

Final Report

Ceriodaphnia dubia **Whole Sediment Survival and Reproduction Toxicity Test Results**

TVA, Kingston Monitoring and Analysis Project Clinch River Sediment Sample CRM3.0

Prepared for:

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EXECUTIVE SUMMARY

A whole sediment toxicity test was conducted by Environmental Enterprises USA, Inc. (EE USA) to determine potential toxicity of a Clinch River site sediment sample to *Ceriodaphnia dubia* neonates. Three samples were used in this test: Clinch River site sediment identified as BULKSED-CRM3.0-EEUSA (CRM3.0); Clinch River reference sediment identified as BULKSED-CLINCHREFERENCE-EEUSA (CRS); and Clinch River water identified as BULKSW-CRM7.0-EEUSA (CRW). Several dilutions of CRM3.0 prepared with CRS were tested with four concurrent controls. A moderately hard synthetic freshwater (MHSW) only control was included to assess test organism health. Separate laboratory control sediment (LCS) exposures with either MHSW or CRW were included to assess test acceptability requirements.

C. dubia survival and reproduction in the CRM3.0 treatments were compared to survival and reproduction in a CRS control with CRW. Survival and reproduction of *C. dubia* neonates in the CRM3.0 treatments were not reduced when compared to survival and reproduction in the CRS control. Test results are shown in Table 1.

Table 1. *Ceriodaphnia dubia* Chronic Survival and Reproduction Test Results for CRM3.0.

| SURVIVAL | REPRODUCTION |
|--------------------------------|--------------------------------|
| NOEC / LOEC = 100.0 / > 100.0% | NOEC / LOEC = 100.0 / > 100.0% |
| IC_{25} > 100.0% | IC_{25} > 100.0% |

INTRODUCTION

EE USA was contracted by Tennessee Valley Authority (TVA) to complete whole sediment toxicity tests with Clinch River sediment and water samples using *C. dubia* neonates. The project is described in TVA's Sediment Toxicity Study Design [1]. Site sediment samples were collected from eight representative locations on the Clinch River. Clinch River reference sediment samples and river water samples were collected upstream of the site sediment locations. The two reference sediment samples were mixed together 50:50 and homogenized in the field. TVA's contractors, Jacobs Engineering and Restoration Services, Inc., coordinated sample collection in the field and delivery of the samples to EE USA. The samples were delivered to EE USA on February 12, 2011, on ice and with custody seals intact (Appendix D).

This test, which was performed on one (CRM3.0) of the eight sediment samples obtained from the Clinch River, was conducted in accordance with American Society for Testing and Materials (ASTM) [2] and U. S. EPA [3] toxicity testing methods. Test organisms were cultured at EE USA and were 16.0 to 21.0 hours old when this test was initiated. Ten replicates of each control treatment and six CRM3.0 concentrations were prepared the day before the test was initiated. CRM3.0 dilutions were prepared with CRS. CRM3.0 concentrations tested were 10.0, 20.0, 40.0, 60.0, 80.0, and 100.0%. A portion of the overlying water in each replicate was replaced daily. This test was initiated February 17, 2011, at 1500 and completed February 23 at 1525.

MATERIALS AND METHODS

C. dubia was cultured and maintained in MHSW at $25 \pm 1^{\circ}\text{C}$. Test organisms were selected from adults producing at least ten in their third or subsequent brood. Only ten neonates from any one adult were used so that one replicate in each treatment was populated with a neonate from the same adult. Test organisms were fed *Selenastrum capricornutum* (SCAP) and Yeast-Cerophyl-digested Tetramin (YCT) daily at the rate of 0.1 mL each per 15 mL of water.

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On February 12, 2011, the CRM3.0, CRS, and CRW samples were delivered to EE USA and stored at 0.1 to 6°C (Appendix D & Table 2). Two 1-liter containers each of CRM3.0 and CRS were put into separate mixing bowls and large bark pieces, rocks, and leaves were removed with forceps. Each 2-liter sediment sample was mixed for approximately five minutes with a KitchenAid Model KHM7TGCS hand-held mixer set at position "3", 580 ± 5 rpm. On February 16th (Day -1), the density of each sediment, LCS, CRS, and CRM3.0, was measured and the test treatments were prepared (Appendix A, page 1). Eight-dram shell vials were used as test chambers. The vials were washed with soap and water and rinsed with acetone, 10% HCl, deionized water, and MHSW prior to being used as test chambers. Test chambers were labeled with test concentration, replicate, and EE USA's project number. Dilutions of CRM3.0 were made with CRS according to the calculations on page 1 of Appendix A. For each treatment, 400 mL of sediment was prepared, homogenized, and then 5 mL were transferred to 11 test replicates. The 11th replicate of each treatment was used for water quality only.

Table 2. Clinch River Site Sediment, Reference Sediment, and Water Samples.

| TVA Sample ID | EE USA Sample ID | Date Collected | Date Received |
|--|------------------|-----------------------------|-----------------------------|
| BULKSED-CRM3.0-EEUSA Composite Sample, Site Sediment (CRM3.0) | E-091-11 | February 8, 2011 @ 0921 | February 12, 2011 @ 1240 |
| BULKSED-CLINCHREFERENCE-EEUSA Composite Sample, Clinch Reference Sediment (CRS) | E-095-11 | February 9, 2011 @ 0940 | |
| BULKSW-CRM7.0-EEUSA Grab Sample, Clinch River Water (CRW) | E-086-11 | February 11, 2011 @ 1057 | |

SCAP and YCT were added to aliquots of the overlying waters, MHSW and CRW; 6.0 mL each of SCAP and YCT was added to 900 mL of MHSW and 12 mL each of SCAP and YCT was added to 1800 mL CRW. The MHSW and CRW aliquots were warmed up to 25 ± 1°C. Twenty mL of MHSW were transferred to 11 test replicates of the MHSW only control. LCS, No. 5 sand supplied by EE USA and wetted to saturation with MHSW, was homogenized with a stainless steel spoon and five mL were transferred to 22 test replicates. Eleven LCS + MHSW replicates received 20 mL MHSW and 11 LCS + CRW replicates received 20 mL CRW. Twenty mL of CRW were added to each replicate of the CRS and CRM3.0 treatments.

After dispensing the sediments and water, the test chambers were placed in an environmental chamber at 25 ± 1°C with a photoperiod of 16 hours light and 8 hours dark. The test was not aerated. Initial water quality parameters (dissolved oxygen (DO) and temperature) were measured daily in the 11th replicate of each treatment. At the end of each 24-hour exposure period, prior to renewal, the ending DO and temperature in each treatment were recorded (Appendix A, pages 9 – 12 & Table 3). Alkalinity, hardness, conductivity, pH, DO, total residual chlorine, and ammonia were measured in CRW and each batch of MHSW (Appendix A, page 2 & Table 4).

**Table 3. Initial and Final Temperature and Dissolved Oxygen Data for Each Treatment:
Mean, Minimum, and Maximum.**

| Water Quality Summary for Test Exposures February 17 – 23, 2011 | | | | | | | Mean | | | |
|--|-----------------|------|-------|------|------------------------|-----|-------|-----|-----|-----|
| % Sample | Temperature, °C | | | | Dissolved Oxygen, mg/L | | | | Min | Max |
| | Initial | | Final | | Initial | | Final | | | |
| MHSW | 25.0 | | 24.9 | | 7.4 | | 7.5 | | 7.3 | 7.7 |
| | 24.7 | 25.4 | 24.4 | 25.0 | 7.1 | 7.5 | 7.3 | 7.7 | | |
| LCS + MHSW | 25.0 | | 24.9 | | 7.4 | | 7.4 | | 7.2 | 7.5 |
| | 24.4 | 25.5 | 24.5 | 25.1 | 7.1 | 7.6 | 7.2 | 7.5 | | |
| LCS + CRW | 25.0 | | 24.9 | | 7.3 | | 7.2 | | 7.0 | 7.4 |
| | 24.5 | 25.5 | 24.5 | 25.2 | 6.8 | 7.5 | 7.0 | 7.4 | | |
| CRS + CRW | 25.0 | | 24.9 | | 6.2 | | 6.0 | | 5.8 | 6.5 |
| | 24.7 | 25.4 | 24.4 | 25.1 | 6.0 | 6.5 | 5.8 | 6.5 | | |
| 10.0 | 25.0 | | 24.9 | | 6.1 | | 6.0 | | 5.6 | 6.4 |
| | 24.4 | 25.5 | 24.2 | 25.0 | 5.8 | 6.4 | 5.6 | 6.4 | | |
| 20.0 | 24.9 | | 25.0 | | 6.0 | | 6.1 | | 5.9 | 6.3 |
| | 24.4 | 25.6 | 24.4 | 25.3 | 5.6 | 6.2 | 5.9 | 6.3 | | |
| 40.0 | 24.9 | | 24.8 | | 6.0 | | 6.0 | | 5.7 | 6.1 |
| | 24.7 | 25.4 | 24.3 | 25.0 | 5.5 | 6.3 | 5.7 | 6.1 | | |
| 60.0 | 25.0 | | 24.9 | | 6.0 | | 5.9 | | 5.5 | 6.3 |
| | 24.6 | 25.3 | 24.3 | 25.2 | 5.6 | 6.3 | 5.5 | 6.3 | | |
| 80.0 | 25.0 | | 24.8 | | 6.0 | | 5.9 | | 5.6 | 6.1 |
| | 24.6 | 25.5 | 24.5 | 25.2 | 5.8 | 6.2 | 5.6 | 6.1 | | |
| 100.0 | 24.9 | | 24.9 | | 5.9 | | 5.8 | | 5.6 | 6.0 |
| | 24.6 | 25.3 | 24.5 | 25.0 | 5.8 | 6.0 | 5.6 | 6.0 | | |

Table 4. Water Quality Data for CRW and Each Batch of MHSW.

| | CRW | MHSW | MHSW | MHSW |
|------------------------|---------------------|---------------------------------|------------------------|------------------------|
| Collected | 2/11/2011 | | | |
| Batch Number | BULKSW-CRM7.0-EEUSA | FW-016-11 ¹ | FW-018-11 ² | FW-019-11 ³ |
| Alkalinity, mg/l | 140 | 60 | 76 | 64 |
| Hardness, mg/l | 140 | 100 | 84 | 96 |
| Conductivity, µmhos/cm | 313 | 299 | 320 | 312 |
| pH, su | 8.2 | 8.1 | 8.1 | 8.2 |
| Dissolved Oxygen, mg/l | 8.3 | 8.3 | 8.3 | 8.3 |
| TRC, mg/l | 0.04 | 0.0 | 0.0 | 0.0 |
| Total Ammonia, mg/l | 0.04 | < 0.02 | < 0.02 | < 0.02 |
| | | ¹ used 02/16-18/2011 | | |
| | | ² used 02/19-21/2011 | | |
| | | ³ used 02/22/2011 | | |

The test was initiated February 17th (Day 0) after 15 mL of water were removed from each replicate of each treatment and replaced with water into which proper aliquots of food had been added. One *C. dubia* neonate was transferred to each replicate, and then the test chambers were placed in an environmental chamber. On Days 1-5, the test exposures were renewed as follows:

1. The *C. dubia* in each replicate and approximately 5 mL of the water in the replicate were transferred to a 30-mL disposable plastic cup.
2. Additional water equivalent to a total of 15 mL was removed from the replicate.
3. 15 mL of fresh MHSW or CRW as appropriate was transferred to the replicate.
4. The *C. dubia* was transferred back to the replicate.

Water was removed from and added to each replicate with a 25-mL pipette. *C. dubia* were transferred with disposable 3.5-mL transfer pipettes. Survival was recorded daily (Appendix A, pages 3 - 8). Reproduction was also recorded and newly produced neonates discarded before renewal. The test was terminated after six days, after \geq 60% of each set of control organisms released their third brood.

The endpoints for the chronic test were survival and neonate production. The test acceptability criteria were 80% or greater survival in the LCS + MHSW control and an average of 15 or more young per surviving female in the control solutions (60% of surviving control females must produce three broods).

The response used in the statistical analysis of the survival data was the proportion of test organisms surviving in each treatment chamber after six days. Fisher's Exact test was used to test for a significant difference between survival in the CRS + CRW control and each CRM3.0 concentration. The response used in the reproduction data analysis was the total number of neonates produced per replicate. Reproduction data were tested for normal distribution and homogeneity of variance using the Kolmogorov D and Bartlett's tests, respectively. Reproduction data were not normally distributed; thus, the data were evaluated by Steel's Many-One Rank Test. The statistical tests were performed using ToxCalc Version 5.0.32 at a probability level of 0.05 [4].

Sensitivity of test organisms to a known toxicant was determined by performing a chronic Standard Reference Toxicant (SRT) test, CD1102, with potassium chloride (Sigma Chemical, Lot 099K0202). The most recent SRT test was initiated on February 10, 2011, with less than 24-hour-old *C. dubia* neonates.

RESULTS AND DISCUSSION

The control *C. dubia* met the test acceptability criteria of 80% or greater survival and an average of 15 or more young per surviving female in the LCS + MHSW control solution. One hundred percent survival occurred in the LCS + MHSW control. All (100%) of the control females produced three broods; the mean brood size was 22.8.

The No Observed Effect Concentration (NOEC) for survival was 100% CRM3.0. The Lowest Observed Effect Concentration (LOEC) was > 100.0% CRM3.0. The IC₂₅, a point estimate of the concentration that causes a 25% reduction in survival was > 100.0% CRM3.0 (Appendix B, page 1 & Table 5).

The NOEC for reproduction was 100.0% CRM3.0. The LOEC was > 100.0% CRM3.0. The Minimum Significant Difference percent for this reproduction data set was 20.4% (Appendix B, page 2). The IC₂₅, a point estimate of the concentration that causes a 25% reduction in reproduction was > 100.0% CRM3.0 (Appendix B, page 2 & Table 5).

Table 5. Summary of Percent Survival, Mean Reproduction, and Survival and Reproduction NOECs, LOECs, and IC₂₅s for CRM3.0.

| | LCS + CRW | CRS + CRW | 10% CRM3.0 | 20% CRM3.0 | 40% CRM3.0 | 60% CRM3.0 | 80% CRM3.0 | 100% CRM3.0 |
|--------------------------|-------------|-----------|---------------|------------|------------------------|------------|------------|-------------|
| % Survival | 100 | 100 | 100 | 100 | 100 | 100 | 100 | 100 |
| Mean Reproduction | 28.3 | 31.9 | 36.8 | 35.2 | 33.1 | 32.8 | 29.6 | 27.9 |
| | NOEC | | LOEC | | IC₂₅ | | | |
| Survival | 100% CRM3.0 | | > 100% CRM3.0 | | > 100% CRM3.0 | | | |
| Reproduction | 100% CRM3.0 | | > 100% CRM3.0 | | > 100% CRM3.0 | | | |

In summary, *C. dubia* survival and reproduction were not significantly reduced in any control or CRM3.0 treatment. Survival and reproduction statistical data for the MHSW only, LCS + MHSW, and LCS + CRW controls are presented on pages 3 and 4 of Appendix B.

The neonates used in the potassium chloride SRT met all of the quality control test parameters. The following SRT control charts are given in Appendix C:

- Survival IC₂₅ with ± 2 SD Control Limits
- Survival IC₂₅ %CV with 75th and 90th Percentile Warning Limits
- Survival PMSD
- Reproduction IC₂₅ with ± SD Control Limits
- Reproduction IC₂₅ %CV with 75th and 90th Percentile Warning Limits
- Control Reproduction with Lower Limit
- Control Reproduction %CV with TVA Limit
- Reproduction PMSD

REFERENCES

1. Tennessee Valley Authority. 2011. Kingston Monitoring and Analysis Project Non-Time-Critical Removal Action Sampling and Analysis Plan Sediment Toxicity Study Design. Chattanooga, TN.
2. American Society for Testing and Materials. 2005. Standard Test Method for Measuring the Toxicity of Sediment-Associated Contaminants with Freshwater Invertebrates. Annex A2. Guidance for Conducting Sediment Toxicity Tests with *D. magna* and *C. dubia*. E 1706-05. West Conshohocken, PA.
3. U.S. Environmental Protection Agency. 2002. Short-term methods for estimating the chronic toxicity of effluents and receiving waters to freshwater organisms, 4th ed. EPA-821-R-02-013. Office of Water, Washington, DC.
4. Tidepool Scientific Software. 2007. ToxCalc™ Toxicity Data Analysis Software. Version 5.0.32. McKinleyville, CA.

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REPORT TEST REVIEW

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APPENDIX A

Cladoceran, *Ceriodaphnia dubia***Whole Sediment Survival and Reproduction Test**

ASTM E 1706 – 05, Standard Test Method for Measuring the Toxicity of Sediment-Associated Contaminants with Freshwater Invertebrates: A2.
 GUIDANCE FOR CONDUCTING SEDIMENT TOXICITY TESTS WITH
DAPHNIA MAGNA (*D. magna*) AND *CERIODAPHNIA DUBIA* (*C. dubia*)

TVA, Kingston Monitoring and Analysis Project
CRM 3.0 Sediment & CRS Reference Sediment

| | Density | | |
|---------|---------------|---------------|---------------|
| | LCS | CRS | CRM 3.0 |
| 1 | 72.58 g/40 ml | 62.92 g/40 ml | 51.17 g/40 ml |
| 2 | 72.97 g/40 ml | 63.34 g/40 ml | 51.45 g/40 ml |
| 3 | 72.84 g/40 ml | 63.36 g/40 ml | 51.06 g/40 ml |
| MEAN | 72.80 g/40 ml | 63.21 g/40 ml | 51.23 g/40 ml |
| g/ml | 1.82 | 1.58 | 1.28 |
| g/5 ml | 9.10 | 7.90 | 6.40 |
| Initial | Dup SG | Dup SG | Dup SG |

2/6/11
 Balance ID N1
 Dup 3/9/11

Test Concentrations, % CRM 3.0

| Ceriodaphnia dubia | Total Sediment Vol./ Conc., ml | CRM 3.0 ml / gram | CRS ml / gram | LCS ml / gram | grams sediment/ replicate | CRW/rep | | MHSW/rep | |
|-----------------------|---|----------------------|------------------|------------------|---------------------------------|----------|---------------|----------|---------------|
| | | | | | | Day 0 | Days 1 - 6 | Day 0 | Days 1 - 6 |
| 100.0% | 400 | 400 / 512.3 | 0 | 0 | 6.40 | 20 | 15 | 0 | 0 |
| 80.0% | 400 | 320 / 409.6 | 80 / 126.4 | 0 | 6.70 | 20 | 15 | 0 | 0 |
| 60.0% | 400 | 240 / 307.4 | 160 / 252.8 | 0 | 7.00 | 20 | 15 | 0 | 0 |
| 40.0% | 400 | 160 / 204.9 | 240 / 379.3 | 0 | 7.30 | 20 | 15 | 0 | 0 |
| 20.0% | 400 | 80 / 102.5 | 320 / 505.7 | 0 | 7.60 | 20 | 15 | 0 | 0 |
| 10.0% | 400 | 40 / 51.2 | 360 / 568.9 | 0 | 7.75 | 20 | 15 | 0 | 0 |
| CRS w CRW | 400 | 0 | 400 / 632.1 | 0 | 7.90 | 20 | 15 | 0 | 0 |
| LCS w CRW | 400 | 0 | 0 | 400 / 728.0 | 9.10 | 20 | 15 | 0 | 0 |
| LCS w MHSW | 400 | 0 | 0 | 400 / 728.0 | 9.10 | 0 | 0 | 20 | 15 |
| MHSW | n/a | 0 | 0 | 0 | n/a | 0 | 0 | 20 | 15 |

Data pages & Calculations by: Patricia QA/QC Check by: Jennifer Griffith

Overlying waters + sediments dispensed on 2/16/11, Day -1.

Dup 3/9/11

MHSW = Moderately Hard Synthetic Freshwater
 CRW = Clinch River Reference Water

| | MHSW | MHSW | MHSW | CRW | Meter # |
|--------------|------------|------------|------------|----------------------|---------|
| Date | 02/16/2011 | 2/19/2011 | 2/22/2011 | 2/16/2011 | III |
| Batch # | FW-016-11 | FW-018-11 | FW-019-11 | Delivered 02/12/2011 | |
| Alkalinity | 60 | 76 | 64 | 140 | |
| Hardness | 100 | 84 | 96 | 140 | |
| Conductivity | 299 | 320 | 312 | 313 | A46 |
| pH | 8.154 | 8.154 | 8.254 | 8.254 | Q8 |
| DO | 8.3 mg/L | 8.3 mg/L | 8.3 mg/L | 8.3 mg/L | 57 |
| TRC | 0.0 mg/L | 0.0 mg/L | 0.0 mg/L | 0.04 mg/L | A27 |
| Ammonia | <0.02 mg/L | <0.02 mg/L | <0.02 mg/L | 0.04 mg/L | |
| Initial | 0.00 | 0.00 | 0.00 | 0.00 | |

TRC: mg/l Alkalinity: mg/l as CaCO₃ Conductivity: µS/cm Hardness: mg/l as CaCO₃

Comments: CRS sandy (w) some bark and a few leaves.

CRM 3.0 very clean. DWD 2/16/11

Cladoceran, Ceriodaphnia dubia**Whole Sediment Survival and Reproduction Test**

ASTM E 1706 – 05, Standard Test Method for Measuring the Toxicity of Sediment-Associated Contaminants with Freshwater Invertebrates:
 A2. GUIDANCE FOR CONDUCTING SEDIMENT TOXICITY TESTS WITH
 DAPHNIA MAGNA (D. magna) AND CERIODAPHNIA DUBIA (C. dubia)

TVA, Kingston Monitoring and Analysis Project
CRM 3.0 Sediment & CRS Reference Sediment
Test Organisms Age: 16.0 - 21.0 Hours OldTest Organisms Source: EE USA Test Initiation At: 1500 on 12/11/2011Counted by: David L. Daniel QC/QA by: J. S.Loaded by: David L. Daniel Organism Lot # CDD2111-04Exposure Chamber: 8 dram vials. Feeding: 0.1 ml S. capricornutum (Lot # S1-11) &
0.1 ml YCT (Lot # Y2-11) / 15 ml.**C. dubia Daily Survival & Reproduction Data****Treatment: MHSW only.**

| REP | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | % Sur. | No. of Neonates Per Day | Tech |
|-----|--------------------------------------|----|----|----|----|----|----|----|----|----|--------|-------------------------|--------|
| 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | /// | /// | D40 |
| 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | D40 |
| 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | D40 |
| 3 | 5 | 6 | 6 | 6 | 5 | 6 | 6 | 5 | 6 | 4 | 100 | 55 | D41 |
| 4 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | D40 |
| 5 | 8 | 8 | 8 | 5 | 9 | 7 | 8 | 8 | 6 | 6 | 100 | 73 | D40 |
| 6 | 12 | 10 | 10 | 8 | 10 | 11 | 10 | 11 | 10 | 9 | 100 | 101 | D40 |
| 7 | | | | | | | | | | | | | |
| | 3rd Brood Reproduction Per Replicate | | | | | | | | | | Mean | CV % | ////// |
| | 25 | 24 | 24 | 19 | 24 | 24 | 24 | 24 | 22 | 19 | 22.9 | 4.53 | |

Comments:

C. dubia Daily Survival & Reproduction Data Cont.**Treatment: LCS w MHSW.**

| | REP | 11 | 12 | 13 | 14 | 15 | 16 | 17 | 18 | 19 | 20 | % Sur. | No. of Neonates Per Day | Tech |
|---|--------------------------------------|----|----|----|----|----|----|----|----|----|------|--------|-------------------------|--------|
| | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | /// | /// | 040 |
| 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | 040 |
| 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | 040 |
| 3 | S | 5 | 6 | 5 | 6 | 6 | 6 | 4 | 6 | 5 | 5 | 100 | 54 | 040 |
| 4 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 2 | 0 | 0 | 0 | 100 | 2 | 040 |
| 5 | 6 | 8 | 9 | 6 | 8 | 8 | 6 | 10 | 8 | 9 | 100 | 78 | 040 | |
| 6 | 10 | 7 | 11 | 11 | 12 | 10 | 8 | 0 | 12 | 13 | 100 | 94 | | |
| 7 | | | | | | | | | | | | | | |
| | 3rd Brood Reproduction Per Replicate | | | | | | | | | | | Mean | CV % | ////// |
| | 21 | 20 | 26 | 22 | 26 | 24 | 20 | 16 | 26 | 27 | 22.8 | 15.7 | | |

Treatment: LCS w CRW.

| | REP | 21 | 22 | 23 | 24 | 25 | 26 | 27 | 28 | 29 | 30 | % Sur. | No. of Neonates Per Day | Tech |
|---|--------------------------------------|----|----|----|----|----|----|----|----|----|------|--------|-------------------------|--------|
| | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | /// | /// | 040 |
| 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | 040 |
| 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | 040 |
| 3 | S | 6 | 6 | 6 | 6 | 6 | 5 | 5 | 6 | 6 | 6 | 100 | 62 | 040 |
| 4 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | 040 |
| 5 | 11 | 10 | 12 | 10 | 13 | 9 | 9 | 7 | 10 | 8 | 100 | 99 | 040 | |
| 6 | 10 | 16 | 0 | 15 | 18 | 15 | 13 | 14 | 12 | 14 | 100 | 127 | 040 | |
| 7 | | | | | | | | | | | | | | |
| | 3rd Brood Reproduction Per Replicate | | | | | | | | | | | Mean | CV % | ////// |
| | 26 | 32 | 18 | 31 | 37 | 30 | 27 | 26 | 28 | 28 | 29.3 | 17.4 | | |

Comments: (A) 57 wrong data JC-022811

28.3
wrong data
JC-022811

C. dubia Daily Survival & Reproduction Data Cont.

| | REP | Treatment: CRS w CRW. | | | | | | | | | | | | % Sur. | No. of Neonates Per Day | Tech |
|--|-----|--------------------------------------|----|----|----|----|----|----|----|----|----|--------------------------------------|------|--------|-------------------------|--------|
| | | 31 | 32 | 33 | 34 | 35 | 36 | 37 | 38 | 39 | 40 | 3rd Brood Reproduction Per Replicate | Mean | CV % | | |
| | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 111 | 111 | 0.00 | | |
| | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | 0.00 | | |
| | 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | 0.00 | | |
| | 3 | 5 | 6 | 6 | 6 | 5 | 7 | 6 | 6 | 5 | 6 | 100 | 58 | 0.00 | | |
| | 4 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | 0.00 | | |
| | 5 | 12 | 10 | 10 | 11 | 12 | 14 | 11 | 13 | 13 | 11 | 100 | 117 | 0.00 | | |
| | 6 | 18 | 15 | 14 | 8 | 17 | 16 | 14 | 5 | 21 | 16 | 100 | 144 | 0.00 | | |
| | 7 | | | | | | | | | | | | | | | |
| | | 3rd Brood Reproduction Per Replicate | | | | | | | | | | | | Mean | CV % | 111111 |
| | | 35 | 31 | 30 | 25 | 34 | 37 | 31 | 24 | 39 | 33 | 31.9 | 15.0 | | | |

| | REP | Treatment: 10% CRM 3.0 w CRW. | | | | | | | | | | | | % Sur. | No. of Neonates Per Day | Tech |
|--|-----|--------------------------------------|----|----|----|----|----|----|----|----|----|--------------------------------------|------|--------|-------------------------|--------|
| | | 41 | 42 | 43 | 44 | 45 | 46 | 47 | 48 | 49 | 50 | 3rd Brood Reproduction Per Replicate | Mean | CV % | | |
| | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 111 | 111 | 0.00 | | |
| | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | 0.00 | | |
| | 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | 0.00 | | |
| | 3 | 7 | 5 | 6 | 5 | 5 | 6 | 5 | 7 | 5 | 5 | 100 | 56 | 0.00 | | |
| | 4 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | 0.00 | | |
| | 5 | 14 | 13 | 14 | 14 | 14 | 13 | 11 | 15 | 16 | 13 | 100 | 137 | 0.00 | | |
| | 6 | 17 | 18 | 20 | 18 | 16 | 15 | 14 | 22 | 23 | 12 | 100 | 175 | 0.00 | | |
| | 7 | | | | | | | | | | | | | | | |
| | | 3rd Brood Reproduction Per Replicate | | | | | | | | | | | | Mean | CV % | 111111 |
| | | 38 | 36 | 40 | 37 | 35 | 34 | 30 | 44 | 44 | 42 | 36.8 | 13.4 | | | |

Comments:

30
wrong data JG-022811

C. dubia Daily Survival & Reproduction Data Cont.

| | REP | Treatment: 20% CRM 3.0 w CRW. | | | | | | | | | | | | No. of Neonates Per Day | Tech |
|---|--------------------------------------|-------------------------------|----|----|----|----|----|----|----|----|------|--------|------|-------------------------|--------|
| | | 51 | 52 | 53 | 54 | 55 | 56 | 57 | 58 | 59 | 60 | % Sur. | | | |
| | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | /// | /// | 000 | |
| 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | 000 | |
| 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | 000 | |
| 3 | 7 | 5 | 6 | 6 | 5 | 4 | 6 | 5 | 7 | 6 | 100 | 57 | 000 | | |
| 4 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | 000 | |
| 5 | 12 | 11 | 12 | 13 | 8 | 14 | 12 | 16 | 15 | 11 | 100 | 124 | 000 | | |
| 6 | 20 | 19 | 0 | 15 | 23 | 22 | 16 | 18 | 21 | 17 | 100 | 171 | 000 | | |
| 7 | | | | | | | | | | | | | | | |
| | 3rd Brood Reproduction Per Replicate | | | | | | | | | | | | Mean | CV % | ////// |
| | 39 | 35 | 18 | 34 | 36 | 40 | 44 | 39 | 43 | 34 | 35.2 | 19.3 | | | |

34 wrung data JG-022811

| | REP | Treatment: 40% CRM 3.0 w CRW. | | | | | | | | | | | | No. of Neonates Per Day | Tech |
|---|--------------------------------------|-------------------------------|----|----|----|----|----|----|----|----|------|--------|------|-------------------------|--------|
| | | 61 | 62 | 63 | 64 | 65 | 66 | 67 | 68 | 69 | 70 | % Sur. | | | |
| | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | /// | /// | 000 | |
| 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | 124 | |
| 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | 000 | |
| 3 | 6 | 4 | 5 | 6 | 6 | 6 | 5 | 6 | 6 | 6 | 100 | 56 | 000 | | |
| 4 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | 000 | |
| 5 | 14 | 11 | 12 | 12 | 14 | 12 | 13 | 14 | 14 | 12 | 100 | 128 | 000 | | |
| 6 | 22 | 18 | 0 | 16 | 21 | 20 | 15 | 0 | 22 | 13 | 100 | 147 | 000 | | |
| 7 | | | | | | | | | | | | | | | |
| | 3rd Brood Reproduction Per Replicate | | | | | | | | | | | | Mean | CV % | ////// |
| | 42 | 33 | 17 | 34 | 41 | 38 | 33 | 20 | 42 | 31 | 33.1 | 26.3 | | | |

Comments:

C. dubia Daily Survival & Reproduction Data Cont.

| Treatment: 60% CRM 3.0 w CRW. | | | | | | | | | | | | | | |
|-------------------------------|--------------------------------------|----|----|----|----|----|----|----|----|----|------|--------|-------------------------|------|
| | REP | 71 | 72 | 73 | 74 | 75 | 76 | 77 | 78 | 79 | 80 | % Sur. | No. of Neonates Per Day | Tech |
| | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 111 | 111 | D40 |
| | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | D40 |
| | 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | D40 |
| | 3 | 5 | 4 | 6 | 5 | 6 | 6 | 6 | 6 | 5 | 5 | 100 | 55 | D40 |
| | 4 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | D40 |
| | 5 | 13 | 15 | 13 | 12 | 13 | 13 | 12 | 14 | 13 | 12 | 100 | 130 | D40 |
| | 6 | 16 | 12 | 16 | 14 | 17 | 20 | 18 | 0 | 15 | 15 | 100 | 143 | D40 |
| | 7 | | | | | | | | | | | | | |
| | 3rd Brood Reproduction Per Replicate | | | | | | | | | | | | Mean | CV % |
| | 34 | 44 | 45 | 31 | 36 | 39 | 36 | 20 | 34 | 32 | 32.8 | 15.7 | | |
| | 31 | 35 | | | | | | | | | | | | |

wrong data JG-022811

| Treatment: 80% CRM 3.0 w CRW. | | | | | | | | | | | | | | |
|-------------------------------|--------------------------------------|----|----|----|----|----|----|----|----|----|------|--------|-------------------------|------|
| | REP | 81 | 82 | 83 | 84 | 85 | 86 | 87 | 88 | 89 | 90 | % Sur. | No. of Neonates Per Day | Tech |
| | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 111 | 111 | D40 |
| | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | D40 |
| | 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | D40 |
| | 3 | 5 | 4 | 6 | 5 | 6 | 4 | 5 | 5 | 5 | 5 | 100 | 50 | D40 |
| | 4 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | D40 |
| | 5 | 13 | 13 | 13 | 14 | 11 | 11 | 13 | 12 | 14 | 12 | 100 | 126 | D40 |
| | 6 | 14 | 20 | 0 | 18 | 13 | 14 | 15 | 0 | 17 | 9 | 100 | 120 | D40 |
| | 7 | | | | | | | | | | | | | |
| | 3rd Brood Reproduction Per Replicate | | | | | | | | | | | | Mean | CV % |
| | 32 | 37 | 19 | 37 | 30 | 29 | 33 | 17 | 36 | 26 | 29.6 | 24.0 | | |

Comments:

C. dubia Daily Survival & Reproduction Data Cont.

| | Treatment: 100% CRM 3.0 w CRW. | | | | | | | | | | | | | |
|--------------------------------------|--------------------------------|----|----|----|----|----|----|----|----|----|-----|--------|-------------------------|-------|
| | REP | 91 | 92 | 93 | 94 | 95 | 96 | 97 | 98 | 99 | 100 | % Sur. | No. of Neonates Per Day | Tech |
| 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 111 | DCD |
| | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | DCD |
| | 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | DCD |
| | 3 | 5 | 5 | 6 | 5 | 6 | 5 | 6 | 5 | 5 | 5 | 100 | 53 | DCD |
| | 4 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 | DCD |
| | 5 | 10 | 11 | 12 | 10 | 12 | 13 | 12 | 10 | 12 | 10 | 100 | 112 | DCD |
| | 6 | 9 | 15 | 14 | 12 | 0 | 15 | 14 | 15 | 7 | 13 | 100 | 114 | DCD |
| | 7 | | | | | | | | | | | | | |
| 3rd Brood Reproduction Per Replicate | | | | | | | | | | | | Mean | CV % | 11111 |
| | | 24 | 31 | 32 | 27 | 18 | 33 | 32 | 30 | 24 | 28 | 27.9 | 11.0 | |

Calculations by: Bethany QA/QC by: Jennifer Diffrith
 Data Entry by: Bethany
 Double Data Entry by: Bethany or
 QA/QC Officer: M/A

Comments: Day 0: water very clear prior to renewal/turbid after but settled fast. DCD
 Day 1: no problem finding neonates. DCD

C. dubia Water Quality Data

All Treatments: Initial Temp.: 23.5 to 26.4°C; Initial DO: 4.0 to 8.3 mg/l

| Day -1 | Controls | | | Treatment % CRM 3.0 | | | | | | | Meter # |
|----------------|--------------------|------------|------------|---------------------|-------|-------|-------|-------|-------|--------|---------------|
| | MHSW | | LCS + MHSW | Clinch River Water | | | | | | | |
| 02/16/11 | MHSW | LCS + MHSW | LCS + CRW | CRS | 10.0% | 20.0% | 40.0% | 60.0% | 80.0% | 100.0% | |
| DO | I | 8.1 | 8.0 | 8.3 | 7.3 | 7.2 | 7.1 | 7.2 | 7.5 | 7.6 | 7.7 |
| Temp | I | 25.4 | 25.3 | 24.3 | 24.3 | 24.4 | 24.4 | 24.4 | 24.3 | 24.4 | 24.5 |
| Tech. Initials | Initials: DCD | | | | | | | | | | DO 2/16/11 |
| Times | Initial Time: 1545 | | | | | | | | | | |

| Day 0 | Controls | | | Treatment % CRM 3.0 | | | | | | | Meter # | |
|----------------|------------------|------------|------------|---------------------|-------|-------|-------|--------------------|-------|--------|---------|-----|
| | MHSW | | LCS + MHSW | LCS + CRW | CRS | 10.0% | 20.0% | 40.0% | 60.0% | 80.0% | 100.0% | |
| 02/17/11 | MHSW | LCS + MHSW | LCS + CRW | CRS | 10.0% | 20.0% | 40.0% | 60.0% | 80.0% | 100.0% | | |
| DO | F | 7.6 | 7.5 | 7.5 | 6.4 | 5.9 | 5.6 | 5.6 | 4.9 | 5.4 | 5.7 | 57 |
| | I | 7.4 | 7.5 | 7.5 | 6.5 | 6.4 | 6.2 | 6.3 | 6.3 | 6.0 | 5.8 | 57 |
| Temp | F | 25.0 | 24.9 | 24.8 | 24.8 | 25.0 | 25.0 | 24.8 | 25.0 | 24.8 | 24.7 | A46 |
| | I | 25.0 | 25.0 | 25.1 | 25.4 | 25.2 | 25.0 | 24.9 | 25.1 | 25.1 | 25.1 | A46 |
| Tech. Initials | Finals: DCD | | | | | | | Initials: DCD | | | | |
| Times | Final Time: 1235 | | | | | | | Initial Time: 1542 | | | | |

| Day 1 | Controls | | | Treatment % CRM 3.0 | | | | | | | Meter # | |
|----------------|------------------|------------|------------|---------------------|-------|-------|-------|--------------------|-------|--------|---------|-----|
| | MHSW | | LCS + MHSW | LCS + CRW | CRS | 10.0% | 20.0% | 40.0% | 60.0% | 80.0% | 100.0% | |
| 02/18/11 | MHSW | LCS + MHSW | LCS + CRW | CRS | 10.0% | 20.0% | 40.0% | 60.0% | 80.0% | 100.0% | | |
| DO | F | 7.5 | 7.5 | 7.4 | 6.5 | 6.0 | 6.0 | 6.1 | 5.8 | 5.9 | 6.0 | 57 |
| | I | 7.5 | 7.5 | 7.5 | 6.4 | 6.2 | 6.2 | 6.2 | 6.2 | 6.2 | 6.0 | 57 |
| Temp | F | 24.9 | 25.1 | 25.0 | 24.8 | 25.0 | 25.3 | 25.0 | 25.0 | 24.8 | 25.0 | A46 |
| | I | 24.8 | 24.8 | 24.8 | 24.8 | 24.9 | 24.6 | 24.8 | 24.9 | 24.8 | 24.7 | A46 |
| Tech. Initials | Finals: DCD | | | | | | | Initials: DCD | | | | |
| Times | Final Time: 0946 | | | | | | | Initial Time: 1530 | | | | |

TVA, CRM 3.0, Site Sediment
TVA, CRS, Reference Sediment
TVA, CRW, River Water

C. dubia 7-day Chronic.
9 of 12

E-091-11
E-095-11
E-086-11

C. dubia Water Quality Data Cont.

All Treatments: Initial Temp.: 23.5 to 26.4°C; Initial DO: 4.0 to 8.3 mg/l

| Day 2 | Controls | | | | Treatment % CRM 3.0 | | | | | | | Meter # |
|----------------|------------------|------------|--------------------|------|---------------------|-------|--------------------|-------|-------|--------|-----|---------|
| | MHSW | | Clinch River Water | | | | | | | | | |
| 02/19/11 | MHSW | LCS + MHSW | LCS + CRW | CRS | 10.0% | 20.0% | 40.0% | 60.0% | 80.0% | 100.0% | | |
| DO F | 7.7 | 7.5 | 7.3 | 5.9 | 6.0 | 5.9 | 5.7 | 5.5 | 5.6 | 5.8 | 57 | |
| I | 7.5 | 7.6 | 7.5 | 6.0 | 5.8 | 5.6 | 5.5 | 5.6 | 5.8 | 5.9 | 57 | |
| Temp F | 24.9 | 24.9 | 25.0 | 25.1 | 25.0 | 25.0 | 24.9 | 25.2 | 24.9 | 25.0 | A46 | |
| I | 25.1 | 25.3 | 25.2 | 25.2 | 25.4 | 25.2 | 25.0 | 25.1 | 25.3 | 25.2 | A46 | |
| Tech. Initials | Finals: DWD | | | | | | Initials: DWD | | | | | |
| Times | Final Time: 0835 | | | | | | Initial Time: 1533 | | | | | |

| Day 3 | Controls | | | | Treatment % CRM 3.0 | | | | | | | Meter # |
|----------------|------------------|------------|--------------------|------|---------------------|-------|--------------------|-------|-------|--------|-----|---------|
| | MHSW | | Clinch River Water | | | | | | | | | |
| 02/20/11 | MHSW | LCS + MHSW | LCS + CRW | CRS | 10.0% | 20.0% | 40.0% | 60.0% | 80.0% | 100.0% | | |
| DO F | 7.6 | 7.4 | 7.3 | 5.9 | 6.4 | 6.2 | 6.1 | 6.0 | 6.0 | 5.9 | 57 | |
| I | 7.5 | 7.4 | 6.8 | 6.1 | 5.9 | 5.9 | 5.9 | 5.8 | 6.2 | 5.9 | 57 | |
| Temp F | 25.0 | 25.1 | 25.2 | 24.8 | 24.9 | 25.1 | 24.8 | 24.9 | 24.9 | 25.0 | A46 | |
| I | 25.4 | 25.5 | 25.5 | 25.3 | 25.5 | 25.6 | 25.4 | 25.3 | 25.5 | 25.3 | A46 | |
| Tech. Initials | Finals: DWD | | | | | | Initials: DWD | | | | | |
| Times | Final Time: 1003 | | | | | | Initial Time: 1500 | | | | | |

Comments:

C. dubia Water Quality Data Cont.

All Treatments: Initial Temp.: 23.5 to 26.4°C; Initial DO: 4.0 to 8.3 mg/l

| Day 5 | Controls | | | | Treatment % CRM 3.0 | | | | | | Meter # |
|----------------|------------------|------------|--------------------|------|---------------------|-------|--------------------|-------|-------|--------|---------|
| | MHSW | | Clinch River Water | | | | | | | | |
| 02/22/11 | MHSW | LCS + MHSW | LCS + CRW | CRS | 10.0% | 20.0% | 40.0% | 60.0% | 80.0% | 100.0% | |
| DO F | 7.3 | 7.2 | 7.3 | 6.0 | 6.2 | 6.3 | 6.1 | 6.3 | 6.1 | 5.6 | 57 |
| I | 7.1 | 7.1 | 7.1 | 6.2 | 6.1 | 6.0 | 6.0 | 6.0 | 5.9 | 5.8 | 57 |
| Temp F | 24.4 | 24.5 | 24.5 | 24.4 | 24.2 | 24.4 | 24.3 | 24.3 | 24.5 | 24.5 | A46 |
| I | 24.7 | 24.4 | 24.5 | 24.7 | 24.4 | 24.4 | 24.7 | 25.0 | 24.8 | 24.7 | A46 |
| Tech. Initials | Finals: DCD | | | | | | Initials: OCO | | | | |
| Times | Final Time: 0748 | | | | | | Initial Time: 1530 | | | | |

Comments:

TVA, CRM 3.0, Site Sediment
TVA, CRS, Reference Sediment
TVA, CRW, River Water

C. dubia 7-day Chronic.
11 of 12

E-091-11
E-095-11
E-086-11

C. dubia Water Quality Data Cont.

All Treatments: Initial Temp.: 23.5 to 26.4°C; Initial DO: 4.0 to 8.3 mg/l

Comments:

TVA, CRM 3.0, Site Sediment
TVA, CRS, Reference Sediment
TVA, CRW, River Water

C. dubia 7-day Chronic.
12 of 12

E-091-11
E-095-11
E-086-11

Environmental Enterprises USA, Inc.

APPENDIX B

Ceriodaphnia Survival and Reproduction Test-7 Day Survival

Start Date: 2/17/2011 Test ID: cd09111 Sample ID: CRM3.0
 End Date: 2/23/2011 Lab ID: EE USA Sample Type: Whole Sediment
 Sample Date: 2/8/2011 Protocol: ASTM E1706-05 Annex A2 Test Species: CD-Ceriodaphnia dubia
 Comments: LCS=Lab Control Sediment; CRW=Clinch River Water; CRS=Clinch Reference Sediment

| Conc-% | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---------|--------|--------|--------|--------|--------|--------|--------|--------|--------|--------|
| LCS+CRW | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 |
| CRS+CRW | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 |
| 10 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 |
| 20 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 |
| 40 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 |
| 60 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 |
| 80 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 |
| 100 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 |

| Conc-% | Mean | N-Mean | Resp | Not Resp | Total | N | Fisher's Exact P | 1-Tailed Critical | Isotonic | |
|---------|--------|--------|------|----------|-------|----|------------------|-------------------|----------|--------|
| | | | | | | | | | Mean | N-Mean |
| LCS+CRW | 1.0000 | 1.0000 | 0 | 10 | 10 | 10 | 0.6238 | * | 1.0000 | 1.0000 |
| CRS+CRW | 1.0000 | 1.0000 | 0 | 10 | 10 | 10 | | | 1.0000 | 1.0000 |
| 10 | 1.0000 | 1.0000 | 0 | 10 | 10 | 10 | 1.0000 | 0.0500 | 1.0000 | 1.0000 |
| 20 | 1.0000 | 1.0000 | 0 | 10 | 10 | 10 | 1.0000 | 0.0500 | 1.0000 | 1.0000 |
| 40 | 1.0000 | 1.0000 | 0 | 10 | 10 | 10 | 1.0000 | 0.0500 | 1.0000 | 1.0000 |
| 60 | 1.0000 | 1.0000 | 0 | 10 | 10 | 10 | 1.0000 | 0.0500 | 1.0000 | 1.0000 |
| 80 | 1.0000 | 1.0000 | 0 | 10 | 10 | 10 | 1.0000 | 0.0500 | 1.0000 | 1.0000 |
| 100 | 1.0000 | 1.0000 | 0 | 10 | 10 | 10 | 1.0000 | 0.0500 | 1.0000 | 1.0000 |

Hypothesis Test (1-tail, 0.05) NOEC LOEC ChV TU

Fisher's Exact Test 100 >100 1

Treatments vs CRS+CRW

Linear Interpolation (200 Resamples)

| Point | % | SD | 95% CL | Skew |
|-------|------|----|--------|------|
| IC05 | >100 | | | |
| IC10 | >100 | | | |
| IC15 | >100 | | | |
| IC20 | >100 | | | |
| IC25 | >100 | | | |
| IC40 | >100 | | | |
| IC50 | >100 | | | |

Ceriodaphnia Survival and Reproduction Test-Reproduction

Start Date: 2/17/2011 Test ID: cd09111 Sample ID: CRM3.0
 End Date: 2/23/2011 Lab ID: EE USA Sample Type: Whole Sediment
 Sample Date: 2/8/2011 Protocol: ASTM E1706-05 Annex A2 Test Species: CD-Ceriodaphnia dubia
 Comments: LCS=Lab Control Sediment; CRW=Clinch River Water; CRS=Clinch Reference Sediment

| Conc-% | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---------|--------|--------|--------|--------|--------|--------|--------|--------|--------|--------|
| LCS+CRW | 26.000 | 32.000 | 18.000 | 31.000 | 37.000 | 30.000 | 27.000 | 26.000 | 28.000 | 28.000 |
| CRS+CRW | 35.000 | 31.000 | 30.000 | 25.000 | 34.000 | 37.000 | 31.000 | 24.000 | 39.000 | 33.000 |
| 10 | 38.000 | 36.000 | 40.000 | 37.000 | 35.000 | 34.000 | 30.000 | 44.000 | 44.000 | 30.000 |
| 20 | 39.000 | 35.000 | 18.000 | 34.000 | 36.000 | 40.000 | 34.000 | 39.000 | 43.000 | 34.000 |
| 40 | 42.000 | 33.000 | 17.000 | 34.000 | 41.000 | 38.000 | 33.000 | 20.000 | 42.000 | 31.000 |
| 60 | 34.000 | 31.000 | 35.000 | 31.000 | 36.000 | 39.000 | 36.000 | 20.000 | 34.000 | 32.000 |
| 80 | 32.000 | 37.000 | 19.000 | 37.000 | 30.000 | 29.000 | 33.000 | 17.000 | 36.000 | 26.000 |
| 100 | 24.000 | 31.000 | 32.000 | 27.000 | 18.000 | 33.000 | 32.000 | 30.000 | 24.000 | 28.000 |

| Conc-% | Transform: Untransformed | | | | | | Rank Sum | 1-Tailed Critical | Isotonic | | |
|---------|--------------------------|--------|--------|--------|--------|--------|----------|-------------------|----------|--------|--------|
| | Mean | N-Mean | Mean | Min | Max | CV% | | | Mean | N-Mean | |
| LCS+CRW | 28.300 | 0.8871 | 28.300 | 18.000 | 37.000 | 17.395 | 10 | * | 34.633 | 1.0000 | |
| CRS+CRW | 31.900 | 1.0000 | 31.900 | 24.000 | 39.000 | 15.030 | 10 | | 34.633 | 1.0000 | |
| 10 | 36.800 | 1.1536 | 36.800 | 30.000 | 44.000 | 13.423 | 10 | 129.50 | 74.00 | 34.633 | 1.0000 |
| 20 | 35.200 | 1.1034 | 35.200 | 18.000 | 43.000 | 19.259 | 10 | 129.00 | 74.00 | 34.633 | 1.0000 |
| 40 | 33.100 | 1.0376 | 33.100 | 17.000 | 42.000 | 26.278 | 10 | 115.50 | 74.00 | 33.100 | 0.9557 |
| 60 | 32.800 | 1.0282 | 32.800 | 20.000 | 39.000 | 15.665 | 10 | 114.00 | 74.00 | 32.800 | 0.9471 |
| 80 | 29.600 | 0.9279 | 29.600 | 17.000 | 37.000 | 23.952 | 10 | 97.00 | 74.00 | 29.600 | 0.8547 |
| 100 | 27.900 | 0.8746 | 27.900 | 18.000 | 33.000 | 17.018 | 10 | 82.00 | 74.00 | 27.900 | 0.8056 |

Auxiliary Tests

| | | | | |
|---|----------|----------|----------|----------|
| Kolmogorov D Test indicates non-normal distribution (p <= 0.05) | 1.243287 | 0.895 | -0.98536 | 0.882873 |
| Bartlett's Test indicates equal variances (p = 0.39) | 6.315244 | 16.81189 | | |
| The control means are not significantly different (p = 0.11) | 1.656644 | 2.100922 | | |

Hypothesis Test (1-tail, 0.05) NOEC LOEC ChV TU

Steel's Many-One Rank Test 100 >100 1

Treatments vs CRS+CRW

Linear Interpolation (200 Resamples)

| Point | % | SD | 95% CL | Skew |
|-------|--------|--------|---------------|--------|
| IC05 | 53.222 | 16.786 | 16.860 78.790 | 0.0080 |
| IC10 | 70.188 | | | |
| IC15 | 81.902 | | | |
| IC20 | >100 | | | |
| IC25 | >100 | | | |
| IC40 | >100 | | | |
| IC50 | >100 | | | |

| Hypothesis Test (1-tail, 0.05) | NOEC | LOEC | ChV | TU | MSDu | MSDp | MSB | MSE | F-Prob | df |
|--------------------------------|------|------|-----|----|----------|----------|----------|----------|----------|-------|
| Dunnett's Test | 100 | >100 | | 1 | 6.497464 | 0.203682 | 93.59048 | 38.31587 | 0.034656 | 6, 63 |
| Treatments vs CRS+CRW | | | | | | | | | | |

Ceriodaphnia Survival and Reproduction Test-7 Day Survival

Start Date: 2/17/2011 Test ID: cd09111 Sample ID: CRM3.0
 End Date: 2/23/2011 Lab ID: EE-Environmental Enterprises U Sample Type: Whole Sediment
 Sample Date: 2/8/2011 Protocol: ASTM E1706-05 Annex A2 Test Species: CD-Ceriodaphnia dubia
 Comments: MHSW=Mod Hard Synthetic Water; LCS=Lab Control Sediment; CRW=Clinch River Water

| Conc-% | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|----------|--------|--------|--------|--------|--------|--------|--------|--------|--------|--------|
| MHSW | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 |
| LCS+MHSW | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 |
| LCS+CRW | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 |

| Conc-% | Mean | N-Mean | Resp | Not Resp | Total | N | Fisher's | 1-Tailed |
|----------|--------|--------|------|----------|-------|----|----------|----------|
| | | | | | | | Exact P | Critical |
| MHSW | 1.0000 | 1.0000 | 0 | 10 | 10 | 10 | 0.6238 | |
| LCS+MHSW | 1.0000 | 1.0000 | 0 | 10 | 10 | 10 | * | |
| LCS+CRW | 1.0000 | 1.0000 | 0 | 10 | 10 | 10 | 1.0000 | 0.0500 |

Hypothesis Test (1-tail, 0.05)

Fisher's Exact Test indicates no significant differences

Treatments vs LCS+MHSW

Ceriodaphnia Survival and Reproduction Test-Reproduction

Start Date: 2/17/2011 Test ID: cd09111 Sample ID: CRM3.0
 End Date: 2/23/2011 Lab ID: EE USA Sample Type: Whole Sediment
 Sample Date: 2/8/2011 Protocol: ASTM E1706-05 Annex A2 Test Species: CD-Ceriodaphnia dubia
 Comments: MHSW=Mod Hard Synthetic Water; LCS=Lab Control Sediment; CRW=Clinch River Water

| Conc-% | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|----------|--------|--------|--------|--------|--------|--------|--------|--------|--------|--------|
| MHSW | 25.000 | 24.000 | 24.000 | 19.000 | 24.000 | 24.000 | 24.000 | 24.000 | 22.000 | 19.000 |
| LCS+MHSW | 21.000 | 20.000 | 26.000 | 22.000 | 26.000 | 24.000 | 20.000 | 16.000 | 26.000 | 27.000 |
| LCS+CRW | 26.000 | 32.000 | 18.000 | 31.000 | 37.000 | 30.000 | 27.000 | 26.000 | 28.000 | 28.000 |

| Conc-% | Transform: Untransformed | | | | | | | 1-Tailed | | |
|----------|--------------------------|--------|--------|--------|--------|--------|----|----------|----------|-------|
| | Mean | N-Mean | Mean | Min | Max | CV% | N | t-Stat | Critical | MSD |
| MHSW | 22.900 | 1.0044 | 22.900 | 19.000 | 25.000 | 9.534 | 10 | | | |
| LCS+MHSW | 22.800 | 1.0000 | 22.800 | 16.000 | 27.000 | 15.719 | 10 | * | | |
| LCS+CRW | 28.300 | 1.2412 | 28.300 | 18.000 | 37.000 | 17.395 | 10 | -2.856 | 1.734 | 3.339 |

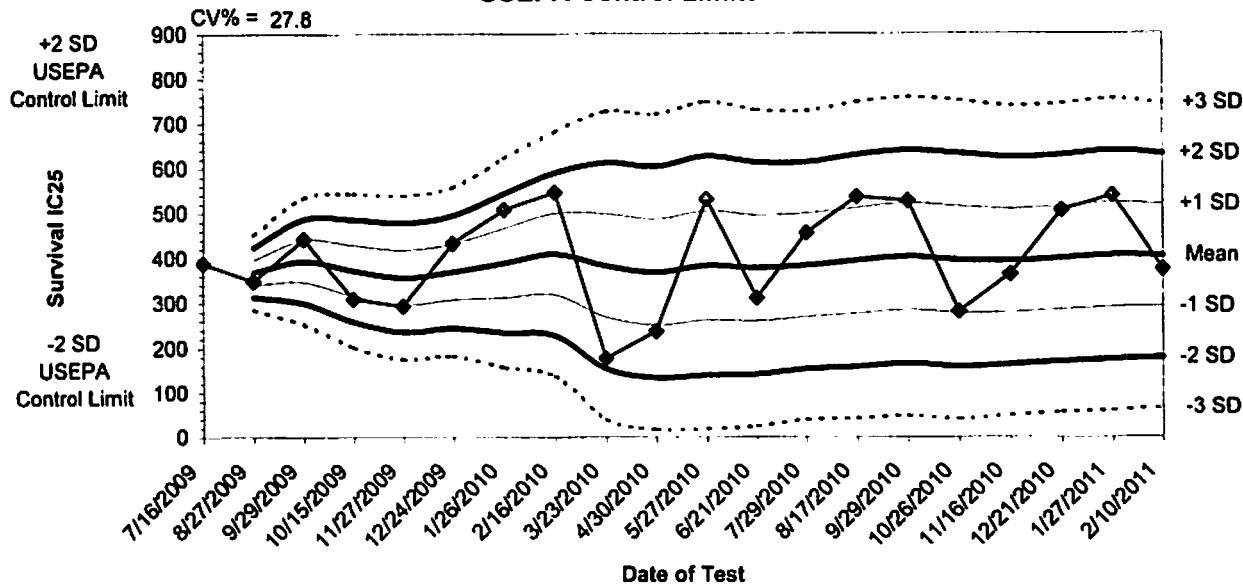
Auxiliary Tests

| | Statistic | Critical | Skew | Kurt |
|--|-----------|----------|----------|----------|
| Shapiro-Wilk's Test indicates normal distribution ($p > 0.05$) | 0.955834 | 0.905 | -0.45773 | 1.189465 |
| F-Test indicates equal variances ($p = 0.36$) | 1.886678 | 6.54109 | | |
| The control means are not significantly different ($p = 0.94$) | 0.075354 | 2.100922 | | |
| Hypothesis Test (1-tail, 0.05) | MSDu | MSDp | MSB | MSE |
| Homoscedastic t Test indicates no significant differences | 3.339042 | 0.146449 | 151.25 | 18.53889 |
| Treatments vs LCS+MHSW | | | 0.010488 | 1, 18 |

Environmental Enterprises USA, Inc.

APPENDIX C

***C. dubia* SRT, Survival IC25, mg/L KCl
USEPA Control Limits**



| Test # | Test Date | Survival IC25 | Mean IC25 | -1 SD | -2 SD | +1 SD | +2 SD | -3 SD | +3 SD | Toxicant Lot # |
|--------|------------|---------------|-----------|-------|-------|-------|-------|-------|-------|----------------|
| CD0909 | 7/16/2009 | 389 | | | | | | | | 029K0050 |
| CD0910 | 8/27/2009 | 350 | 370 | 342 | 314 | 397 | 425 | 287 | 452 | 029K0050 |
| CD0911 | 9/29/2009 | 443 | 394 | 347 | 301 | 441 | 487 | 254 | 534 | 029K0050 |
| CD0912 | 10/15/2009 | 310 | 373 | 316 | 260 | 430 | 486 | 203 | 543 | 029K0050 |
| CD0913 | 11/27/2009 | 294 | 357 | 297 | 236 | 418 | 478 | 176 | 539 | 049K0305 |
| CD0915 | 12/24/2009 | 433 | 370 | 307 | 245 | 432 | 495 | 183 | 557 | 049K0305 |
| CD1002 | 1/26/2010 | 508 | 390 | 312 | 235 | 467 | 544 | 158 | 621 | 079K0011 |
| CD1003 | 2/16/2010 | 546 | 409 | 319 | 228 | 500 | 590 | 138 | 680 | 079K0011 |
| CD1004 | 3/23/2010 | 178 | 383 | 269 | 155 | 498 | 612 | 40 | 727 | 049K0305 |
| CD1006 | 4/30/2010 | 238 | 369 | 252 | 134 | 486 | 603 | 17 | 721 | 049K0305 |
| CD1007 | 5/27/2010 | 531 | 384 | 262 | 141 | 505 | 627 | 19 | 748 | 049K0305 |
| CD1009 | 6/21/2010 | 312 | 378 | 260 | 142 | 495 | 613 | 25 | 731 | 049K0305 |
| CD1010 | 7/29/2010 | 456 | 384 | 269 | 154 | 498 | 613 | 39 | 728 | 079K0011 |
| CD1011 | 8/17/2010 | 536 | 395 | 277 | 160 | 512 | 630 | 42 | 747 | 079K0011 |
| CD1012 | 9/29/2010 | 527 | 403 | 285 | 167 | 522 | 640 | 49 | 758 | 079K0011 |
| CD1013 | 10/26/2010 | 281 | 396 | 277 | 159 | 514 | 632 | 41 | 751 | 099K0202 |
| CD1014 | 11/16/2010 | 364 | 394 | 279 | 164 | 509 | 623 | 49 | 738 | 099K0202 |
| CD1015 | 12/21/2010 | 506 | 400 | 286 | 171 | 515 | 629 | 57 | 744 | 099K0202 |
| CD1101 | 1/27/2011 | 539 | 407 | 292 | 176 | 523 | 639 | 60 | 755 | 099K0202 |
| CD1102 | 2/10/2011 | 376 | 406 | 293 | 180 | 519 | 632 | 67 | 744 | 099K0202 |

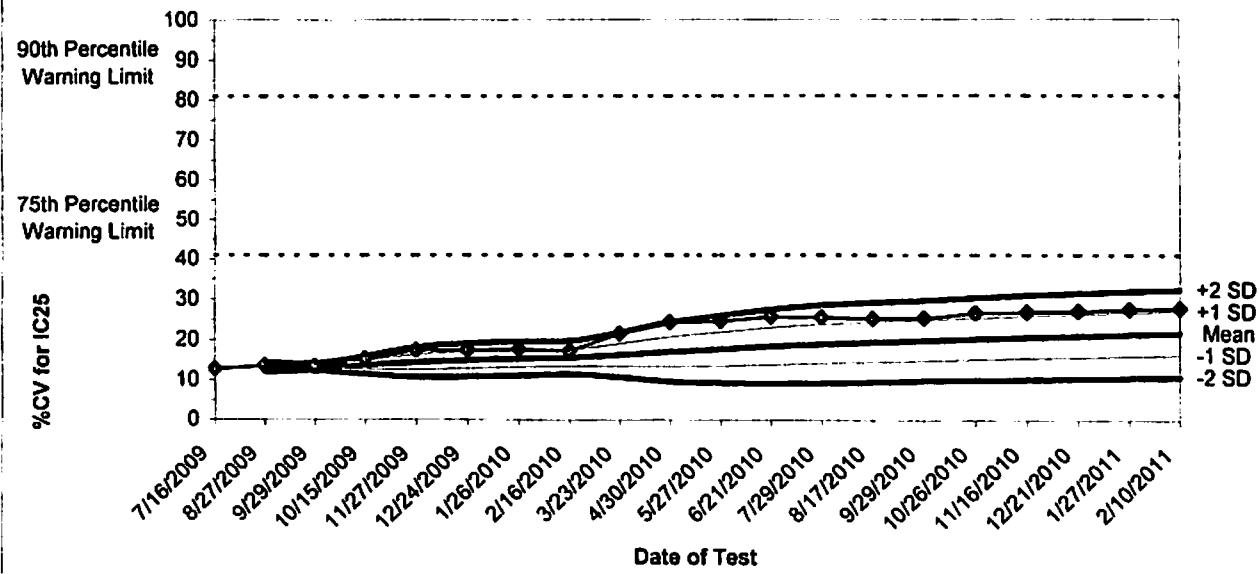
CD1005 - IC25 less than lowest concentration tested and could not be graphed

CD1002 - Widened dilution series to 140, 212, 316, 476, & 716 mg/L KCl

CD1001 - Training lab tech

CD0914 - Training lab tech

**C. dubia SRT, Survival IC25,
USEPA Within Lab %CV Warning and Control Limits**



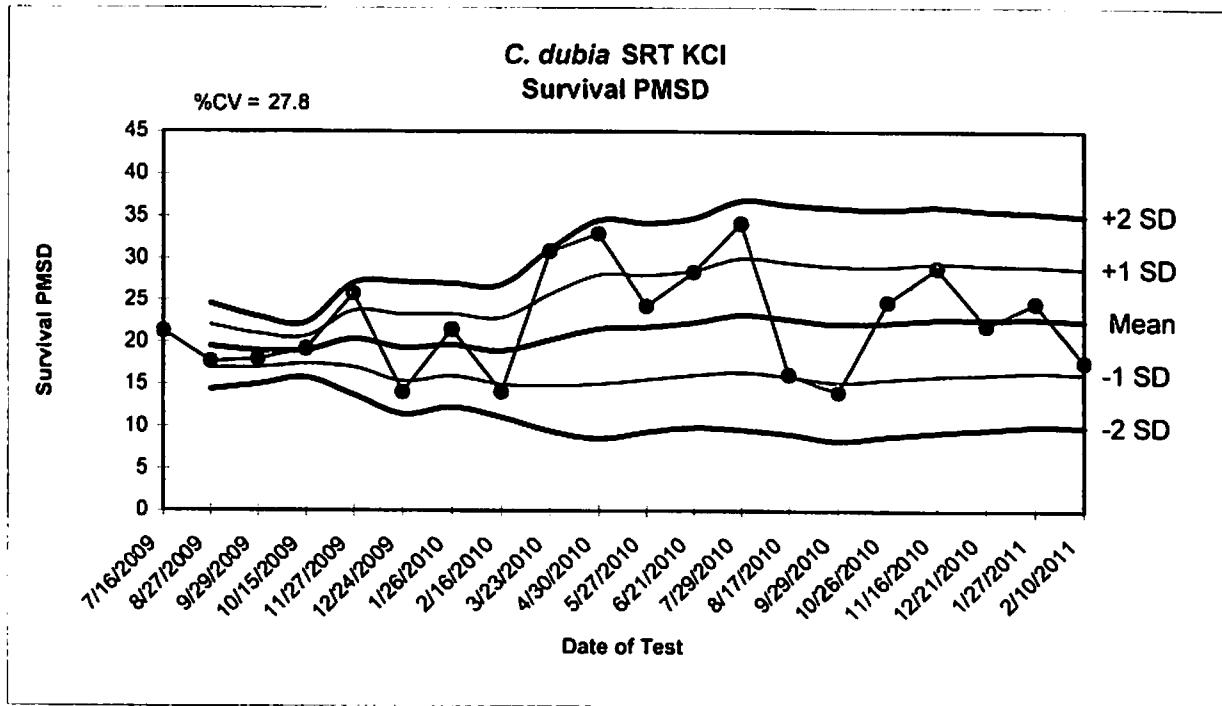
| Test # | Test Date | %CV for IC25 | Mean %CV | -1 SD | -2 SD | +1 SD | +2 SD | 75th Warning Limit | 90th Warning Limit | Toxicant Lot # |
|--------|------------|--------------|----------|-------|-------|-------|-------|--------------------|--------------------|----------------|
| CD0909 | 7/16/2009 | 12.8 | | | | | | 41.0 | 81.0 | 029K0050 |
| CD0910 | 8/27/2009 | 13.6 | 13.2 | 12.6 | 12.0 | 13.8 | 14.4 | 41.0 | 81.0 | 029K0050 |
| CD0911 | 9/29/2009 | 13.4 | 13.3 | 12.8 | 12.4 | 13.7 | 14.1 | 41.0 | 81.0 | 029K0050 |
| CD0912 | 10/15/2009 | 15.3 | 13.8 | 12.7 | 11.6 | 14.9 | 15.9 | 41.0 | 81.0 | 029K0050 |
| CD0913 | 11/27/2009 | 17.4 | 14.5 | 12.6 | 10.8 | 16.3 | 18.2 | 41.0 | 81.0 | 049K0305 |
| CD0915 | 12/24/2009 | 17.4 | 15.0 | 12.9 | 10.9 | 17.0 | 19.0 | 41.0 | 81.0 | 049K0305 |
| CD1002 | 1/26/2010 | 17.6 | 15.3 | 13.2 | 11.1 | 17.5 | 19.6 | 41.0 | 81.0 | 079K0011 |
| CD1003 | 2/16/2010 | 17.2 | 15.6 | 13.5 | 11.5 | 17.6 | 19.7 | 41.0 | 81.0 | 079K0011 |
| CD1004 | 3/23/2010 | 21.5 | 16.2 | 13.5 | 10.7 | 19.0 | 21.7 | 41.0 | 81.0 | 049K0305 |
| CD1006 | 4/30/2010 | 24.4 | 17.0 | 13.4 | 9.7 | 20.7 | 24.4 | 41.0 | 81.0 | 049K0305 |
| CD1007 | 5/27/2010 | 24.7 | 17.7 | 13.6 | 9.4 | 21.9 | 26.1 | 41.0 | 81.0 | 049K0305 |
| CD1009 | 6/21/2010 | 25.7 | 18.4 | 13.8 | 9.2 | 23.0 | 27.6 | 41.0 | 81.0 | 049K0305 |
| CD1010 | 7/29/2010 | 25.5 | 19.0 | 14.1 | 9.3 | 23.8 | 28.6 | 41.0 | 81.0 | 079K0011 |
| CD1011 | 8/17/2010 | 25.2 | 19.4 | 14.5 | 9.6 | 24.3 | 29.2 | 41.0 | 81.0 | 079K0011 |
| CD1012 | 9/29/2010 | 25.3 | 19.8 | 14.8 | 9.8 | 24.8 | 29.8 | 41.0 | 81.0 | 079K0011 |
| CD1013 | 10/26/2010 | 26.6 | 20.2 | 15.1 | 10.0 | 25.3 | 30.4 | 41.0 | 81.0 | 099K0202 |
| CD1014 | 11/16/2010 | 26.8 | 20.6 | 15.4 | 10.2 | 25.8 | 31.0 | 41.0 | 81.0 | 099K0202 |
| CD1015 | 12/21/2010 | 27.1 | 21.0 | 15.7 | 10.4 | 26.2 | 31.5 | 41.0 | 81.0 | 099K0202 |
| CD1101 | 1/27/2011 | 27.6 | 21.3 | 16.0 | 10.6 | 26.7 | 32.0 | 41.0 | 81.0 | 099K0202 |
| CD1102 | 2/10/2011 | 27.8 | 21.6 | 16.2 | 10.8 | 27.0 | 32.4 | 41.0 | 81.0 | 099K0202 |

CD1005 - IC25 less than lowest concentration tested and could not be graphed

CD1002 - Widened dilution series to 140, 212, 316, 476, & 716 mg/L KCl

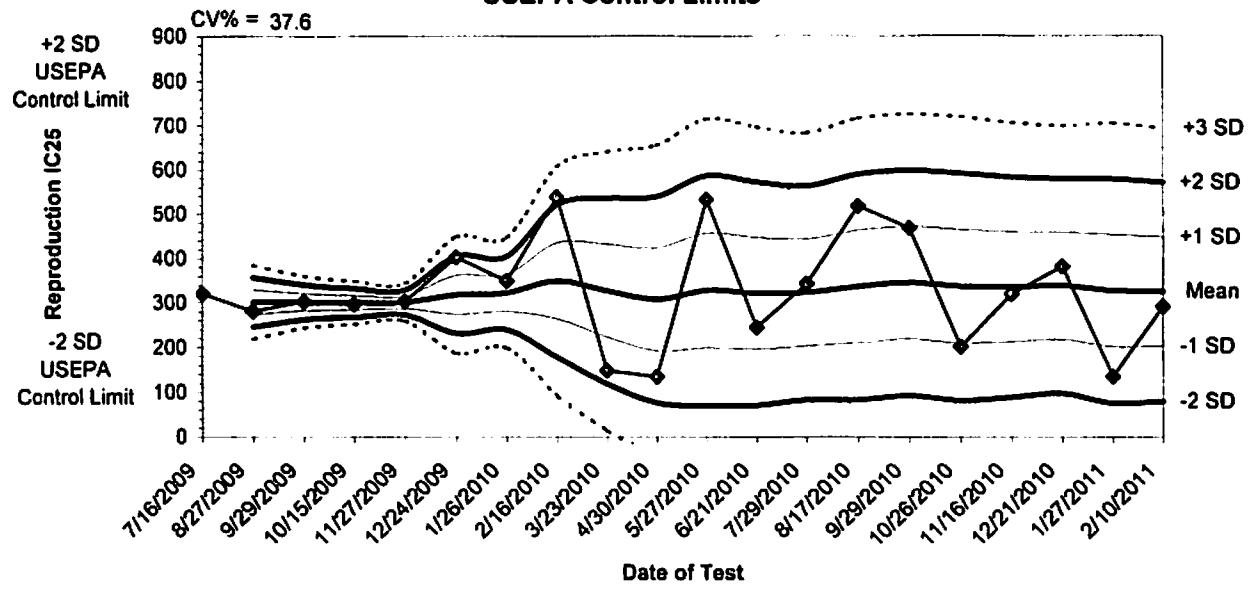
CD1001 - Training lab tech

CD0914 - Training lab tech



| Test # | Test Date | Survival PMSD | Mean | -1 SD | -2 SD | +1 SD | +2 SD | Toxicant Lot | # |
|--------|------------|---------------|---------|---------|---------|---------|---------|--------------|---|
| CD0909 | 7/16/2009 | 21.3 | | | | | | 029K0050 | |
| CD0910 | 8/27/2009 | 17.7 | 19.5000 | 16.9544 | 14.4088 | 22.0456 | 24.5912 | 029K0050 | |
| CD0911 | 9/29/2009 | 18.0 | 19.0000 | 17.0025 | 15.0050 | 20.9975 | 22.9950 | 029K0050 | |
| CD0912 | 10/15/2009 | 19.2 | 19.0500 | 17.4160 | 15.7820 | 20.6840 | 22.3180 | 029K0050 | |
| CD0913 | 11/27/2009 | 25.8 | 20.4000 | 17.0661 | 13.7322 | 23.7339 | 27.0678 | 049K0305 | |
| CD0915 | 12/24/2009 | 14.1 | 19.3500 | 15.4121 | 11.4742 | 23.2879 | 27.2258 | 049K0305 | |
| CD1002 | 1/26/2010 | 21.5 | 19.6571 | 15.9717 | 12.2862 | 23.3426 | 27.0281 | 079K0011 | |
| CD1003 | 2/16/2010 | 14.1 | 18.9625 | 15.0252 | 11.0878 | 22.8998 | 26.8372 | 079K0011 | |
| CD1004 | 3/23/2010 | 30.9 | 20.2889 | 14.8668 | 9.4448 | 25.7109 | 31.1330 | 049K0305 | |
| CD1006 | 4/30/2010 | 33.0 | 21.5600 | 15.0570 | 8.5540 | 28.0630 | 34.5660 | 049K0305 | |
| CD1007 | 5/27/2010 | 24.3 | 21.8091 | 15.5847 | 9.3603 | 28.0335 | 34.2579 | 049K0305 | |
| CD1009 | 6/21/2010 | 28.4 | 22.3583 | 16.1261 | 9.8939 | 28.5906 | 34.8228 | 049K0305 | |
| CD1010 | 7/29/2010 | 34.2 | 23.2692 | 16.4582 | 9.6471 | 30.0803 | 36.8914 | 079K0011 | |
| CD1011 | 8/17/2010 | 16.2 | 22.7643 | 15.9531 | 9.1420 | 29.5754 | 36.3866 | 079K0011 | |
| CD1012 | 9/29/2010 | 14.1 | 22.1867 | 15.2525 | 8.3183 | 29.1208 | 36.0550 | 079K0011 | |
| CD1013 | 10/26/2010 | 24.8 | 22.3500 | 15.6192 | 8.8884 | 29.0808 | 35.8116 | 099K0202 | |
| CD1014 | 11/16/2010 | 28.9 | 22.7353 | 16.0274 | 9.3195 | 29.4432 | 36.1511 | 099K0202 | |
| CD1015 | 12/21/2010 | 22.0 | 22.6944 | 16.1845 | 9.6746 | 29.2044 | 35.7143 | 099K0202 | |
| CD1101 | 1/27/2011 | 24.7 | 22.8000 | 16.4568 | 10.1135 | 29.1432 | 35.4865 | 099K0202 | |
| CD1102 | 2/10/2011 | 17.7 | 22.5450 | 16.2665 | 9.9880 | 28.8235 | 35.1020 | 099K0202 | |

***C. dubia* SRT, Reproduction IC25, mg/L KCl
USEPA Control Limits**



| Test # | Test Date | Repro. IC25 | Mean IC25 | -1 SD | -2 SD | +1 SD | +2 SD | -3 SD | +3 SD | Toxicant Lot # |
|--------|------------|-------------|-----------|-------|-------|-------|-------|-------|-------|----------------|
| CD0909 | 7/16/2009 | 321 | | | | | | | | 029K0050 |
| CD0910 | 8/27/2009 | 282 | 302 | 274 | 246 | 329 | 357 | 219 | 384 | 029K0050 |
| CD0911 | 9/29/2009 | 303 | 302 | 282 | 263 | 322 | 341 | 243 | 361 | 029K0050 |
| CD0912 | 10/15/2009 | 297 | 301 | 285 | 268 | 317 | 333 | 252 | 349 | 029K0050 |
| CD0913 | 11/27/2009 | 304 | 301 | 287 | 273 | 315 | 329 | 259 | 344 | 049K0305 |
| CD0915 | 12/24/2009 | 403 | 318 | 275 | 232 | 362 | 405 | 188 | 448 | 049K0305 |
| CD1002 | 1/26/2010 | 349 | 323 | 281 | 240 | 364 | 405 | 199 | 446 | 079K0011 |
| CD1003 | 2/16/2010 | 539 | 350 | 264 | 179 | 435 | 521 | 93 | 606 | 079K0011 |
| CD1004 | 3/23/2010 | 149 | 327 | 223 | 119 | 432 | 536 | 15 | 640 | 049K0305 |
| CD1006 | 4/30/2010 | 135 | 308 | 193 | 77 | 424 | 539 | -39 | 655 | 049K0305 |
| CD1007 | 5/27/2010 | 533 | 329 | 200 | 71 | 458 | 586 | -58 | 715 | 049K0305 |
| CD1009 | 6/21/2010 | 246 | 322 | 197 | 71 | 447 | 572 | -54 | 697 | 049K0305 |
| CD1010 | 7/29/2010 | 343 | 323 | 203 | 83 | 443 | 563 | -37 | 683 | 079K0011 |
| CD1011 | 8/17/2010 | 518 | 337 | 211 | 84 | 464 | 590 | -42 | 717 | 079K0011 |
| CD1012 | 9/29/2010 | 469 | 346 | 220 | 93 | 473 | 599 | -34 | 726 | 079K0011 |
| CD1013 | 10/26/2010 | 202 | 337 | 210 | 82 | 465 | 592 | -45 | 719 | 099K0202 |
| CD1014 | 11/16/2010 | 320 | 336 | 213 | 89 | 460 | 583 | -34 | 707 | 099K0202 |
| CD1015 | 12/21/2010 | 382 | 339 | 218 | 98 | 459 | 579 | -22 | 699 | 099K0202 |
| CD1101 | 1/27/2011 | 136 | 328 | 202 | 76 | 454 | 580 | -49 | 705 | 099K0202 |
| CD1102 | 2/10/2011 | 292 | 326 | 203 | 81 | 449 | 572 | -42 | 694 | 099K0202 |

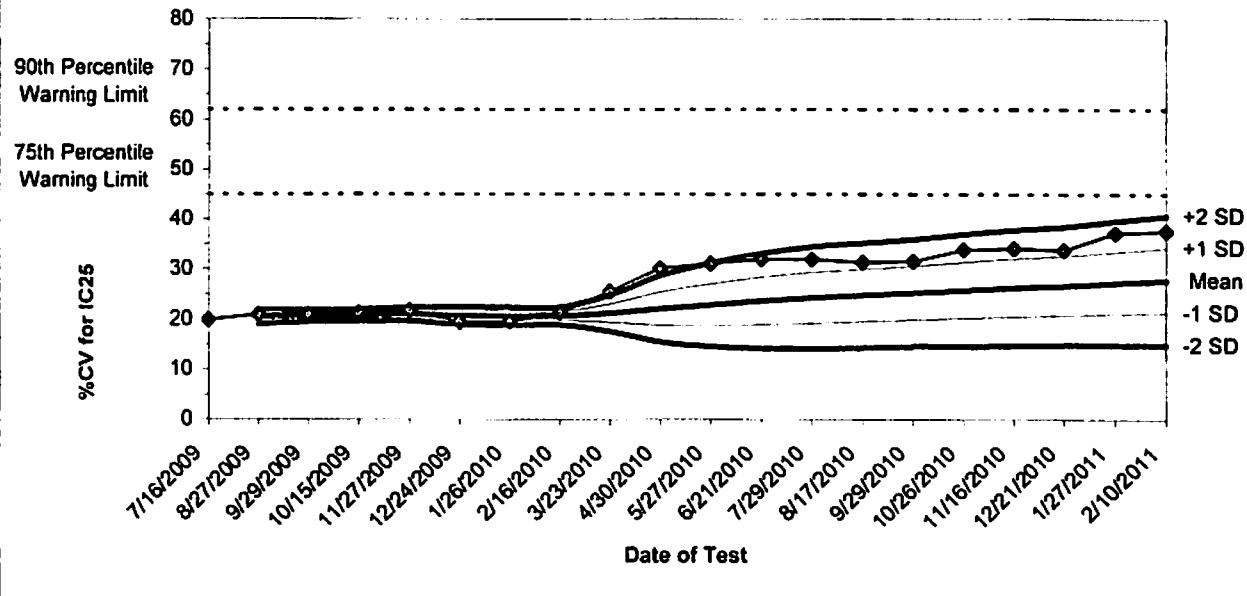
CD1005 - IC25 less than lowest concentration tested and could not be graphed

CD1002 - Widened dilution series to 140, 212, 316, 476, & 716 mg/L KCl

CD1001 - Training lab tech

CD0914 - Training lab tech

C. dubia SRT, Reproduction IC25
USEPA Within Lab %CV Warning and Control Limits



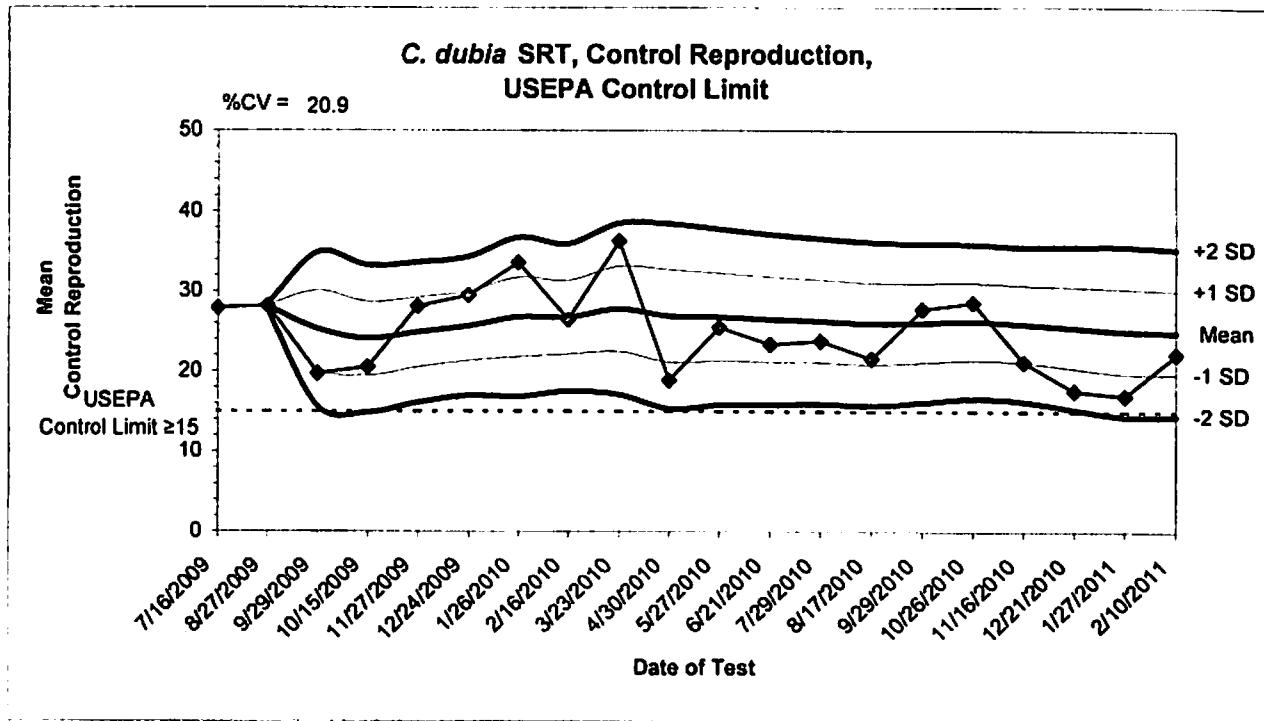
| Test # | Test Date | %CV for IC25 | Mean %CV | -1 SD | -2 SD | +1 SD | +2 SD | 75th Warning Limit | 90th Warning Limit | Toxicant Lot # |
|--------|------------|--------------|----------|-------|-------|-------|-------|--------------------|--------------------|----------------|
| CD0909 | 7/16/2009 | 19.9 | | | | | | 45.0 | 62.0 | 029K0050 |
| CD0910 | 8/27/2009 | 21.0 | 20.4 | 19.7 | 19.0 | 21.2 | 21.9 | 45.0 | 62.0 | 029K0050 |
| CD0911 | 9/29/2009 | 21.0 | 20.6 | 20.0 | 19.4 | 21.2 | 21.8 | 45.0 | 62.0 | 029K0050 |
| CD0912 | 10/15/2009 | 21.3 | 20.8 | 20.2 | 19.6 | 21.4 | 21.9 | 45.0 | 62.0 | 029K0050 |
| CD0913 | 11/27/2009 | 21.8 | 21.0 | 20.3 | 19.6 | 21.7 | 22.3 | 45.0 | 62.0 | 049K0305 |
| CD0915 | 12/24/2009 | 19.4 | 20.7 | 19.9 | 19.0 | 21.6 | 22.5 | 45.0 | 62.0 | 049K0305 |
| CD1002 | 1/26/2010 | 19.6 | 20.6 | 19.6 | 18.7 | 21.5 | 22.4 | 45.0 | 62.0 | 079K0011 |
| CD1003 | 2/16/2010 | 21.3 | 20.7 | 19.8 | 18.9 | 21.5 | 22.4 | 45.0 | 62.0 | 079K0011 |
| CD1004 | 3/23/2010 | 25.5 | 21.2 | 19.4 | 17.6 | 23.0 | 24.8 | 45.0 | 62.0 | 049K0305 |
| CD1006 | 4/30/2010 | 30.1 | 22.1 | 18.8 | 15.5 | 25.4 | 28.7 | 45.0 | 62.0 | 049K0305 |
| CD1007 | 5/27/2010 | 31.1 | 22.9 | 18.8 | 14.6 | 27.0 | 31.2 | 45.0 | 62.0 | 049K0305 |
| CD1009 | 6/21/2010 | 31.9 | 23.7 | 18.9 | 14.2 | 28.4 | 33.1 | 45.0 | 62.0 | 049K0305 |
| CD1010 | 7/29/2010 | 31.9 | 24.3 | 19.2 | 14.1 | 29.4 | 34.4 | 45.0 | 62.0 | 079K0011 |
| CD1011 | 8/17/2010 | 31.3 | 24.8 | 19.6 | 14.3 | 30.0 | 35.2 | 45.0 | 62.0 | 079K0011 |
| CD1012 | 9/29/2010 | 31.6 | 25.2 | 19.9 | 14.6 | 30.6 | 35.9 | 45.0 | 62.0 | 079K0011 |
| CD1013 | 10/26/2010 | 33.9 | 25.8 | 20.2 | 14.6 | 31.4 | 36.9 | 45.0 | 62.0 | 099K0202 |
| CD1014 | 11/16/2010 | 34.1 | 26.3 | 20.5 | 14.7 | 32.0 | 37.8 | 45.0 | 62.0 | 099K0202 |
| CD1015 | 12/21/2010 | 33.8 | 26.7 | 20.8 | 14.9 | 32.6 | 38.4 | 45.0 | 62.0 | 099K0202 |
| CD1101 | 1/27/2011 | 37.2 | 27.2 | 21.0 | 14.9 | 33.4 | 39.6 | 45.0 | 62.0 | 099K0202 |
| CD1102 | 2/10/2011 | 37.6 | 27.8 | 21.3 | 14.8 | 34.2 | 40.7 | 45.0 | 62.0 | 099K0202 |

CD1005 - IC25 less than lowest concentration tested and could not be graphed

CD1002 - Widened dilution series to 140, 212, 316, 476, & 716 mg/L KCl

CD1001 - Training lab tech

CD0914 - Training lab tech



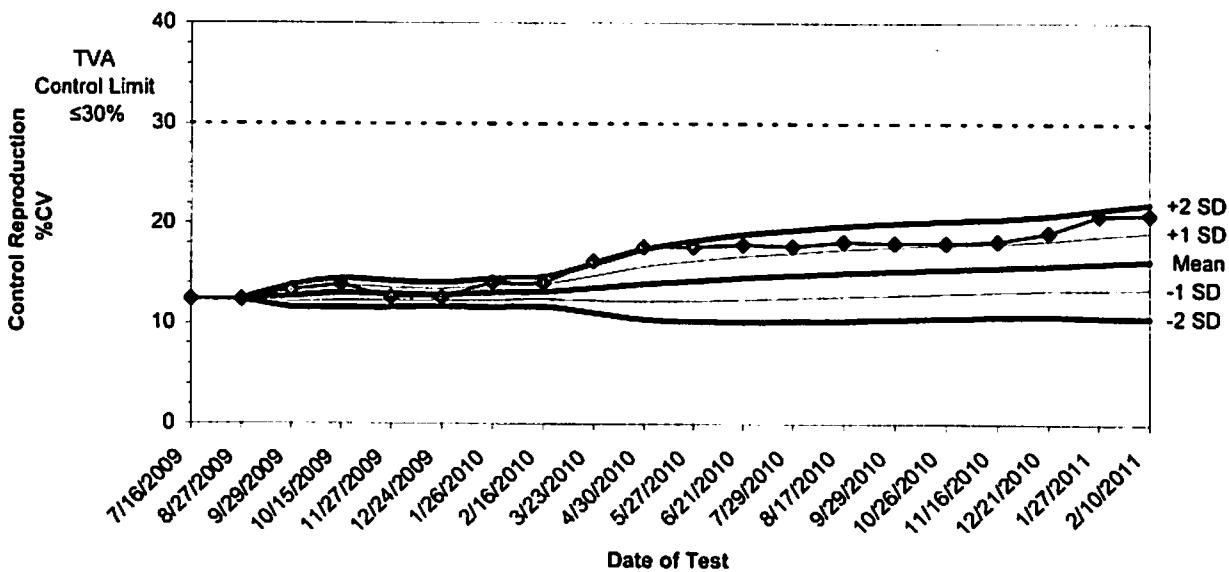
| Test # | Test Date | Mean Control Repro. | Mean | -1 SD | -2 SD | +1 SD | +2 SD | | Control Limit | Toxicant Lot # |
|--------|------------|---------------------|------|-------|-------|-------|-------|--|---------------|----------------|
| CD0909 | 7/16/2009 | 27.9 | | | | | | | 15.0 | 029K0050 |
| CD0910 | 8/27/2009 | 28.2 | 28.1 | 27.8 | 27.6 | 28.3 | 28.5 | | 15.0 | 029K0050 |
| CD0911 | 9/29/2009 | 19.7 | 25.3 | 20.4 | 15.6 | 30.1 | 34.9 | | 15.0 | 029K0050 |
| CD0912 | 10/15/2009 | 20.5 | 24.1 | 19.5 | 14.9 | 28.7 | 33.3 | | 15.0 | 029K0050 |
| CD0913 | 11/27/2009 | 28.1 | 24.9 | 20.5 | 16.1 | 29.3 | 33.6 | | 15.0 | 049K0305 |
| CD0915 | 12/24/2009 | 29.5 | 25.7 | 21.3 | 17.0 | 30.0 | 34.3 | | 15.0 | 049K0305 |
| CD1002 | 1/26/2010 | 33.6 | 26.8 | 21.8 | 16.8 | 31.8 | 36.7 | | 15.0 | 079K0011 |
| CD1003 | 2/16/2010 | 26.4 | 26.7 | 22.1 | 17.5 | 31.3 | 36.0 | | 15.0 | 079K0011 |
| CD1004 | 3/23/2010 | 36.3 | 27.8 | 22.4 | 17.1 | 33.2 | 38.5 | | 15.0 | 049K0305 |
| CD1006 | 4/30/2010 | 18.9 | 26.9 | 21.1 | 15.3 | 32.7 | 38.5 | | 15.0 | 049K0305 |
| CD1007 | 5/27/2010 | 25.5 | 26.8 | 21.3 | 15.8 | 32.3 | 37.8 | | 15.0 | 049K0305 |
| CD1009 | 6/21/2010 | 23.4 | 26.5 | 21.2 | 15.8 | 31.8 | 37.2 | | 15.0 | 049K0305 |
| CD1010 | 7/29/2010 | 23.8 | 26.3 | 21.1 | 16.0 | 31.5 | 36.6 | | 15.0 | 079K0011 |
| CD1011 | 8/17/2010 | 21.6 | 26.0 | 20.8 | 15.7 | 31.1 | 36.2 | | 15.0 | 079K0011 |
| CD1012 | 9/29/2010 | 27.8 | 26.1 | 21.1 | 16.2 | 31.0 | 36.0 | | 15.0 | 079K0011 |
| CD1013 | 10/26/2010 | 28.6 | 26.2 | 21.4 | 16.6 | 31.1 | 35.9 | | 15.0 | 099K0202 |
| CD1014 | 11/16/2010 | 21.2 | 25.9 | 21.1 | 16.3 | 30.8 | 35.6 | | 15.0 | 099K0202 |
| CD1015 | 12/21/2010 | 17.6 | 25.5 | 20.4 | 15.3 | 30.6 | 35.6 | | 15.0 | 099K0202 |
| CD1101 | 1/27/2011 | 17.0 | 25.0 | 19.7 | 14.4 | 30.3 | 35.7 | | 15.0 | 099K0202 |
| CD1102 | 2/10/2011 | 22.2 | 24.9 | 19.7 | 14.5 | 30.1 | 35.3 | | 15.0 | 099K0202 |

CD1002 - Widened dilution series to 140, 212, 316, 476, & 716 mg/L KCl

CD1001 - Training lab tech

CD0914 - Training lab tech

C. dubia SRT, Control Reproduction %CV
TVA Control Limit



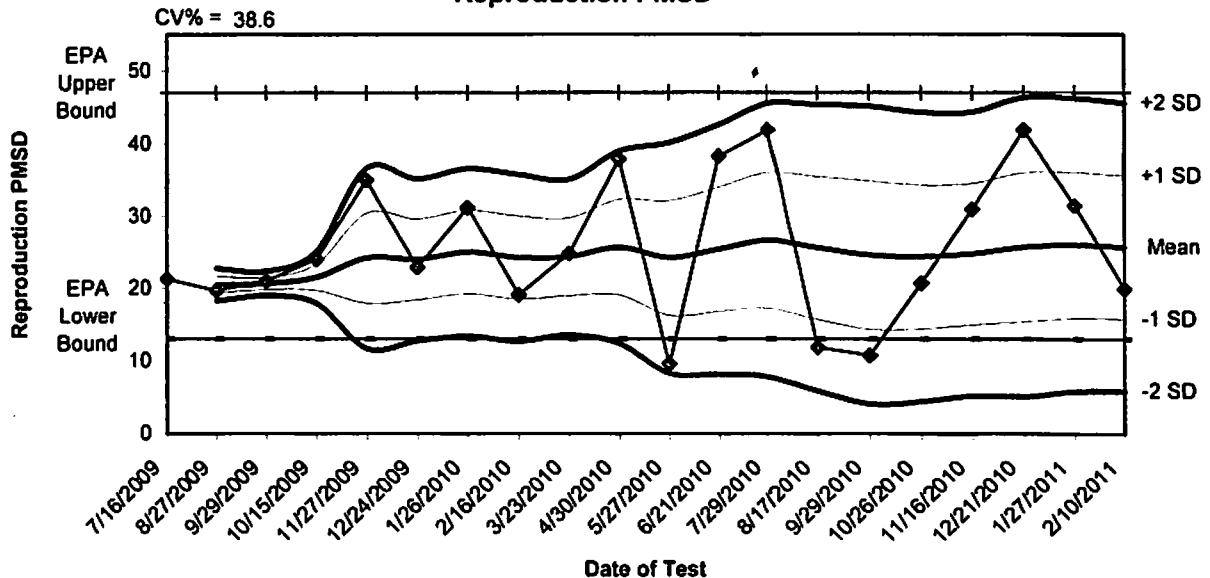
| Test # | Test Date | Control Repro. %CV | Mean %CV | -1 SD | -2 SD | +1 SD | +2 SD | | Control Limit | Toxicant Lot # |
|--------|------------|--------------------|----------|-------|-------|-------|-------|--|---------------|----------------|
| CD0909 | 7/16/2009 | 12.4 | | | | | | | 30.0 | 029K0050 |
| CD0910 | 8/27/2009 | 12.4 | 12.4 | 12.4 | 12.3 | 12.4 | 12.4 | | 30.0 | 029K0050 |
| CD0911 | 9/29/2009 | 13.3 | 12.7 | 12.1 | 11.6 | 13.2 | 13.8 | | 30.0 | 029K0050 |
| CD0912 | 10/15/2009 | 13.8 | 13.0 | 12.3 | 11.5 | 13.7 | 14.4 | | 30.0 | 029K0050 |
| CD0913 | 11/27/2009 | 12.6 | 12.9 | 12.2 | 11.6 | 13.6 | 14.2 | | 30.0 | 049K0305 |
| CD0915 | 12/24/2009 | 12.5 | 12.8 | 12.2 | 11.6 | 13.4 | 14.0 | | 30.0 | 049K0305 |
| CD1002 | 1/26/2010 | 14.1 | 13.0 | 12.3 | 11.6 | 13.7 | 14.5 | | 30.0 | 079K0011 |
| CD1003 | 2/16/2010 | 14.0 | 13.1 | 12.4 | 11.6 | 13.9 | 14.6 | | 30.0 | 079K0011 |
| CD1004 | 3/23/2010 | 16.2 | 13.5 | 12.2 | 11.0 | 14.7 | 15.9 | | 30.0 | 049K0305 |
| CD1006 | 4/30/2010 | 17.6 | 13.9 | 12.1 | 10.4 | 15.6 | 17.4 | | 30.0 | 049K0305 |
| CD1007 | 5/27/2010 | 17.6 | 14.2 | 12.2 | 10.2 | 16.2 | 18.2 | | 30.0 | 049K0305 |
| CD1009 | 6/21/2010 | 17.9 | 14.5 | 12.3 | 10.2 | 16.7 | 18.9 | | 30.0 | 049K0305 |
| CD1010 | 7/29/2010 | 17.7 | 14.8 | 12.5 | 10.2 | 17.0 | 19.3 | | 30.0 | 079K0011 |
| CD1011 | 8/17/2010 | 18.2 | 15.0 | 12.7 | 10.3 | 17.4 | 19.7 | | 30.0 | 079K0011 |
| CD1012 | 9/29/2010 | 18.0 | 15.2 | 12.8 | 10.4 | 17.6 | 20.0 | | 30.0 | 079K0011 |
| CD1013 | 10/26/2010 | 18.0 | 15.4 | 13.0 | 10.5 | 17.8 | 20.2 | | 30.0 | 099K0202 |
| CD1014 | 11/16/2010 | 18.2 | 15.6 | 13.1 | 10.7 | 18.0 | 20.5 | | 30.0 | 099K0202 |
| CD1015 | 12/21/2010 | 19.1 | 15.8 | 13.2 | 10.7 | 18.3 | 20.8 | | 30.0 | 099K0202 |
| CD1101 | 1/27/2011 | 20.8 | 16.0 | 13.3 | 10.6 | 18.7 | 21.4 | | 30.0 | 099K0202 |
| CD1102 | 2/10/2011 | 20.9 | 16.3 | 13.4 | 10.6 | 19.1 | 22.0 | | 30.0 | 099K0202 |

CD1002 - Widened dilution series to 140, 212, 316, 476, & 716 mg/L KCl

CD1001 - Training lab tech

CD0914 - Training lab tech

***C. dubia* SRT KCI
Reproduction PMSD**



| Test # | Test Date | Reprod. PMSD | Mean PMSD | -1 SD | -2 SD | +1 SD | +2 SD | Upper PMSD Bound | Lower PMSD Bound | Toxicant Lot # |
|--------|------------|--------------|-----------|---------|---------|---------|---------|------------------|------------------|----------------|
| CD0909 | 7/16/2009 | 21.3 | | | | | | 47 | 13 | 029K0050 |
| CD0910 | 8/27/2009 | 19.7 | 20.5000 | 19.3686 | 18.2373 | 21.6314 | 22.7627 | 47 | 13 | 029K0050 |
| CD0911 | 9/29/2009 | 21.0 | 20.6667 | 19.8162 | 18.9657 | 21.5172 | 22.3676 | 47 | 13 | 029K0050 |
| CD0912 | 10/15/2009 | 24.0 | 21.5000 | 19.6945 | 17.8889 | 23.3055 | 25.1111 | 47 | 13 | 029K0050 |
| CD0913 | 11/27/2009 | 35.0 | 24.2000 | 17.9634 | 11.7268 | 30.4366 | 36.6732 | 47 | 13 | 049K0305 |
| CD0915 | 12/24/2009 | 22.9 | 23.9833 | 18.3800 | 12.7766 | 29.5867 | 35.1901 | 47 | 13 | 049K0305 |
| CD1002 | 1/26/2010 | 31.2 | 25.0143 | 19.2173 | 13.4204 | 30.8112 | 36.6082 | 47 | 13 | 079K0011 |
| CD1003 | 2/16/2010 | 19.1 | 24.2750 | 18.5151 | 12.7552 | 30.0349 | 35.7948 | 47 | 13 | 079K0011 |
| CD1004 | 3/23/2010 | 24.8 | 24.3333 | 18.9426 | 13.5519 | 29.7241 | 35.1148 | 47 | 13 | 049K0305 |
| CD1006 | 4/30/2010 | 37.9 | 25.6900 | 19.0389 | 12.3879 | 32.3411 | 38.9921 | 47 | 13 | 049K0305 |
| CD1007 | 5/27/2010 | 9.6 | 24.2273 | 16.2681 | 8.3090 | 32.1864 | 40.1456 | 47 | 13 | 049K0305 |
| CD1009 | 6/21/2010 | 38.3 | 25.4000 | 16.7923 | 8.1846 | 34.0077 | 42.6154 | 47 | 13 | 049K0305 |
| CD1010 | 7/29/2010 | 41.9 | 26.6692 | 17.2426 | 7.8160 | 36.0958 | 45.5224 | 47 | 13 | 079K0011 |
| CD1011 | 8/17/2010 | 11.9 | 25.6143 | 15.7347 | 5.8551 | 35.4939 | 45.3734 | 47 | 13 | 079K0011 |
| CD1012 | 9/29/2010 | 10.8 | 24.6267 | 14.3668 | 4.1069 | 34.8865 | 45.1464 | 47 | 13 | 079K0011 |
| CD1013 | 10/26/2010 | 20.7 | 24.3813 | 14.4208 | 4.4603 | 34.3417 | 44.3022 | 47 | 13 | 099K0202 |
| CD1014 | 11/16/2010 | 31.0 | 24.7706 | 14.9937 | 5.2168 | 34.5475 | 44.3243 | 47 | 13 | 099K0202 |
| CD1015 | 12/21/2010 | 41.9 | 25.7222 | 15.4137 | 5.1052 | 36.0307 | 46.3392 | 47 | 13 | 099K0202 |
| CD1101 | 1/27/2011 | 31.5 | 26.0263 | 15.9209 | 5.8156 | 36.1317 | 46.2371 | 47 | 13 | 099K0202 |
| CD1102 | 2/10/2011 | 19.9 | 25.7200 | 15.7892 | 5.8584 | 35.6508 | 45.5816 | 47 | 13 | 099K0202 |

Environmental Enterprises USA, Inc.

APPENDIX D

BIOMONITORING CHAIN OF CUSTODY RECORD

Page 1 of 1

COC No. **BULKSED-021111-EEUSA**

| | | | | | |
|--|--|--|--|--|--|
| Client: TVA | Environmental Enterprises USA, Inc. 58485 Pearl Acres Road, Suite D Slidell, LA 70461 Attn: David L. Daniel Office 800.966.2788 Cell 985.707.5442 | | | | |
| Project Name: KIF Ash Toxicity Study | | | | | |
| Date of Sample Collection: 02/07/11, 02/08/11, 02/09/11 | | | | | |
| Location: CRM0.0, CRM1.5, CRM2.0, CRM2.5, CRM3.0, CRM3.5, CRM4.0, CRM4.5, CRM6.5, CRM7.5 | | | | | |
| Collected By: R. Josefczyk (RSI), L. Jackson (TVA), R. Vance (RSI), E. Arnold (RSI), M. Greer (RSI), D. Mathis (RSI) | | | | | |

| Field Identification / Sample Description | Grab/ Comp | Collection Date/Time | Number of Containers & Volume Collected | Depth (ft) | Rain Event? (Mark as Appropriate) | | | Laboratory Use (as applicable). | | | | |
|--|------------|----------------------|---|----------------|---|---------------------------------------|-------|---------------------------------|--------------------|-----------------|--|--|
| | | | | | If Yes, inches | No | Trace | Log # | Arrival Temp. (°C) | By | | |
| Example: BULKSED-CRM0.0-EEUSA | — | Date | Time | — | — | — | — | E-087-11 | 1.0 ± 1.1 | 0-0 | | |
| BULKSED-CRM0.0-EEUSA | C | 02/07/11 | 1014 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | 1320/1430 | Sediment | | |
| BULKSED-CRM1.5-EEUSA | G | 02/07/11 | 1243 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | 1320/1305 | “ | | |
| BULKSED-CRM2.0-EEUSA | G | 02/07/11 | 0955 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | 1515/1345 | “ | | |
| BULKSED-CRM2.5-EEUSA | G | 02/07/11 | 1340 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | 1355/1520 | “ | | |
| BULKSED-CRM3.0-EEUSA | G | 02/08/11 | 0921 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | 1530/1610 | “ | | |
| BULKSED-CRM3.5-EEUSA | G | 02/08/11 | 1000 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | E-092-11 | 0.9 | 0-0 | | |
| BULKSED-CRM4.0-EEUSA | G | 02/08/11 | 1235 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | E-093-11 | 0.8 | | |
| BULKSED-CRM4.5-EEUSA | G | 02/08/11 | 1058 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | E-094-11 | 1.9 | | |
| BULKSED-CLINCHREFERENCE-EEUSA | C | 02/09/11 | 0940 | 32 (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | E-095-11 | 0.8, 2, 3, 4, 6 | | |
| Sample Custody – Fill In From Top Down | | | | | Date/Time | Received By (Signature)/ Affiliation: | | | Date/Time | | | |
| Relinquished By (Signature)/Affiliation: | | Date/Time | | | Received By (Signature)/ Affiliation: | | | Date/Time | | | | |
| <i>Ramon Joseph / RJS</i> <i>150210, 021111</i> | | 02/07/11 / 0920 | | | <i>Kotin Johnson / TVA</i> <i>02/08/11 / 100</i> | | | 02/08/11 / 0920 | | | | |
| <i>Ramon Joseph / RJS</i> <i>150210, 021111</i> | | 02/08/11 / 100 | | | <i>Darrell B. Smith / DBS</i> <i>02/09/11 / 0940</i> | | | 02/09/11 / 1240 | | | | |

Associated UPS Tracking #s (if applicable):

Laptopbook: TUA-K1F-N76D YUX-003

CHAINS OF CHORDS RECORD

Page 1 of 1

COK No. BULKSW-0211-EUSN

| | | | | | | | | | | | | |
|---|--|----------------------|---|-----------|-----------------------------------|--------------------------------|-------|-------|--------------------|-----|------|------------|
| Client: TVA | Environmental Enterprises USA, Inc. 58485 Pearl Acres Road, Suite D Slidell, LA 70461 Attn: David L. Daniel Office 800.966.2788 Cell 985.707.5442 | | | | | | | | | | | |
| Project Name: KIF Ash Toxicity Study | Date of Sample Collection: 02/11/2011 | | | | | | | | | | | |
| Location: CRM7.0 | TVA-KIF-NTC-TOX-ΦΦ3 | | | | | | | | | | | |
| Collected By: M. Greer (RSI), C.L. Jackson (TVA), R. Josefczyk (RSI), A. Jones (RSI), T. Wall (ESI), S.C. Greer (RSI), C. Tauras (RSI), J. Van Kirk (ESI) | General Comments: Bulk Clinch River reference water for sediment toxicity study collected in 2.5 gallon (10L) cubitainers. | | | | | | | | | | | |
| Description | Grab/ Comp. | Collection Date/Time | Number of Containers & Volume Collected | Depth (m) | Rain Event? (Mark as Appropriate) | Laboratory Use (as applicable) | | | | | | |
| | — | Date Time | — | — | Yes If Yes, Inches | No | Trace | Log # | Arrival Temp. (°C) | By | Time | Appearance |
| BULKSW-CRM7.0-EEUSA | G | 02/11/2011 | 1054 (2) 10L cubitainers | 8.10 | NA | NA | NA | | 2.9°C | 010 | 1300 | c/leaf r |
| | | | | | | | | | | | | |
| | | | | | | | | | | | | |
| | | | | | | | | | | | | |
| | | | | | | | | | | | | |
| | | | | | | | | | | | | |
| Sample Custody – Fill In From Top Down | | | | | | | | | | | | |
| Relinquished By (Signature)/Affiliation: | Received By (Signature)/Affiliation: | | | | | | | | | | | |
| <i>John H. Gray (RSI)</i> | <i>Karen Goss / TVA</i> | | | | | | | | | | | |
| Associated UPS Tracking #'s (if applicable): | <i>02/11/11 / 1230</i> | | | | | | | | | | | |
| | <i>02/11/11 / 1300</i> | | | | | | | | | | | |
| | <i>02/11/11 / 1230</i> | | | | | | | | | | | |
| | <i>02/11/11 / 1240</i> | | | | | | | | | | | |

COURIER TRANSPORT DOCUMENTATION

DATE: 02/11/2011

COURIER COMPANY:

Sonic Subcontractor

| From: | To: |
|--|--|
| TVA c/o Katie Gassaway 189 Lakeshore Drive Harriman, TN 37748 865-803-4503 | Environmental Enterprises USA, Inc. 58485 Pearl Acres Road, Suite D Slidell, LA 70461 Attn: David L. Daniel 1-800-966-2788 985-707-5442 |

| No. of Items: | Description: |
|---------------|---|
| 7 | Cooler(s) taped and custody sealed. Coolers are batched 1 of 1, 2 of 2, and 4 of 4 containing water and sediment. |

Shippers Name/Company: Sonic ^{KG02111} Brian Wooten Katie Gassaway/Ti

Date / Time: 02/11/11 / 1400

Courier Signature/Company: R. Wooten

Date / Time: 2-11-11 1400

Receipt Signature/Company: R. J. Daniel / EE USA

Date / Time: 2/12/11 1240 David Daniel

Corresponding Chains of Custody:

| | |
|----------------------------------|--|
| BULKSED-021111-EEUSA page 1 of 1 | |
| BULKSW-021111-EEUSA page 1 of 1 | |
| | |
| | |
| | |
| | |
| | |

BIOMONITORING CHAIN OF CUSTODY RECORD

Page 1 of 1

COC No. BULKSED-021111-EEUSA

| | | | | | | | | | | |
|--|---|--|--|--|--|--|--|--|--|--|
| Client: TVA | <i>Non-Record copy</i> | | | | | | | | | |
| Project Name: KIF Ash Toxicity Study | Delivered By (Circle One): <input checked="" type="checkbox"/> FedEx <input type="checkbox"/> UPS <input type="checkbox"/> Bus <input type="checkbox"/> Client <input checked="" type="checkbox"/> Courier | | | | | | | | | |
| Date of Sample Collection: 02/07/11, 02/08/11, 02/09/11 | Other (specify): _____ | | | | | | | | | |
| Location: CRM0.0, CRM1.5, CRM2.0, CRM2.5, CRM3.0, CRM3.5, CRM4.0, CRM4.5, CRM6.5, CRM7.5 | General Comments: Homogenized sediment from the Clinch River | | | | | | | | | |
| Collected By: R. Josefczyk (RSI), L. Jackson (TVA), R. Vance (RSI), E. Arnold (RSI), M. Greer (RSI), D. Mathis (RSI) | "CLINCHREFERENCE" is a composite sample of CRM6.5 and CRM7.5. | | | | | | | | | |

| Field Identification / Sample Description | Grab/ Comp | Collection Date/Time | Number of Containers & Volume Collected | Depth (ft) | Rain Event? (Mark as Appropriate) | Laboratory Use (as applicable) | | | | | | |
|---|------------|----------------------|---|------------|-----------------------------------|--------------------------------|-------|----------|--------------------|-----|------|------------|
| Example: BULKSED-CRM0.0-EEUSA | | Date Time | | | Yes If Yes - Inches | No | Trace | Log # | Arrival Temp. (°C) | By | Time | Appearance |
| BULKSED-CRM0.0-EEUSA 0 | G | 02/07/11 1014 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | | | | | |
| BULKSED-CRM1.5-EEUSA 2 | G | 02/07/11 1243 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | E-098-II | 1.1 | 020 | 1505 | sediment |
| BULKSED-CRM2.0-EEUSA 2 | G | 02/07/11 0955 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | E-089-II | 0.8 | 020 | 1515 | " |
| BULKSED-CRM2.5-EEUSA 2 | G | 02/07/11 1340 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | E-092-II | 0.7 | 020 | 1520 | " |
| BULKSED-CRM3.0-EEUSA 2 | G | 02/08/11 0921 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | E-091-II | 1.3 | 020 | 1530 | 1 |
| BULKSED-CRM3.5-EEUSA 0 | G | 02/08/11 1000 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | | | | | |
| BULKSED-CRM4.0-EEUSA 0 | G | 02/08/11 1235 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | | | | | |
| BULKSED-CRM4.5-EEUSA 0 | G | 02/08/11 1058 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | | | | | |
| BULKSED-CLINCHREFERENCE-EEUSA/2 | C | 02/09/11 0940 | 3L (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | E-095-II | 1.6 | 020 | 1545 | " |

Sample Custody – Fill In From Top Down

| Relinquished By (Signature)/Affiliation: | Date/Time | Received By (Signature)/Affiliation: | Date/Time |
|--|---------------|--------------------------------------|---------------|
| <i>John Jackson, TVA</i> | 02/07/11 1014 | <i>John Jackson, TVA</i> | 02/08/11 0920 |
| <i>John Jackson, TVA</i> | 02/08/11 1000 | <i>John Jackson, TVA</i> | 02/08/11 1240 |

Associated UPS Tracking #'s (if applicable):

A/af

Logbooks: TVA-K1F-NTC-TCX-001, 0002, 0003

BIOMONITORING CHAIN OF CUSTODY RECORD

Page 1 of 1

COC No. BULKSED-021111-EEUSA

| | | | | | | |
|--|--|--|--|--|--|--|
| Client: TVA | Environmental Enterprises USA, Inc. 58485 Pearl Acres Road, Suite D Slidell, LA 70461 Attn: David L. Daniel Office 800.966.2788 Cell 985.707.5442 | | | | | |
| Project Name: K1F Ash Toxicity Study | | | | | | |
| Date of Sample Collection: 02/07/11, 02/08/11, 02/09/11 | | | | | | |
| Location: CRM0.0, CRM1.5, CRM2.0, CRM2.5, CRM3.0, CRM3.5, CRM4.0, CRM4.5, CRM6.5, CRM7.5 | | | | | | |
| Collected By: R. Josefczyk (RSI), L. Jackson (TVA), R. Vance (RSI), E. Arnold (RSI), M. Greer (RSI), D. Mathis (RSI) | | | | | | |

| Field Identification / Sample Description | Grab/ Comp | Collection Date/Time | Number of Containers & Volume Collected | Depth (ft) | (Mark as Appropriate) | Laboratory Use (as applicable) | | | | | | |
|---|------------|----------------------|---|----------------|-----------------------|--------------------------------|-----|-------|-------|--------------------|-----|------|
| | | | | | | If Yes, Rain Event? Inches | No | Trace | Log # | Arrival Temp. (°C) | By | Time |
| Example: BULKSED-CRM0.0-EEUSA | — | Date | Time | — | — | — | — | — | — | — | — | — |
| BULKSED-CRM0.0-EEUSA | G | 02/07/11 | 1014 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | N/A | N/A | N/A | N/A |
| BULKSED-CRM1.5-EEUSA | G | 02/07/11 | 1243 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | N/A | N/A | N/A | N/A |
| BULKSED-CRM2.0-EEUSA | G | 02/07/11 | 0955 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | N/A | N/A | N/A | N/A |
| BULKSED-CRM2.5-EEUSA | G | 02/07/11 | 1340 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | N/A | N/A | N/A | N/A |
| BULKSED-CRM3.0-EEUSA | G | 02/08/11 | 0921 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | N/A | N/A | N/A | N/A |
| BULKSED-CRM3.5-EEUSA | G | 02/08/11 | 1000 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | N/A | N/A | N/A | N/A |
| BULKSED-CRM4.0-EEUSA | G | 02/08/11 | 1235 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | N/A | — | 0.8 | D.0 |
| BULKSED-CRM4.5-EEUSA | G | 02/08/11 | 1058 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | N/A | E-094-11 | 1.9 | D.0 |
| BULKSED-CLINCHREFERENCE-EEUSA | C | 02/09/11 | 0940 | 32 (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | N/A | E-095-11 | | |

Sample Custody - Fill In From Top Down

| Relinquished By (Signature)/Affiliation: | Date/Time | Received By (Signature)/ Affiliation: | Date/Time |
|--|--------------------------------|---|--------------------------------|
| Ramon Joseph /K251 Ramon Joseph /TV | 02/07/11 0920 02/07/11 1100 | Kotie Yannas / TVA Kotie Yannas / TV | 02/08/11 0920 02/08/11 1240 |
| Patricia Jones / TVA | 02/08/11 1058 | | |

Associated UPS Tracking #'s (if applicable):

32

BIOMONITORING CHAIN OF CUSTODY RECORD

Page 1 of 1

COC No. BULKSED-021111-EEUSA

| | | | | | | |
|--|--|--|--|--|--|---|
| Client: TVA | Environmental Enterprises USA, Inc. 58485 Pearl Acres Road, Suite D Slidell, LA 70461 Attn: David L. Daniel Office 800.966.2788 Cell 985.707.5442 | | | | | Delivered By (Circle One): <input checked="" type="checkbox"/> FedEx <input type="checkbox"/> UPS <input type="checkbox"/> Bus <input type="checkbox"/> Client <input checked="" type="checkbox"/> Courier |
| Project Name: KIF Ash Toxicity Study | | | | | | Other (specify): _____ |
| Date of Sample Collection: 02/07/11, 02/08/11, 02/09/11 | | | | | | General Comments: Homogenized sediment from the Clinch River |
| Location: CRM0.0, CRM1.5, CRM2.0, CRM2.5, CRM3.0, CRM3.5, CRM4.0, CRM4.5, CRM6.5, CRM7.5 | | | | | | "CLINCHREFERENCE" is a composite sample of CRM6.5 and CRM7.5. |
| Collected By: R. Josefczyk (RSI), L. Jackson (TVA), R. Vance (RSI), E. Arnold (RSI), M. Greer (RSI), D. Mathis (RSI) | | | | | | |

| Field Identification / Sample Description | Grab/ Comp | Collection Date/Time | Number of Containers & Volume Collected | Depth (ft) | Rain Event? (Mark as Appropriate) | Laboratory Use (as applicable) |
|---|------------|----------------------|---|----------------|-----------------------------------|---|
| Example: BULKSED-CRM0.0-EEUSA | — | Date | Time | — | Yes If Yes, Inches | No Trace Log # Arrival Temp. (°C) By Time Appearance |
| BULKSED-CRM0.0-EEUSA | Z | 02/07/11 | 1014 | (4) 1000 mL | 0.0-0.5 N/A | N/A N/A 1.1 1430 sediment |
| BULKSED-CRM1.5-EEUSA | D | 02/07/11 | 1243 | (4) 1000 mL | 0.0-0.5 N/A | N/A N/A |
| BULKSED-CRM2.0-EEUSA | D | 02/07/11 | 0955 | (4) 1000 mL | 0.0-0.5 N/A | N/A N/A |
| BULKSED-CRM2.5-EEUSA | D | 02/07/11 | 1340 | (4) 1000 mL | 0.0-0.5 N/A | N/A N/A |
| BULKSED-CRM3.0-EEUSA | D | 02/08/11 | 0921 | (4) 1000 mL | 0.0-0.5 N/A | N/A N/A |
| BULKSED-CRM3.5-EEUSA | D | 02/08/11 | 1000 | (4) 1000 mL | 0.0-0.5 N/A | N/A N/A |
| BULKSED-CRM4.0-EEUSA | D | 02/08/11 | 1235 | (4) 1000 mL | 0.0-0.5 N/A | N/A N/A |
| BULKSED-CRM4.5-EEUSA | D | 02/08/11 | 1058 | (4) 1000 mL | 0.0-0.5 N/A | N/A N/A |
| BULKSED-CLINCHREFERENCE-EEUSA | Y | 02/09/11 | 0940 | 32 (4) 1000 mL | 0.0-0.5 N/A | N/A N/A E-095-# 2.3 200 1450 sediment |

Sample Custody - Fill In From Top Down

| Relinquished By (Signature)/Affiliation: | Date/Time | Received By (Signature)/Affiliation: | Date/Time |
|--|---------------------------------|--------------------------------------|-----------------|
| <i>Demonee Josefczyk / RSI</i> | 02/07/11 / 0920 150211 / 100 | <i>Kotini Johnson / TVA</i> | 02/08/11 / 0920 |
| <i>Kits Johnson / TVA</i> | 02/08/11 / 100 | <i>Bob Johnson</i> | 02/08/11 / 1240 |

Associated UPS Tracking #'s (if applicable):

S/

Logbooks: TVA-K1F-NTC-TCX-001, 002, 003

BIOMONITORING CHAIN OF CUSTODY RECORD

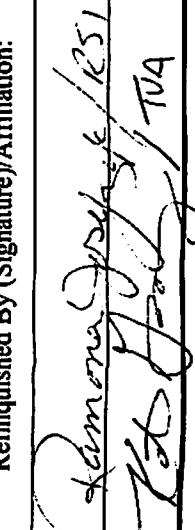
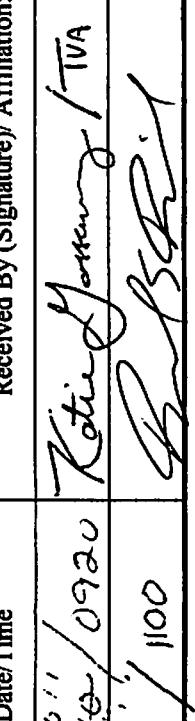
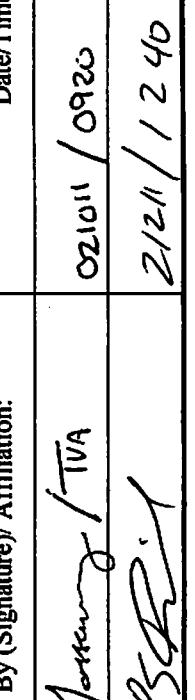
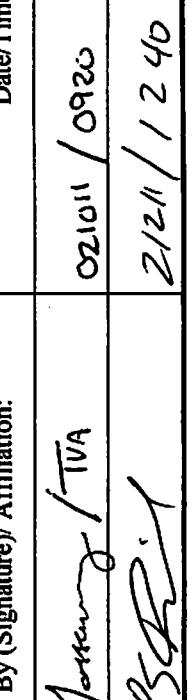
Page 1 of 1

COC No. BULKSED-021111-EEUSA

| | | | | | | | | | | |
|--|--|--|--|--|--|--|--|--|--|--|
| Client: TVA | Environmental Enterprises USA, Inc. 58485 Pearl Acres Road, Suite D Slidell, LA 70461 Attn: David L. Daniel Office 800.966.2788 Cell 985.707.5442 | | | | | | | | | |
| Project Name: KIF Ash Toxicity Study | Delivered By (Circle One): <input checked="" type="radio"/> Courier | | | | | | | | | |
| Date of Sample Collection: 02/07/11, 02/08/11, 02/09/11 | FedEx UPS Bus Client Other (specify): _____ | | | | | | | | | |
| Location: CRM0.0, CRM1.5, CRM2.0, CRM3.0, CRM3.5, CRM4.0, CRM4.5, CRM6.5, CRM7.5 | General Comments: Homogenized sediment from the Clinch River | | | | | | | | | |
| Collected By: R. Joscfczyk (RSI), L. Jackson (TVA), R. Vance (RSI), E. Arnold (RSI), M. Greer (RSI), D. Mathis (RSI) | "CLINCHREFERENCE" is a composite sample of CRM6.5 and CRM7.5. | | | | | | | | | |

| Field Identification / Sample Description | Grab/ Comp | Collection Date/Time | Number of Containers & Volume Collected | Depth (ft) | Rain Event? (Mark as Appropriate) | Laboratory Use (as applicable) | | | | | | |
|---|------------|----------------------|---|----------------|-----------------------------------|--------------------------------|-----|-------|----------|--------------------|----|---------------|
| Example BULKSED-CRM6.0-EEUSA | — | Date | Time | — | Yes | If Yes, Inches | No | Trace | Log # | Arrival Temp. (°C) | By | Time |
| BULKSED-CRM0.0-EEUSA | Z | 02/07/11 | 1014 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | E-087-11 | 1.0 | 09 | 1330 sediment |
| BULKSED-CRM1.5-EEUSA | Z | 02/07/11 | 1243 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | E-098-11 | 0.3 | 09 | 1340 " |
| BULKSED-CRM2.0-EEUSA | Z | 02/07/11 | 0955 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | E-089-11 | 1.9 | 09 | 1345 " |
| BULKSED-CRM2.5-EEUSA | Z | 02/07/11 | 1340 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | E-090-11 | 0.9 | 09 | 1355 " |
| BULKSED-CRM3.0-EEUSA | Z | 02/08/11 | 0921 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | E-091-4 | 1.8 | 09 | 1410 " |
| BULKSED-CRM3.5-EEUSA | 4 | 02/08/11 | 1000 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | E-092-11 | 0.9 | 09 | 1425 " |
| BULKSED-CRM4.0-EEUSA | 0 | 02/08/11 | 1235 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | | | | |
| BULKSED-CRM4.5-EEUSA | 0 | 02/08/11 | 1058 | (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | E-093-11 | 0.8 | 09 | 1320 sediment |
| BULKSED-CLINCHREFERENCE-EEUSA | 6 | 02/09/11 | 0940 | 32 (4) 1000 mL | 0.0-0.5 | N/A | N/A | N/A | E-094-11 | 0.8 | 09 | 1320 sediment |

Sample Custody - Fill In From Top Down

| Relinquished By (Signature)/Affiliation: | Date/Time | Received By (Signature)/Affiliation: | Date/Time |
|---|-----------------|---|-----------------|
|  Karen Joscfczyk / R51 TVA | 02/07/11 / 0920 |  Katie Yost / TVA | 02/07/11 / 0920 |
|  Bob Schaefer / TVA | 02/08/11 / 1000 |  Bob Schaefer / TVA | 02/08/11 / 1240 |

Associated UPS Tracking #'s (if applicable):

ED/00