

# A Multi-phased Toxicity Study for Evaluating Potential Risks of Kingston Fossil Plant Fly Ash Exposures to Benthic and Aquatic Biota

R. Sherrard, N. Carriker — Tennessee Valley Authority, Kingston Ash Recovery Project, Kingston, TN

## ABSTRACT

TVA initiated multi-phase laboratory toxicity studies in March 2009 to evaluate potential risks to biota from exposure to fly ash from the Kingston ash release to the Emory River and subsequent dredging. The overall objective is to evaluate toxicity (survival, growth, reproduction) and metals bioaccumulation elicited by exposure of benthic and aquatic species to whole ash, elutriates, dredge plume water, and ash stilling pond effluent. Various acute and chronic test protocols were used in the first phase with (1) two 3.1-m Vibracore® ash composite samples collected March 17; (2) two 3.1-m Vibracore® ash composite samples collected June 11-12; and, (3) monthly Emory River dredge plume and stilling pond effluent samples collected April – June. Results from the March 17 ash composite samples indicated no appreciable bioaccumulation of metals in *Corbicula fluminea* exposures (28-d) to whole ash nor any toxic effects in exposures of *Ceriodaphnia dubia* (96-h), *Pimephales promelas* (96-h), or *Lampisilis siliquioidea* (10-d) to ash elutriates. *Lumbriculus variegatus* exposures (4-d) to whole ash showed no effects on survival, but worms did not burrow so bioaccumulation was not assessed. No effects on survival were noted for *L. siliquioidea* 5-d exposures to whole ash, but 10-d exposures to one of the whole ash samples did result in significant effects on survival relative to laboratory control sediment. *Hyalella azteca* exposures (10-d) to both whole ash samples indicated adverse effects on survival. No effects (survival, reproduction) were observed in 7-d chronic exposures with *C. dubia* to plume or stilling pond effluent samples collected April – June. Results with identical exposures to the April and May samples by *P. promelas* (survival, growth) were invalidated due to confirmed pathogen interference. *P. promelas* chronic tests with Ultraviolet-treated plume and stilling pond effluent samples collected in June resulted in no adverse effects. The second phase of testing (96-h *C. dubia* and *P. promelas*) involves monitoring of dredge plume and stilling pond effluent samples (August - present) in response to increased dredging rates. Plume test results (16 samples) have shown no effects on survival of either species. Stilling pond effluent tests (17 samples) have resulted in no effects on survival with one exception: exposure to a single sample resulted in decreased survival by *C. dubia*. Test results from the June 11-12 ash composite samples are currently being evaluated. A third phase of this study focusing on the bioavailability of metals in ash and evaluating resin-treatment of ash to provide a suitable reference control is underway.

## INTRODUCTION

- Prior to Kingston ash release, virtually no toxicological data existed for aquatic and benthic organism exposures to fly ash in a natural system
- TVA identified exposure-response scenarios that required early laboratory study
- Are benthic animals in direct contact with ash affected, and if so, how?
- Are benthic and aquatic animals affected by dredging, and if so, how?
- Phase 1 — characterize toxicity in the major routes of exposure (whole ash, dredge plume, dredge dewatering and treated water discharge)
- Phase 2 — more intensive monitoring of dredging in response to increasing dredging rates
- Phase 3 — focus on bioavailability of metals in whole ash

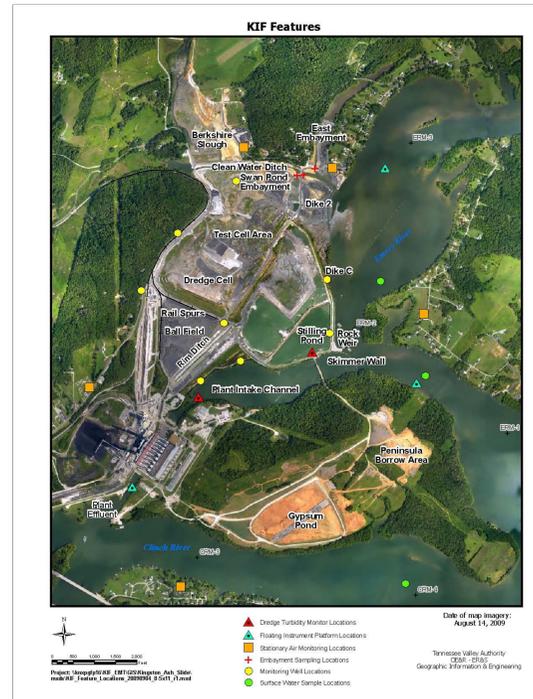
## OBJECTIVES

Comprehensively evaluate toxicity (survival, growth, reproduction) and metals bioaccumulation from exposure of benthic and aquatic species to:

- Whole ash
- Elutriates
- Dredge plume water
- Ash Stilling Pond effluent

## MATERIALS AND METHODS

- Multiple 3.1-m Vibracore® samples collected immediately upstream (Vb.1) and downstream (Vb.2) of dredging on March 17, composited into 2 discrete samples, individually homogenized, placed in 19-L plastic buckets and kept in cold storage (4°C) until use
- Multiple 3.1-m Vibracore® samples (Vb.3 & Vb.4) collected and composited June 11—12, homogenized, placed in 19-L plastic buckets and kept in cold storage (4°C) until use
- All whole ash and elutriate exposures conducted with Vibracore® ash composites that required re-homogenization upon each use
- Emory River dredge plumes visually located; Hydrolab used to delineate most turbid depth; peristaltic pump used to collect aqueous plume sample, held at 4°C until use
- 24-h composite stilling pond effluent samples collected with ISCO sampler, held at 4°C until use
- Reference control and dilution water collected from unaffected areas of Emory River
- Laboratory control sediment collected from Clinch River mile 189.0



## Toxicity Test Conditions and Acceptance Criteria

Test Organism	Whole Ash				Elutriate			Plume	Stilling Pond
	<i>H. azteca</i>	Freshwater Juvenile Mussel	<i>L. variegatus</i>	<i>C. fluminea</i>	<i>C. dubia</i>	<i>P. promelas</i>	Freshwater Juvenile Mussel	<i>C. dubia</i> <i>P. promelas</i>	<i>C. dubia</i> <i>P. promelas</i>
<b>Test Type</b>	static renewal (water only)	5-d no renewal 10-d renewal @ 6	static renewal (water only)	static renewal (water only)	static non-renewal	static non-renewal	static renewal	static renewal	static renewal
<b>Test Duration</b>	10 d	5 d and 10 d	4 d	28 d	96 h	96 h	10 d	Apr-Jun 7 d Aug-Oct 96 h	Apr-Jun 7 d Aug-Oct 96 h
<b>Temperature</b>	23 ± 1° C	24 ± 1° C	23 ± 1° C	Mean 20 ± 1° C Inst. 20 ± 3° C	25 ± 1° C	25 ± 1° C	24 ± 1° C	25 ± 1° C	25 ± 1° C
<b>Photoperiod</b>	16 h light 8 h dark	24 h dark	16 h light 8 h dark	16 h light 8 h dark	16 h light 8 h dark	16 h light 8 h dark	24 h dark	16 h light 8 h dark	16 h light 8 h dark
<b>Chamber Size</b>	300 mL	5-cm cylinder in 250 mL dish	300 mL	16.4 L	30 mL (min)	250 mL (min)	5-cm cylinder in 250 mL dish	30 mL Cd 250 mL Pp	30 mL Cd 250 mL Pp
<b>Sediment Volume</b>	100 mL	20 mL	100 mL	250 g sediment / g wet tissue	N/A	N/A	20 mL Lab control sed.	N/A	N/A
<b>Overlying Water Volume</b>	175 mL	200 mL	175 mL	15 L	15 mL (min)	200 mL (min)	200 mL (min)	15 mL Cd 200 mL Pp	15 mL Cd 200 mL Pp
<b>Water Renewal</b>	2 vol. additions/d	5-d no renewal 10-d renewal @ 6	2 vol. additions/d	50% 3x per wk	None	None	Renewal @ 6 Sed & Water	Apr-Jun daily Aug-Oct @ 48h	Apr-Jun daily Aug-Oct @ 48h
<b>Age</b>	7- 14 d	<8 d	Adults	0.5 - 1.5 g wet tis- sue	<24 h	<24 h	<8 d	<24 h	<24 h
<b>Organisms/Rep</b>	10	10	10	Total 150 g wet tissue	5	10	10	Apr-JunCd1Pp10 Aug-OctCd5Pp10	Apr-JunCd1Pp10 Aug-OctCd5Pp10
<b>No. Replicates</b>	8	5	4	5	5	5	5	Apr-JunCd10Pp4 Aug-OctCd5Pp5	Apr-JunCd10Pp4 Aug-OctCd5Pp5
<b>Feeding</b>	YCT 1mL/d	6 mL algal conc. @ 0, 3, 6, 9 d	None	None	YCT + Algae 0 and 48 h	<i>Artemia</i> 0 and 48 h	6 mL algal conc. @ 0, 3, 6, 9 d	Apr-Jun daily Aug-Oct @ 48h	Apr-Jun daily Aug-Oct @ 48h
<b>Aeration</b>	If <2.5 mg/L	If <5 mg/L	None	Moderate, as needed	None	If <4 mg/L	If <5 mg/L	Cd None Pp if <4 mg/L	Cd None Pp if <4 mg/L
<b>Test Concentrations</b>	N/A	N/A	N/A	N/A	No-cen:100-0%,5 Cent: 100%	No-cen:100-0%,5 Cent: 100%	No-cen:100-0%,5 Cent: 100%	Apr-Jun 0 & 100% Aug-Oct 100-0,5	Apr-Jun 0 & 100% Aug-Oct 100-0,5
<b>Endpoints</b>	Survival, Growth	Survival (foot & ciliary action)	Burrowing Survival	Bioaccumulation	Survival	Survival	Survival (foot & ciliary action)	Apr-Jun SR&G Aug-Oct Survival	Apr-Jun SR&G Aug-Oct Survival
<b>Acceptability Criteria</b>	Survival ≥ 80% Meas. Growth	Survival ≥ 90%	Survival ≥ 90%	Adequate Mass	Survival ≥ 90%	Survival ≥ 90%	Survival ≥ 90%	EPA Methods	EPA Methods

## RESULTS - PHASE 1

### Whole Ash Exposure — *Hyalella azteca*

- Significant effects (survival, growth, and biomass) relative to laboratory control sediment
- Vb.1 survival = 11.3%; Vb.2 survival = 25%; Lab control sediment survival = 89%

### Whole Ash Exposure — 5-d Freshwater Juvenile Mussel

- No effect on survival relative to laboratory control sediment
- Vb.1, Vb.2, & Lab control sediment survival = 100%

### Whole Ash Exposure — 10-d Freshwater Juvenile Mussel

- Vb.1: Significant effect (survival) relative to laboratory control sediment
- Vb.1 survival = 48%; Vb.2 survival = 96%; Lab control sediment survival = 92%

### Whole Ash Exposure — *Lumbriculus variegatus*

- No effect on survival relative to laboratory control sediment; Vb.1 survival = 97.5%; Vb.2 survival = 100%; Lab control sediment survival = 100%
- Significant effect (burrowing) relative to laboratory control sediment; Vb.1 burrowing = 0%; Vb.2 burrowing = 60%; Lab control sediment burrowing = 100%
- 28-day bioaccumulation test is not appropriate for this species

### Whole Ash Exposure — *Corbicula fluminea* Bioaccumulation

- No apparent effects on survival
- Bioaccumulation Factors (BAFs) negligible; maximum BAF among metals was for zinc (mean = 0.343, range = 0.146 - 0.591 kg sediment (dry wt) / kg body weight (wet wt))

### Elutriate Exposure — *Ceriodaphnia dubia*

- No effect on survival relative to Emory River control
- Vb.1 100%-elutriate survival = 100%; Vb.2 100%-elutriate survival = 92%; Emory River control survival = 100%

### Elutriate Exposure — *Pimephales promelas*

- No effect on survival relative to Emory River control
- Vb.1 100%-elutriate survival = 84%; Vb.2 100%-elutriate survival = 96%; Emory River control survival = 98%

### Elutriate Exposure — 10-d Freshwater Juvenile Mussel

- No effect on survival relative to Emory River control
- Vb.1 100%-elutriate survival = 98%; Vb.2 100%-elutriate survival = 94%; Emory River control survival = 96%

### Plume Exposure — *Ceriodaphnia dubia* and *Pimephales promelas*

- No effect on *C. dubia* survival or reproduction relative to Emory River control with April through June samples (3 total)
- April & May tests with *P. promelas* invalid due to pathogen interference; no effect on survival or growth with June UV-treated plume water relative to UV-treated Emory River control

### Stilling Pond Effluent Exposure — *C. dubia* and *P. promelas*

- No effect on *C. dubia* survival or reproduction relative to Emory River control with April through June samples (3 total)
- April & May tests with *P. promelas* invalid due to pathogen interference; no effect on survival or growth with June UV-treated effluent relative to UV-treated Emory River control

Vb.3 and Vb.4 test results await final evaluation to be conducted upon completion of Phase 3 bioavailability study.

## RESULTS - PHASE 2

### Plume Exposure — *Ceriodaphnia dubia* and *Pimephales promelas*

- No effect on *C. dubia* or *P. promelas* survival relative to Emory River control with August through February 2010 samples (16 total)

### Stilling Pond Effluent Exposure — *C. dubia* and *P. promelas*

- No effect on *P. promelas* survival relative to Emory River control with August through February 2010 samples (17 total)
- In *C. dubia* exposures, only 1 of 17 samples resulted in decreased survival

## DISCUSSION

- No definitive conclusions can be drawn from the few results that have indicated statistically significant effects on laboratory organisms
- Results from tests with Vb.1 and Vb.2 were statistically evaluated relative to a natural depositional sediment located 185 river miles upstream of Kingston Fossil Plant, as no reference sediment was available near the site
- Laboratory staff reported no observed test organism behavior which indicated stress or avoidance of ash

## PHASE 3 - NEXT STEPS

- Fly ash in Emory River possesses unique physical properties that present unfamiliar challenges to sediment toxicologists
- Within a day or two of collecting whole ash and storing in containers, the solids settle out into a very compacted state while porewater separates and surfaces; this same behavior occurs in toxicity test chambers
- With each use, the compacted ash and separated porewater must be homogenized to ensure consistent exposures



- The key to evaluating ash toxicity appears to lie in the development of a suitable reference control material that has similar properties
- To date, TVA has (1) attempted to formulate a reference sediment; (2) considered "ash washing" with acids followed by porewater reconstitution; and (3) treated ash with resins to decrease the bioavailable fraction of metals in porewater
- Coupled with follow-up studies with resin treatment of ash, TVA will focus on experimental approaches to better understand bioavailability of metals in ash

Experimental Design of Ash & Porewater Bioavailability Study with *H. azteca* and *C. dubia*

