

Mercury accumulation in amphibians of the South River: Nondestructive indices of exposure, maternal transfer, and reproductive effects



Christine Bergeron

Jason Unrine, Ph.D.

Cathy Bodinof

Outline

Part 1 – Mercury Accumulation and Nondestructive Indices

Objectives:

1) Determine whether amphibians accumulate high concentrations of Hg in their tissues, making them important to the fate and transport of Hg within the South River foodweb.

2) Determine whether accumulation of Hg in amphibians follows the same spatial pattern as observed in other biota along the South River.

3) Determine if tail tissue is a useful nondestructive index of Hg exposure for amphibians that exhibit tail autonomy.

Part 2 – Maternal Transfer of Mercury and Reproductive Effects

Why are Amphibians Critical Components of the South River Ecological Study?

High Density

High Conversion Efficiency

Diverse Trophic Levels

Diverse Habitat Requirements

Important Predators/Prey

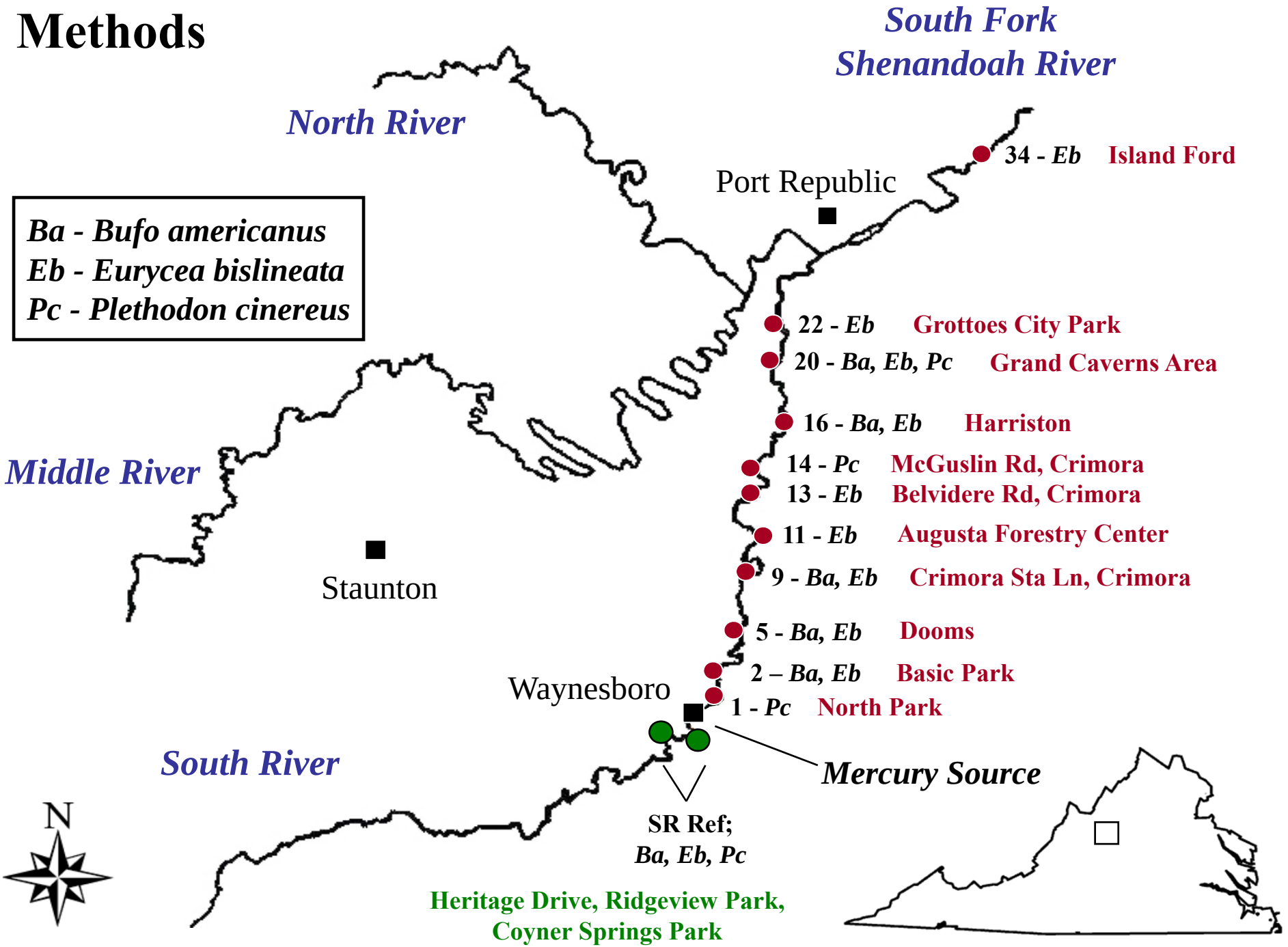
Important in cycling contaminants???


Eurycea bislineata
Two-lined Salamanders
Photo: J.D. Willson


Plethodon cinereus
Red-Backed Salamanders

Bufo americanus
American Toad

Methods


South Fork
Shenandoah River
North River
Port Republic
34 - Eb Island Ford
Ba - Bufo americanus
Eb - Eurycea bislineata
Pc - Plethodon cinereus
22 - Eb Grottoes City Park
20 - Ba, Eb, Pc Grand Caverns Area
16 - Ba, Eb Harriston
Middle River
14 - Pc McGuslin Rd, Crimora
13 - Eb Belvidere Rd, Crimora
11 - Eb Augusta Forestry Center
Staunton
9 - Ba, Eb Crimora Sta Ln, Crimora
5 - Ba, Eb Dooms
2 – Ba, Eb Basic Park
Waynesboro
1 - Pc North Park
South River
Mercury Source
N
SR Ref;
Ba, Eb, Pc
Heritage Drive, Ridgeview Park,
Coyner Springs Park

Methods

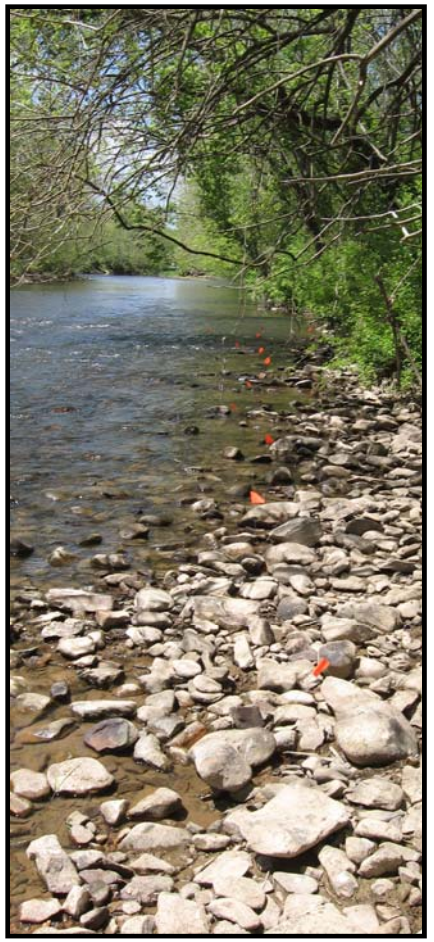
- Collected salamanders by flipping rocks and logs

- Collected *B. americanus* adults and larvae at breeding ponds

- Held individuals for 48 hours

- Sacrificed individuals, separated tails and bodies in salamanders, obtained blood in *B. americanus*

- Froze samples until analyses



Sample Sizes

Site	*E. bislineata* adult	*E. bislineata* larvae	*P. cinereus* adult	*B. americanus* adult	*B. americanus* larvae*
Reference Subsites					
SR REF	5	5	6	13	7
CS REF	5	5	-	-	-
Reference (totals)	**10**	**10**	**6**	**13**	**7**
Contaminated subsites					
SR RM 1	-	-	6	-	-
SR RM 2	5	5	-	1	4
SR RM 5	5	5	-	12	4
SR RM 9	5	5	-	12	4
SR RM 11	1	1	-	-	-
SR RM 13	5	5	-	-	-
SR RM 14	-	-	6	-	-
SR RM 16	5	5	-	4	4
SR RM 20	5	5	6	6	3
SR RM 22	5	5	-	-	-
South River (totals)	**36**	**36**	**18**	**35**	**19**
South Fork Shenanndoah River					
SFSR RM 34	5	4	-	-	-
Species Total	**51**	**50**	**24**	**48**	**26**

* composite samples of 4-7 tadpoles each

Analytical methods

Total Hg:

Direct Hg Analyzer

Methyl Hg/Total Hg:

gas chromatographic cold-vapor atomic fluorescence spectrometry (GC-CVAFS) & ICPMS

Selenium:

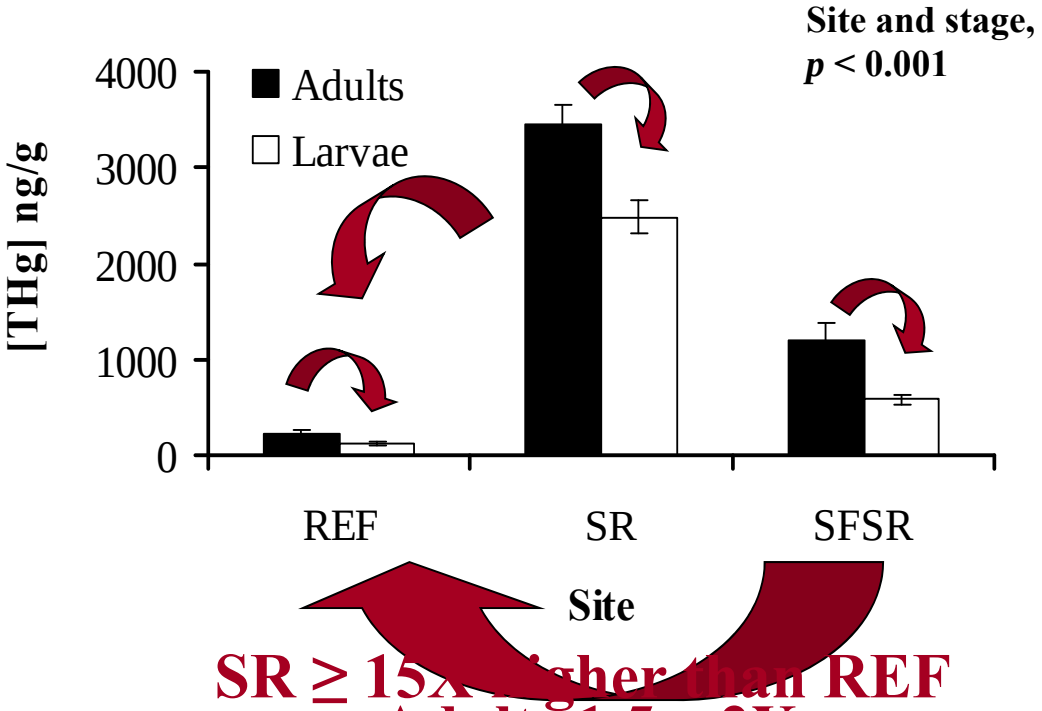
ICPMS

The University of Georgia
Savannah River Ecology Laboratory

Advanced Analytical Center for Environmental Sciences

Eurycea bislineata

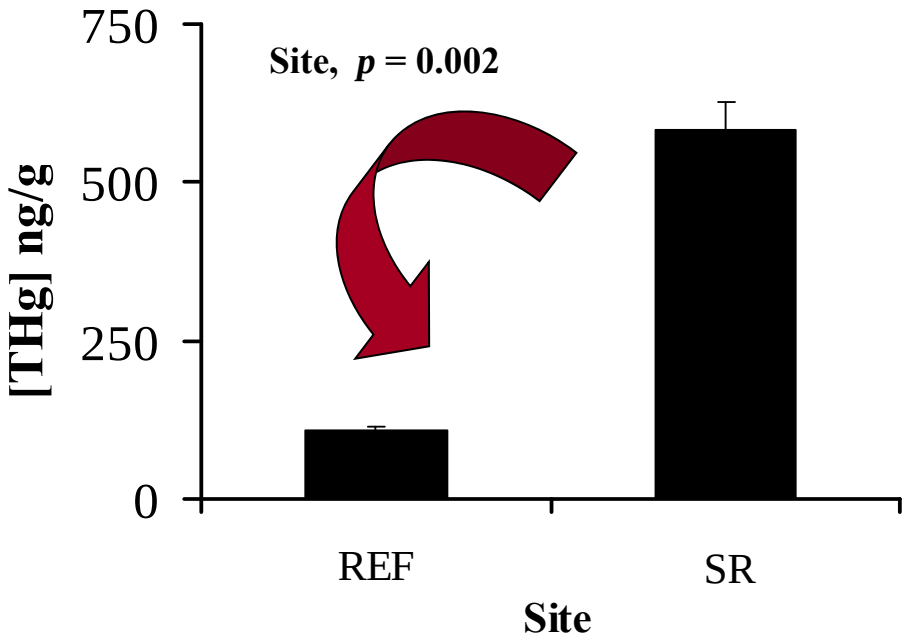
Bergeron et al., in prep



SR ≥ 15X higher than REF
Adults 1.5 – 2X
higher than Larvae
SFSR 5X higher than REF

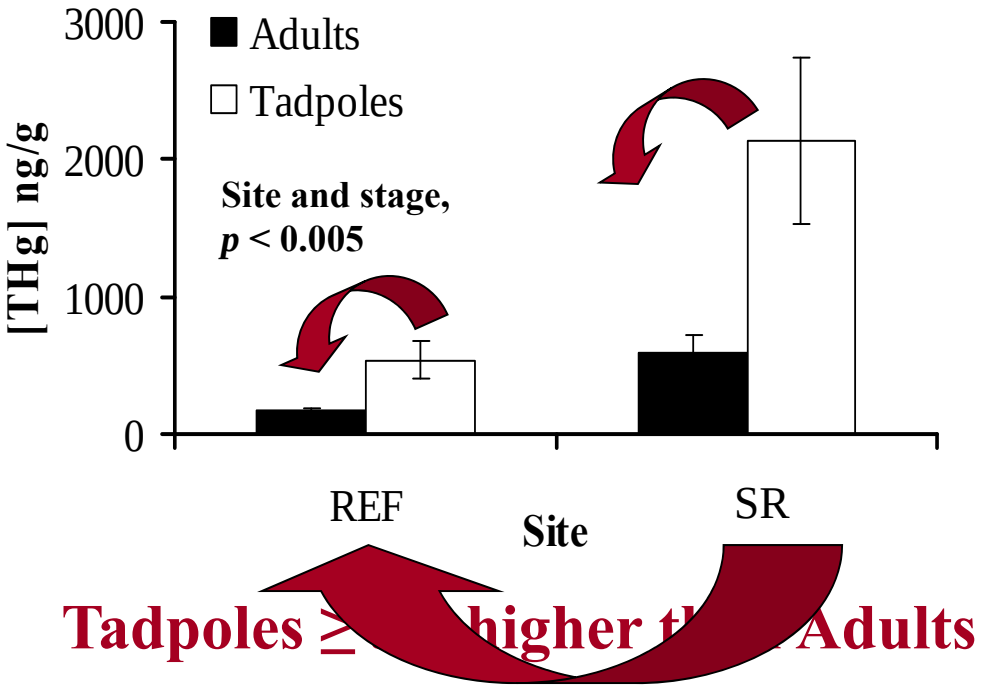
Plethodon cinereus

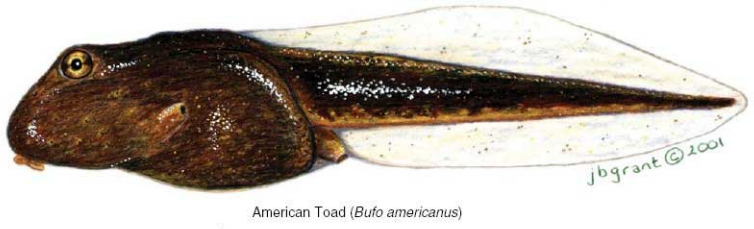
Bergeron et al., in prep



SR 5.4X higher than REF

Bufo americanus



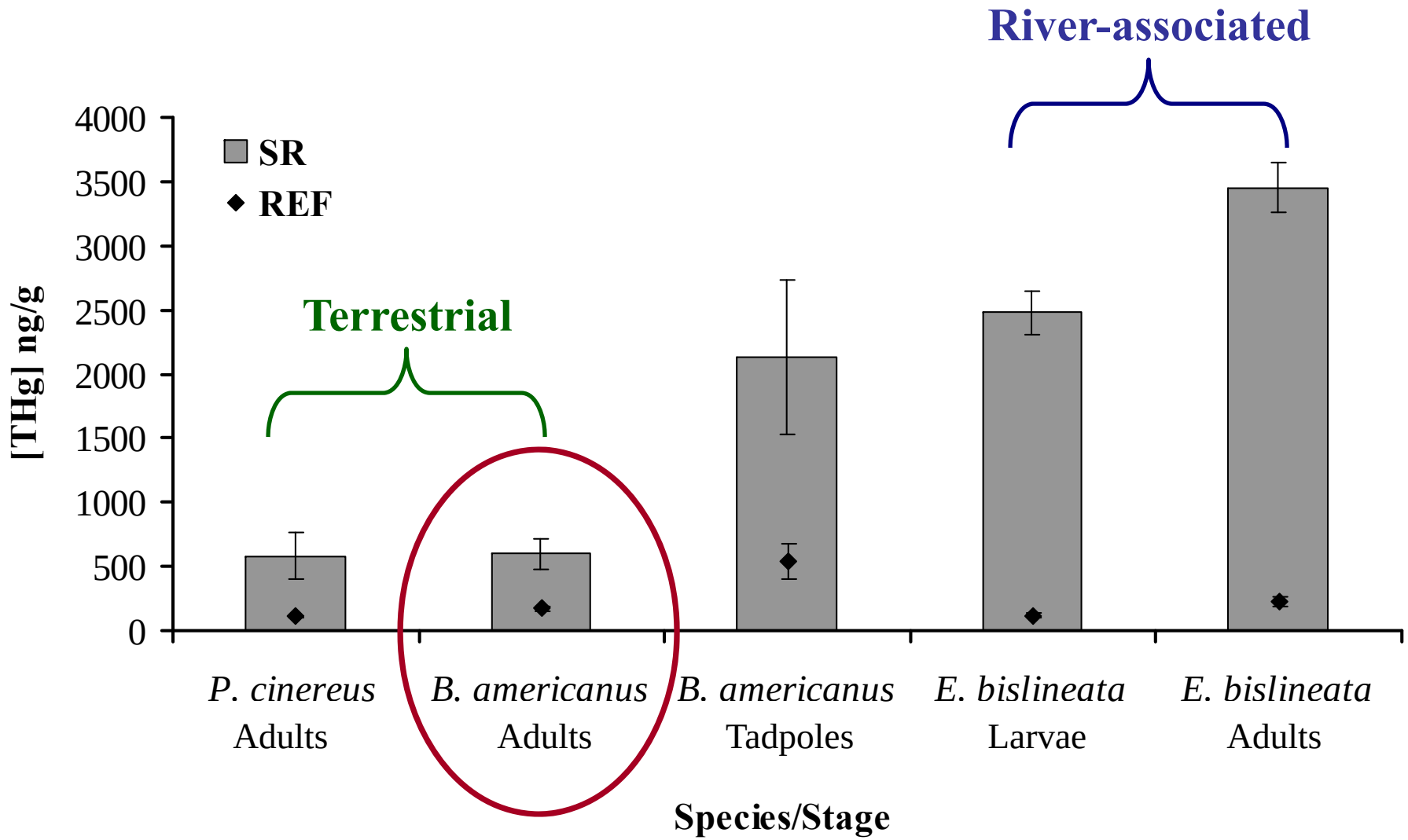


American Toad (*Bufo americanus*)

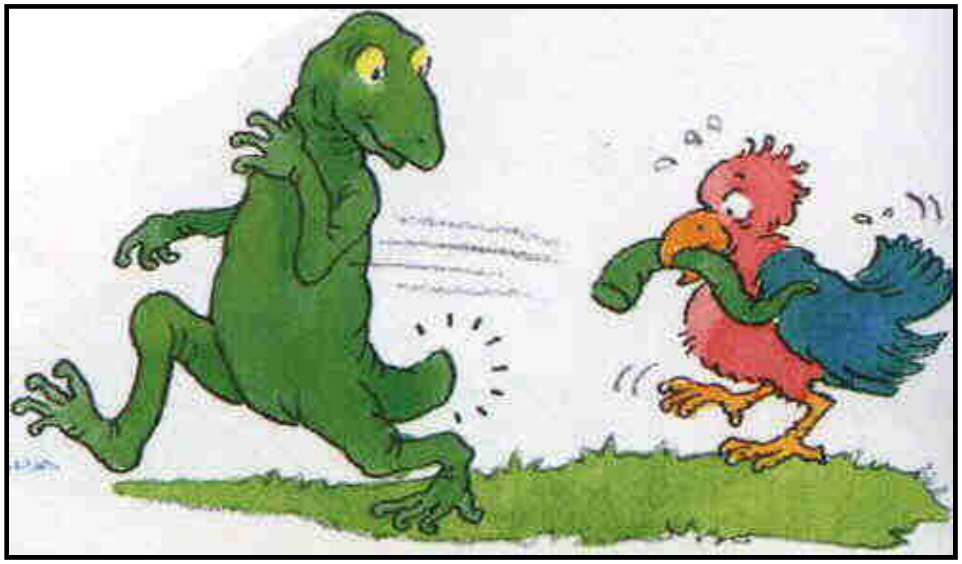
SR ≥ 3.5X higher than REF

Bergeron et al., in prep

Species Summary

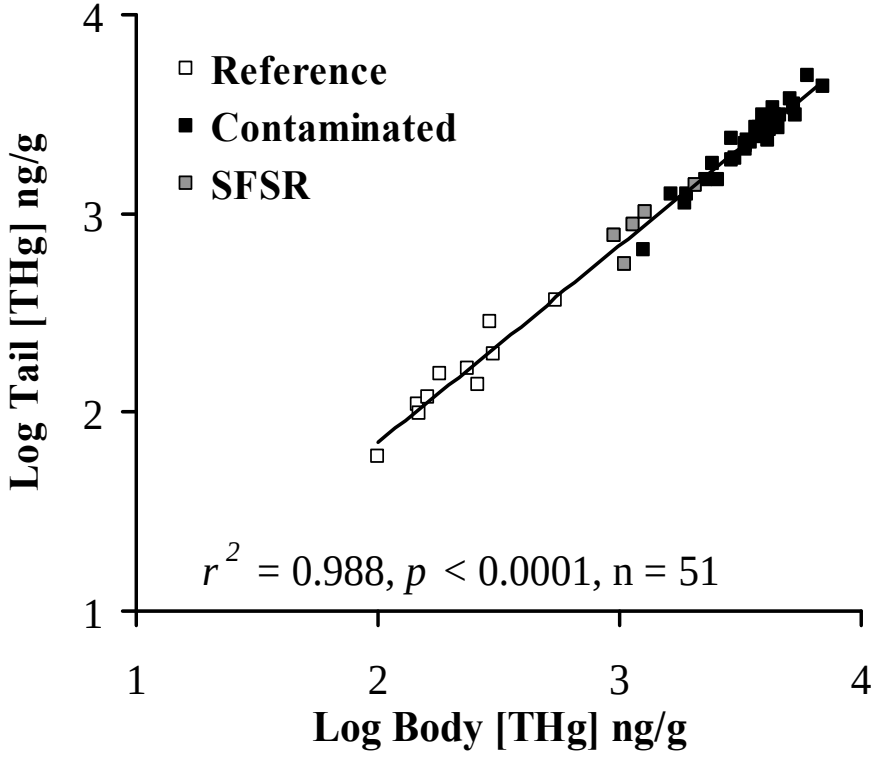


Nondestructive Indices of Hg Exposure

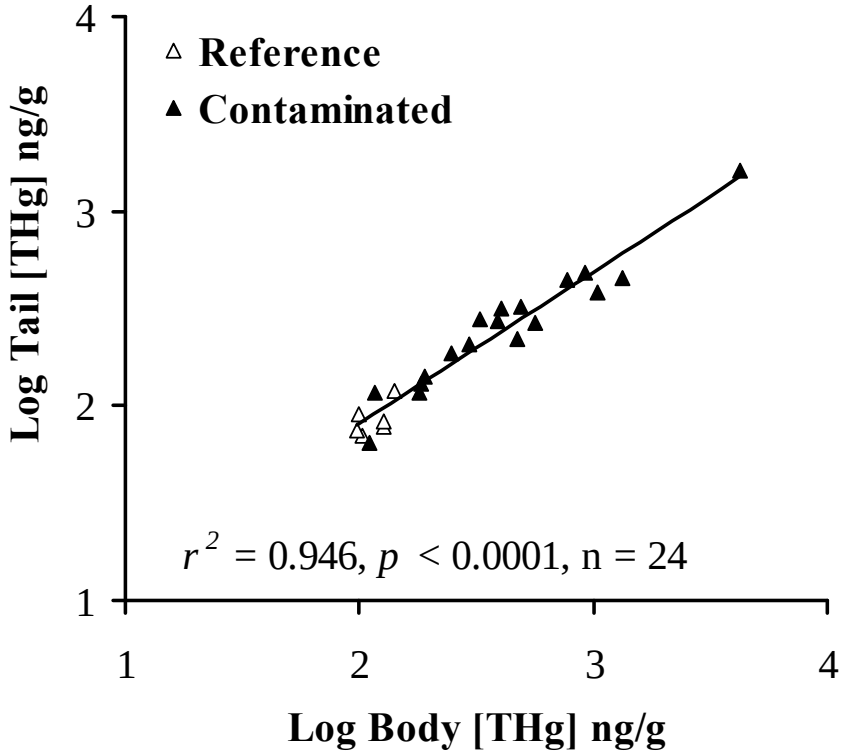


Nondestructive Indices of Hg Exposure

Eurycea bislineata



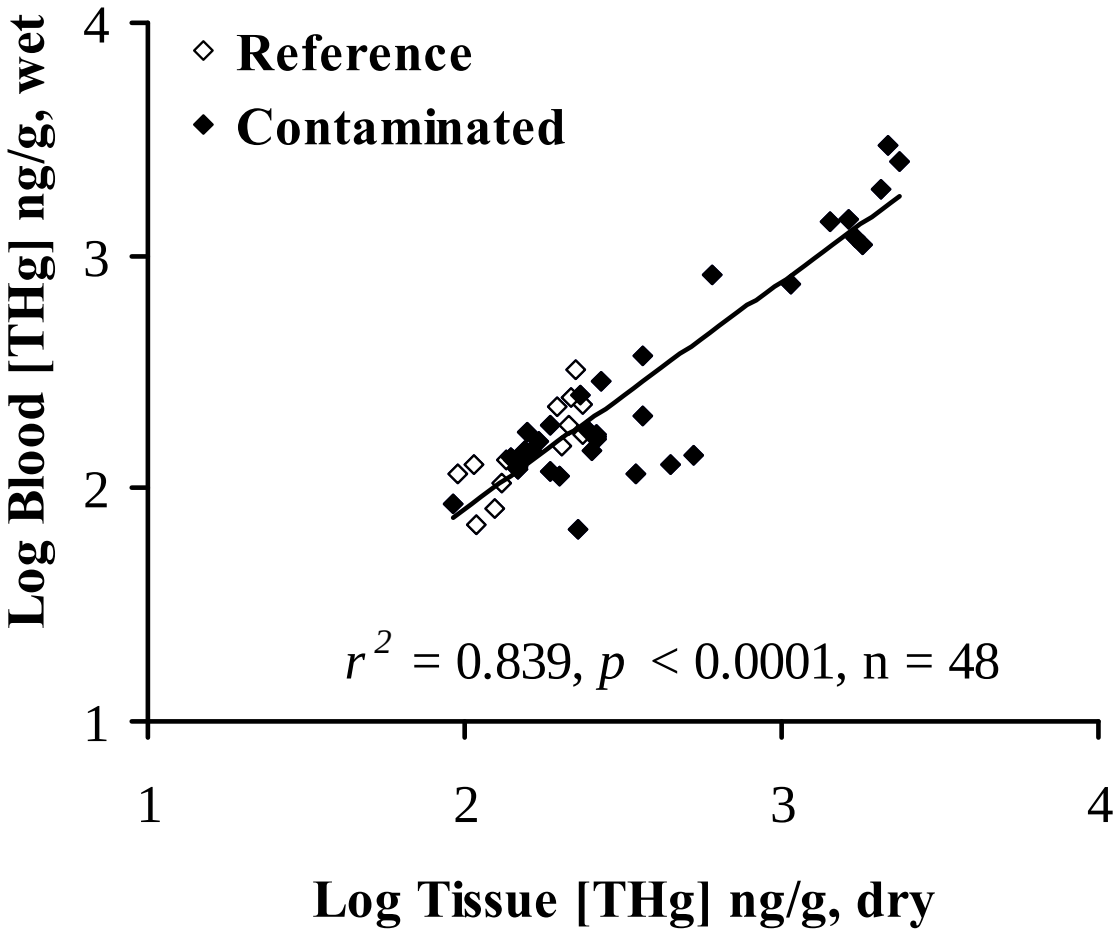
Plethodon cinereus



Bergeron et al., in prep

Nondestructive Indices of Hg Exposure

Bufo americanus



Bergeron et al., in prep

Part 1 - Summary

- **Amphibians inhabiting the South River have elevated Hg in their tissue compared to reference amphibians, but accumulation is species- and stage-dependent**

- **In general, *E. bislineata* adults and larvae had the highest [THg] and adult *B. americanus* and *P. cinereus* had the lowest [THg]**

- ***E. bislineata* adults had higher [THg] than larvae**
  ***B. americanus* larvae had higher [THg] than adults**

- **Mean % MMHg ranged from 45-60% in carcasses and were ~70% in *B. americanus* blood in both contaminated and reference sites**

- **Tails in salamanders and blood in *B. americanus* are excellent non-lethal alternatives for examining Hg in these species**

Part 2: Maternal Transfer and Reproductive Effects

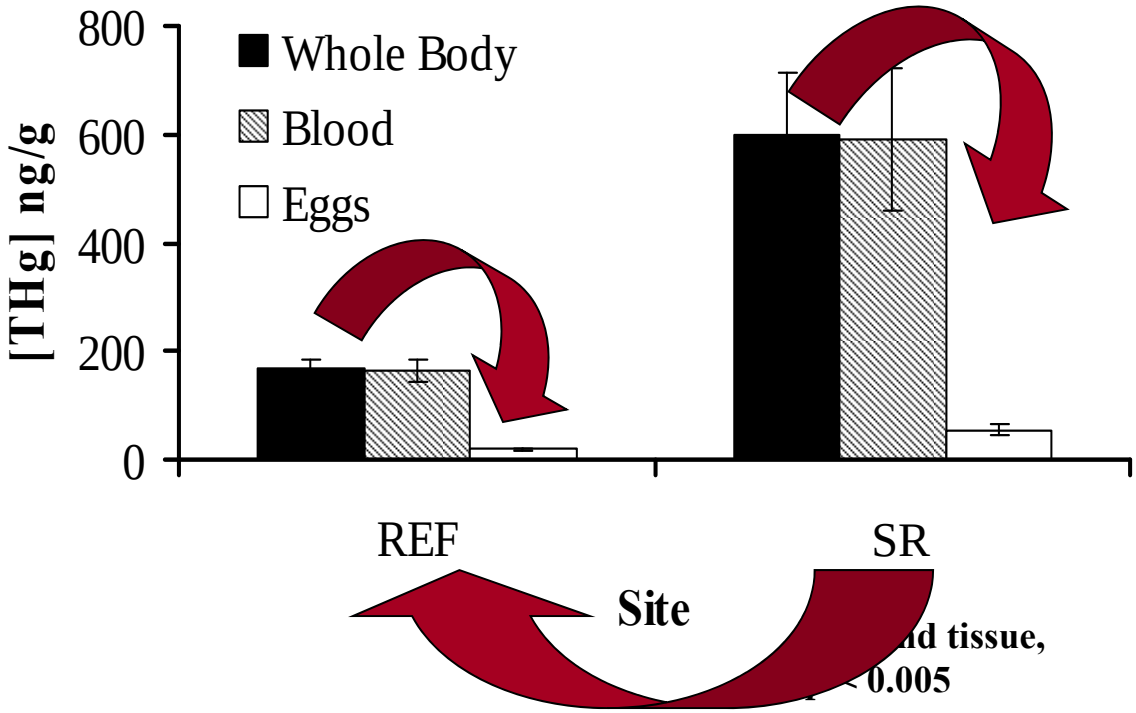
Objectives:

1) Determine whether female amphibians maternally transfer Hg to their offspring in a tissue – residue dependent manner.

2) Determine whether maternal transfer of Hg negatively impacts hatching success, growth, and development in a tissue – residue dependent manner.

Part 2: Methods

- Collected breeding pairs from ponds
- Injected with hCG to facilitate egg laying
- Counted eggs
  - Subset for hatching success/ developmental assessment
  - Subset Hg analyses
- Released males, held females for 48 hours
- Obtained blood from females, sacrificed for Hg analyses



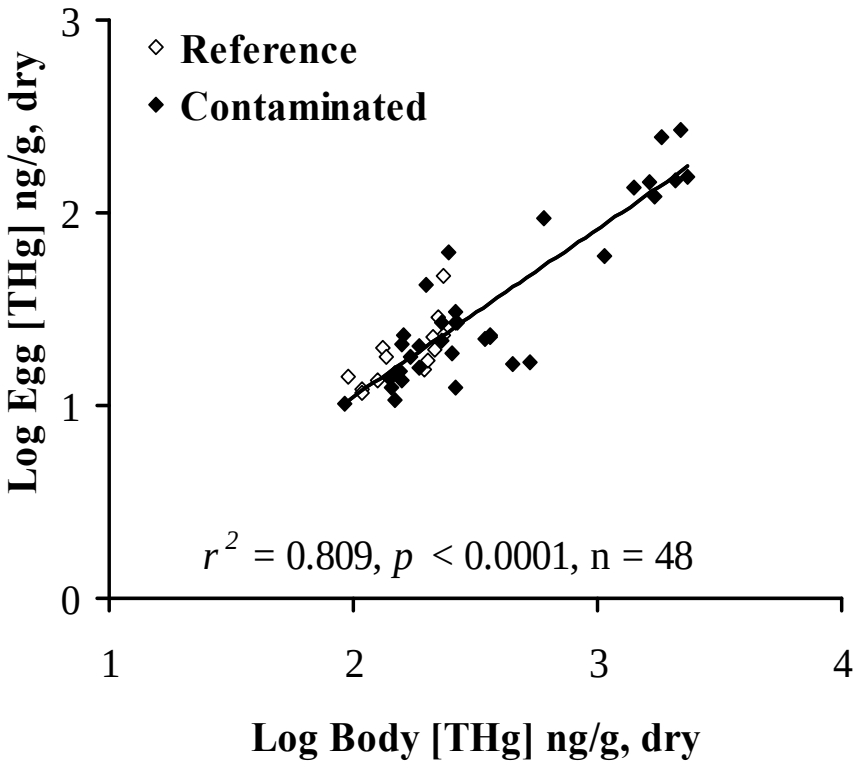
Bufo americanus – Egg concentrations

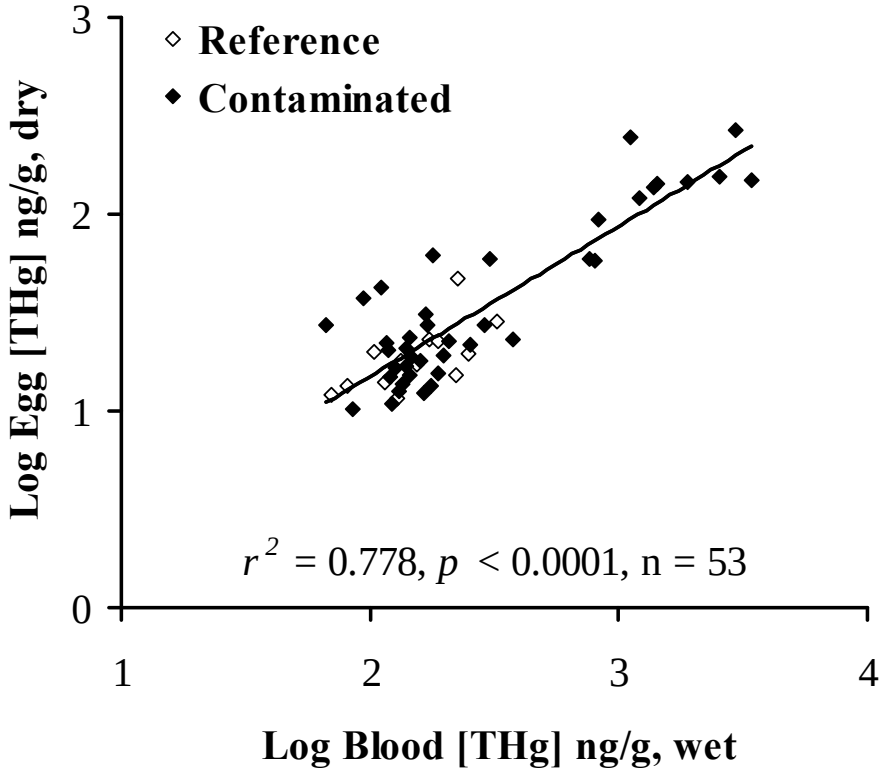
Body ~ 10X higher than Eggs

SR ≥ 3X higher than REF

Bergeron et al., in prep

Bufo americanus – Egg relationships





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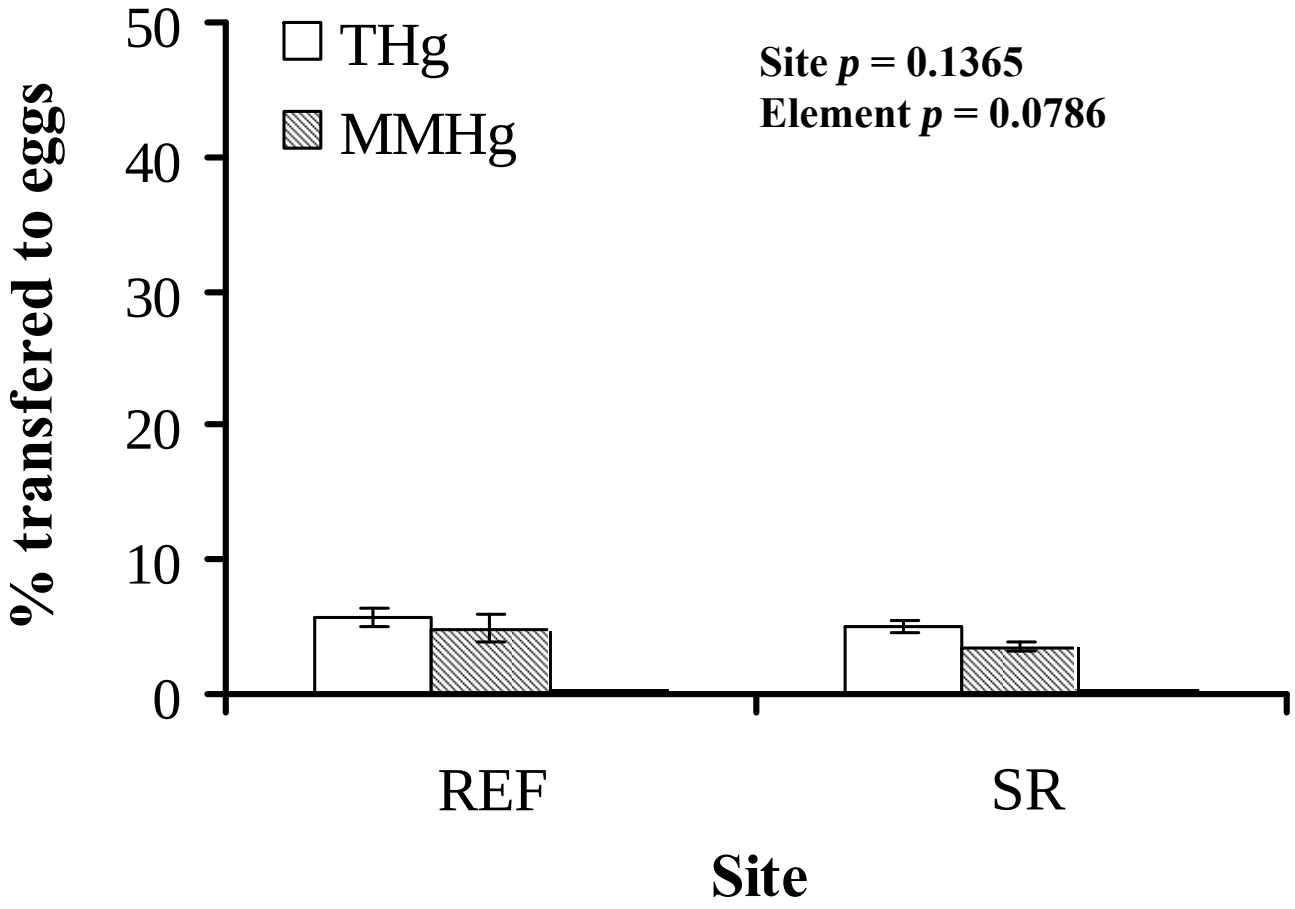
Bufo americanus – Maternal Transfer



Site p = 0.1365
Element p = 0.0786

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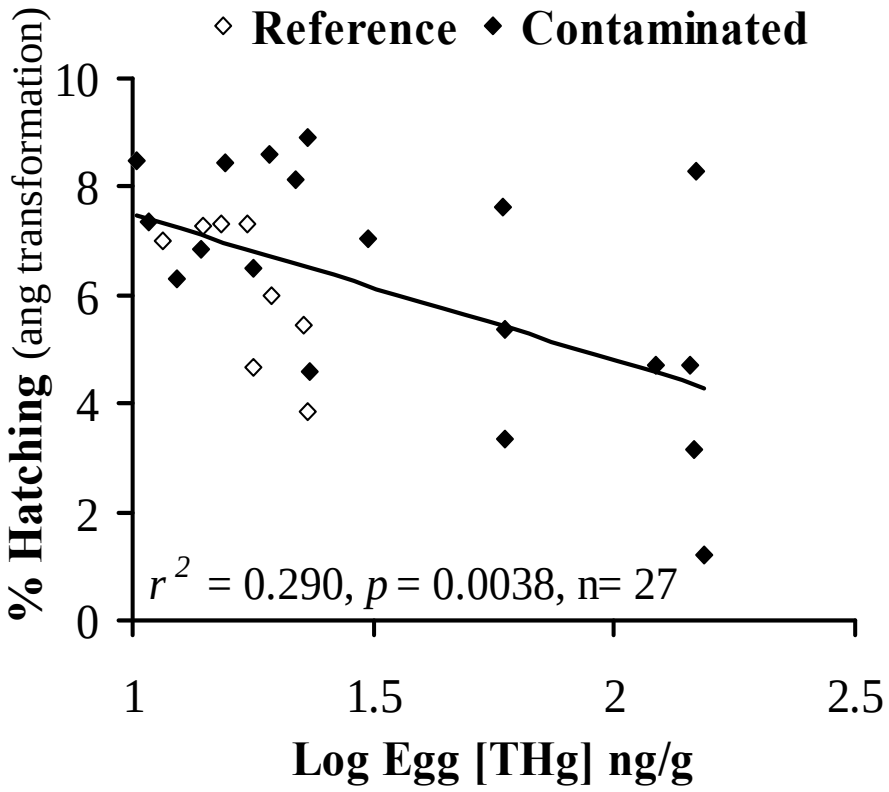
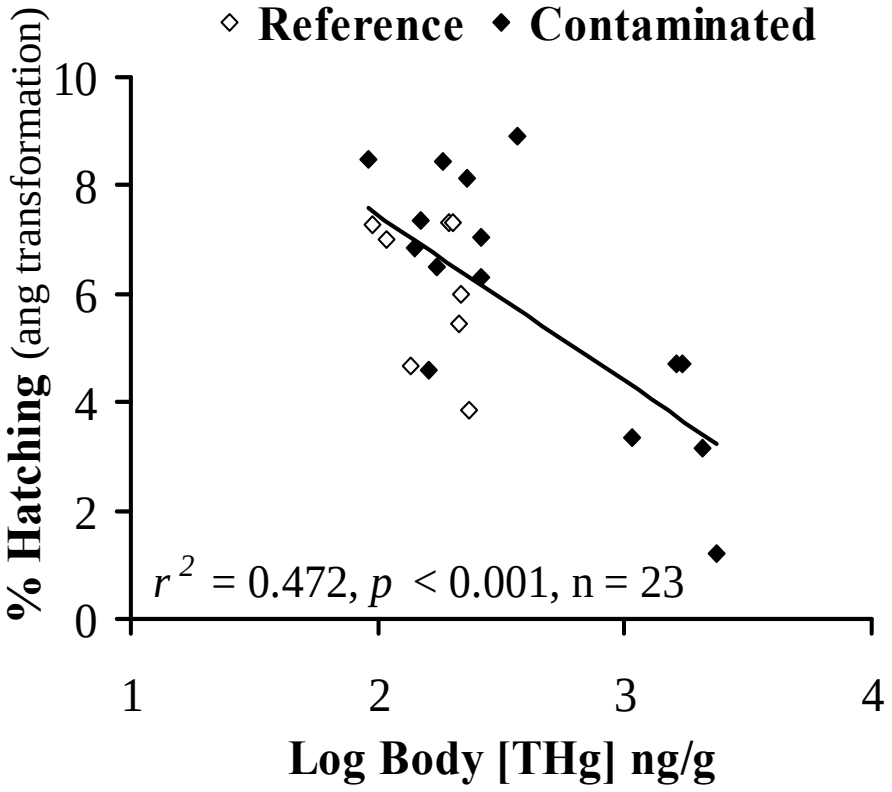
Bufo americanus – Reproductive Effects





% Hatching vs Log Blood [THg] – r^2 = 0.178, p = 0.029, n =27

First time residue-specific changes in reproductive function have been shown

Part 2 - Summary

- **Egg [THg] are ≥ 10x lower than female whole body and blood**
- **Body and blood [THg] are a good predictors of egg [THg]**
- **Females transfer ~ 5% of their THg body burden to their eggs**
- **Maternally transferred Hg is negatively correlated with hatching success**
- **Further research is needed to clarify the impact of Hg bioaccumulation on:**
  - **embryonic development (e.g., malformation frequencies)**
  - **larval development**
  - **female reproductive success**
  - **ecological significance of compromised reproductive success**
  - **other species ???**

Acknowledgements

Hopkins Lab: Sarah Budischak, Sarah DuRant,
Sarah Orlofske, Haruka Wada
SREL: Heather Brandt, Diane Addis
Dan Cristol and Lab
Scott Gregory, SRST office
Mike Liberati, SRST
Ralph Stahl, SRST
Erin Mack, SRST
John Schmerfeld, US Fish and Wildlife
Don Kain, VA DEQ

Landowners along the South River
Waynesboro Parks and Recreation Department