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Higher and more variable methylmercury biomagnification factors for floodplain than the contiguous river (South River, Virginia USA)

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ABSTRACT

Extending previous trophic transfer studies of the mercury-contaminated South River watershed, predictive models were built for mercury biomagnification in floodplain food webs at two more locations (North Park and Grand Cavern). Four of five models built to date based on methylmercury and $\delta^{15}N$ met the *a priori* requirement for useful prediction (prediction $r^2 \approx 0.80$). An additional factor included in models was organism thermoregulatory strategy (poikilothermy or homeothermy). The methylmercury food web biomagnification factors (FWMFs, fold increase per trophic level) for the North Park and Grand Cavern locations were 17.4 (95% CI of 9.5–31.6) and 6.2 (95% CI of 3.5–11.0) respectively. FWMF calculated in 2009 were 9.3 (95% CI of 5.4–16.2) for the Augusta Forestry Center and 25.1 (95% CI of 12.6–50.1) for Grottoes Town Park. The overall South River floodplain FWMF generated by meta-analysis of the four locations was 12.4 (95% CI of 6.8–22.3). These results supported previous findings that the South River floodplain food webs had higher biomagnification factors than the contiguous aquatic food web (4.6, 95% CI of 3.6–5.7). Floodplain FWMFs were also more variable than those of the river.

1. Introduction

Mercury, specifically methylmercury, can be elevated in some species due to biomagnification. Consequently, an ecosystem with modestly elevated mercury concentrations in soils or sediments might still have high body burdens of mercury in apex predators (dos Santos et al., 2006; Macedo-Sousa et al., 2009). This being the case, effective natural resource management and decision making requires tools for predicting mercury concentrations in apex predators via biomagnification (Tom et al., 2010).

Mercury biomagnification is influenced by community structure (Chasar et al., 2009), food source (Gorski et al., 2003; Chételat et al., 2011), food chain length (Cabana et al., 1994), trophic position (Newman et al., 2011) and other factors; however, trophic position is the most widely studied of these factors. Trophic position is commonly characterized with stable nitrogen isotope quotients ($\delta^{15}N$). Mercury biomagnification models have been produced for diverse aquatic food webs based on $\delta^{15}N$ (Campbell et al., 2008; Chasar et al., 2009; Tom et al., 2010). Far fewer have been produced for terrestrial food webs (Gaines et al., 2002; Choy et al., 2010; Newman et al., 2011) despite suggestions from recent studies that members of terrestrial food webs might experience similar or even higher mercury exposure (e.g., Cristol et al., 2008).

This study extended previous trophic transfer studies of a mercury-contaminated reach of the South River (Virginia USA). In a 2007 sampling of aquatic organisms at six locations along a river reach extending downriver 23 miles from the historic site of release, Tom et al. (2010) found that a $\delta^{15}N$ based trophic transfer model could predict methylmercury concentrations in members of aquatic food webs. The methylmercury food web biomagnification factor (FWMF) calculated from that model was 4.6 fold increase per trophic level (TL) (95% CI of 3.6–5.8) assuming that $\delta^{15}N$ increased 3.4‰ per TL (Newman et al., 2011; Chasar et al., 2009). Because several studies (Brasso and Cristol, 2008; Cristol et al., 2008) suggested that wildlife on the South River floodplain might be experiencing harmful mercury exposure, mercury biomagnification in two terrestrial locations on the South River floodplain, Augusta Forestry Center (AFC, Crimora, VA, 11.8 river miles (RM) below historic point of input) and Grottoes Town Park (GTP, Grottoes, VA, RM=22.4), was studied in 2009 (Newman et al., 2011). The 2009 floodplain study built models for each site, reinforcing the findings of the previous aquatic study that a $\delta^{15}N$–based model had better predictive capability for methylmercury concentration than for total mercury, and that the FWMFs from these floodplain locations (9.3, 95% CI of 5.4–16.2 and 25.1, 95% CI of 12.6–50.1 for AFC and GTP respectively) were

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higher than that of the contiguous aquatic food webs. Models for more floodplain locations were deemed necessary due to the material difference between floodplain and aquatic food webs, and the large difference between the two modeled floodplain sites. In May 2010, two more floodplain locations were studied (1) to assess whether the floodplain food webs had consistently higher FWMF than the contiguous aquatic food webs; and (2) to explore factors that might produce the differences observed among floodplain locations.

2. Materials and methods

2.1. Sampling

Two locations (AFC and GTP) were sampled during the summer of 2009 and another two were sampled in the same 23 mile river reach (North Park (NP, RM=2.0, about 10 acres) and Grand Cavern (GC, RM=20.0, about 30 acres)) during the first two weeks of May 2010. General sampling locations related to recent terrestrial studies can be found in Brasso and Cristol (2008). These two locations were added to collect samples between the historic point source and AFC, and between AFC and GTP, so that the four locations were relatively evenly distributed along the 23 mile river reach. Also, the locations selected were based on accessibility and coordination with another South River bird study. In each location, three sites were randomly selected within 50 m of the river bank. Terrestrial invertebrates were collected using either pitfall traps or sweep net. Samples from each site were pooled together for each species to form one replicate with at least two individuals (for invertebrates) in each pooled sample based on their sizes and availability. Triplicate samples were collected whenever possible. Mice and voles were captured by baited snap trap. Unfortunately, only a few small mammals were caught during the sampling period, so only three deer mice in NP, two pine voles in GC and one deer mouse in GC were available for analysis. Birds were captured using mist nets in each site. Again, the number of replicates depended largely on the availability of each species. Triplicate samples of emergent aquatic insects and crayfish were collected along the river bank. More details about sampling procedures can be found in Tom et al. (2010) and Newman et al. (2011). Species sampled in these two locations were shown in Table 1.

Table 1
Organisms from the two floodplain locations in South River watershed (VA, USA).

Locations	Common name	Latin name	Sample type	Symbol
Abiotic				
NP, GC	Soil			A
NP, GC	Leaf litter			B
Aquatic emergent insect				
NP, GC	Mayfly	*Ephemeroptera*	Adult—whole body	C
NP, GC	Midge	*Diptera*	Adult—whole body	D
NP, GC	Caddisfly	*Trichoptera*	Adult—whole body	E
Aquatic invertebrate				
NP, GC	Crayfish	*Astacoidea*	Whole body	F
Plant				
NP, GC	Grass	*Festuca elatior*	Green tissue	G
NP, GC	Honey suckle	*Lonicera japonica*	Green tissue	H
NP, GC	Violet	*Viola striata*	Green tissue	I
Detritivore				
NP, GC	Earthworm	*Lumbricus rubellus*	Whole body	J
NP, GC	Isopod	*Microcerberidae*	Whole body	K
NP	Slug	*Prophysaon dubium*	Whole body	L
Insect				
NP, GC	Ladybug	*Harmonia axyridis*	Adult—whole body	M
GC	Ground beetle	*Harpalus pensylvanicus*	Adult—whole body	N
GC	Caterpillar	*Lepidoptera*	Whole body	O
NP, GC	Eastern tent caterpillar	*Malacosoma americanum*	Whole body	P
NP	Asiatic garden beetle	*Maladera castanea*	Adult—whole body	Q
NP, GC	Common black ground beetle	*Pterostichus melanarius*	Adult—whole body	R
GC	Sawflies	*Tenthredinidae*	Larvae—whole body	S
Spider				
NP, GC	Wolf spider	*Lycosidae*	Whole body	T
Small mammal				
GC	Pine vole	*Microtus pinetorum*	Liver, muscle	U1,U2
NP, GC	Deer mouse	*Peromyscus maniculatus*	Liver, muscle	V1, V2
Bird				
NP, GC	Eastern tufted titmouse	*Baeolophus bicolor*	Blood, feather	BA1, BA2
NP, GC	Northern cardinal	*Cardinalis cardinalis*	Blood, feather	BB1, BB2
GC	Eastern wood-pewee	*Contopus virens*	Blood, feather	BC1, BC2
NP	Gray catbird	*Dumetella carolinensis*	Blood, feather	BD1, BD2
GC	Wood thrush	*Hylocichla mustelina*	Blood, feather	BE1, BE2
NP, GC	Eastern song sparrow	*Melospiza melodia*	Blood, feather	BF1, BF2
GC	Great crested flycatcher	*Myiarchus crinitus*	Blood, feather	BG1, BG2
NP, GC	Eastern screech-owl	*Otus asio*	Blood, feather	BH1, BH2
NP, GC	Downy woodpecker	*Picoides pubescens*	Blood, feather	BI1, BI2
GC	Scarlet tanager	*Piranga olivacea*	Blood, feather	BJ1, BJ2
GC	Eastern phoebe	*Sayornis phoebe*	Blood, feather	BK1, BK2
GC	White-breasted nuthatch	*Sitta carolinensis*	Blood, feather	BL1, BL2
GC	American goldfinch	*Spinus tristis*	Blood, feather	BM1, BM2
NP, GC	American robin	*Turdus migratorius*	Blood, feather	BN1, BN2
NP, GC	Carolina wren	*Thryothorus ludovicianus*	Blood, feather	BO1, BO2
GC	Red-eyed vireo	*Vireo olivaceus*	Blood, feather	BP1, BP2
GC	Mourning dove	*Zenaida macroura*	Blood, feather	BQ1, BQ2

2.2. Sample analysis

All results were reported on a dry weight basis unless otherwise indicated. Sample preparation and analytical procedure could be found in Tom et al. (2010). Briefly, freeze dried samples were weighed, homogenized, and a portion of each was sent to the commercial analytical laboratory, CEBAM Inc. (Bothell, WA, USA) for total mercury (THg) and methylmercury (MeHg) analyses. Another portion was sent to the Stable Isotope Facility at the University of California–Davis (Davis, CA, USA) for $\delta^{15}N$ and $\delta^{13}C$ analyses. Carbon isotopic ratios were gathered only to facilitate qualitative grouping of species. Given this use, we did not extract lipids before $\delta^{13}C$ analysis. Although tissue lipids can be ^{13}C depleted (DeNiro and Epstein, 1977), the extraction technique can elevate sample ^{15}N (Post et al., 2007). Regardless, some samples were too small to split for separate isotope analyses. Also, our previous study (Newman et al., 2011) showed that $\delta^{13}C$ information was not as important as $\delta^{15}N$ in modeling mercury biomagnification. Finally, there were little empirical data for mathematical correction for terrestrial samples. Consequently, no mathematical correction was made. Although ^{15}N accumulation in arthropods exoskeletons could cause overestimation of bioavailable $\delta^{15}N$ in whole animals, the difficulty of determining the digestibility of exoskeleton and the common use of whole insects in the literature resulted in our use of whole insects here. The resulting data are consistent with our previous study and comparable to those of many other studies using natural abundance of $\delta^{15}N$.

The mercury analytical quality at CEBAM was gauged with laboratory sample splits, laboratory spiked samples and certified reference materials (CRMs) (BCR-580, DORM-2, IAEA350, IAEA142). The mean differences between sample splits were 1.2% (SD=5.8%, n=36) for THg and 0.1% (SD=4.7%, n=36) for MeHg. The mean recoveries for spiked analysis were 100.1% (SD=7.9%, n=28) for THg and 101.6% (SD=5.5%, n=24) for MeHg. The mean recoveries for CRMs analysis were 100. 6% (SD=2.9%, n=9) for THg and 98.0% (SD=3.4%, n=9) for MeHg. Analytical quality analysis for stable isotopes at UC-Davis was assessed using replicate analyses for five standard materials, G-11 Nylon, G-12 Glutamic Acid-Enriched, G-13 Bovine Liver, G-7 Peach leaves, and G-9 Glutamic acid. The mean recoveries of $\delta^{13}C$ for G-11, G-12, G-13, G-7 and G-9 were 100.0% (SD=0.1%, n=52), 100.1% (SD=0.4%, n=11), 100.0% (SD=0.2%, n=5), 100.2% (SD=0.1%, n=4) and 100.1% (SD=0.2%, n=13), respectively. The mean recoveries of $\delta^{15}N$ for G-11, G-12, G-13, G-7 and G-9 were 100.0% (SD=1.0%, n=50), 100.1% (SD=0.4%, n=8), 100.0% (SD=3.0%, n=5), 103.5% (SD=15.7%, n=4) and 93.6% (SD=6.6%, n=11), respectively. All results from the above procedures indicated excellent analytical accuracy and precision that was adequate for the intended modeling.

2.3. Model construction and selection

The approach to model construction was reported previously (Newman et al., 2011). Briefly, a predictive model was constructed,

$$Y_i = a + bX_{1i} + cX_{2i} + dX_{3i} + \cdots + \varepsilon, \quad (1)$$

where Y is the response variable. Estimated model parameters were a (intercept), and b,c,d (estimated regression coefficients for factors, X_{1i}, X_{2i}, $X_{3i}\ldots$). The X_i were the values of different factors associated with the sampled organisms and ε was the unexplained error associated with the response.

Previous research on two floodplain sites revealed that mercury concentration could be predicted using $\delta^{15}N$ and organism thermoregulatory strategy (abbreviated Therm). Given the sampling methods, it is important to remember that Therm could also include a confounding influence of tissue type (Newman et al., 2011). Adding data from two more sites allowed further exploration of factors that might have a material influence on biomagnification and potentially improve model predictive capability. The candidate response variables (Y) included THg concentration, MeHg concentration, and percentage of total mercury that was methylmercury (%MeHg). Predictors (X_i) included $\delta^{15}N$, $\delta^{13}C$, site, and Therm. Site was treated in initial modeling as a categorical variable with four categories (NP, AFC, GC and GTP) and Therm was also treated as a categorical variable with two categories (poikilotherm and homeotherm).

Best models were selected based on PROC GLMSELECT of the SAS® package (SAS Institute Inc., Cary, NC, USA) with forward selection and Akaike's Information Criterion (AIC) as the stopping criterion. A cross validation coefficient ($r^2_{\text{prediction}}$) was used to gauge model predictive capability. An *a priori* criterion for useful prediction was set to an $r^2_{\text{prediction}}$ of approximately 0.80 (Tom et al., 2010; Newman et al., 2011). Akaike's Information Criterion (AIC) was calculated in the SAS program using the following equation,

$$\text{AIC} = n \times \ln(\text{MSE}) + 2k, \quad (2)$$

where n is the number of total non-missing observations, MSE is model mean squared error, and k is the number of explanatory variables. The applied minimum AIC estimation (MAICE) method favors model with the fewest explanatory variables (lowest k) and best goodness-of-fit (lowest MSE).The best model was refit using trophic level (TL) in place of $\delta^{15}N$. This was done by converting $\delta^{15}N$ to TL using the following equation (Newman et al., 2011),

$$\text{TL} = (\delta^{15}N_i - \delta^{15}N_{\text{pp}})/3.4‰ + 1 \quad (3)$$

where $\delta^{15}N_i$ and $\delta^{15}N_{\text{pp}}=\delta^{15}N$ of sample i and primary producer.

2.4. Meta-analysis

Modeling with both continuous and categorical variables should be done carefully because a crucial assumption of such a covariance model is slope constancy (Neter et al., 1990). Our previous study fitted separate models for the two floodplain locations because data failed to meet this assumption. This assumption was checked again for the four floodplain locations. If the assumption of consistency of slopes again was not met, a meta-analysis would be performed based on the separate models to develop an overall estimate of floodplain mercury biomagnification. Results from different studies could share a common FWMF in which case a fixed model would be applied. A random model would be appropriate if locations had similar but distinct FWMF (Cumming, 2012). A Q statistic will be calculated to examine the heterogeneity of these FWMFs using Eq. (4) (Cochran, 1954; Borenstein et al., 2009)

$$Q = \sum_{i=1}^{k} W_i Y_i^2 - \left(\sum_{i=1}^{k} W_i Y_i\right)^2 \Big/ \sum_{i=1}^{k} W_i \quad (4)$$

where W_i is the location weight, Y_i is the location FWMF, k is the number of locations. W_i is the inverse of location variance, $1/V_i$. V_i is calculated from standard error of FWMF in each location by SE_i^2. If locations are homogeneous, Q should follow a central χ^2 distribution with degrees of freedom $k-1$ and expected value of $k-1$(Borenstein et al., 2009). Otherwise, if Q significantly deviates from its degrees of freedom, this indicates that locations are heterogeneous. The composite estimate of FWMFs across locations for a random effect model is calculated with Eq. (5):

$$Y^* = \sum W_i^* Y_i / \sum W_i^* \quad (5)$$

where W_i^* is corrected location weight calculated from corrected location variance V_i^* by $W_i^*=1/V_i^*$. The corrected variance $V_i^*=V_i+T^2$. T^2 quantifies the deviation of Q from its degree of freedom using Eq. (6):

$$T^2 = \frac{Q - df}{\sum W_i - \left(\sum W_i^2 / \sum W_i\right)} \quad (6)$$

3. Results

Stable isotopes were used to estimate relative trophic position of food web members ($\delta^{15}N$) and to suggest food sources of organisms ($\delta^{15}N$ and $\delta^{13}C$). Previous study of the AFC and GTP sites (Newman et al., 2011) indicated that members of South River floodplain food webs generally fell in one of three groups: terrestrial species that were detritivory-based (earthworm, slug and isopod), aquatic species that were also primarily detritivory-based (emergent insects), and terrestrial species that were primarily herbivory-based (all other species). Stable isotope data in this study were consistent with this categorization as shown in Fig. 1. At both sites, $\delta^{15}N$ of herbivory-based species steadily increased with increasing trophic status as gauged from their feeding habitats. Emergent insects from the river (mayfly, caddisfly and midge) clustered below the general terrestrial herbivory-based trend. Crayfish, an aquatic prey item of the Eastern screech owl, was also below the general trend. Terrestrial detritivores (earthworm, slug and isopod) were not obviously different from the general trend but tended to have similar $\delta^{15}N$ (interpreted as occupying similar trophic status) and $\delta^{13}C$ (interpreted as having similar carbon source). Because these aquatic species and terrestrial detritivores had feeding pathways distinct from the herbivory-based species, they were omitted from biomagnification modeling.

Using $\delta^{15}N$ to quantify trophic position, the relationship was explored between mercury concentration of terrestrial herbivory-based species and trophic position. Clear trends were observed between $\log_{10}$ THg concentration or $\log_{10}$ MeHg concentration and trophic status at both locations. Many previous studies suggested that %MeHg would increase with organism trophic

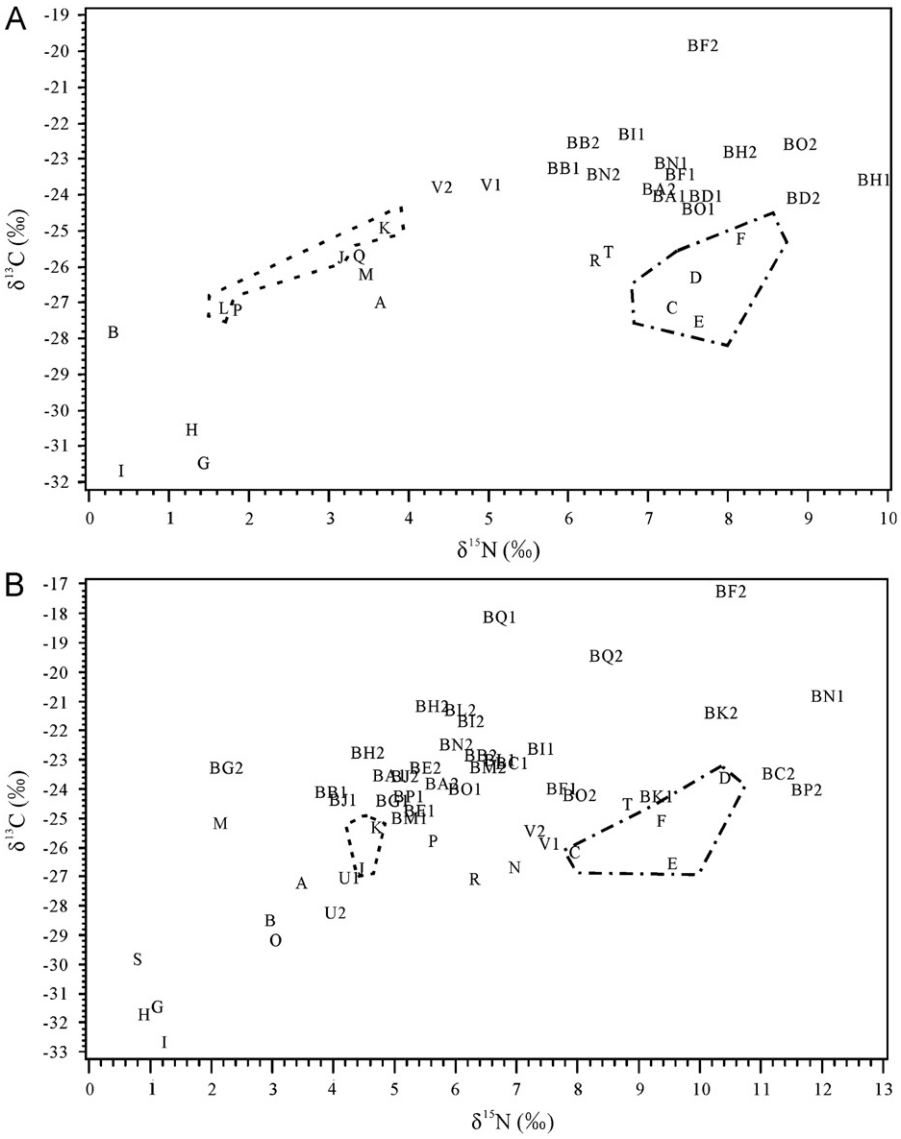

Fig. 1. Isotopic patterns from North Park (RM 2.0, Panel A) and Grand Cavern (RM 20.0, Panel B) based on averages for each sample type. The dotted lines enclose the samples coming from trophic pathways other than that of herbivory, i.e., terrestrial and aquatic detritivory-based paths. Soil and leaf litter were included for reference.

position (e.g., Hill et al., 1996; Tom et al., 2010), but our results only showed a clear pattern for the North Park food web (Fig. 2). Generally, plant tissues and herbivorous insects contained less than 10% of their mercury body burden as MeHg; bird tissues (blood and feather) contained consistently higher percentages of MeHg. If data for all birds were pooled, %MeHg of blood samples was 93.5% (95% CI=92.1–94.9%), slightly higher than that of the feather samples (88.5%, 95% CI=85.8–91.2%).

Use of feather mercury as an indicator of mercury exposure has been criticized recently by other researchers (e.g., Evers et al., 2005; Bond and Diamond, 2008). These new feather data, as well as those of previous research (Newman et al., 2011) were characterized by substantial variation so feather mercury results were omitted during model construction.

After pooling data from all four terrestrial sites and omitting terrestrial detritivores, members from aquatic food webs and bird feathers, PROC GLMSELECT was applied to examine the models that used $\log_{10}$ THg, $\log_{10}$ MeHg, or %MeHg as response variables. Using the AIC, forward selection picked δ^{15}N first and then Therm as predictive variables for all three response variables. The MeHg model also included δ^{13}C as a predictor and the model with %MeHg included both δ^{13}C and site as well. Previous research (Newman et al., 2011) discussed the differences that might exist among different sites and modeled each location separately. Including more data allowed us to statistically examine the validity of including continuous and categorical variables in the same model by testing the significance of their interaction terms. An ANCOVA test showed that only δ^{15}N and site interaction term had significant effect on MeHg concentrations with an F-value of 3.77 and p-value of 0.01, which brought into question the assumption of parallel slopes. As a result, separate models were built for each site to get estimates of FWMF and to predict mercury concentration in food web members.

Fitting full models (including δ^{15}N, δ^{13}C and Therm) to the three response variables for the individual sites showed that δ^{15}N was significant in all models; whereas Therm was significant in most models except for models of THg and MeHg for NP, models of THg for GTP, and models of %MeHg for AFC. The value of δ^{13}C was not significant in most models except that for MeHg for NP, %MeHg for GC and THg for GTP. Consequently, and consistent with Newman et al. (2011), δ^{15}N and Therm were finally selected to model mercury concentrations for individual sites (Eq. (7)).

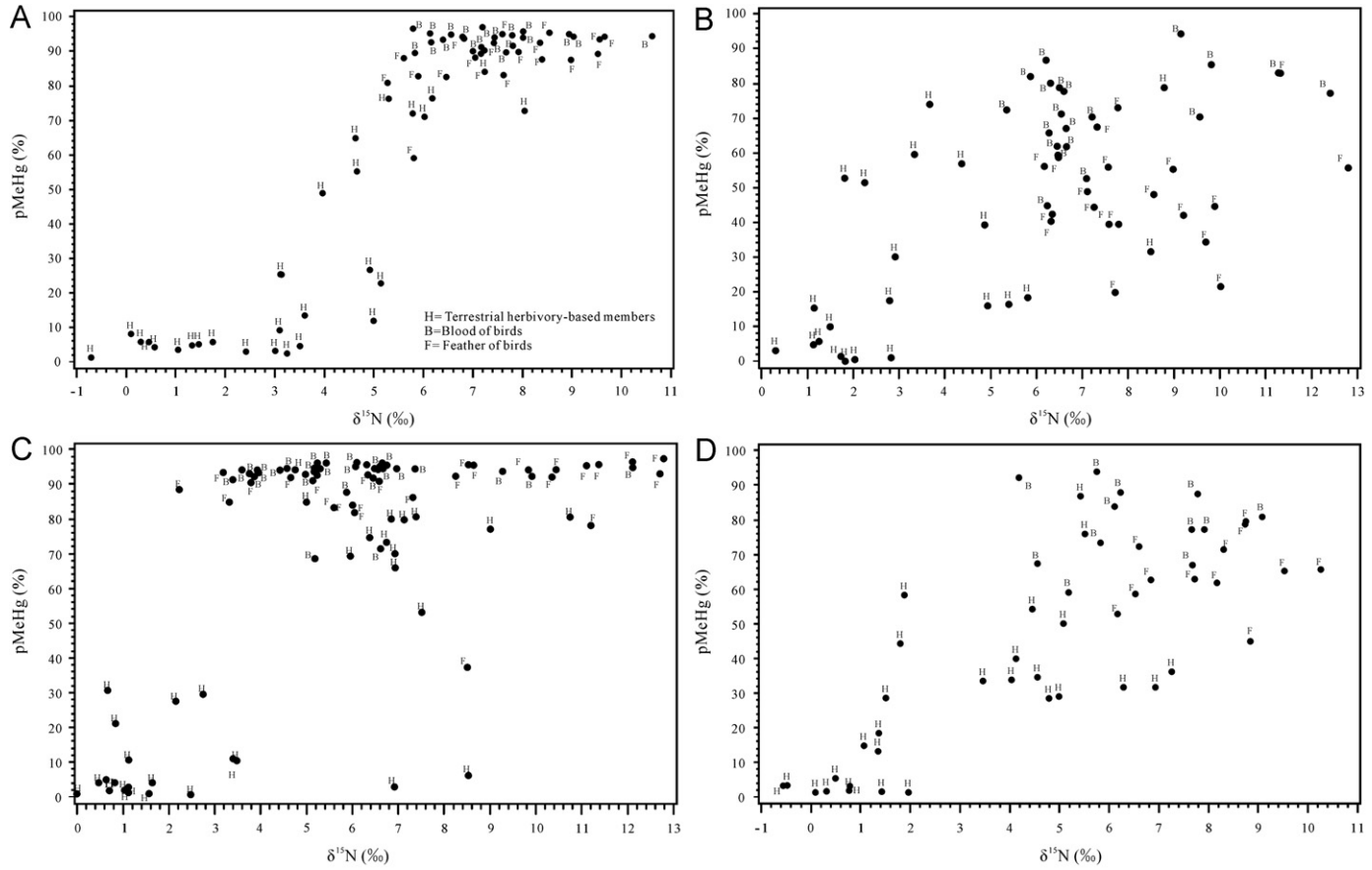

Fig. 2. Examination of the relationship between percentage of total mercury that was methylmercury (%MeHg) with $\delta^{15}N$ for North Park (RM 2.0, Panel A), Augusta Forestry Center (RM 11.8, Panel B), Grand Cavern (RM 20.0, Panel C) and Grottoes Town Park (RM 22.4, Panel D). Only terrestrial herbivory-based food web members were included.

Model results with $\delta^{15}N$ and calculated TL were given in Table 2. Model parameters and statistics for AFC and GTP reported previously (Newman et al., 2011) were included in Table 2 for comparison.

$$\log_{10}\text{THg or }\log_{10}\text{MeHg or \%MeHg} = a + b\delta^{15}N + c(\text{Therm}) + \varepsilon. \quad (7)$$

Given the current results, models with MeHg, but not THg and %MeHg, had sufficient predictive capabilities (Fig. 3) that met the *a priori* criterion. Consequently, models with THg and %MeHg were not analyzed further. The only exception was the MeHg model for GC which failed to meet the *a priori* criteria for adequate prediction. Overall, the biomagnification models based on $\delta^{15}N$ effectively predicted MeHg on most sites (three of floodplain sites and one aquatic model for the 23 mile river reach). Models based on calculated TL were built for the four sites (Table 2). Prediction of mercury concentration could be made by back transforming to the arithmetic scale with a proper correction (Newman, 1993; Newman et al., 2011).

The MeHg food web biomagnification factor (FWMF, fold increase per TL) was calculated using the estimated model parameter b, i.e., 10^b. A Q statistics was calculated to examine the heterogeneity of the four FWMF, which turned out to be 12.327 compared to its df of 3. In this situation, a p-value for Q was recommended by Cumming (2012) for judging the significance of Q. The p-value was 0.0063 suggesting that FWMF for the four locations were heterogeneous and a random effect model appropriate. For meta-analysis, the FWMF for North Park (17.4, 95% CI of 9.5–31.6) and Grand Cavern (6.2, 95% CI of 3.5–11.0) were combined with those of the previous study. Overall MeHg FWMF for the terrestrial food webs was 12.4 (95% CI of 6.8–22.3), which was higher than that of the contiguous river food web (4.6, with 95% CI of 3.6–5.8), supporting the previous findings of Newman et al. (2011) (Fig. 4).

4. Discussion

These trophic transfer studies were prompted by river manager concerns about potentially harmful mercury exposure of high trophic level species inhabiting the contaminated reach of the South River. Results from this study were consistent with those of previous studies (Tom et al., 2010; Newman et al., 2011) that MeHg was more amenable to biomagnification modeling than THg, and that %MeHg generally increased with trophic level. This was the case for both, the aquatic and floodplain food webs. After further examining GC data, we found that more bird species collected in GC were migratory species compared to NP and our previous studies. The mercury concentrations of these non-resident species might not reflect mercury exposure in GC and might contribute to the model variation. After omitting these species, the model r^2 increased to 0.71 and the $r^2_{\text{prediction}}$ increased to 0.67; the new FWMF was calculated to be 8.31 (95% CI of 4.57, 15.14) for GC.

Regardless, FWMFs calculated in this study were consistent with Newman et al. (2011). Methylmercury concentration increased more during movement through the floodplain food webs than in the contiguous aquatic food web of the South River. The mercury biomagnification in the South River floodplain food webs were also greater than that noted for the aquatic food

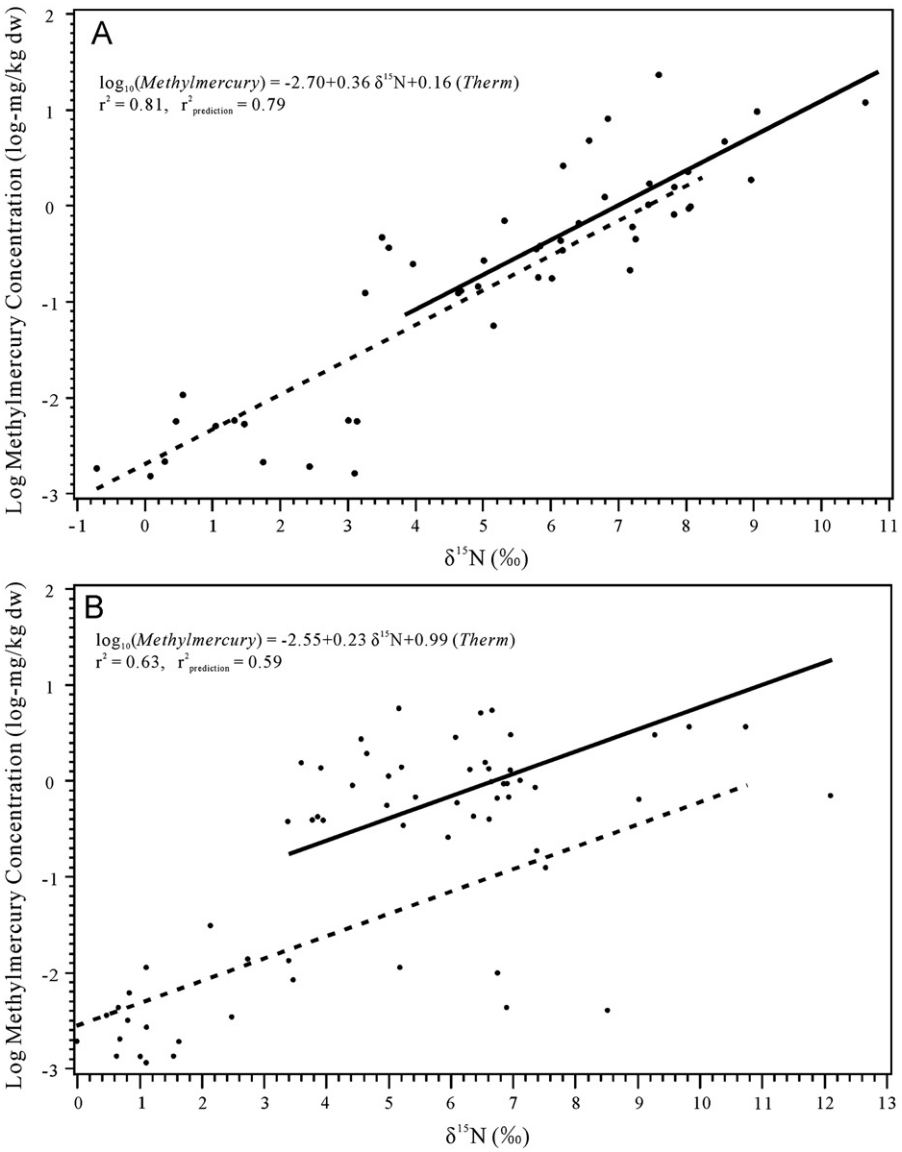

Fig. 3. Methylmercury biomagnification with $\delta^{15}N$ on North Park (RM 2.0, Panel A) and Grand Cavern (RM 20.0). The solid line indicates predictions for homeotherms and the dotted line indicates predictions for poikilotherms.

web of a similar contaminated Virginia river, the Holston River (Fig. 4; Tom et al., 2010). Although many studies explored factors that might influence mercury biomagnification, no clear general consensus has been reached to date. Wong et al. (1997) studied two aquatic communities with similar physical and chemical qualities, finding that food web structure might have influenced mercury biomagnification. Although this research was based on aquatic systems, it might provide some insight about the differences between floodplain and aquatic food webs in this study. The aquatic food web was composed of poikilotherms only but many of the floodplain food web members were homeotherms that consume more mercury-contaminated food than pokilotherms to meet their metabolic requirements. Also, compared to the adjacent river, samples from the terrestrial floodplain included more omnivores such as beetles, small mammals, and small birds that increased the complexity of the terrestrial food web structure. A more complex food web might lead to higher biomagnification rates (Vander Zanden and Rasmussen, 1996). Finally, the mercury assimilation efficiency of the terrestrial species might be higher than of their aquatic counterparts, resulting in higher biomagnification rates (Morel et al., 1998).

Although the overall FWMF calculated using meta-analysis in the terrestrial food webs was higher than the studied aquatic food webs, variation among terrestrial sites was substantial which could be seen from Fig. 4 and the Q statistic. Including more locations allowed a further examination of differences among floodplain food webs. Because of the relative lipophilicity of MeHg, it moves more readily through trophic webs than inorganic mercury. Therefore, %MeHg would increase consistently with increasing trophic positions for a food web with an uncomplicated structure, e.g., driven by a progressive movement from primary producer, to primary consumer, to secondary consumer, to higher consumers. However, only North Park in the current study showed a clear and consistent relationship between %MeHg and $\delta^{15}N$ that was comparable to that of the aquatic food web (Tom et al., 2010). Grottoes Town Park had a general relationship but it was not as clear as that of North Park. Augusta Forestry Center and Grand Cavern showed the least clear relationships. North Park is relatively small and has a relatively uniform topography compared to Augusta Forestry Center and Grand Cavern. More habitat heterogeneity might obscure the relationship between %MeHg and trophic position because higher habitat

Table 2
Summary of model parameters for four floodplain locations.[a]

	r^2	*a* (95% CL)	*b* (95% CL)	*c* (95% CL)	MSE	$r^2_{prediction}$
Total mercury (THg)[b]						
NP	0.47	−1.23 (−1.59, −0.88)	0.20 (0.12, 0.28)	−0.07 (−0.52, 0.39)	0.329	0.41
AFC	0.71	−1.47 (−1.76, −1.18)	0.20 (0.14, 0.27)	0.37 (−0.04, 0.78)	0.274	0.67
GC	0.39	−1.14 (−1.43, −0.85)	0.12 (0.06, 0.17)	0.36 (0.06, 0.66)	0.296	0.33
GTP	0.75	−1.83 (−2.11, −1.54)	0.29 (0.21, 0.37)	0.14 (−0.31, 0.58)	0.241	0.71
Methylmercury (MeHg)						
$\delta^{15}N$						
NP	0.81	−2.70 (−3.02, −2.37)	0.36 (0.29, 0.44)	0.16 (−0.26, 0.58)	0.279	0.79
AFC	0.83	−2.66 (−2.99, −2.34)	0.29 (0.21, 0.36)	0.89 (0.43, 1.35)	0.343	0.80
GC	0.63	−2.55 (−2.94, −2.15)	0.23 (0.16, 0.31)	0.99 (0.58, 1.40)	0.549	0.59
GTP	0.87	−3.12 (−3.42, −2.82)	0.41 (0.32, 0.50)	0.55 (0.08, 1.03)	0.273	0.85
TL						
NP	0.81	−3.71 (−4.21, −3.21)	1.24 (0.98, 1.50)	0.16 (−0.26, 0.58)	0.279	0.79
AFC	0.83	−3.45 (−3.91, −3.00)	0.97 (0.73, 1.21)	0.89 (0.43, 1.35)	0.343	0.80
GC	0.63	−3.19 (−3.75, −2.63)	0.79 (0.54, 1.04)	0.99 (0.58, 1.40)	0.549	0.59
GTP	0.87	−4.26 (−4.74, −3.78)	1.40 (1.10, 1.70)	0.55 (0.08, 1.03)	0.273	0.85
Percentage of methylmercury in total mercury (%MeHg)[b]						
NP	0.81	−8.32 (−19.22, 2.58)	11.08 (8.51, 13.64)	14.27 (0.16, 28.39)	313.336	0.79
AFC	0.57	14.56 (0.51, 28.62)	5.26 (2.22, 8.29)	28.65 (8.77, 48.52)	640.073	0.49
GC	0.70	7.13 (−4.28, 18.54)	6.25 (4.13, 8.36)	42.93 (31.16, 54.71)	456.481	0.67
GTP	0.70	6.47 (−4.66, 17.60)	6.26 (3.00, 9.53)	29.16 (11.66, 46.65)	375.512	0.66

[a] The number of observations for North Park (NP), Augusta Forestry Center (AFC), Grand Cavern (GC) and Grottoes Town Park (GTP) were 49, 43, 63 and 40 respectively.
[b] Models based on trophic level (TL) for THg and %MeHg were not constructed. Parameters were from models based on $\delta^{15}N$ unless otherwise indicated.

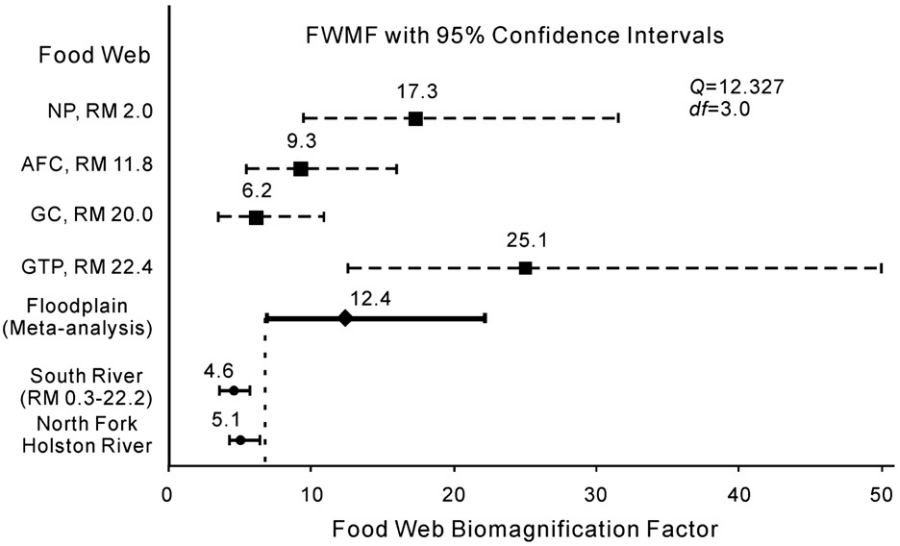

Fig. 4. Comparison of methylmercury FWMF in South River watershed (Augusta Forestry Center (RM 11.8), Grottoes Town Park (RM 22.4) from Newman et al. (2011) and North Park (RM 2.0) and Grand Cavern (RM 20.0) and meta-analysis from the current study). South River FWMFs were calculated from six pooled sites within the contaminated 23 mile reach, Holston River FWMFs were calculated from sites 46 miles downriver of the historical site of discharge (Tom et al. 2010). FWMF with its confidence interval were presented for each food webs, with numbers above each symbol represent estimates of FWMF.

heterogeneity could result in higher biodiversity (Benton et al., 2003).The impact of biodiversity on %MeHg could involve two features. Different species from the same trophic level might have different methylmercury assimilation efficiencies. Also, predators or competitors might be attracted to sites with higher biodiversity (Benton et al., 2003), which could bring species with %MeHg signatures reflective of other locations.

Avian species were the high position predators of interest on the South River floodplain. The present study documented increased mercury concentration with increasing trophic position, and produced models to predict the mercury concentrations for higher position members of South River food webs. Previous South River studies suggested adverse effects due to mercury exposure of tree swallow (Brasso and Cristol, 2008; Hallinger and Cristol, 2011) and Eastern bluebird (Condon and Cristol, 2009). Combining our 2009 and 2010 data, blood mercury concentrations of 82 birds and feather mercury concentrations of 81 birds were available to compare with the avian biomonitoring literature. Unfortunately, minimal information about toxicity thresholds could be found based on avian blood and feather concentration. Evers et al. (2008) suggested that captive loons with blood mercury concentrations (ww, THg) over 3 μg/g were at high risk of adverse effects. Their corresponding threshold based on feathers was 40 μg/g (ww, THg). Ignoring uncertainties associated with variation in sensitivity between loon and the avian species in this study, only two out of 82 blood samples (2.4%) were greater than 3 μg/g and one out of 81 feather samples (1.2%) was greater than 40 μg/g in this study. Using the mercury concentrations at which adverse effects were noted for tree swallows (Brasso and Cristol, 2008) (3.56 μg/g (ww, THg) for blood and 13.55 μg/g (ww, THg) for feather), two

blood samples (2.4%) and seven feather samples (8.5%) from this study were above the corresponding values. A recent publication (Jackson et al., 2011) modeling the relationship between reproductive successes of Carolina wren in South River and mercury exposure suggested that as low as 0.7 μg/g ww THg in female blood would cause 10% reduction of nest success. Another recent paper (Cristol et al., 2012) discussed the threshold of using feather mercury as the indicator of mercury effects, suggesting that the threshold might be lower than 9–14 μg/g ww THg. Generally and contrary to our initial assumptions, evidence from this study does not support the hypothesis that the bird populations sampled in this study were exposed to harmful mercury concentrations.

5. Conclusions

Extending the previous trophic transfer study of the mercury-contaminated South River watershed, the current study assessed models for making useful predictions of mercury concentration in members of the floodplain food webs at two additional locations. Acceptable prediction from each model was gauged with an *a priori* established prediction r^2 of approximately 0.80. Overall, the models predicting methylmercury were superior to models for total mercury or the percentage of the mercury present as methylmercury. Including previous models for other river sites, four of five attempted methylmercury models based on $\delta^{15}N$ met the criterion for useful prediction (A sixth model built by Tom et al. (2010) for another mercury-contaminated Virginia river also met this criterion.). For floodplain models, thermoregulatory status was included. The only location failing to produce a model that met a priori criteria, Grand Caverns, included more migratory avian species which could have contributed to the wide variation in the associated data. The food web biomagnification factor of North Park and Grand Cavern were 17.4 (95% CI of 9.5–31.6) and 6.2 (95% CI of 3.5–11.0) respectively, and the overall food web biomagnification factor of South River floodplain generated using meta-analysis was 12.4 (95% CI of 6.8–22.3), supporting previous findings that the South River floodplain food webs had higher biomagnification factors than the contiguous aquatic food web.

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