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Mercury Bioaccumulation in Northern Two-lined Salamanders from Streams in the Northeastern United States

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Abstract. Mercury (Hg) bioaccumulation in salamanders has received little attention despite widespread Hg contamination of aquatic ecosystems and worldwide amphibian declines. Here we report concentrations of methyl Hg (MeHg) and total Hg in larval northern two-lined salamanders (*Eurycea bislineata bislineata*) collected from streams in Acadia National Park (ANP), Maine, and Bear Brook Watershed, Maine (BBWM; a paired, gauged watershed treated with bimonthly applications (25 kg/ha/yr) of ammonium sulfate [$(NH_4)_2SO_4$]) since 1989), and Shenandoah National Park (SNP), Virginia. MeHg comprised 73–97% of total Hg in the larval salamander composite samples from ANP. At BBWM we detected significantly higher total Hg levels in larvae from the $(NH_4)_2SO_4$ treatment watershed. At ANP total Hg concentrations in salamander larvae were significantly higher from streams in unburned watersheds in contrast with larval samples collected from streams located in watersheds burned by the 1947 Bar Harbor fire. Additionally, total Hg levels were significantly higher in salamander larvae collected at ANP in contrast with SNP. Our results suggest that watershed-scale attributes including fire history, whole-catchment $(NH_4)_2SO_4$ additions, wetland extent, and forest cover type influence mercury bioaccumulation in salamanders inhabiting lotic environments. We also discuss the use of this species as an indicator of Hg bioaccumulation in stream ecosystems.

Keywords: amphibian; *Eurycea bislineata bislineata*; mercury; salamander; stream; watershed

Introduction

Mercury (Hg) enters aquatic ecosystems via atmospheric deposition (Mierle, 1990; Wiener et al., 1990) and is biologically and chemically converted to methylmercury (MeHg), a biologically active and toxic form. Acidified freshwater ecosystems with high temperatures and dissolved organic carbon levels facilitate MeHg bioaccumulation and biomagnification (Wiener et al., 2003 and references therein). MeHg can be toxic to fish and other aquatic biota, impairing productivity, growth and development, eliciting

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aberrant behavior, and potentially causing death (Wiener and Spry, 1996). Fish and their long-lived predators commonly have elevated Hg concentrations even in undeveloped areas with no local pollution sources (Lucotte et al., 1999). Toxic chemicals and metals may interact synergistically with other anthropogenic stressors such as habitat degradation, UV-B radiation, and global climate change to affect amphibian populations (Stebbins and Cohen, 1995; Sparling et al., 2000). Hg contamination of amphibians has received little attention despite worldwide population declines (Houlahan et al., 2000).

Complexity in watershed, trophic, and within stream mercury sorption likely influences the bioavailability, bioaccumulation, and biomagnification of mercury in lotic ecosystem biota. Watershed and trophic complexities encompass interactions from both within streams and between watercourses and adjacent terrestrial landscapes, including riparian zones (Naiman and Décamps, 1997). Despite many investigations of mercury contamination levels in fish (Wiener et al., 2003 and references therein), no previous study has analyzed the effects of fire history, chronic acidification or regional heterogeneity on Hg bioaccumulation for a stream-dwelling amphibian species.

Northern two-lined salamanders (*Eurycea bislineata bislineata*) are relatively common in northeastern North America and inhabit streams from Virginia to Ohio to southern Ontario, Canada. These salamanders exist as aquatic larvae for 1–3 years although 3 year old larvae are considered rare. Larvae nocturnally forage for invertebrates and diurnally seek refuge under rocks and debris (Petranka, 1984; 1998). Adults occupy riparian habitats and return to streams to reproduce. The specific objectives of this study were to evaluate the effects of: (1) watershed fire history at ANP, (2) chronic whole catchment $(NH_4)_2SO_4$ additions (a realistic surrogate for acid rain) at BBWM, and (3) regional landscape heterogeneity on northern two-lined salamander Hg bioaccumulation rates. We also compare salamander larvae Hg concentrations with similarly aged brook trout (*Salvalinus fontinalis*) and adult salamanders, and evaluate relationships between larval salamander length and weight measurements with total Hg concentrations.

Methods

Study sites

We report concentrations of total Hg in larval northern two-lined salamanders from 3 principal locations: (1) Acadia National Park (ANP), Maine, (2) Bear Brook Watershed (BBWM), Maine, and (3) Shenandoah National Park (SNP), Virginia. Acadia National Park (44°21′ N, 68°13′ W) encompasses over 15,233 ha, with 12,260 ha on Mount Desert Island and 2973 ha in surrounding parcels. ANP has 26 mountains, and approximately 20% of the park is classified as wetland habitat including marshes, lakes, ponds, streams (elevation range 50–250 m), vernal pools, swamps, and bogs (Calhoun et al., 1994). The park also contains salt marshes, marine aquatic beds, and intertidal shellfish flats. Terrestrial habitats include peatlands, coniferous forest, and upland and riparian deciduous forests. The area is dominated by white spruce (*Picea glauca*), red spruce (*Picea rubens*), and balsam fir (*Abies balsamea*). Dominant deciduous tree species include birch (*Betula* spp.), aspen (*Populus* spp.), maple (*Acer* spp.), and red oak (*Quercus rubra*). In 1947, a human-caused fire swept through Bar Harbor, Maine, severely burning 6880 ha of Northeast Mount Desert Island. We collected larval two-lined salamanders from 14 headwater streams in ANP, including those located in burned (dominated by hardwood forest) and unburned (dominated by coniferous forest) watersheds.

Shenandoah National Park encompasses 79,382 ha in the Blue Ridge Mountains of Virginia and is $>95\%$ forested. Over half of the land is dominated by either chestnut (*Castanea dentata*) or red oak forests situated at high elevations. Maple (*Acer* spp.), birch (*Betula* spp.), ash (*Fraxinus* spp.), and basswood (*Tilia americana*) trees are common at mid-elevations, and tulip poplar (*Liriodendron tulipifera*) forests are found along stream corridors. Unlike ANP, SNP streams (elevation range 290–700 m) contain a variety of other stream salamander species besides northern two-lined salamanders, including the northern red salamander (*Pseudotriton ruber ruber*), northern spring salamander (*Gyrinophilus porphyriticus porphyriticus*), northern dusky salamander (*Desmognathus fuscus*), seal salamander (*Desmognathus monticola*), long-tailed salamander (*Eurycea longicauda longicauda*), and the southern two-lined salamander (*Eurycea cirrigera*) which occurs in the southern region of the park (Witt, 1993).

BBWM is a paired, gauged watershed treated since 1989 with bi-monthly helicopter applications (25 kg/ha/yr) of ammonium sulfate [$(NH_4)_2SO_4$] to study watershed responses to chronic acidification. BBWM has two first order streams (East Bear Brook EBB and West Bear Brook WBB) with approximately 300 m of stream channel above the gauging weirs. Ammonium sulfate was applied to WBB, whereas EBB watershed served as a reference site and receives no treatment. Both streams have similar watershed areas and different chemical characteristics resulting from the chronic acidification treatment (Table 1). The EBB reference stream unit models a stream recovering from acidification. Forest vegetation at BBWM is comprised of softwood (25%), hardwood (35%), and mixed (40%) forest stands. Dominant hardwood species include American beech (*Fagus grandifolia*), maple, and birch. The upper regions of each watershed are comprised of nearly pure softwood stands of red spruce, balsam fir, and hemlock (*Tsuga canadensis*).

Field methods

Two-lined salamander larvae (1–3 years old, $n = 3$–17 per stream) were collected opportunistically using a small aquarium dip net from 14 headwater streams (1st and 2nd order) park-wide in ANP in November 2000 and May–June 2001 and 2002. Salamanders were also collected from 12 randomly selected streams ($n = 5$ per stream) distributed park-wide in SNP during May 2002 and 2 streams in BBWM (EBB $n = 7$, WBB $n = 6$) during June 2002 using the same methods. Captured salamander larvae were rinsed with stream water, placed in a sterile sample bag (Whirl-Pak, M-Tech Diagnostics Ltd., Cheshire, England), measured (total length, snout-vent-length, tail length), and immediately decapitated. ANP salamanders were collected from 4 streams within the 1947 Bar Harbor fire perimeter and 10 streams in unburned regions in the park. We also simultaneously collected salamander adult two-lined salamanders, and juvenile 1–2 year old brook trout from Hadlock Brook in ANP to evaluate differences in Hg bioavailability among species and salamander age classes. Haines (unpubl. data) captured brook trout via electro-shocking. All samples were stored in a freezer prior to mercury analysis. During field collections of salamanders, we evaluated whether individuals were lethargic. Lethargic behavior was recorded when we observed salamanders rising to the surface, turning on their side, and not swimming away after we lifted the cover object to capture the individual.

Table 1. Physical and chemical data for Bear Brook Watershed, Beddington, Maine, November 2000–October 2001

Parameter	West Bear (WBB) $(NH_4)_2SO_4$ treatment stream	East Bear (EBB) reference stream
Drainage area (ha)	10.2	10.7
Discharge (cubic feet per second)	0.042 (0–1.84)	0.045 (0–2.50)
Annual temperature (°C)	4.2 (0–10)	4.9 (0–10)
Specific conductance (*u*s/cm)	43 (31–52)	23 (20–28)
pH	4.90 (4.54–6.25)	5.58 (4.90–6.66)
Acid neutralizing capacity (*u*eq/l)	−5.0 (−12.0–17.9)	3.90 (−8.0–31.7)
SO_4 (mg/l)	9.02 (5.42–10.08)	4.18 (3.79–4.80)
NO_3 (mg/l)	2.48 (0.01–4.15)	0.06 (0.01–0.23)
NH_4 (mg/l)	0.02 (0.05–0.13)	0.01 (0.03–0.05)
Total Al (mg/l)	0.61 (0.082–1.09)	0.17 (0.065–0.256)
Ca (mg/l)	1.98 (1.62–2.40)	1.14 (0.66–1.64)
Mg (mg/l)	0.45 (0.40–0.57)	0.27 (0.18–0.45)
Dissolved organic carbon (mg/l)	2.3 (1.1–7.5)	2.7 (1.3–4.5)

Values represent means with ranges (min–max) in parentheses. Discharge data are from the USGS Water Resources of Maine (*http://water.usgs.gov/me/nwis/rt*). All other data are from the Senator George J. Mitchell Center for Environmental and Watershed Research at the University of Maine, Orono, ME (Kahl et al. unpublished data).

Mercury analyses

All biotic samples were processed for total Hg using acid digestion and atomic absorption spectrometry, based on EPA Method 245.6 (USEPA 1991). Wet weights of samples were recorded in the laboratory and composite samples consisting of 8 larvae (randomly selected) from each of the 4 ANP streams were used for total Hg:MeHg ratio analyses. For MeHg analyses, composites were freeze-dried, freeze-fractured, and homogenized and then analyzed for both total Hg and MeHg content with a modified version of Draft EPA Method 1630, combined with alkaline digestion (USEPA 2001). Accuracy of analytical techniques was evaluated by analyzing certified reference materials (International Atomic Energy Agency-356 and TORT-2 {National Research Council Canada} for MeHg; DOLT-2 {National Research Council Canada} for total Hg) with each sample batch and by determining the Hg recovery from spiked homogenates. Precision of Hg analytical techniques was evaluated using one duplicate sample for each composite analysis run. For each sample we used reagent blanks to document any laboratory contamination. All mercury analyses were conducted at the Environmental Chemistry Laboratory at the University of Maine. We also evaluated ANP and SNP Hg deposition data from the Mercury Deposition Network (MDN) database. Atmospheric Hg deposition (wet only) is monitored weekly at both ANP and SNP (Fig. 1).

Statistical analyses

Data normality was evaluated using Lillefor's test (Zar, 1999), and three outliers were removed prior to analyses. We removed outliers due to suspected lab contamination ($n = 2$) or if the data point was > 4 SE ($n = 1$) from the mean. Log-transformed mercury concentration data were used in mixed-model nested analysis of variance (ANOVA) tests to assess within and among stream variation of larval salamander total Hg concentrations. We also compared salamander body measurements (wet weights, total lengths, snout-vent lengths, tail lengths) among burned and unburned streams in ANP and among parks using mixed-model nested ANOVA tests. We used *t*-tests on log-transformed data to compare total Hg concentrations, wet weights, total lengths, snout-vent lengths, and tail lengths of salamander larvae among the reference and treatment sites at BBWM. We used a one-way ANOVA (unbalanced design) and Tukey's test for post-hoc pair-wise comparisons to test for differences among Hg concentration levels in salamander larvae, salamander adults, and brook trout collected from Hadlock Brook, ANP. We used a Pearson correlation matrix to evaluate relationships between total Hg and salamander

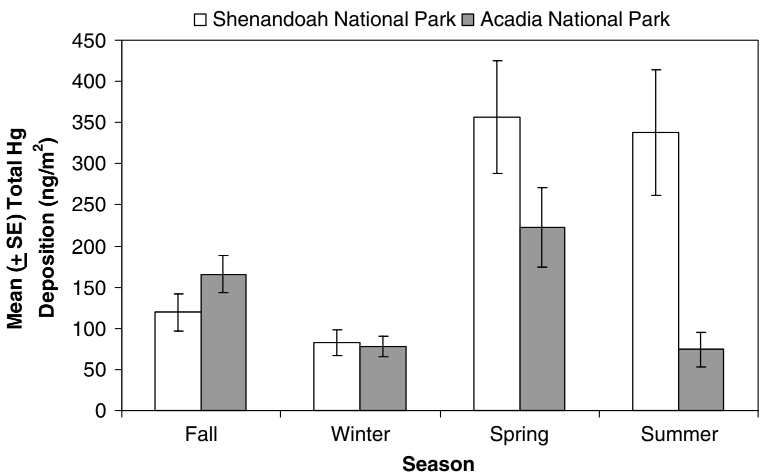

Figure 1. Seasonal variability of total Hg deposition (ng/m^2) at Acadia National Park, Maine, and Shenandoah National Park, Virginia, November 2002–December 2003. Data are from the National Atmospheric Deposition Program / Mercury Deposition Network. (NADP/MDN, 2004).

measurements. Pearson product-moment correlation analyses were conducted on data pooled among streams and analyzed separately for ANP and SNP, respectively. Hg deposition (ng/m^2) data were analyzed using two-factor ANOVA with deposition as the dependent variable and season and park as factors. All statistical analyses were conducted using SYSTAT 10.2 (SYSTAT, 2002). Statistical significance was accepted at $p \leq 0.05$.

Results

Methyl Hg comprised 73–97% of total Hg in the larval composites ($n_{salamanders} = 8$ per stream, $n_{streams} = 4$) from ANP. Total Hg concentrations in northern two-lined salamander larvae from ANP exceeded ($ANOVA_{Nested}$ $F = 34.86$; $df = 1, 27$; $p < 0.001$) those from SNP (Table 2), despite significantly higher total Hg deposition (ng/m^2) during November 2002-December 2003 at SNP ($F = 13.29$; $df = 1$; $p = 0.0004$, Fig. 1). Total Hg deposition was significantly higher during spring compared to other seasons ($F = 13.69$; $df = 3$; $p < 0.00001$, Fig. 1), and the season by park interaction was also significant ($F = 7.75$; $df = 3$; $p < 0.000$, Fig. 1).

Salamander larvae from unburned ANP watersheds had greater total Hg than those collected from burned watersheds ($ANOVA_{Nested} F = 5.63$; $df = 1,12$; $p = 0.035$; Table 2). Total Hg concentrations in salamander larvae

Table 2. Mean (±SE) total Hg concentrations (based on wet weights), wet weights, total length, snout-vent length, and tail length of larval two-lined salamander larvae from ANP ($n_{streams} = 14$; 4 burned, 10 unburned), SNP ($n_{streams} = 12$), and BBWM ($n_{streams} = 1$ reference, $n_{streams} = 1$ treatment)

Site Location	Total Hg (ng/g)[a]	Wet weight (g)	Total length (cm)	Snout-vent length (cm)[b]	Tail length (cm)	(*n*)
Acadia National Park	66.1 ± 3.4	0.30 ± 0.01	4.7 ± 0.07	2.6 ± 0.04	2.1 ± 0.04	116
Burned Watersheds	47.7 ± 1.9	0.33 ± 0.02	4.8 ± 0.12	2.7 ± 0.06	2.1 ± 0.06	49
Unburned Watersheds	79.5 ± 5.2	0.28 ± 0.02	4.7 ± 0.10	2.6 ± 0.04	2.1 ± 0.06	67
Shenandoah National Park	26.8 ± 1.6	0.29 ± 0.02	4.4 ± 0.12	2.4 ± 0.06	2.1 ± 0.07	60
BBWM-WBB $(NH_4)_2SO_4$ Treatment	64.5 ± 3.1	0.11 ± 0.02	3.2 ± 0.16	2.0 ± 0.08	1.3 ± 0.07	6
BBWM-EBB Reference	21.4 ± 3.8	0.18 ± 0.05	4.0 ± 0.50	2.1 ± 0.22	1.8 ± 0.23	7

Samples were collected during May–June 2001 and 2002 for ANP, and during May–June 2002 for SNP and BBWM. Sample size (*n*) in the table refers to the number of salamanders.
[a] $p < 0.05$ for ANP vs. SNP, ANP Burned vs. ANP Unburned, and BBWM Reference vs. BBWM Treatment.
[b] $p < 0.01$ for ANP vs. SNP only.

Table 3. Pearson product–moment correlation coefficients for the variables: salamander wet weight (WETWT), total length (TL), snout-vent-length (SVL), tail, and logarithm of Total Hg concentrations (LOGTOTALHG).

	WETWT	TL	SVL	TAIL	LOGTOTALHG
(A) Acadia ($n = 116$)					
WETWT	1.000				
TL	0.864*	1.000			
SVL	0.811*	0.918*	1.000		
TAIL	0.750*	0.944*	0.736*	1.000	
LOGTOTALHG	0.038	0.121	0.042	0.172	1.000
(B) Shenandoah ($n = 60$)					
WETWT	1.000				
TL	0.921*	1.000			
SVL	0.892*	0.894*	1.000		
TAIL	0.800*	0.930*	0.666*	1.000	
LOGTOTALHG	−0.564*	−0.547*	−0.501*	−0.499*	1.000

Data collected during May–June 2001, 2002 in Acadia National Park, Maine, USA (A) and in May 2002 in Shenandoah National Park, Virginia, USA (B). Asterik indicates $p < 0.05$.

collected from the BBWM $(NH_4)_2SO_4$ treatment stream exceeded those sampled from the reference stream ($t = 6.15$; $df = 11$; $p < 0.001$; Table 2). Other than snout-vent length comparison between ANP and SNP salamanders ($\text{ANOVA}_{\text{Nested}} F = 10.98$; $df = 1{,}24$; $p = 0.003$; Table 2), larval salamander measurements did not differ significantly between parks nor between fire history categories (burned vs. unburned) at ANP (Table 2). At BBWM three of the six salamander larvae collected from the stream in the treated watershed were noticeably lethargic.

No significant correlations were found between larval salamander total Hg concentrations (log-transformed data) and length and weight data from ANP samples (Table 3). However, there were significant negative relationships between salamander larvae Hg levels and all measurement variables for SNP samples (Table 3).

Larvae and adult two-lined salamanders had significantly higher total Hg concentrations ($F = 7.31$; $df = 2, 29$; $p = 0.003$, Fig. 2) than juvenile (1–2 years of age) brook trout collected from the same stream in ANP. Although average adult total Hg concentrations were higher than those measured in larval salamanders, this difference was not significant ($p > 0.05$, Fig. 2).

Discussion

Our results indicate that total Hg at the ng/g level bioaccumulates within 1–3 years in larval northern two-lined salamanders and that these concentrations vary regionally. The majority of Hg in salamanders at ANP was in the MeHg (73–97%) form. The long larval period and invertebrate diet of this species likely contributes to its susceptibility to mercury bioaccumulation. Two-lined salamander larvae live and forage, often at high densities ($> 1/m^2$), at the water column-streambed interface near stream edges where total sediment Hg and MeHg levels are highest (Morel et al., 1998; Petranka 1998). Unlike fish, recently metamorphosed and adult two-lined salamanders disperse during autumn into riparian habitats where they may be consumed by terrestrial vertebrates (Petranka, 1998), transporting Hg between aquatic and terrestrial environments.

Patterns of mercury bioaccumulation are spatially complex. Hg concentrations in salamander larvae may be affected by a variety of factors including geographic location and watershed conditions such as fire history and post-fire succession patterns, acidification, wetland extent, and forest type (Grigal, 2002 and references therein;

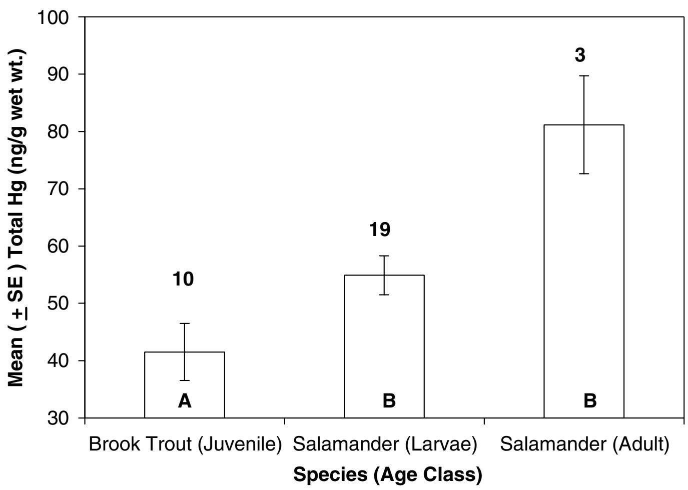

Figure 2. Comparison of mean (+SE) total mercury concentrations (ng/g wet wt.) in brook trout (1–2 years of age), two-lined salamander adults (≥ 3 years of age) and larvae (1–3 years of age) from Hadlock Brook in Acadia National Park, Maine, 2000–2002. Number above graph bars represents sample size (n). Data analysis was performed on log-transformed data. Raw data are presented in the graph, and graph bars with non-identical letters were significantly different ($p > 0.05$) in pairwise comparisons.

Wiener et al., 2003 and references therein). Data from the BBWM ammonium sulfate treatment site indicate that Hg bioaccumulation was higher (Table 2) in salamander larvae collected from this stream, presumably as a result of increased MeHg production by enhanced anaerobic, sediment based, sulfate reducing bacteria activity and efficient transfer through the food web (Compeau and Bartha, 1985; Gilmour and Henry, 1991; Gilmour et al., 1992; Devereux et al., 1996; Morel et al., 1998).

Conditions in the reference stream at BBWM may possibly reflect conditions of a watershed recovering from the effects of acid rain. Our findings that larval salamander mercury concentration levels were significantly lower at EBB (Table 2) may have important implications for air and water quality policy. The sulfate additions at BBWM created environmental conditions that were likely favorable for MeHg biomagnification and bioaccumulation (Gilmour and Henry, 1991; Gilmour et al., 1992; Morel et al., 1998; Harmon et al., 2003). Harmon et al. (2003) reported that sulfate additions in wetland mesocosms significantly increased Hg bioaccumulation in periphyton, eastern mosquitofish (*Gambusia holbrooki*), and lake chubsuckers (*Erimyzon sucetta*). Therefore, reductions in acid pollutants such as nitrogen oxides and sulfur dioxides may potentially mitigate mercury contamination in lotic biota. However, it is important to point out that salamander Hg levels from unburned ANP watersheds exceeded Hg concentrations from the treatment watershed at BBWM (Table 2). The higher Hg levels recorded in salamanders from the unburned ANP watersheds were likely a result of localized differences in wetland extent, methylation capacity, soil retention capabilities, trophic complexity, labile organic matter abundance, sediment dynamics, watershed processes, forest types, or other biotic and abiotic factors including marine inputs of Hg and sulfate.

ANP salamander larvae had greater total Hg concentrations than SNP larvae despite the fact that SNP received higher total Hg deposition (measured as wet-only). These findings suggest that recovery from mercury pollution may likely have a high degree of spatial and temporal variability. Aquatic ecosystems with different watershed conditions, and varying sources of Hg deposition, levels of wetland extent and food web complexity, Hg methylation capacity and overall ecosystem sensitivity to Hg contamination (Brigham et al., 2003), will likely respond differently to potential reductions in atmospheric pollutants that influence Hg bioavailability. At ANP, accumulation rates of Hg to the sediment at two locations were 100–200 $\mu g/m^2$-yr in the 1980s, suggesting that a large amount of dry Hg input is not measured by the wet-only MDN collector. Therefore, we recommend using caution in interpreting these data since the MDN collectors only measure wet deposition and dry deposition is not quantified. Future research should address the linkage between watershed condition and recovery from mercury pollution across a gradient of land-use practices and for different surface water types in the northeastern United States including lakes, bogs, vernal pools, ponds, and streams.

During this investigation, lethargic salamander larvae were observed only at WBB (50%, $n = 6$). Although obvious lethargy was not noted for salamanders from any other stream location during the investigation, effects from contaminants, including mercury, on biota may be extremely subtle and difficult to detect (Webber and Haines, 2003). Other researchers have observed negative effects of both nitrate (Rouse et al., 1999) and aluminum (Clark and Hall, 1985; Clark and LaZerte, 1985) on palustrine amphibian species at levels comparable to or lower than the nitrate and aluminum water concentrations (Table 1) recorded at the BBWM ammonium sulfate treatment stream (WBB), however it is unknown which chemical attributes or interactions were primarily responsible for causing lethargy in these individuals.

Differences in total Hg concentrations in larvae collected from streams in burned and unburned watersheds suggest that watershed fire history also affects total Hg bioaccumulation at ANP. Our results are consistent with those of a three year, paired-gauged watershed investigation in ANP, reporting greater total Hg flux, total Hg export, and higher total Hg levels in throughfall precipitation in the unburned, conifer-dominated basin (Kahl et al., 2002; Johnson et al., In Press, Bank et al., In Press). Coniferous forest canopies may be more efficient at intercepting atmospheric Hg (Grigal, 2002 and references therein), and coniferous watersheds also likely have a more labile

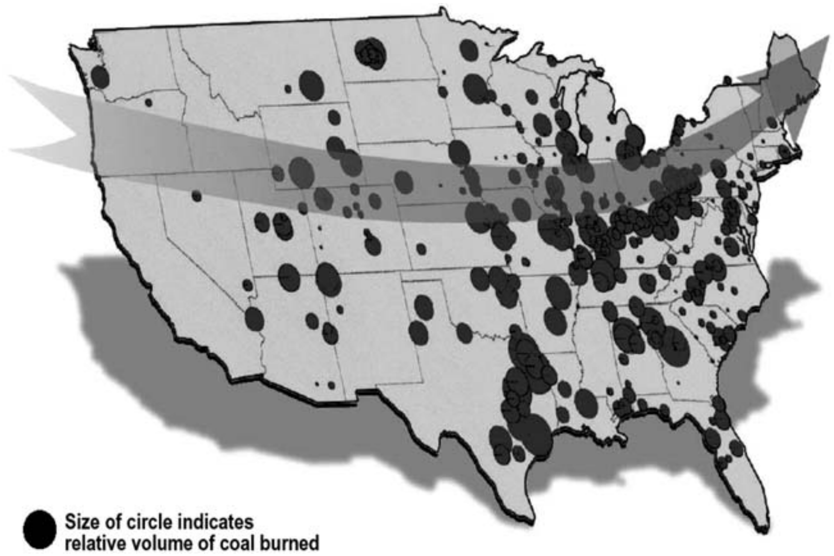

Figure 3. Spatial distribution of coal-fired plants and volume of coal burned (black dots) and predominant wind patterns in the conterminous United States. Map courtesy of Portland Press Herald, Portland, Maine, USA. Source: United States Energy Information Administration, E-3 Ventures (USDOE, 1999).

organic matter base that likely enhances mobility of Hg (Amirbahman et al., 2004). Additionally, Hg volatilization from burned soils may also explain the lower Hg export in streams draining regenerating, deciduous forests (Kahl et al., 2002; Amirbahman et al., 2004).

The high bioaccumulation of total Hg in salamander larvae collected from ANP also may be influenced by the park's abundance of wetlands (approximately 20% of ANP total area; Calhoun et al., 1994), soil properties (i.e., ANP glaciated vs. SNP non-glaciated) and distribution of coniferous forests. In contrast, SNP contains $<1\%$ wetland and little ($<5\%$) coniferous forest (D. Hurlbert, personal communication). Additionally, ANP is a coastal park situated in the extreme Northeast United States and unlike SNP receives marine inputs of Hg and sulfate as well as other pollutants. Total atmospheric Hg deposition at SNP was higher in comparison to ANP (Fig. 1), possibly reflecting the interaction between the distribution and abundance of coal consumption, and the predominant wind patterns and long range transport capabilities of Hg pollution likely originating from the Midwest (Fig. 3). These data do not reflect variation in total Hg dry deposition and therefore should be interpreted with caution.

No consistent trends in salamander total Hg concentrations with size were observed for ANP salamander larvae, although significant negative relationships were detected at SNP (Table 2). This difference among parks may reflect regional or geographical differences in diet (although Petranka 1998 notes that Chirononmidae larvae are an important source of prey throughout the range where food habits of this species have been studied) or local abiotic factors. Other possible explanations include regional differences in toxicokinetics, growth rates and depuration efficiency. However, detoxification mechanisms for salamanders, and for amphibians in general, are largely unknown (Sparling et al., 2000), and therefore future research is required to evaluate these hypotheses.

Salamander larvae Hg concentrations were significantly greater than brook trout samples collected from the same stream. These differences may reflect differences in the diets, growth rates, toxicokinetics, and general physiology of these two species. Brook trout often incorporate terrestrial prey items into their diet (Wipfli, 1997), whereas salamander larvae focus on benthic sources of prey. The higher Hg levels in adult salamanders compared to larvae Hg concentrations may reflect higher bioaccumulation as a function of age. However, at some streams salamander adults and larvae had similar total Hg concentrations potentially as a result of the semi-aquatic habitat use patterns of adult salamanders (Petranka, 1998) and their incorporation of terrestrial prey into their diet. These terrestrial prey items likely had substantially lower total Hg concentrations than the lotic prey encountered by the strictly aquatic larvae. Future research should make comparisons of fish and salamander mercury concentrations using a higher number of sampling streams to further evaluate this hypothesis.

Assessing Hg pollution and its effects on Northeastern North American aquatic ecosystems should be a conservation priority, especially considering that fish consumption advisories are widespread throughout the United States due to elevated Hg concentrations (NAS, 2000). Our research indicates that biota at lower trophic levels also have elevated Hg concentrations, potentially signaling a cascading, system-wide effect. Larval northern two-lined salamanders may be useful monitors of regional Hg contamination in stream ecosystems from the Northeast United States. Salamander larvae are likely more representative of local stream conditions than some fish species, which often feed on terrestrial drift (Wipfli, 1997), are more mobile, and are commonly stocked from unknown origins. The abundance, broad distribution, low mobility, and ease and cost effectiveness of field collection make two-lined salamanders ideal for characterizing Hg bioavailability in stream ecosystems. Use of this species in toxicology studies is possible but may be somewhat limited due to their long larval period (1–3 years). We recommend that toxicology studies use realistic MeHg exposure regimes, and address the long-term and sub-lethal effects of this contaminant in relation to the interactive or additive effects from the presence of predators and/or predator cues (Relyea and Mills, 2001).

Monitoring population trends as well as the extent of Hg contamination may reflect amphibian responses to co-occurring, multiple anthropogenic stressors (Sparling et al., 2000). However, monitoring will only document the degree of contamination. Reduced environmental Hg accumulation is dependent on sound environmental policies that improve air and water quality by reducing Hg emissions and addressing pollution issues at local, regional and national scales.

Future research should address the role of diet, trophic complexity, and energy sources on Hg bioavailability in stream ecosystems as well as the influence of drying and re-wetting of stream margin habitats on sulfate reducing bacteria production and Hg methylation dynamics. In coastal areas, effects of marine derived Hg inputs should also be evaluated. Additionally, evaluating interactions between other stressors and low level, chronic exposure of mercury on stream salamanders and other amphibians may aid in the risk assessment of local populations. A more synthesized perspective on patterns and processes related to aquatic and terrestrial biogeochemistry linkages (Grimm et al., 2003) related to the fate, transport, and bioavailability of mercury in aquatic ecosystems will result from long term, broad-scale studies that address variation in mercury cycling across a broad gradient of physical, climatic, and biotic conditions (Grigal, 2002).

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