the malformation threshold. Depending on species however, Hg concentrations in herpetofauna from the Adirondack Park were generally greater than those found in other established studies. While effect levels are relatively unknown, evidence from other studies indicates that further investigations in New York are recommended.

#### 18.0 INTRODUCTION

Recent research indicates that northeastern United States contains some of the highest levels of MeHg in North America (Evers et al. 1998, 2005, 2007). Furthermore, levels in the Northeast exceed those associated with behavioral and reproductive impacts in wildlife (Evers et al. 2008). While most studies have emphasized Hg impacts in avian, mammalian, and fish food webs, few studies have addressed Hg impacts in herpetofauna, despite worldwide population declines (Houlahan et al. 2000). Amphibian declines have been reported in both eastern and western Canada (Wyman, 1990). Declines in Australia have been associated with habitat destruction and mercury pollution as a result of gold mining (Wyman, 1990).

Since amphibians are known to accumulate heavy metals, they can be useful indicators of pollution levels in the environment (Punzo 1993, Bank et al. 2007a, Wolfe et al. 2007). As such, amphibians can serve an important role in assessing aquatic and even terrestrial environments of contaminant stress. River frogs (*Rana heckscheri*) showed a 69% decrease in fertilization success when gametes were exposed to 2.5mg/L of Hg (Punzo (1993). Unrine et al (2004) found that high Hg levels in southern leopard frog larvae (*Rana sphenocephala*) decreased the rate of tail absorption. Decreased tail absorption can lead to an increase in predation and malformation, impacting the survival rate of tadpoles.

Bank et al. (2007) found that some streams in the Acadia National Park (ANP) experience episodic acidification events that enhance leaching of toxic aluminum and Hg, degrading habitat conditions for sensitive aquatic biota such as salamanders. He suggested that atmospheric pollutants and the enhanced mobilization of toxic substances are the best explanation for declines of dusky salamanders (*Desmognathus ochrophaeus*) in the Park (Bank et al. 2006). In another study, Bank et al. (2005) found patterns of Hg bioaccumulation in two-lined salamanders (*Eurycea bislineta*) were influenced by geographical location and watershed conditions. Mercury concentrations in salamander larvae observed in the ANP, feeding on benthic sources, were significantly greater than brook trout samples collected from the same stream.

Bank et al. (2007) found that bioaccumulation of Hg in the North American bullfrog (*Rana catesbeiana*) and green frog (*Rana clamitans*) tadpoles varied across ponds in Acadia National Park. In addition, tadpole Hg correlated with Hg concentrations in water. A statistical significance was detected between tadpole length and Hg levels, showing a possible relationship between bioaccumulation and age.

Bergeron et al. (2007) documented similar blood Hg concentrations in painted turtles (*Chrysemys picta*), snapping turtles (*Chelydra serpentina*), eastern red-bellied (*Pseudemys rubriventris*), and common musk turtles (*Sternotherus odoratus*). Golet and Haines (2001) found snapping turtle scutes contained high Hg

levels, which were correlated with levels found in muscle tissue and blood. Blanvillain et al. (2007) studied temporal fluctuations in Hg levels in the blood and scutes of diamondback terrapins (*Malaclemys terrapin*). Scute Hg concentration did not vary seasonally, but Hg blood concentrations were lowest in August. Significant relationships were documented between blood and scute Hg concentrations, particularly in June, August and October. Blanvillain et al. (2007) also found that female terrapins had significantly higher levels of Hg.

These and other studies provide compelling evidence to investigate the availability of methylmercury in herpetofauna of Adirondack Park, New York. This study is an exploratory effort to advance that need.

## 19.0 STUDY AREA

Samples were collected from a subset of lakes previously sampled in 2005 for Hg in Common Loons and other biota. Thirteen lakes were opportunistically chosen, based on their east-west spatial distribution as well as varying elevations (Figure 19-1). Tissues were successfully sampled within the Black River, Oswegatchie River, Raquette River, Hudson River, and St. Lawrence River watersheds.

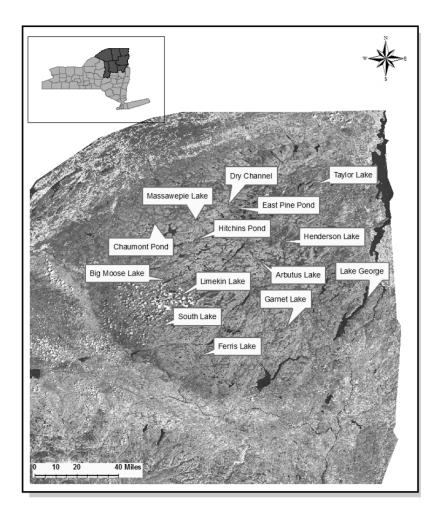


Figure 19-1. Herpetofauna sample locations in the Adirondack Park, NY, 2006.

## 20.0 METHODS

#### 20.1 CAPTURE METHOD FOR TADPOLES

Salamander and frog tadpole searches were performed by walking the perimeter of the available shoreline at each study site, focusing on vegetative areas. To catch tadpoles by dip net we used 0.125"-mesh dip nets or 0.25"-mesh two-pole seines. Dip net surveys were conducted at night using spotlights or during the day by carefully rooting around aquatic vegetation. Two-pole seines were also used. All potential aquatic habitats, especially the deeper sections of pools, were intensively sampled.

## 20.1.1 Tadpole Processing

All captured tadpoles were examined visually while holding the net or trap just under water. Tadpoles requiring further examination were only handled with wet hands frequently irrigated with site water in order to prevent the skin from drying. Tips of the tadpole tail were snipped just posterior of the last vertebra, using surgical scissors sterilized in Betadine®. A subset of individuals was preserved as whole animal tissue specimens; all others were released directly at the site. All individuals were caught by using minnow traps and dip nets. Tadpoles were individually bagged in reclosable plastic bags, sorted by species and iced for transport. Samples were shipped to Texas A&M University, Trace Element Research Laboratory, College Station, Texas, in individually labeled reclosable plastic, and refrigerated until processing.

## 20.2 CAPTURE METHOD FOR TURTLES

All samples were caught by hoop trap, net or by hand catching. Painted and map turtles (*Graptemys geographica*) were located congregating on logs or at the water's edge. Snapping turtles were primarily captured in shallow water with abundant vegetative cover. Same species were preferred, but mixing of species was acceptable as long as all individuals were identified to species.

## 20.2.1 Hoop and Minnow Traps

Hoop traps were suspended in water and baited with a small amount of canned cat food placed in a perforated reclosable plastic bag. Stakes and ropes were used for suspension in order to insure an air pocket at all times (air pockets are essential to prevent suffocation of trapped animals). Baiting occurred past 1400 hours and traps were checked by 0800 hours. All traps locations were recorded by GPS coordinates.

## 20.2.2 Catching by Net

Coarsely meshed fish landing nets were used to capture turtles. Basking turtles were visually identified from a distance, using binoculars. Individuals were approached from a direction away from the head and carefully netted. If the turtle dove into the water, net surveys were conducted by carefully netting the bottom adjacent to the basking site.

## 20.2.3 Catching by Hand

Catching by hand was possible by using a boat and snorkel. Basking turtles were approached underwater. The swimmers drifted toward the turtle without allowing their fins to break the surface of the water. The boat was also used to create a sight barrier. Once the turtle was startled into the water by the proximity of the swimmer, it could then be hand-caught.

## 20.2.4 Turtle Processing

Post-capture, scute samples were taken from the outermost edge of the posterior marginal scutes, starting with the post central scutes until a large enough sample size was gathered from each individual. Sample weight preference was 0.4g, with no less than 0.1g collected from each individual. Samples were taken using large nail clippers or surgical scissors sterilized with Betadine®. Care was taken to insure the bone was not nicked or included in the sample. Each sample was bagged separately and sorted by species. All individuals were kept cool throughout the processing and returned to the water at the capture location.

#### 20.3 STATISTICAL ANALYSIS

Due to the small sample size for each species, statistical analysis was limited. Using JMP software, we conducted multivariate correlations using nonparametric Spearman's rho test between Hg and weight, snout to vent length, and snout to tail length on salamander tadpoles. Using SigmaPlot software we conducted a linear regression model between weight and  $log_{10}$  of Hg in map turtles.

#### 20.4 LABORATORY ANALYSIS

Laboratory analysis was conducted by Texas A&M Trace Element Research Lab (TERL), College Station, Texas. All tissue samples were analyzed for total mercury Cold Vapor Atomic Absorption (CVAA) methods. Mercury concentrations are presented as wet weight (ww) values. Total Hg was analyzed instead of MeHg because of limited funding for this exploratory study, total Hg levels provide adequate comparisons in the context of this study, and other comparable studies are based on total Hg measurements.

## 21.0 RESULTS AND DISCUSSION

#### 21.1 SAMPLING EFFORT

Seventy-six samples were collected from 13 lakes between May and September, 2006. A total of seven species were collected, including: eastern red-backed salamander (*Plethodon cinereus*) (n = 19), eastern newt (*Notophthalmus viridescens*) (n = 20), North American bullfrog (n = 7), green frog (n = 4), and three species of turtles: snapping turtle (n = 2), painted turtle (n = 1), map turtle (n = 24; Table 4-1).

Table 21-1. Location and herpetological species captured during 2006 effort (species code and common name provided\*).

Lake	RBSA*	EANE*	NABU*	GRFR*	SNTU*	PATU*	MATU*	Total
Arbutus Lake	8	0	0	3	0	0	0	10
Big Moose Lake	2	0	0	0	0	0	0	2
Chavmont Pond	0	0	1	0	0	0	0	1
Dry Channel	2	0	0	0	0	0	0	2
East Pine Lake	3	0	0	0	1	0	0	4
Ferris Lake	0	10	4	0	0	0	0	14
Garnet Lake	0	2	0	0	0	1	0	3
Henderson Lake	0	3	0	0	0	0	0	3
Hitchins Pond	2	0	0	0	0	0	0	2
Lake George	0	0	0	0	0	0	24	24
Limekin Lake	2	0	0	0	0	0	0	2
Massawapie Lake	0	0	0	0	1	0	0	1
South Lake	0	5	0	1	0	0	0	6
Taylor Pond	0	0	2	0	0	0	0	2
Total	19	20	7	4	2	1	24	76

<sup>\*</sup> RBSA= red-backed salamander, EANE = Eastern newt, NABU = North American bullfrog, GRFR = green frog, SNTU = snapping turtle, PATU = painted turtle, MATU = map turtle.

## 21.2 TURTLE ANALYSIS

Map turtles in the 100-300g weight class had the lowest mean Hg levels at 0.22 ug/g (dw), while snapping turtles in the highest weight class (11-13kg) had the greatest Hg exposure with a mean of 4.76 ug/g (dw). We found a significant relationship between weight and Hg levels (P < 0.001,  $r^2 = 0.57$ ) in map turtles (Figure 21-1). With the exception of snapping turtles, Bergeron (2007) found body mass did not explain varying amounts of Hg levels in turtles ( $r^2 = 0.05$ ). Because of the small sample sizes of snapping and painted turtles in this study, comparative statistical analyses could not be conducted.

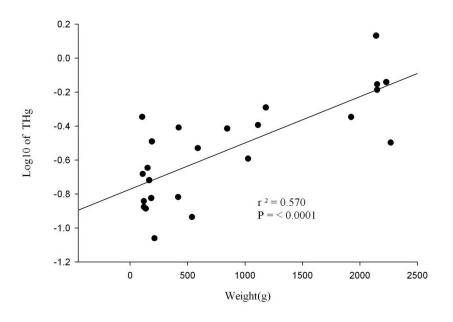


Figure 21-1. Map Turtle (Graptemys geographica) Log10 THg vs. weight.

Variation in Hg levels found in turtles could be a result of weight, trophic level, and prey items. Golet and Haines (2001) found 0.5-3.3 ug/g (dw; n = 26) in snapping turtle scutes from southeastern Connecticut. Bergeron et al. (2007) found levels up to 3.6 ug/g (dw; n = 552) in whole blood in four turtle species. Blanvillain et al. (2007) found scute Hg concentration ranging between 0.26-4.42 ug/g (dw; n = 34) in diamondback terrapin females. Scute Hg levels in this study ranged from 0.10-6.83 ug/g dw ( Table 21-2, 21-3).

Table 21-2. Comparison of Hg (ug/g, dw) in turtle scutes with other studies.

Study	Location	Species	n	Mean	SD	Range
BRI	Lake Geoge, NY	Map turtle	24	0.40	0.31	0.10 - 1.49
BRI	Adirondacks, NY	Snapping turtle	2	4.76	2.93	2.68 - 6.83
Golet and Haines (2001)	Southeastern, CT	Snapping turtle	26	-	-	0.50 - 3.30
Blanvillain et al. (2007)	South Carolina	Diamondback terrapin*	35	-	-	0.26 - 4.42

<sup>\*</sup>female

Table 21-3. Summary statistics of Hg (ug/g, dw) in turtle scutes.

Species	Weight class (g)	n	mean	SD	range
Map turtle	100-300	10	0.22	0.12	0.10 - 0.50
Map turtle	301-600	4	0.26	0.14	0.13 - 0.44
Painted turtle	301-601	1	1.58		1
Map turtle	601-900	1	0.43		•
Map turtle	901-1300	3	0.43	0.14	0.28 - 0.57
Map turtle	1900-2300	6	0.77	0.39	0.35 - 1.49
Snapping turtle	11000-13000	2	4.76	2.93	2.68 - 6.83

## 21.3 TADPOLE ANALYSIS

Tadpole Hg exposure was measured in whole body and tail tips for frogs and salamanders. The highest mean Hg level was found in the eastern newt at 0.10 ug/g (ww) for whole body and 0.12 ug/g (ww) for tail (Table 21-4). The lowest Hg exposure was found in green frogs at 0.03 ug/g for whole body and 0.02 ug/g (ww) for tail. (Table 21-4,). A subset of tadpole samples were measured for both tail tip and whole body in the same individuals. Tadpole tail tips represented 45% to 94% of the whole body Hg (Figure 21-2).

Table 21-4. BRI and reference Hg (ug/g,dw) in tadpole whole body and tail tip samples.

				Tail Tip			Wholebody			
Species	Study	Location	n	Mean	SD	Range	n	Mean	SD	Range
		Adirondack Mtns,								
Eastern newt	BRI	NY	17	0.13	0.30	0.00 - 1.3	10	0.10	0.06	0.02 - 0.23
Eastern red-backed		Adirondack Mtns,								
salamander	BRI	NY	18	0.04	0.03	0.01 - 0.10	3	0.05	0.03	0.02 - 0.07
		Adirondack Mtns,								
Green frog	BRI	NY	4	0.02	0.01	0.01 - 0.03	3	0.03	0.01	0.02 - 0.03
	Bank et al.	Acadia National								
	(2007)	Park, ME	•	-	-	-	60	0.07	-	0.03 - 0.11
		Adirondack Mtns,								
North American bullfrog	BRI	NY	6	0.02	0.01	0.01 - 0.04	3	0.05	0.01	0.03 - 0.06
	Bank et al.	Acadia National								
	(2007)	Park, ME	-	-	-	=	30	0.06	-	0.02 - 0.08
Northern two-lined	Bank et al.	Acadia National								
salamander	(2007)	Park, ME	-	-	-	-	116	0.07	0.00	-
	Bank et al.	Shenandoah NP,								
	(2007)	VA	-	-	-	-	60	0.03	0.00	-

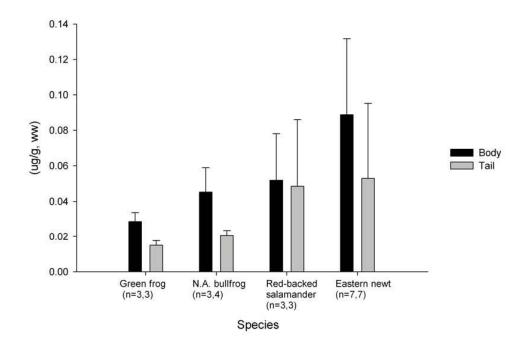


Figure 21-2. Body and tail comparison for Hg (ug/g, ww) in same individuals.

The Spearman's rhos nonparametric correlation analysis (Table 21-5) indicated that length and weight of red-backed salamander and eastern newt tadpoles influences Hg levels. Among salamander tadpoles, we found a significant correlation between these morphometrics and Hg in red-backed salamanders, but there was no such relationship in eastern newts. This could be due to different life cycles, behaviors, or sample sizes between the two species. Our Hg data also did vary between salamander species, which could be due to trophic level, prey selection, and habitat. Whole body frog tadpole Hg levels did not exceed the 0.30 to 0.66 malformation effects levels established by Unrine et al. (2004). Whole body frog tadpole Hg levels ranged from 0.02 - 0.06 ug/g (ww; Table 21-4).

Compared to Bank et al. (2005) who reported a mean of 0.06 ug/g (ww) in whole body northern two-lined salamander (*Eurycea bislineata*) larvae inhabiting the Acadia National Park, our whole body Hg mean for eastern red-backed salamanders was less (0.05 ug/g, ww) and for eastern newts was greater (0.10 ug/g, ww; Table 21-4). The differences could be related to differing habitats, geography, and food webs.

 $Table\ 21-5.\ Correlation\ analysis\ between\ measurement\ and\ total\ Hg\ (ug/g,\ ww)\ in\ tail\ tips\ for\ eastern\ newt\ (Notophthalmus\ vividescens)\ and\ eastern\ red-backed\ salamander\ (Plethodon\ cinereus).$ 

Species	Measurement correlated with Hg		R value	P value
Eastern newt				
	Weight	20	0.25	0.29
	Snout to Vent Length	20	0.01	0.92
	Snout to Tail tip Length	20	0.20	0.45
Eastern red-backed salamander				
	Weight	18	0.79	< 0.01
	Snout to Vent Length	18	0.49	0.04
	Snout to Tail Tip Length	18	0.55	0.02

## 22.0 CONCLUSION

Our findings show that amphibians from the Black River, Oswegatchie River, Raquette River, Hudson River, and St. Lawrence River watersheds have Hg levels below the malformation threshold. Depending on species however, our results were greater than those found in other established studies (Bank et al. 2004, Bank et al. 2007a,b, Blanvillain et al. 2007, BRI unpubl. data, Golet and Haines 2001, Unrine et al. 2004). Among turtles, we found lower Hg levels in map turtles when compared to Bergeron et al. (2007) and Golet and Haines (2001). Mercury levels in this species also varied with weight. When comparing Hg levels in two snapping turtles, we found levels higher than those found in Bergeron et al. (2007) and Golet and Haines (2001). Overall, data from this study suggest that length and weight of amphibian tadpoles and reptiles could influence Hg levels. Furthermore, our Hg data did vary between species, which could be related to trophic levels, prey selection, habitat and morphometrics. While effect levels are relatively unknown, evidence from other studies indicates that further investigations in New York are recommended.

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Appendix 1. Habitat Description of New York Study Sites, 2006.

			Elevation	
Site	Township	County	(ft)	   Habitat
		,	( -/	Moderate submergent and emergent vegetation
Arbutus Lake	Newcomb	Essex	1727	with adjacent hardwood forest upland
				Heavy amounts of submergent vegetation and
				moderate amounts of emergent vegetation, with
Big Moose Lake	Webb	Herkimer	1841	adjacent hardwood forest upland
				Moderate submergent and emergent vegetation
Chaumont Pond	Clifton	St. Lawrence	1454	with adjacent hardwood forest upland
				Moderate submergent and emergent vegetation
Dry Channel Pond	Altamont	Franklin	1625	with adjacent hardwood forest upland
				Moderate amount of submergent and emergent
				vegetation with adjacent hardwood forest
East Pine Lake	Tupper Lake	Franklin	1594	upland
				Moderate amount of algae with little emergent
				vegetation and a moderate amount of emergent
				vegetation with adjacent upland of hardwood
Ferris Lake	Arietta	Hamilton	1701	forest
				Moderate amount of submergent vegetation and
				extensive amounts of emergent vegetation with
Garnet Lake	Thurman	Warren	1494	adjacent hardwood forest upland
				Moderate amount of submergent vegetation
Hitchins Pond	Colton	St. Lawrence	1746	with adjacent hardwood forest upland
				Small amount of submergent and emergent
				vegetation at some areas of the lake with
Henderson Lake	Newcomb	Essex	1892	surrounding mixed hardwood white cedar forest
				Small amount of emergent vegetation and algae
Limekiln lake	Ohio	Herkimer	1898	with adjacent hardwood forest upland
				Small amount of submergent and emergent
				vegetation with adjacent upland of evergreen
Massawepie Lake	Piercefield	St. Lawrence	1554	forest
				Moderate submergent and emergent vegetation
South Lake	Ohio	Herkimer	2088	with adjacent hardwood forest upland
				Extensive submergent and emergent vegetation
Taylor Pond	Black Brook	Clinton	1425	with adjacent hardwood forest upland

Appendix 2. Common and Scientific Species Names

Common Name	Species				
Frogs					
Green frog	Rana clamitans				
North American Bullfrog	Rana catesbeiana				
River frog	Rana heckscheri				
Southern leopard frog	Rana sphenocephala				
Turtles					
Common musk turtle	Sternotherus odoratus				
Diamondback terrapin	Malaclemys terrapin				
Eastern red-bellied turtle	Pseudemys rubriventris				
Map turtle	Graptemys geographica				
Painted turtle	Chrysemys picta				
Snapping turtle	Chelydra serpentina				
Salamanders and Newts					
Allegheny mountain dusky salamander	Desmognathus ochrophaeus Notophthalmus				
Eastern newt	viridescens				
Eastern red-backed salamander	Plethodon cinereus				
Northern two-lined salamander	Eurycea bislineta				

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# AN EXPLORATORY STUDY OF METHYLMERCURY AVAILABILITY IN TERRESTRIAL WILDLIFE OF NEW YORK AND PENNSYLVANIA, 2005-2006

FINAL REPORT 10-03

STATE OF NEW YORK DAVID A. PATERSON, GOVERNOR

NEW YORK STATE ENERGY RESEARCH AND DEVELOPMENT AUTHORITY VINCENT A. DEIORIO, ESQ., CHAIRMAN FRANCIS J. MURRAY, JR., PRESIDENT AND CHIEF EXECUTIVE OFFICER

