An Overview of Ecological Studies of Wildlife Potentially Impacted by the Kingston Ash Spill

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ABSTRACT

The release of fly ash at the Tennessee Valley Authority (TVA) Kingston Fossil Plant (KIF) on December 22, 2008 discharged approximately 5.4 million cubic yards of ash slurry, which contains metals and metalloids that might pose a risk to ecological receptors within the adjacent aquatic and terrestrial systems. TVA and collaborating organizations have continued and expanded the studies initiated in the aftermath of the release in order to assess impacts of the spill to wildlife resources in the area and to document whether the ash spill has exposed wildlife in the region to trace elements.

Wildlife groups in these studies include piscivorous, insectivorous, and herbivorous birds; spring breeding amphibians; aquatic reptiles; and terrestrial mammals. Wildlife species selected include osprey (*Pandion haliaetus*), great blue heron (*Ardea herodias*), Canada goose (*Branta canadensis*), tree swallow (*Tachycineta bicolor*), upland chorus frog (*Pseudacris feriarum*), spring peeper (*P. crucifer*), American toad (*Bufo americanus*), snapping turtle (*Chelydra serpentina*), common musk turtle (*Sternotherous ordoratus*), and raccoon (*Procyon lotor*). Tissues tested include contents of eggs (birds), whole body (amphibian and bird nestlings), blood (turtles and mammals), and hair and organs (mammals). While bioaccumulation was not noted or expected in the first year of studies for higher trophic levels, the continued studies are the first results indicating whether these receptors are accumulating constituents of concern. Trends for constituents and species are presented.

OBJECTIVES

The goal of these studies is to document whether the ash spill has exposed wildlife in the region to trace elements found in coal ash.

Objective 1: Determine whether trace elements are bioavailable to wildlife occupying different trophic levels/ feeding ecologies.

Objective 2: Determine whether wildlife are maternally transferring trace elements to young.

- Wildlife resources were identified in 2009, and multiple species were selected based upon their availability and potential exposure to 26 trace elements.
- Elements tested include aluminum (Al), antimony (Sb), arsenic (As), barium (Ba), beryllium (Be), boron (B), cadmium (Cd), calcium (Ca), chromium (Cr), cobalt (Co), copper (Cu), iron (Fe), lead (Pb), magnesium (Mg), manganese (Mn), mercury (Hg), molybdenum (Mo), nickel (Ni), potassium (K), selenium (Se), silver (Ag), sodium (Na), strontium (Sr), thallium (Tl), vanadium (V), and zinc (Zn). Only Se concentrations have been quantitatively compared at this point.
- Tissues tested include contents of eggs (birds), whole body (amphibian and bird nestlings), blood (turtles and mammals), hair and organs (mammals).
- Samples were collected from potentially impacted sites, as well as from locations upstream and downstream of the Kingston Fossil Plant (KIF) for comparison. Reference sites include the upstream Tennessee River, Fort Loudoun Dam, Tellico Dam, Timberlake Pond, the Hugo Property (Figure 2). Melton Hill Dam was used as a historically ash-impacted site.

Objective 3: Determine whether bioaccumulation and maternal transfer result in adverse effects.

- Based on results from biomonitoring efforts in 2009-2010, full scale effects-based studies have been funded by TVA and will be initiated in Spring 2011 on tree swallows and aquatic turtles. Studies will focus on maternal transfer of trace elements, reproductive success, and metrics of physiological performance

COLLECTION SITES

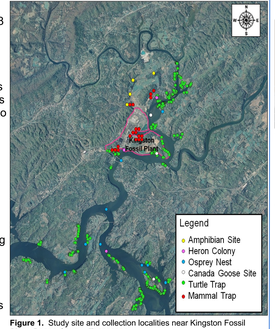

Figure 1. Study site and collection localities near Kingston Fossil Plant, Roane County, Tennessee.

OVERVIEW MAP WITH REFERENCE SITES

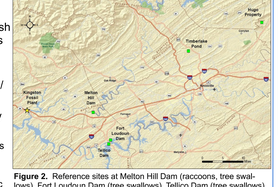

Figure 2. Reference sites at Melton Hill Dam (raccoons, tree swallows), Fort Loudoun Dam (tree swallows), Tellico Dam (tree swallows), Timberlake Pond (amphibians, turtles), Hugo Property (raccoons).

Osprey nest on the Emory River immediately following spill. Photo credit: T. Hill Henry.

PISCIVOROUS AND HERBIVOROUS BIRDS

GREAT BLUE HERON, OSPREY, CANADA GOOSE

TVA Transmission Linemen sampling heron eggs on the upstream TN River.

Osprey on nest at Kingston Fossil Plant intake. Photo credit: T. Hill Henry.

Canada geese feeding around ash pools on site.

GREAT BLUE HERON AND OSPREY

- Eggs of great blue heron (*Ardea herodias*) and osprey (*Pandion haliaetus*) were collected from available nests (1 egg per nest) at potentially impacted sites around KIF and at the upstream Tennessee (TN) River reference site. Eggs were weighed, measured, and frozen (≤10°C) for analysis.
- 2009 Collections: 10 heron eggs (5 reference, 5 impacted); 9 osprey eggs (3 reference, 6 impacted).
- 2010 Collections: 13 heron eggs (4 reference, 9 impacted); 10 osprey eggs (5 reference, 5 impacted).
- Selenium concentrations were compared between locations for each species (Figure 3). Selenium concentrations were significantly higher in heron eggs collected from impacted sites (3.26 ± 0.18) than in heron eggs from the reference site (2.63 ± 0.15; t-test, p=0.031). In contrast, no significant difference was found in Se concentrations detected in osprey eggs from the impacted area (2.33 ± 0.12) compared to osprey eggs from the reference site (2.60 ± 0.44; t-test, p=0.928).

Harriman Utility Board crew sampling heron eggs.

Field biologist measuring an osprey egg collected from a nest on the Emory River.

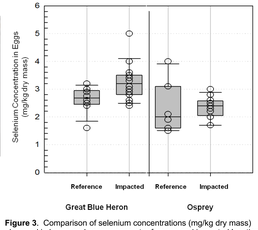

Figure 3. Comparison of selenium concentrations (mg/kg dry mass) observed in heron and osprey eggs at reference and impacted locations (2009 and 2010 data combined).

CANADA GOOSE

- Eggs of Canada geese (*Branta canadensis*) were collected from available nests (1 egg per nest) at potentially impacted sites around KIF and upstream in the Tennessee River. Eggs were weighed, measured, and frozen (≤10°C) for analysis.
- 2009 Collections: 4 eggs (4 impacted).
- 2010 Collections: 25 eggs (10 reference, 15 impacted).
- Selenium concentrations were compared between locations (Figure 4). Based on Wilcoxon-Mann Whitney (WMW) test, Se concentrations were significantly higher in goose eggs from the impacted area (2.12 ± 0.31) compared to eggs from the reference site (0.62 ± 0.04; p<0.0001).

Field biologist collecting a Canada goose egg from a nest on the Emory River.

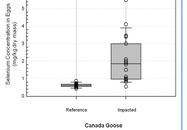

Figure 4. Comparison of selenium concentrations (mg/kg dry mass) observed in Canada geese eggs at reference and impacted locations (2009 and 2010 data combined).

AMPHIBIANS

AMERICAN TOAD, SPRING PEEPER, UPLAND CHORUS FROG

American toad captured at sample site. Photo credit: H. LeGrand

Spring peeper. Photo credit: Dr. J.D. Wilson.

Upland chorus frog. Photo credit: Dr. J.D. Wilson.

FROGS AND TOADS

- American toads (*Bufo americanus*), upland chorus frogs (*Pseudacris feriarum*), and spring peepers (*P. crucifer*) were collected at two potentially impacted sites near KIF, two unimpacted reference sites located north and upstream of KIF, and one nearby reference site in an adjacent county. Amphibians were allowed to void gut contents before being euthanized and frozen (≤10°C) for analysis.
- 2009 Collections: 34 spring peepers (23 reference, 11 impacted), 11 chorus frogs (11 impacted), and 31 American toads (10 reference, 21 impacted).
- 2010 Collections: 53 spring peepers (35 reference, 18 impacted), 35 chorus frogs (13 reference, 22 impacted), and 33 American toads (13 reference, 20 impacted).
- Selenium concentrations were compared between location type and species (Figure 5) using a 2-way ANOVA, WMW indicated that all reference sites were similar (p=0.629), so they were pooled for comparisons.
- Whole body Se concentrations were significantly higher in amphibians from impacted sites compared to reference sites (p<0.001). Toads had higher Se concentrations than the other two species (p=009). There was no significant interaction between location and species (p=0.672).
- Statistical comparisons of other elements have not yet been conducted, but several elements of interest are reported below in Tables 1 and 2.

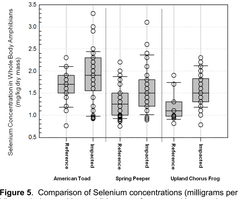

Figure 5. Comparison of Selenium concentrations (milligrams per kilogram) observed in amphibians at reference and impacted locations (2009 and 2010 data combined).

Table 1. Comparisons of Total Body Trace Element Concentrations (mg/kg dry mass) in Adult American Toads

Constituent	Reference[1] n=23	Impacted[2] n=41	Reference[3] n=2-6	Impacted[3] n=7-10
Arsenic	1.28 ± 0.33 (7)	1.14 ± 0.62 (25)	0.23 ± 0.05	1.58 ± 0.27
Barium	78.67 ± 5.58 (0)	80.65 ± 5.60 (0)	77.20 ± 11.25	133.64 ± 32.26
Cadmium	0.26 ± 0.034 (10)	0.30 ± 0.025 (23)	0.12 ± 0.02	0.27 ± 0.050
Copper	6.60 ± 0.53 (4)	7.19 ± 0.43 (8)	20.80 ± 1.67	29.50 ± 7.19
Lead	1.00 ± 0.15 (1)	0.93 ± 0.23 (13)	1.06 ± 0.09	0.70 ± 0.14
Selenium	1.65 ± 0.074 (0)	1.89 ± 0.089 (0)	2.10 ± 0.09	17.40 ± 3.32
Strontium	82.73 ± 7.39 (0)	103.88 ± 7.31 (0)	132.10 ± 13.92	386.70 ± 74.31
Zinc	125.08 ± 6.03 (0)	113.78 ± 2.71 (0)	194.30 ± 93.38	99.20 ± 5.82

Note: Concentrations for 2009 and 2010 combined, presented as mean ± 1 SE (number of samples below detection limit).
[1] Reference concentrations include data from Timberlake Pond, Dawson's Farm, and Rocky Top Farm.
[2] Impacted concentrations include data from the North Embayment and West Embayment.
[3] Literature values cited in Hopkins et al. (1998) for adult male southern toads collected during breeding around a coal ash settling pond.

Table 2. Comparisons of Total Body Trace Element Concentrations (mg/kg dry mass) in Adult Spring Peepers and Adult Chorus Frogs

	Spring Peepers		Upland Chorus Frogs	
Constituent	Reference[1] n=58	Impacted[2] n=29	Reference[1] n=13	Impacted[2] n=33
Arsenic	0.29 ± 0.015 (54)	0.26 ± 0.017 (27)	0.22 ± 0.0067 (8)	0.45 ± 0.055 (15)
Barium	48.87 ± 3.64 (0)	60.75 ± 3.63 (0)	53.80 ± 8.56 (0)	58.12 ± 3.33 (0)
Cadmium	0.21 ± 0.022 (34)	0.18 ± 0.023 (16)	0.079 ± 0.0078 (14)	0.20 ± 0.024 (6)
Copper	9.86 ± 0.77 (2)	15.02 ± 1.57 (0)	14.09 ± 2.62 (0)	12.75 ± 0.94 (0)
Lead	0.38 ± 0.041 (30)	0.69 ± 0.026 (13)	0.34 ± 0.052 (3)	0.35 ± 0.027 (16)
Selenium	1.30 ± 0.045 (0)	1.61 ± 0.097 (0)	1.18 ± 0.088 (0)	1.58 ± 0.065 (0)
Strontium	40.28 ± 2.37 (0)	58.34 ± 4.25 (0)	51.03 ± 5.07 (0)	58.11 ± 3.41 (0)
Zinc	98.73 ± 2.73 (0)	104.09 ± 3.62 (0)	153.81 ± 15.75 (0)	133.62 ± 6.26 (0)

Note: Concentrations for 2009 and 2010 combined, presented as mean ± 1 SE (number of samples below detection limit).
[1] Reference concentrations include data from Timberlake Pond, Dawson's Farm, and Rocky Top Farm.
[2] Impacted concentrations include data from the North Embayment and West Embayment.

REPTILES

SNAPPING TURTLE, COMMON MUSK, EASTERN SPINY SOFTSHELL

Snapping turtle captured on the upstream Tennessee River (reference location).

Musk turtle captured on the Emory River.

Eastern spiny softshell. Photo credit: http://www.paherps.com.

TURTLES

- Snapping turtles (*Chelydra serpentine*), musk turtles (*Sternotherus sp.*), eastern mud turtles (*Kinosternon subrubrum*), and eastern spiny softshell turtles (*Apalone spinifera spinifera*) were collected from potentially impacted sites around KIF and at the upstream TN River reference site. Blood, toenails and carapace (softshells only) were sampled and frozen (≤10°C) for analysis. Only blood has been analyzed at this point.
- 2009 Trapping (350 traps set): 26 musk/mud turtles (26 impacted), 22 snapping turtles (13 reference, 9 impacted), 582 non-target species collected.
- 2010 Trapping (1,347 traps set): 350 musk/mud turtles (51 reference, 299 impacted), 58 snapping turtles (29 reference, 29 impacted), 89 eastern spiny softshell turtles (11 reference, 78 impacted), 3,310 non-target species.
- Selenium concentrations in turtle blood were compared between locations and species (Figure 6, preliminary 2010 data only). Concentrations reported below detection limit (BDL) were graphed at the detection limit. Mean concentrations (± standard error) are presented in Table 3.
- Statistical comparisons of other elements have not yet been conducted. A total of 23 of 26 metals were detected in turtle blood. Al, Be, Cr, and Mo were below detection limits. Sb, As, B, Cd, Ag, Tl, and V were detected in less than 5% of samples.

Field biologists checking hoop turtle trap on TN River, baited with raw meat..

PRELIMINARY DATA

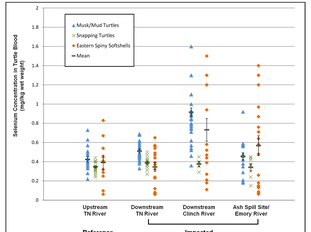

Figure 6. Selenium concentrations (mg/kg wet weight) observed in turtle blood at reference (Upstream TN River) and impacted locations at the site (2010 data).

Field biologist comparing eastern mud turtle (left) with musk turtle (right).

Field biologist collecting blood sample from snapping turtle.

Field biologist collecting toenail sample from softshell turtle.

Table 3. Comparisons of Selenium Concentrations (mg/kg wet weight) in Turtles

Location	Musk/Mud Turtles n	[Se][1]	Snapping Turtles n	[Se][1]	Eastern Spiny Softshells n	[Se][1]
Upstream TN River[2]	20	0.42 ± 0.03 (0)	21	0.35 ± 0.01 (0)	10	0.39 ± 0.08 (1)
Downstream TN River	22	0.51 ± 0.02 (0)	17	0.39 ± 0.01 (0)	18	0.35 ± 0.05 (3)
Clinch River	41	0.92 ± 0.04 (0)	5	0.38 ± 0.03 (0)	16	0.73 ± 0.12 (0)
Emory River	21	0.46 ± 0.04 (0)	8	0.34 ± 0.04 (0)	20	0.57 ± 0.10 (1)

[1] Selenium (Se) concentrations in 2010 (mg/kg wet weight), presented as mean ± 1 SE (number of samples below detection limit).
[2] Reference location.

DISCUSSION

Initial review of the results from the first two years of monitoring suggests that most species examined have experienced exposure to Se derived from coal ash. Comparisons of other trace elements are ongoing. Higher concentrations of Se were detected in heron eggs collected from impacted nests than those collected from reference nests; however, Se concentrations in osprey eggs were similar between locations. The differences in Se accumulation could be a result of variation in dietary composition and/or feeding-ecologies between these two piscivores (heron forage primarily in shallow waters and seasonally-exposed sediment compared to osprey which often capture prey items over open water). Selenium concentrations in Canada goose eggs collected from impacted nests were also higher than concentrations of reference eggs, probably because Canada geese are frequently observed eating grasses emerging from the ash.

Although both heron and geese from the impacted area maternally transferred more Se to their eggs than from reference areas, none of the observed Se concentrations were above those typically thought to reduce hatching success or induce deformities in birds (Janz et al. 2010). Interestingly, higher concentrations of Se were observed in insectivorous birds foraging near the site (see comparison poster on tree swallows). Taken together, our findings suggest that lower trophic level birds grazing directly on ash (geese) and those eating invertebrates that developed in the benthos (tree swallows) are at greater risk of exposure than piscivores. This may be related to a significant lag time between introduction of ash into the system and possible bioaccumulation in fish.

Whole body Se concentrations were low in amphibians. For comparison (Table 1), Hopkins et al. documented considerably higher concentrations of Se in toads breeding in a coal ash settling pond in South Carolina. Although statistical tests have not yet been performed, there is qualitative evidence (Table 1) that other elements such as Sr may also be accumulated in toads in the impacted area. Monitoring efforts will continue in the impacted area to determine whether observed patterns remain stable over time.

The preliminary evaluation of Se concentrations in turtle blood indicates higher levels in species whose diet is predominately comprised of benthic invertebrates (musk turtles and eastern spiny softshell), compared to species feeding at a higher trophic level (snapping turtles). These findings are consistent with the bird results, suggesting that up to this point, invertebrate prey represent a more direct trophic pathway of exposure to consumers than do fish prey. Full scale effects-based work will be launched in Spring 2011 to determine whether turtles maternally transfer Se to their eggs and whether reproductive success is affected by the Kingston ash spill.

REFERENCES

Hopkins, W.A., C.L. Rowe, and J.D. Congdon, 1999. Elevated Trace Element Concentrations and Standard Metabolic Rate in Banded Water Snakes (Nerodia fasciata) Exposed to Coal Combustion Wastes. *Environmental Toxicology and Chemistry.* 18(6): 1258-1263.

Hopkins, W.A., M.T. Mendonca, C.L. Rowe, J.D. Congdon. 1998. Elevated Trace Element Concentrations in Southern Toads, *Bufo terrestris*, Exposed to Coal Combustion Waste. *Archives of Environmental Contamination and Toxicology.* 35, 325-329.

Janz, D.M. et al. Chapter 6: Selenium Toxicity to Aquatic Organisms. CRC Press—Taylor & Francis Group, LLC, Boca Raton, FL, 141-231. 2010.

PROGRESS OF OTHER STUDIES

Tree swallow monitoring was initiated in 2009 and expanded in 2010. Results are presented concurrently in this session, including concentrations of trace elements in eggs and 15-day nestlings.

In addition, the University of Tennessee is completing an analysis of trace elements in raccoon tissues along the shoreline at the ash spill. Data collected in 2009 will be analyzed over the next several months and specimen collections are currently underway for the 2010 season. These samples will be analyzed over the next few months.

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