

2. METHODS

2.1 7-D TOPSMELT (*ATHERINOPS AFFINIS*) SURVIVAL AND GROWTH TOXICITY TEST

A static-renewal 7-d toxicity test and reference toxicant test using topsmelt (*A. affinis*) was conducted in accordance with EVS Environment Consultants Standard Operating Procedures (SOP) 1100-5 (EVS, 2004) with modifications based on U.S. Environmental Protection Agency (USEPA, 1995). Test conditions and methods are summarized in Table 1.

This 7-day test exposes topsmelt larvae to different concentrations of a given sample. Fish are fed on a daily basis and both survival and growth endpoints are measured at test termination. These observations are assessed in comparison to the pooled negative and brine controls.

2.2 ECHINODERM (*DENDRASTER EXCENTRICUS*) FERTILIZATION TOXICITY TEST

The echinoderm (*Dendraster excentricus*) fertilization toxicity test was conducted in accordance with Environment Canada (1992 with 1997 amendments). Test conditions and methods summarized in Table 2.

This fertilization test involves exposing echinoderm sperm to a series of test concentrations for ten minutes, echinoderm eggs are then added allowing fertilization to occur for ten minutes. Following the ten minutes exposure time the eggs are preserved and the number of fertilized and unfertilized eggs in each replicate are counted. These observations are assessed in comparison to the pooled negative and brine controls.

2.3 STATISTICAL ANALYSIS

Statistical analyses for all tests were conducted using the computer software program TOXCALC (version 5.0.23; Tidepool Scientific Software, 1994).

2.4 QUALITY ASSURANCE/QUALITY CONTROL (QA/QC)

This study followed a comprehensive QA/QC Program to ensure full documentation and minimize possible errors in computation and reporting of results. The following general

QA/QC guidelines were applied in this test: use of negative controls, use of positive controls, use of brine controls, replication, instrument calibration, water quality maintenance and record-keeping, and use of standard operating procedures (SOPs). To ensure the highest quality of data and reporting, all data and statistical analyses were reviewed by a member of our QA/QC Committee prior to reporting the results.

Table 1. 7-d Topsmelt (*Atherinops affinis*) survival and growth toxicity test methods

TEST PARAMETER	TEST CONDITION
Test type	Static-renewal
Test duration	7 d
Test chamber	600-mL beaker
Test solution volume	200 mL
Number of replicate chambers per treatment	5
Number of organisms per test chamber	5
Age of test organisms at test initiation	10 d
Food	Newly hatched <i>Artemia</i> nauplii (<24 hours old)
Feeding Regime	Fed 0.5 mL/ beaker twice daily of concentrated nauplii suspension (prepared to provide 200 nauplii in 0.5 mL); no feeding at test termination
Sample manipulations (e.g. pre-aeration, pH adjustment, filtration)	Salinity-adjusted
Control/dilution water	UV-sterilized and 0.5µm-filtered natural sea water from Vancouver Aquarium, BC
Dilutions	4.4, 9.0, 18.0, 36.0, 71.0% (v/v)
Renewal of dilutions	Daily
Aeration	None
Water quality parameters and frequency	Temperature, pH, dissolved oxygen, and salinity daily
Temperature	20 ± 1°C
Salinity	30 ± 2 (sample adjusted with hypersaline brine [HSB]. Preparation of HSB and salinity adjustment as per EC guidance document on salinity adjustment –July 1997)
Lighting	Overhead full-spectrum fluorescent lights; 538 – 1076 lux; 16:8 light:dark photoperiod
Reference toxicant	Initiated concurrently with sample using copper to generate LC50 and IC50 values; results compared to lab mean ± 2 SD
Endpoints	Survival and growth (dry weight)
Test validity	≥ 80% mean control survival; ≥ 0.85 mg/fish mean dry weight for surviving control fish
Reference protocol	US EPA (1995), EPA/600/R-95/136

Table 2. Echinoderm (*Dendraster excentricus*) fertilization toxicity test methods

TEST PARAMETER	TEST CONDITION
Test type	Static
Test duration	10:10 min
Test chamber	16 X 125 mm test tubes
Test solution volume	10 mL
Number of replicate chambers per treatment	4
Number of eggs per test chamber	2000
Age of test organisms	< 4 hours after spawning
Sample manipulations (e.g. pre-aeration, pH adjustment, filtration)	Salinity adjusted
Control/dilution water	UV-sterilized and 0.5µm-filtered natural sea water from Vancouver Aquarium, BC
Dilutions	4.7, 9.4, 18.8, 37.5, 75.1% (v/v)
Renewal of dilutions	None
Aeration	None during testing
Water quality parameters and frequency	Temperature, pH, dissolved oxygen, and salinity
Temperature	15 ± 1°C
Salinity	30 ± 2 (sample adjusted with hypersaline brine [HSB]. Preparation of HSB and salinity adjustment as per EC guidance document on salinity adjustment –July 1997)
Lighting	Ambient laboratory illumination (moderate intensity)
Reference toxicant	Initiated concurrently with test; same test methods as above using SDS to generate an EC50 value; results compared to lab mean ± 2 SD
Endpoint	Fertilization of eggs
Test validity	≥ 50% and ≤ 100% mean control fertilization
Reference protocols	Environment Canada (1992), (EPS/1/RM/27 with 1997 amendments)

3. RESULTS

3.1 7-D TOPSMELT (*ATHERINOPS AFFINIS*) SURVIVAL AND GROWTH TOXICITY TEST

The test results are summarized in Table 1 and the raw statistical analyses are provided in Appendix A.

The highest concentration tested was approximately 71.0% due to salinity adjustment. The mean survival in both the negative and brine controls was 100%. Mean dry weight in the pooled controls was 0.95mg. The negative and brine controls were not significantly different for the growth and survival endpoints ($p=1.00$ and $p=0.72$, respectively).

The *A. affinis* survival and growth toxicity test showed no adverse effects on survival or growth in all tested concentrations relative to the pooled controls ($p\leq 0.05$). For the survival and growth endpoints the NOEC was 71.0, and the LOEC was $>71.0\%$ (v/v). The LC50 for survival was $>71.0\%$ (v/v). The IC50 and IC25 for growth were both $>71.0\%$ (v/v).

3.2 ECHINODERM (*DENDRASTER EXCENTRICUS*) FERTILIZATION TOXICITY TEST

The test results are summarized in Table 2 and the raw statistical analyses are provided in Appendix B.

The highest concentration tested was 75.1% due to salinity adjustment. Mean fertilization in the pooled controls was 76.6%. The negative and brine controls were not significantly different ($p=0.68$).

The *D. excentricus* fertilization toxicity test exhibited adverse effects on egg fertilization in all except the lowest [4.7%(v/v)] test concentrations relative to the pooled controls ($p\leq 0.05$). The NOEC was 4.7 and LOEC was 9.4 %(v/v). The IC50 and IC25 (95% confidence limits) values were 50.7 (48.1 – 53.6) and 17.5 (11.6 – 22.6) %(v/v), respectively.

3.2 QUALITY ASSURANCE/QUALITY CONTROL (QA/QC)

The tests met all passing criteria for test validity as outlined in the respective protocols. Water quality parameters during the test were all within the acceptable range of values. Point estimates for the reference toxicant tests were all within the laboratory mean ± 2 standard deviations, indicating that the tests were within acceptable limits of variability.

Table 3. Summary of results for the 7-d Topsmelt (*Atherinops affinis*) survival and growth toxicity test

TEST CONCENTRATION (% v/v)	SURVIVAL (%)(MEAN \pm SD)	GROWTH (DRY WEIGHT MG) (MEAN \pm SD)
D-Control	100.0 \pm 0.0	0.93 \pm 0.06
Brine Control	100.0 \pm 0.0	0.96 \pm 0.18
Pooled Controls	100.0 \pm 0.0	0.95 \pm 0.13
4.4	100.0 \pm 0.0	1.06 \pm 0.26
9.0	100.0 \pm 0.0	0.87 \pm 0.06
18.0	100.0 \pm 0.0	0.97 \pm 0.17
36.0	96.0 \pm 8.9	0.92 \pm 0.13
71.0	100.0 \pm 0.0	0.87 \pm 0.16
TEST ENDPOINT	SURVIVAL (% v/v)	GROWTH (% v/v)
NOEC	71.0	71.0
LOEC	>71.0	>71.0
LC50	>71.0	na
IC50	na	>71.0
IC25	na	>71.0

*Indicates significant difference ($p \leq 0.05$) relative to the pooled controls.
SD – Standard Deviation; na – not applicable.

Table 4. Summary of results for the Echinoderm (*Dendraster excentricus*) fertilization toxicity test

TEST CONCENTRATION (% v/v)	PROPORTION FERTILIZED (%) (MEAN \pm SD)
Negative Control	77.0 \pm 2.4
Brine Control	76.2 \pm 2.5
Pooled Control	76.6 \pm 2.3
4.7	75.0 \pm 2.6
9.4	61.2 \pm 1.0*
18.8	57.0 \pm 1.8*
37.5	49.2 \pm 1.0*
75.1	25.8 \pm 1.5*
TEST ENDPOINT	PROPORTION FERTILIZED %(v/v)
NOEC	4.7
LOEC	9.4
IC50 (95% CL)	50.7 (48.1 – 53.6)
IC25 (95% CL)	17.5 (11.6 – 22.6)

*Indicates significant difference ($p \leq 0.05$) relative to the pooled controls.
SD – Standard Deviation; CL – Confidence Limits.

4. REFERENCES

- Environment Canada. 1992. Biological test method: fertilization of echinoids (sea urchins and sand dollars). Environmental Protection Series, Report EPS 1/RM/27, December 1992. Environment Canada, Conservation and Protection, Ottawa, ON. 68 pp + appendices. Amended November 1997.
- EVS (EVS Environment Consultants). 2004. Topsmelt (*Atherinops affinis*) 7-d larval survival and growth test. EVS SOP 1100-5. In: EVS Consultants Laboratory Standard Operating Procedures (SOP) Manual. Volume II: Water Toxicity Tests. EVS Environment Consultants, North Vancouver, BC.
- US EPA (U.S. Environmental Protection Agency). 2002. Short-term methods for estimating the chronic toxicity of effluents and receiving waters to marine and estuarine organisms. 3rd edition. US Environmental Protection Agency, Office of Water (4303T). US Environmental Protection Agency, Washington, DC. EPA/821/R-02/014. 464 pp.
- US EPA. 1995. Short term methods for estimating the chronic toxicity of effluents and receiving waters to marine and estuarine organisms. 2nd edition. US Environmental Protection Agency, Environmental Monitoring Systems Laboratory, Office of Research and Development, Washington, DC. EPA/600/R-95/136. 563 pp.
- Tidepool Scientific Software. 1994. TOXCALC: Comprehensive Toxicity Data Analysis and Database Software, Version 5.0.23. Tidepool Scientific Software, McKinleyville, CA. 80 pp.

APPENDIX A

Raw Data and Statistical Analyses:

Atherinops affinis

EVS ENVIRONMENT CONSULTANTS
7-d *Atherinops affinis* SURVIVAL AND GROWTH TEST DATA SUMMARY

Client Azimuth (Polans)
 EVS Project No. 09-0302-54
 EVS Work Order No. 0400384

EVS Analysts AWD, JxS
 Test Initiation Date Aug 27, 2004

Sample	Initial Sample	Refresh Samples	
	Day 0	Day 2	Day 4
Identification	G - Creek 082404	→	→
Amount Received	1 x 20 L	→	→
Date Collected	24-Aug-04	→	→
Date Received	27-Aug-04	→	→
Temperature (°C)	20.0 ^B 20.0	20.0 ^D 20.0	20.0 ^D 20.0
pH	7.6 ^B 8.1	7.6 ^D 8.2	7.5 ^D 8.3
DO (mg/L)	10.1 ^D 7.4	10.0 ^D 7.5	11.0 ^D 7.5 ²
Conductivity (μmhos/cm)	9230	—	—
Salinity (ppt)	5.2 ^D 28	5.1 ^D 28	5.2 ^D 28
Ammonia (mg/L N)			
Chlorine (mg/L Cl)			
Other	D salinity adj to 30 ± 2 ppt w/ 90 ppt HSB		

DILUTION/CONTROL WATER (initial water quality)

Water Type UV Sterilized S.W.
 Temperature (°C) 20.0
 pH 7.8
 Dissolved Oxygen (mg/L) 7.4
 Salinity 28

TEST CONDITIONS

Temperature Range (°C) 20.0 - 21.0
 pH Range 7.8 - 8.3
 Dissolved Oxygen Range (mg/L) 6.4 - 7.6
 Salinity (ppt) 28
 Photoperiod (L:D h) 16:8
 Aeration Provided? No
 Other —

TEST SPECIES INFORMATION

Source ABS (Aquatic Bio Systems)
 Date Received Aug 26, 2004
 Age (on Day 0) 10-d
 Reference Toxicant Copper
 Current Reference Toxicant Result (incl. 95% CL)

Reference Toxicant Test Date Aug 27, 2004
 7-d survival LC50 121 (107-137) ⁸⁵⁻¹⁵⁰ μg/L
 7-d growth IC50 128 (111-153) ⁸⁵⁻¹⁵⁰ μg/L

Reference Toxicant Warning Limits (mean ± 2SD) and CV

7-d survival LC50 132 ± 48 ⁸⁵⁻¹⁵⁰ μg/L CV: 18%
 7-d growth IC50 138 ± 51 ⁸⁵⁻¹⁵⁰ μg/L CV: 20%

TEST RESULTS

Endpoint	Conc. Units	NOEC	LOEC	LC50 (95% CL)	IC50 (95% CL)	IC25 (95% CL)
Survival	% (✓/✓)	71	> 71	> 71		
Growth		71	> 71		> 71	> 71

Other _____

Data Verified By Quilley Date Verified Oct-27/04

EVS ENVIRONMENT CONSULTANTS

7-d *Atherinops affinis* SURVIVAL AND GROWTH TEST – WATER QUALITY DATA

Client Azimuth (Polaris)
 EVS Project No. 09-0302-54
 EVS Work Order No. 0400384

Sample ID G-Creek @ 082404
 Test Initiation Date/Time Aug 27, 2004 @ 1230 L
 Source/Date Received ABS / Aug 28, 2004

Concentration ‰ (v/v)	Temperature (°C)													
	0	old	1 New	2		3		4		5		6		7
Control	20.0	20.5	20.0	20.5	20.0	20.5	20.0	20.5	21.0	20.0	20.5	20.0	20.0	20.5
Brine Control	20.0	20.5	20.0	20.5	20.0	20.5	20.0	20.5	21.0	20.0	20.5	20.0	20.5	20.0
4.4	20.0	20.5	20.0	20.5	20.0	20.5	20.0	20.5	21.0	20.0	20.5	20.0	20.5	20.5
9.0	20.0	20.5	20.0	20.5	20.0	20.5	20.0	20.5	21.0	20.0	20.5	20.0	20.5	20.5
18	20.0	20.5	20.0	20.5	20.0	20.5	20.0	20.5	21.0	20.0	20.5	20.0	20.5	20.5
36	20.0	20.5	20.0	20.5	20.0	20.5	20.0	20.5	21.0	20.0	20.5	20.0	20.5	20.5
71	20.0	20.5	20.0	20.5	20.0	20.5	20.0	20.5	21.0	20.0	20.5	20.0	20.5	20.5
Tech. Initials	~	~	~	~	~	~	~	~	~	~	~	~	~	~

Concentration ‰ (v/v)	pH													
	0	1		2		3		4		5		6		7
Control	7.8	8.0	8.0	8.1	7.9	8.0	7.9	8.0	7.9	7.9	7.9	7.9	7.9	7.9
Brine Control	7.8	8.0	8.0	8.1	7.9	8.0	7.9	8.0	7.9	7.9	7.9	7.9	7.9	7.9
4.4	7.7	8.1	8.1	8.1	8.1	8.0	8.1	8.0	8.1	8.0	8.1	7.9	7.9	8.0
9.0	7.8	8.3	8.1	8.2	8.1	8.0	8.2	8.0	8.1	8.1	8.1	8.0	7.9	8.0
18	7.8	8.2	8.1	8.2	8.2	8.0	8.2	8.0	8.1	8.1	8.1	8.0	8.0	8.1
36	7.9	8.3	8.2	8.2	8.2	8.0	8.2	8.1	8.2	8.1	8.1	8.1	8.0	8.1
71	8.1	8.3	8.3	8.3	8.3	8.0	8.3	8.2	8.3	8.3	8.2	8.2	8.1	8.1
Tech. Initials	~	~	~	~	~	~	~	~	~	~	~	~	~	~

WQ Instruments Used: Temp. Calibrated Hg Thermometer pH H-A-52
 Comments _____

Test Set Up By AWB Date Verified By Qacph Date Verified Oct 20/04

EVS ENVIRONMENT CONSULTANTS

7-d *Atherinops affinis* SURVIVAL AND GROWTH TEST – WATER QUALITY DATA

Client Azimuth (Polaris)

Sample ID G-Creek 082404

EVS Project No. 09-0302-54

Test Initiation Date/Time Aug 26th, 2004 @ 1230h

EVS Work Order No. 0400384

Source/Date Received ABS / Aug 26th, 2004

Concentration ‰ (ppt)	Salinity (ppt)							
	0	1	2	3	4	5	6	7
Control	28	28	28	28	28	28	28	28
Brine Control	28	28	28	28	28	28	28	28
4.4	28	28	28	28	28	28	28	28
9.0	28	28	28	28	28	28	28	28
18	28	28	28	28	28	28	28	28
36	28	28	28	28	28	28	28	28
71	28	28	28	28	28	28	28	28
Tech. Initials	~	~	~	~	~	~	~	~

Concentration ‰ (ppt)	Dissolved Oxygen (mg/L)													
	0	1	2	3	4	5	6	7	0	1	2	3	4	5
Control	7.4	6.5	7.5	6.9	7.5	7.0	7.5	7.0	7.5	6.4	7.4	7.6	6.5	6.6
Brine Control	7.4	6.6	7.5	6.8	7.5	6.9	7.5	6.9	7.5	6.6	7.4	7.5	6.4	6.4
4.4	7.4	6.7	7.5	6.7	7.5	6.9	7.5	6.9	7.5	6.7	7.5	7.6	6.6	6.5
9.0	7.4	6.6	7.5	6.8	7.5	6.9	7.5	7.0	7.5	6.8	7.5	7.6	6.6	6.5
18	7.4	6.4	7.5	6.8	7.5	6.9	7.5	6.9	7.5	6.8	7.6	7.5	6.7	6.5
36	7.4	6.5	7.6	6.9	7.5	6.9	7.5	6.9	7.5	6.8	7.5	7.5	6.8	6.4
71	7.4	6.5	7.6	6.8	7.5	6.9	7.5	6.9	7.5	6.7	7.4	7.5	6.7	6.5
Tech. Initials	~	~	~	~	~	~	~	~	~	~	~	~	~	~

WQ Instruments Used: Salinity II-A-030304

DO II-A-3

Comments _____

Test Set Up By AWO

Data Verified By Galpin

Date Verified Oct-20/04

EVS ENVIRONMENT CONSULTANTS
7-d *Atherinops affinis* TOXICITY TEST – DAILY SURVIVAL DATA

Client Azimuth (Polaris)
 EVS Project No. 09-0302-54
 EVS Work Order No. 0400384

Sample ID Cr. Creek 082404
 Test Species/Batch A. affinis / Aug 17-04
 Test Initiation Date/Time Aug 23, 2004 @ 1230L
 No. of Organisms/Volume 5 / 200mL

‰ (v/v) Concentration	Rep.	Pan No.	Number of Survivors – Day of Test							Comments
			1	2	3	4	5	6	7	
Control	A	A 31	5	5	5	5	5	5	5	
	B	32	5	5	5	5	5	5	5	
	C	33	5	5	5	5	5	5	5	
	D	34	5	5	5	5	5	5	5	
	E	35	5	5	5	5	5	5	5	
Brine	A	36	5	5	5	5	5	5	5	
	B	37	5	5	5	5	5	5	5	
	C	38	5	5	5	5	5	5	5	
	D	39	5	5	5	5	5	5	5	
	E	40	5	5	5	5	5	5	5	
4.4	A	41	5	5	5	5	5	5	5	
	B	42	5	5	5	5	5	5	5	
	C	43	5	5	5	5	5	5	5	
	D	44	5	5	5	5	5	5	5	
	E	45	5	5	5	5	5	5	5	
9.0	A	546	5	5	5	5	5	5	5	
	B	547	5	5	5	5	5	5	5	
	C	548	5	5	5	5	5	5	5	
	D	549	5	5	5	5	5	5	5	
	E	680	5	5	5	5	5	5	5	
Technician Initials		Jes	m	n	ms	n	n	ms	ms	

Sample Description clear, colorless
 Data Verified By Galt Date Verified Oct 20/04

EVS ENVIRONMENT CONSULTANTS
7-d *Atherinops affinis* TOXICITY TEST – DAILY SURVIVAL DATA

Client Azimuth (Polaris)
 EVS Project No. 09-0302-54
 EVS Work Order No. 0400384

Sample ID G-Creek 082404
 Test Species/Batch *A. affinis* / Aug 7-04
 Test Initiation Date/Time Aug 27, 2004 @ 1220h
 No. of Organisms/Volume 5/200mL

‰ (v/v) Concentration	Rep.	Pan No.	Number of Survivors – Day of Test							Comments
			1	2	3	4	5	6	7	
18	A	²³ A 51 ⁴⁶	5	5	5	5	5	5	5	
	B	²³ 52 ⁴⁷	5	5	5	5	5	5	5	
	C	²³ 53 ⁴⁸	5	5	5	5	5	5	5	
	D	²³ 54 ⁴⁹	5	5	5	5	5	5	5	
	E	²³ 55 ⁵⁰	5	5	5	5	5	5	5	
36	A	²³ 56 ⁵¹	5	5	5	5	5	5	5	
	B	²³ 57 ⁵²	5	5	5	5	5	5	5	
	C	²³ 58 ⁵³	5	5	5	5	5	5	4	
	D	²³ 59 ⁵⁴	5	5	5	5	5	5	5	
	E	²³ 60 ⁵⁵	5	5	5	5	5	5	5	
71	A	61	5	5	5	5	5	²³ 45 ⁵	²³ 45 ⁵	
	B	62	5	5	5	5	5	5	5	
	C	63	5	5	5	5	5	5	5	
	D	64	5	5	5	5	5	5	5	
	E	65	5	5	5	5	5	5	5	
	A	6								
	B	7								
	C	8								
	D	9								
	E	0								
Technician Initials		JCS	m	-	JCS	r	~	JCS	JCS	

Sample Description clear, colorless
 Data Verified By Galpin Date Verified Oct 20/04

EVS ENVIRONMENT CONSULTANTS
7-d *Atherinops affinis* SURVIVAL AND GROWTH TOXICITY TEST – DRY WEIGHT DATA

Client Arimath (Polaris)
 EVS Project No. 09-0302-54
 EVS Work Order No. 0400384

Start Date (Day 0) August 28th, 2009
 Sample ID G-creek 082404
 Balance Type/Serial Number Sartorius BP211D

Sample ID % (v/v)	Rep.	Pan No.	Pan Weight (mg) ¹	Final Weight (mg) (pan + biomass) ¹	Number of Survivors	Number Weighed	Comments (e.g., confirmation weights, organisms lost in transfer)	Tech. Init.
Control	A	A 31	1228.40	1233.24	5	5	pre-wt: 1228.42 mg	<i>[Signature]</i>
	B	32	1229.96	1234.81	5	5		
	C	33	1231.09	1235.98	5	5		
	D	34	1226.23	1230.46	5	5	reweighed 1230.49 mg ✓	
	E	35	1218.59	1223.06	5	5		
Brine Control	A	36	1226.59	1231.11	5	5		
	B	37	1234.73	1239.03	5	5		
	C	38	1220.42	1226.76	5	5		
	D	39	1232.69	1237.55	5	5		
	E	40	1236.53	1240.57	5	5		
4.4 %	A	41	1227.94	1232.63	5	5		
	B	42	1224.25	1231.84	5	5		
	C	43	1235.61	1240.72	5	5	reweighed 1240.68 mg ✓	
	D	44	1219.66	1223.97	5	5		
	E	45	1225.29	1230.00	5	5	pre-wt: 1225.29 mg	<i>[Signature]</i>

1. Re-confirm weights for 10% of final weights and record under "Comments"; relative percent difference (RPD) between pairs of weights should be ≤10% of organism weight.

Data Verified By *[Signature]*

Date Verified Oct. 20/09

EVS ENVIRONMENT CONSULTANTS
7-d *Atherinops affinis* SURVIVAL AND GROWTH TOXICITY TEST – DRY WEIGHT DATA

Client A21m w/b (Polaris)
 EVS Project No. 09-0302-54
 EVS Work Order No. 0400384

Start Date (Day 0) August 27, 2004
 Sample ID G-Creek 082404
 Balance Type/Serial Number Sartorius BP 211D

Sample ID % (J/J)	Rep.	Pan No.	Pan Weight (mg) ¹	Final Weight (mg) (pan + biomass) ¹	Number of Survivors	Number Weighed	Comments (e.g., confirmation weights, organisms lost in transfer)	Tech. Init.
18	A	A 46	1217.86	1222.48	5	5		<i>[Signature]</i>
	B	47	1225.29	1228.77	5	5		
	C	48	1236.54	1242.18	5	5		
	D	49	1239.07	1244.52	5	5		
	E	50	1241.45	1246.42	5	5	reweighed: 1246.43 mg ✓	
36	A	51	1227.42	1230.87	5	5		
	B	52	1224.43	1228.50	5	5		
	C	53	1239.89	1243.83	4	4		
	D	54	1231.71	1236.86	5	5		
	E	55	1236.12	1240.57	5	5		
8.8	A	56	1233.41	1237.89	5	5		
	B	57	1232.69	1236.99	5	5		
	C	58	1218.75	1223.49	5	5		
	D	59	1232.98	1237.03	5	5		
	E	60	1222.96	1227.12	5	5	pre wt: 1222.96 mg	

1. Re-confirm weights for 10% of final weights and record under "Comments"; relative percent difference (RPD) between pairs of weights should be ≤10% of organism weight.

Data Verified By *[Signature]*

Date Verified Oct 20/04

EVS ENVIRONMENT CONSULTANTS
7-d *Atherinops affinis* SURVIVAL AND GROWTH TOXICITY TEST – DRY WEIGHT DATA

Client Arimath (Polaris)
 EVS Project No. 09-0302-54
 EVS Work Order No. 0400384

Start Date (Day 0) August 28, 2004
 Sample ID G-Creek 082404
 Balance Type/Serial Number Sartorius BP211.D

Sample ID % (y/o)	Rep.	Pan No.	Pan Weight (mg) ¹	Final Weight (mg) (pan + biomass) ¹	Number of Survivors	Number Weighed	Comments (e.g., confirmation weights, organisms lost in transfer)	Tech. Init.
²⁷ 72 71	A	A 61	1236.40	1239.74	5	5		JS
	B	62	1236.44	1240.25	5	5		
	C	63	1219.08 1223.84	1224.54	5	5	reweighed 1224.57 mg	
	D	64	1228.47	1233.11	5	5		
	E	65	1243.42	1247.95	5	5	pre-wt: 1243.44 mg	
	A	6						
	B	7						
	C	8						
	D	9						
	E	10						
	A	1						
	B	2						
	C	3						
	D	4						
	E	5						

1. Re-confirm weights for 10% of final weights and record under "Comments"; relative percent difference (RPD) between pairs of weights should be ≤10% of organism weight.

Data Verified By Galpin G

Date Verified Oct 20/04

Test: LF-Larval Fish Growth and Survival Test

Test ID: 0400384

Species: AA-Atherinops affinis

Protocol: EPAW 95-EPA West Coast

Sample ID: G_CREEK

Sample Type: EFF2-Industrial

Start Date: 8/27/2004

End Date: 9/3/2004

Lab ID: BCEVS-EVS Environment Consultants

Pos	ID	Rep	Group	Day 0	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	No. Fish Weighed	Total Wgt(mg)	Tare Wgt(mg)
	1	1	D-Control	5							5	5	1233.24	1228.4
	2	2	D-Control	5							5	5	1234.81	1229.96
	3	3	D-Control	5							5	5	1235.98	1231.09
	4	4	D-Control	5							5	5	1230.46	1226.23
	5	5	D-Control	5							5	5	1223.06	1218.59
	6	1	Brine Control	5							5	5	1231.11	1226.59
	7	2	Brine Control	5							5	5	1239.03	1234.73
	8	3	Brine Control	5							5	5	1226.76	1220.42
	9	4	Brine Control	5							5	5	1237.55	1232.69
	10	5	Brine Control	5							5	5	1240.57	1236.53
	11	1	4.400	5							5	5	1232.63	1227.94
	12	2	4.400	5							5	5	1231.84	1224.25
	13	3	4.400	5							5	5	1240.72	1235.61
	14	4	4.400	5							5	5	1223.97	1219.66
	15	5	4.400	5							5	5	1230	1225.29
	16	1	9.000	5							5	5	1237.89	1233.41
	17	2	9.000	5							5	5	1236.99	1232.69
	18	3	9.000	5							5	5	1223.49	1218.75
	19	4	9.000	5							5	5	1237.03	1233.08
	20	5	9.000	5							5	5	1227.12	1222.96
	21	1	18.000	5							5	5	1222.48	1217.86
	22	2	18.000	5							5	5	1228.77	1225.29
	23	3	18.000	5							5	5	1242.18	1236.54
	24	4	18.000	5							5	5	1244.52	1239.09
	25	5	18.000	5							5	5	1246.42	1241.45
	26	1	36.000	5							5	5	1230.87	1225.42
	27	2	36.000	5							5	5	1228.5	1224.43
	28	3	36.000	5							4	4	1243.83	1239.89
	29	4	36.000	5							5	5	1236.86	1231.71
	30	5	36.000	5							5	5	1240.57	1236.12
	31	1	71.000	5							5	5	1239.74	1236.4
	32	2	71.000	5							5	5	1240.25	1236.44
	33	3	71.000	5							5	5	1224.54	1219.08
	34	4	71.000	5							5	5	1233.11	1228.47
	35	5	71.000	5							5	5	1247.95	1243.42

Comments: Azimuth - Polaris Mine; G-Creek, 09-0302-54 ;0400384

Quality
Oct 27/04

Larval Fish Growth and Survival Test-7 Day Survival

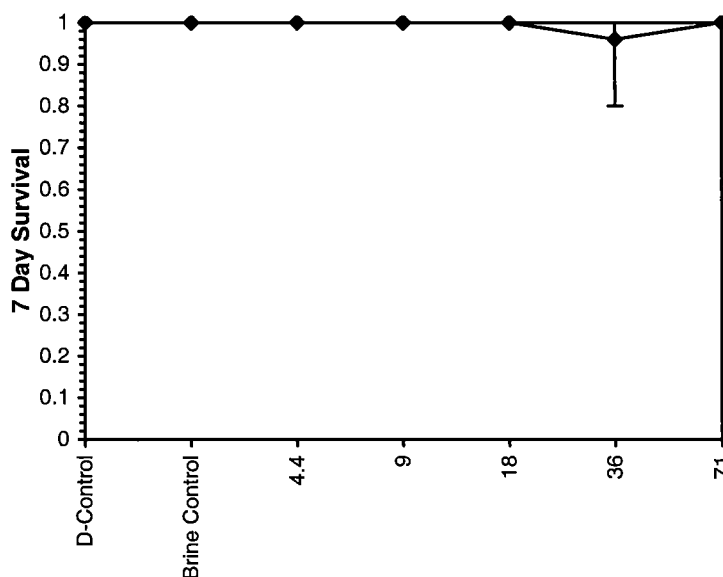
Start Date: 8/27/2004	Test ID: 400384	Sample ID: G_CREEK-082404
End Date: 9/3/2004	Lab ID: BCEVS-EVS Environment Cc	Sample Type: EFF2-Industrial
Sample Date: 8/24/2004	Protocol: EPAW 95-EPA West Coast	Test Species: AA-Atherinops affinis
Comments: Azimuth - Polaris Mine; G-Creek, 09-0302-54 ;0400384		

Conc-%	1	2	3	4	5
D-Control	1.0000	1.0000	1.0000	1.0000	1.0000
Brine Control	1.0000	1.0000	1.0000	1.0000	1.0000
4.4	1.0000	1.0000	1.0000	1.0000	1.0000
9	1.0000	1.0000	1.0000	1.0000	1.0000
18	1.0000	1.0000	1.0000	1.0000	1.0000
36	1.0000	1.0000	0.8000	1.0000	1.0000
71	1.0000	1.0000	1.0000	1.0000	1.0000

Conc-%	Mean	SD	Transform: Arcsin Square Root					Rank Sum	1-Tailed Critical
			Mean	Min	Max	CV%	N		
D-Control	1.0000	0.0000	1.3453	1.3453	1.3453	0.000	5		
Brine Control	1.0000	0.0000	1.3453	1.3453	1.3453	0.000	5		
4.4	1.0000	0.0000	1.3453	1.3453	1.3453	0.000	5	27.50	16.00
9	1.0000	0.0000	1.3453	1.3453	1.3453	0.000	5	27.50	16.00
18	1.0000	0.0000	1.3453	1.3453	1.3453	0.000	5	27.50	16.00
36	0.9600	0.0894	1.2977	1.1071	1.3453	8.207	5	25.00	16.00
71	1.0000	0.0000	1.3453	1.3453	1.3453	0.000	5	27.50	16.00

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates non-normal distribution ($p \leq 0.01$)	0.41613	0.9	-3.8705	19.8512
Equality of variance cannot be confirmed				
The control means are not significantly different ($p = 1.00$)	0	2.30601		
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Steel's Many-One Rank Test	71	>71		1.40845

Dose-Response Plot



Statistical comparison were against the D-control.

Larval Fish Growth and Survival Test-7 Day Survival

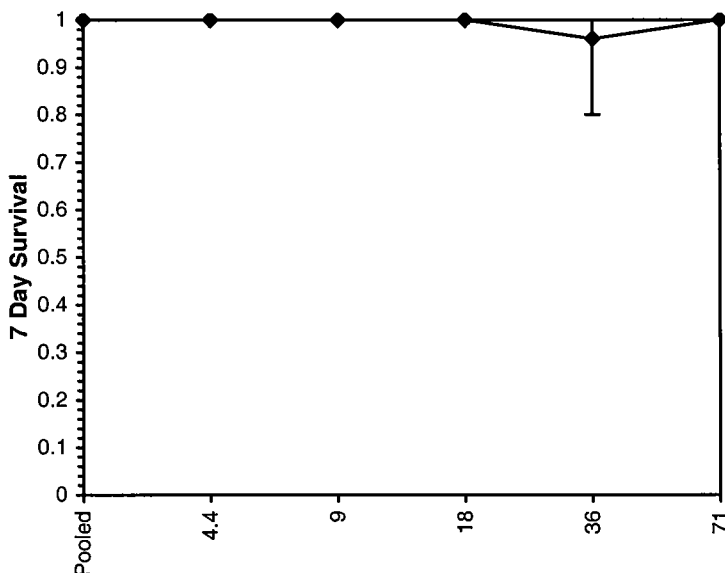
Start Date: 8/27/2004	Test ID: 400384	Sample ID: G_CREEK-082404
End Date: 9/3/2004	Lab ID: BCEVS-EVS Environment Cc	Sample Type: EFF2-Industrial
Sample Date: 8/24/2004	Protocol: EPAW 95-EPA West Coast	Test Species: AA-Atherinops affinis
Comments: Azimuth - Polaris Mine; G-Creek, 09-0302-54 ;0400384		

Conc-%	1	2	3	4	5
D-Control	1.0000	1.0000	1.0000	1.0000	1.0000
Brine Control	1.0000	1.0000	1.0000	1.0000	1.0000
4.4	1.0000	1.0000	1.0000	1.0000	1.0000
9	1.0000	1.0000	1.0000	1.0000	1.0000
18	1.0000	1.0000	1.0000	1.0000	1.0000
36	1.0000	1.0000	0.8000	1.0000	1.0000
71	1.0000	1.0000	1.0000	1.0000	1.0000

Conc-%	Mean	SD	Transform: Arcsin Square Root					Rank Sum	1-Tailed Critical
			Mean	Min	Max	CV%	N		
Pooled	1.0000	0.0000	1.3453	1.3453	1.3453	0.000	10		
4.4	1.0000	0.0000	1.3453	1.3453	1.3453	0.000	5	40.00	21.00
9	1.0000	0.0000	1.3453	1.3453	1.3453	0.000	5	40.00	21.00
18	1.0000	0.0000	1.3453	1.3453	1.3453	0.000	5	40.00	21.00
36	0.9600	0.0894	1.2977	1.1071	1.3453	8.207	5	35.00	21.00
71	1.0000	0.0000	1.3453	1.3453	1.3453	0.000	5	40.00	21.00

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates non-normal distribution ($p \leq 0.01$)	0.38831	0.91	-4.1486	23.0852
Equality of variance cannot be confirmed				
The control means are not significantly different ($p = 1.00$)	0	2.30601		
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Wilcoxon Rank Sum Test	71	>71		1.40845

Dose-Response Plot



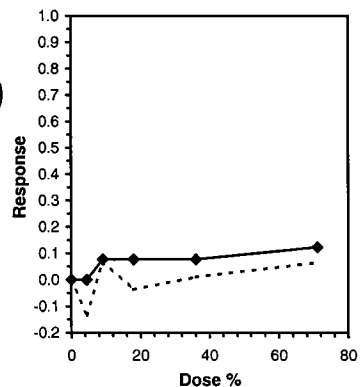
Statistical comparison were against the pooled control.

Larval Fish Growth and Survival Test-7 Day Growth (US)					
Start Date:	8/27/2004	Test ID:	400384	Sample ID:	G_CREEK-082404
End Date:	9/3/2004	Lab ID:	BCEVS-EVS Environment C	Sample Type:	EFF2-Industrial
Sample Date:	8/24/2004	Protocol:	EPAW 95-EPA West Coast	Test Species:	AA-Atherinops affinis
Comments:	Azimuth - Polaris Mine; G-Creek, 09-0302-54 ;0400384				
Conc-%	1	2	3	4	5
D-Control	0.9680	0.9700	0.9780	0.8460	0.8940
Brine Control	0.9040	0.8600	1.2680	0.9720	0.8080
4.4	0.9380	1.5180	1.0220	0.8620	0.9420
9	0.8960	0.8600	0.9480	0.7900	0.8320
18	0.9240	0.6960	1.1280	1.0860	0.9940
36	1.0900	0.8140	0.7880	1.0300	0.8900
71	0.6680	0.7620	1.0920	0.9280	0.9060

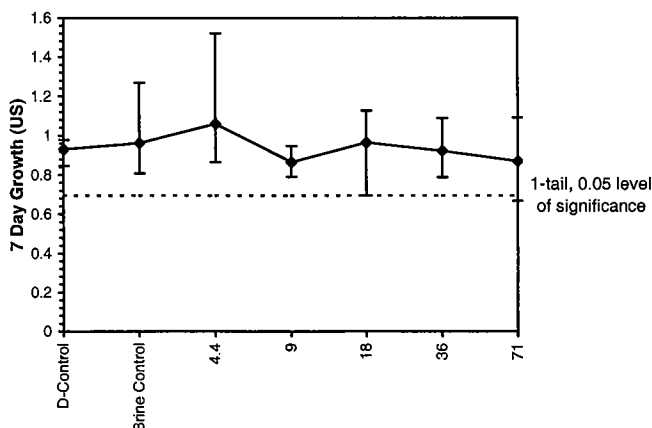
Conc-%	Mean	SD	Transform: Untransformed					t-Stat	1-Tailed			Isotonic	
			Mean	Min	Max	CV%	N		Critical	MSD	Mean	N-Mean	
D-Control	0.9312	0.0585	0.9312	0.8460	0.9780	6.283	5				0.9938	1.0000	
Brine Control	0.9624	0.1811	0.9624	0.8080	1.2680	18.820	5						
4.4	1.0564	0.2642	1.0564	0.8620	1.5180	25.007	5	-1.252	2.360	0.2361	0.9938	1.0000	
9	0.8652	0.0604	0.8652	0.7900	0.9480	6.980	5	0.660	2.360	0.2361	0.9177	0.9235	
18	0.9656	0.1704	0.9656	0.6960	1.1280	17.643	5	-0.344	2.360	0.2361	0.9177	0.9235	
36	0.9224	0.1328	0.9224	0.7880	1.0900	14.396	5	0.088	2.360	0.2361	0.9177	0.9235	
71	0.8712	0.1631	0.8712	0.6680	1.0920	18.721	5	0.600	2.360	0.2361	0.8712	0.8766	

Auxiliary Tests				Statistic	Critical	Skew	Kurt			
Shapiro-Wilk's Test indicates normal distribution ($p > 0.01$)				0.94863	0.9	0.95961	2.51611			
Bartlett's Test indicates equal variances ($p = 0.05$)				11.0427	15.0863					
The control means are not significantly different ($p = 0.72$)				0.36654	2.30601					
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	71	>71		1.40845	0.23609	0.25353	0.02479	0.02502	0.44403	5, 24

Linear Interpolation (200 Resamples)				
Point	%	SD	95% CL(Exp)	Skew
IC05	7.405			
IC10	53.535			
IC15	>71			
IC20	>71			
IC25	>71			
IC40	>71			
IC50	>71			



Dose-Response Plot



Statistical comparison were against the D-control.

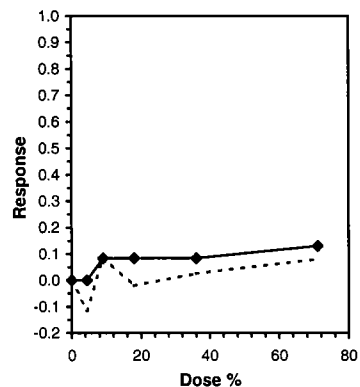
Galpin
Oct-20-104

Larval Fish Growth and Survival Test-7 Day Growth (US)					
Start Date:	8/27/2004	Test ID:	400384	Sample ID:	G_CREEK-082404
End Date:	9/3/2004	Lab ID:	BCEVS-EVS Environment C	Sample Type:	EFF2-Industrial
Sample Date:	8/24/2004	Protocol:	EPAW 95-EPA West Coast	Test Species:	AA-Atherinops affinis
Comments:	Azimuth - Polaris Mine; G-Creek, 09-0302-54 ;0400384				
Conc-%	1	2	3	4	5
D-Control	0.9680	0.9700	0.9780	0.8460	0.8940
Brine Control	0.9040	0.8600	1.2680	0.9720	0.8080
4.4	0.9380	1.5180	1.0220	0.8620	0.9420
9	0.8960	0.8600	0.9480	0.7900	0.8320
18	0.9240	0.6960	1.1280	1.0860	0.9940
36	1.0900	0.8140	0.7880	1.0300	0.8900
71	0.6680	0.7620	1.0920	0.9280	0.9060

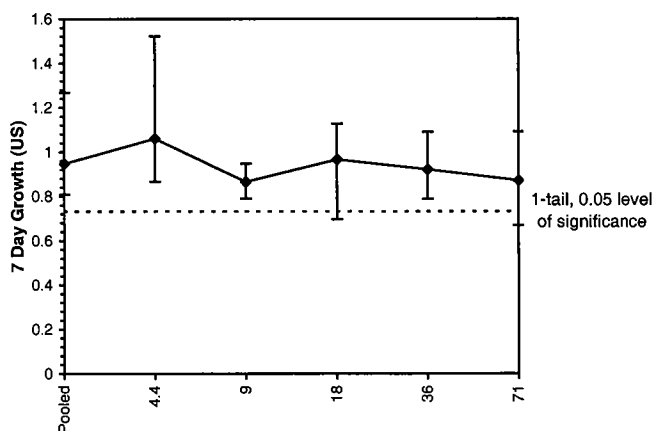
Conc-%	Mean	SD	Transform: Untransformed					N	t-Stat	1-Tailed			Isotonic	
			Mean	Min	Max	CV%	Critical			MSD	Mean	N-Mean		
Pooled	0.9468	0.1280	0.9468	0.8080	1.2680	13.514	10					1.0016	1.0000	
4.4	1.0564	0.2642	1.0564	0.8620	1.5180	25.007	5	-1.258	2.462	0.2146		1.0016	1.0000	
9	0.8652	0.0604	0.8652	0.7900	0.9480	6.980	5	0.936	2.462	0.2146		0.9177	0.9163	
18	0.9656	0.1704	0.9656	0.6960	1.1280	17.643	5	-0.216	2.462	0.2146		0.9177	0.9163	
36	0.9224	0.1328	0.9224	0.7880	1.0900	14.396	5	0.280	2.462	0.2146		0.9177	0.9163	
71	0.8712	0.1631	0.8712	0.6680	1.0920	18.721	5	0.868	2.462	0.2146		0.8712	0.8698	

Auxiliary Tests				Statistic	Critical	Skew	Kurt					
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)				0.94174	0.91	1.04686	2.00659					
Bartlett's Test indicates equal variances (p = 0.18)				7.64107	15.0863							
The control means are not significantly different (p = 0.72)				0.36654	2.30601							
Hypothesis Test (1-tail, 0.05)				NOEC	LOEC	ChV						
				TU	MSDu	MSDp	MSB					
				MSE	F-Prob	df						
Bonferroni t Test				71	>71	1.40845	0.21455	0.22661	0.02493	0.02531	0.44385	5, 29

Linear Interpolation (200 Resamples)				
Point	%	SD	95% CL(Exp)	Skew
IC05	7.147			
IC10	48.255			
IC15	>71			
IC20	>71			
IC25	>71			
IC40	>71			
IC50	>71			



Dose-Response Plot



Statistical comparison were against the pooled control.

Qalif K
Oct 20/04

EVS ENVIRONMENT CONSULTANTS
7-d *Atherinops affinis* SURVIVAL AND GROWTH TEST DATA SUMMARY

Client Azimuth (Polaris)
 EVS Project No. 09-0302-54
 EVS Work Order No. 0400384

EVS Analysts AND. JXS. AXF
 Test Initiation Date Aug 27, 2004

Sample	Initial Sample	Refresh Samples	
	Day 0	Day 2	Day 4
Identification	04-Cu-001	/	
Amount Received	1 x 1 L		
Date Collected	15-March-04		
Date Received			
Temperature (°C)			
pH			
DO (mg/L)			
Conductivity (µmhos/cm)			
Salinity (ppt)			
Ammonia (mg/L N)			
Chlorine (mg/L Cl)			
Other			

DILUTION/CONTROL WATER (initial water quality)

Water Type Filtered, UV sterilized S.W.
 Temperature (°C) 20.0
 pH 7.8
 Dissolved Oxygen (mg/L) 7.4
 Salinity 28

TEST CONDITIONS

Temperature Range (°C) 20.0-21.0
 pH Range 7.8-8.2
 Dissolved Oxygen Range (mg/L) 6.4-7.7
 Salinity (ppt) 28
 Photoperiod (L:D h) 16:8
 Aeration Provided? No
 Other -

TEST SPECIES INFORMATION

Source ABS (Aquatic Biosystems)
 Date Received Aug 26, 2004
 Age (on Day 0) 10-d
 Reference Toxicant Copper
 Current Reference Toxicant Result (incl. 95% CL)

Reference Toxicant Test Date Aug 27, 2004
 7-d survival LC50 121 (107-137) µg/L Cu
 7-d growth IC50 128 (85-150) µg/L Cu

Reference Toxicant Warning Limits (mean ± 2SD) and CV
 7-d survival LC50 132 ± 48 µg/L CV: 18%
 7-d growth IC50 131 ± 52 µg/L CV: 20%

TEST RESULTS

Endpoint	Conc. Units	NOEC	LOEC	LC50 (95% CL)	IC50 (95% CL)	IC25 (95% CL)
Survival	µg/L	100	180	121 (107-137)		
Growth		56	100		128 (85-150)	93 (53-124)

Other _____

Data Verified By Gulick

Date Verified Oct. 27/04

EVS ENVIRONMENT CONSULTANTS

7-d *Atherinops affinis* SURVIVAL AND GROWTH TEST – WATER QUALITY DATA

Client Azimuth (Polaris)
 EVS Project No. 09-0302-54
 EVS Work Order No. 0400384

Sample ID Cu Reflex
 Test Initiation Date/Time Aug 27, 2004 / 1245h
 Source/Date Received ABS / Aug 28, 2004

Concentration µg/L	Temperature (°C)													
	0	1	2	3	4	5	6	7						
Control	20.0	20.5	20.0	20.5	20.0	20.0	20.5	20.0	20.5	20.0	20.5	20.0	21.0	20.5
320	20.0	20.5	20.0	20.5	20.0	20.0	20.5	20.0	20.5	20.0	20.5	20.0	21.0	20.5
560	20.0	20.5	20.0	20.5	20.0	20.0	20.5	20.0	20.5	20.0	20.5	20.0	21.0	20.5
1000	20.0	20.5	20.0	20.5	20.0	20.0	20.5	20.0	20.5	20.0	20.5	20.0	21.0	20.5
1800	20.0	20.5	20.0	20.5	20.0	20.0	20.5	20.0	20.5	20.0	20.5	20.0	21.0	20.5
3200	20.0	20.5	20.0	20.5	20.0	20.0	20.5	20.0	20.5	20.0	20.5	20.0	21.0	20.5
Tech. Initials	~	~	~	~	~	~	~	~	~	~	~	~	~	~

Concentration µg/L	pH													
	0	1	2	3	4	5	6	7						
Control	7.8	8.0	8.0	8.0	7.9	8.0	7.9	7.9	7.9	7.9	7.9	7.9	7.9	7.9
320	7.8	8.0	8.0	8.0	7.9	8.0	7.9	8.0	7.9	7.9	7.9	7.9	7.8	7.8
560	7.8	8.1	8.0	7.9	7.9	8.0	7.9	8.0	7.9	7.9	7.9	7.9	7.8	7.8
1000	7.8	8.1	8.0	8.1	7.9	8.0	7.9	8.0	7.9	8.0	7.9	7.9	7.8	7.9
1800	7.8	8.1	8.0	8.0	7.9	8.0	7.9	8.0	7.9	7.9	7.9	7.9	7.9	7.9
3200	7.8	8.0	8.0	8.0	7.9	8.0	7.9							
Tech. Initials	~	~	~	~	~	~	~	~	~	~	~	~	~	~

WQ Instruments Used: Temp. Calibrated Hg Thermometer pH H-A-52
 Comments _____

Test Set Up By AWB Data Verified By Qalif K Date Verified Oct. 20/04

EVS ENVIRONMENT CONSULTANTS

7-d *Atherinops affinis* SURVIVAL AND GROWTH TEST – WATER QUALITY DATA

Client Azimuth (Polaris)

Sample ID Lu K/L/10

EVS Project No. 09-0302-54

Test Initiation Date/Time Aug 28, 2004 @ 1245L

EVS Work Order No. 0400384

Source/Date Received ABS / Aug 28, 2004

Concentration <u>49/L</u>	Salinity (ppt)							
	0	1	2	3	4	5	6	7
<u>Control</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>
<u>320</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>
<u>560</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>
<u>1000</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>
<u>1800</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>
<u>3200</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>	<u>28</u>
Tech. Initials	<u>^</u>	<u>^</u>	<u>^</u>	<u>JMS</u>	<u>^</u>	<u>^</u>	<u>JMS</u>	<u>JMS</u>

Concentration <u>49/L</u>	Dissolved Oxygen (mg/L)													
	0	1	2	3	4	5	6	7	0	1	2	3	4	5
<u>Control</u>	<u>7.4</u>	<u>6.7</u>	<u>7.5</u>	<u>6.6</u>	<u>7.5</u>	<u>6.9</u>	<u>7.5</u>	<u>7.0</u>	<u>7.7</u>	<u>6.7</u>	<u>7.4</u>	<u>7.6</u>	<u>6.7</u>	<u>6.6</u>
<u>320</u>	<u>7.4</u>	<u>6.4</u>	<u>7.5</u>	<u>6.7</u>	<u>7.5</u>	<u>6.9</u>	<u>7.5</u>	<u>6.9</u>	<u>7.5</u>	<u>6.6</u>	<u>7.4</u>	<u>7.6</u>	<u>6.8</u>	<u>6.7</u>
<u>560</u>	<u>7.4</u>	<u>6.5</u>	<u>7.5</u>	<u>6.6</u>	<u>7.5</u>	<u>6.9</u>	<u>7.5</u>	<u>7.0</u>	<u>7.5</u>	<u>6.6</u>	<u>7.4</u>	<u>7.6</u>	<u>6.9</u>	<u>6.8</u>
<u>1000</u>	<u>7.4</u>	<u>6.5</u>	<u>7.5</u>	<u>6.7</u>	<u>7.5</u>	<u>7.0</u>	<u>7.5</u>	<u>7.1</u>	<u>7.5</u>	<u>6.7</u>	<u>7.4</u>	<u>7.6</u>	<u>6.7</u>	<u>6.7</u>
<u>1800</u>	<u>7.4</u>	<u>6.6</u>	<u>7.5</u>	<u>6.7</u>	<u>7.5</u>	<u>7.0</u>	<u>7.5</u>	<u>7.0</u>	<u>7.5</u>	<u>6.4</u>	<u>7.4</u>	<u>7.7</u>	<u>6.7</u>	<u>6.7</u>
<u>3200</u>	<u>7.4</u>	<u>6.6</u>	<u>7.5</u>	<u>6.7</u>	<u>7.5</u>	<u>7.1</u>	<u>7.5</u>	<u>7.0</u>	<u>7.5</u>	<u>7.0</u>	<u>7.5</u>	<u>7.0</u>	<u>7.5</u>	<u>7.0</u>
Tech. Initials	<u>^</u>	<u>^</u>	<u>^</u>	<u>^</u>	<u>^</u>	<u>JMS</u>	<u>^</u>	<u>^</u>	<u>^</u>	<u>^</u>	<u>^</u>	<u>^</u>	<u>^</u>	<u>JMS</u>

WQ Instruments Used: Salinity J- A-030304 DO J- A-3

Comments _____

Test Set Up By AWD Data Verified By Galpin Date Verified Oct-20/04

EVS ENVIRONMENT CONSULTANTS
7-d *Atherinops affinis* TOXICITY TEST – DAILY SURVIVAL DATA

Client Azimuth (Polaris)
 EVS Project No. 09-0302-54
 EVS Work Order No. 0400384

Sample ID Cu Reflot
 Test Species/Batch *A. affinis* / 1st Aug 2004
 Test Initiation Date/Time Aug 27, 2004 @ 1245L
 No. of Organisms/Volume 5/200mL

Concentration <u>49/L</u>	Rep.	Pan No.	Number of Survivors – Day of Test							Comments
			1	2	3	4	5	6	7	
Control	A	A 1	5	5	5	5	5	5	5	
	B	2	5	5	5	5	5	5	5	
	C	3	5	5	5	5	5	5	5	
	D	4	5	5	5	5	5	5	5	
	E	5	5	5	5	5	5	5	5	
320	A	6	5	5	5	5	5	4	4	
	B	7	5	5	5	5	5	5	5	
	C	8	5	5	5	5	5	5	5	
	D	9	5	5	5	5	5	5	5	
	E	10	5	5	5	5	5	5	5	
560	A	11	5	5	5	5	5	5	5	
	B	12	5	5	5	5	5	5	5	
	C	13	5	5	5	5	5	5	5	
	D	14	5	5	5	5	5	5	5	
	E	15	5	5	5	5	5	5	5	
1000	A	16	4	4	4	4	4	4	4	
	B	17	5	5	5	5	5	5	5	
	C	18	5	5	5	5	5	5	5	
	D	19	4	2	2	2	2	2	2	
	E	20	4	4	4	4	4	3	3	
Technician Initials		Jes	~	~	123	2	2	AXF JXS	Jes	

Sample Description clear, colorless
 Data Verified By Galfrk Date Verified Oct. 27/04

EVS ENVIRONMENT CONSULTANTS
7-d *Atherinops affinis* TOXICITY TEST - DAILY SURVIVAL DATA

Client Azimuth (Polaris)
 EVS Project No. 09-0302-54
 EVS Work Order No. 0400384

Sample ID Cu reflex
 Test Species/Batch A. affinis / Aug 27-04
 Test Initiation Date/Time Aug 27, 2004 @ 12:45
 No. of Organisms/Volume 5 / 200mL

Concentration	Rep.	Pan No.	Number of Survivors - Day of Test							Comments
			1	2	3	4	5	6	7	
49/L 180.0	A	A 21	2	2	2	2	2	2	2	
	B	22	2	0	0	~				
	C	23	5	3	3	3	3	2	0	
	D	24	1	1	1	1	1	0		
	E	25	2	1	1	0				
320.0	A	26	0							
	B	27	0							
	C	28	2	2						
	D	29	15	1						
	E	30	0							
	A	1								
	B	2								
	C	3								
	D	4								
	E	5								
	A	6								
	B	7								
	C	8								
	D	9								
	E	0								
Technician Initials		Jes	~	~	Jes	~	~	AKP/SKS	Jes	

Sample Description clear, colorless
 Data Verified By Qalikh Date Verified Oct. 27/04

EVS ENVIRONMENT CONSULTANTS
7-d *Atherinops affinis* SURVIVAL AND GROWTH TOXICITY TEST – DRY WEIGHT DATA

Client Arimath (Polaris)
 EVS Project No. 09-0302-54
 EVS Work Order No. 0400384

Start Date (Day 0) August 27th, 2004
 Sample ID Cu Pellet
 Balance Type/Serial Number Sartorius BP211.D

Sample ID	Rep.	Pan No.	Pan Weight (mg)	Final Weight (mg) (pan + biomass)	Number of Survivors	Number Weighed	Comments (e.g., confirmation weights, organisms lost in transfer)	Tech. Init.
7916								
Control	A	A 1	1236.92	1240.48	5	5		Awo/SA
	B	2	1234.84	1239.67	5	5		
	C	3	1233.83	1238.43	5	5		
	D	4	1234.81	1239.99	5	5	pre-wt: 1234.82 reweighed: 1240.01 mg ✓	
	E	5	1236.85	1241.27	5	5		
320	A	6	1236.55	1239.45	4	4		
	B	7	1223.58	1227.87	5	5		
	C	8	1228.35	1233.01	5	5		
	D	9	1236.84	1240.60	5	5	reweighed: 1240.62 mg ✓	
	E	10	1230.58	1235.34	5	5		
560	A	11	1240.99	1244.89	5	5		
	B	12	1240.22	1243.80	5	5		
	C	13	1232.03	1236.09	5	5		
	D	14	1222.70	1228.06	5	5	reweighed 1228.01 mg ✓	
	E	15	1227.91	1232.58	5	5		

1. Re-confirm weights for 10% of final weights and record under "Comments"; relative percent difference (RPD) between pairs of weights should be ≤10% of organism weight.

Data Verified By Galfish

Date Verified Oct. 20/04

EVS ENVIRONMENT CONSULTANTS
7-d *Atherinops affinis* SURVIVAL AND GROWTH TOXICITY TEST – DRY WEIGHT DATA

Client Azimuth (Polaris)
 EVS Project No. 09-0302-54
 EVS Work Order No. 0400384

Start Date (Day 0) August 27th, 2004
 Sample ID Cu Re/tox
 Balance Type/Serial Number Sartorius BP 211D

Sample ID	Rep.	Pan No.	Pan Weight (mg) ¹	Final Weight (mg) (pan + biomass) ¹	Number of Survivors	Number Weighed	Comments (e.g., confirmation weights, organisms lost in transfer)	Tech. Init.
49/L								
100.0	A	A 16	1231.06	1234.17	84	84		
	B	17	1232.73	1336.19	5	5		
	C	18	1227.40	1232.10	5	5		
	D	19	1225.62	1227.27	2	2		
	E	20	1229.98	1233.36	3	3		
180.0	A	21	1233.34	1235.48	2	2	pre-wt: 1233.29	
	B	22	1226.36		0			
	C	23	1228.36					
	D	24	1229.37					
	E	25	1226.55					
320.0	A	26	1219.08					
	B	27	1224.60					
	C	28	1229.45					
	D	29	1236.52					
	E	30	1236.61					

1. Re-confirm weights for 10% of final weights and record under "Comments"; relative percent difference (RPD) between pairs of weights should be ≤10% of organism weight.

Data Verified By Galpin

Date Verified Oct 20/04

Test: LF-Larval Fish Growth and Survival Test

Test ID: RTAACu39

Species: AA-Atherinops affinis

Protocol: EPAW 95-EPA West Coast

Sample ID: REF-Ref Toxicant

Sample Type: CU-Copper

Start Date: 8/27/2004

End Date: 9/3/2004

Lab ID: BCEVS-EVS Environment Consultants

Pos	ID	Rep	Group	Day 0	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	No. Fish Weighed	Total Wgt(mg)	Tare Wgt(mg)
	1	1	D-Control	5							5	5	1240.48	1236.92
	2	2	D-Control	5							5	5	1239.67	1234.84
	3	3	D-Control	5							5	5	1238.43	1233.83
	4	4	D-Control	5							5	5	1239.99	1234.81
	5	5	D-Control	5							5	5	1241.27	1236.78
	6	1	32.000	5							4	4	1239.45	1236.55
	7	2	32.000	5							5	5	1227.87	1223.58
	8	3	32.000	5							5	5	1233.01	1228.35
	9	4	32.000	5							5	5	1240.6	1236.84
	10	5	32.000	5							5	5	1235.34	1230.58
	11	1	56.000	5							5	5	1244.89	1240.99
	12	2	56.000	5							5	5	1243.8	1240.22
	13	3	56.000	5							5	5	1236.09	1232.03
	14	4	56.000	5							5	5	1228.06	1222.7
	15	5	56.000	5							5	5	1232.58	1227.91
	16	1	100.000	5							4	4	1234.17	1231.06
	17	2	100.000	5							5	5	1236.19	1232.73
	18	3	100.000	5							5	5	1232.1	1227.4
	19	4	100.000	5							2	2	1227.27	1225.62
	20	5	100.000	5							3	3	1233.36	1229.98
	21	1	180.000	5							2	2	1235.48	1233.34
	22	2	180.000	5							0	0	0	0
	23	3	180.000	5							0	0	0	0
	24	4	180.000	5							0	0	0	0
	25	5	180.000	5							0	0	0	0
	26	1	320.000	5							0	0	0	0
	27	2	320.000	5							0	0	0	0
	28	3	320.000	5							0	0	0	0
	29	4	320.000	5							0	0	0	0
	30	5	320.000	5							0	0	0	0

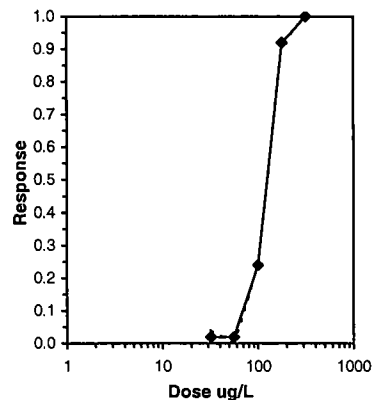
Comments: 09-0302-54, 0400384

Larval Fish Growth and Survival Test-7 Day Survival					
Start Date:	8/27/2004	Test ID:	RTAACu39	Sample ID:	REF-Ref Toxicant
End Date:	9/3/2004	Lab ID:	BCEVS-EVS Environment Cc	Sample Type:	CU-Copper
Sample Date:		Protocol:	EPAW 95-EPA West Coast	Test Species:	AA-Atherinops affinis
Comments:	09-0302-54, 0400384				
Conc-ug/L	1	2	3	4	5
D-Control	1.0000	1.0000	1.0000	1.0000	1.0000
32	0.8000	1.0000	1.0000	1.0000	1.0000
56	1.0000	1.0000	1.0000	1.0000	1.0000
100	0.8000	1.0000	1.0000	0.4000	0.6000
180	0.4000	0.0000	0.0000	0.0000	0.0000
320	0.0000	0.0000	0.0000	0.0000	0.0000

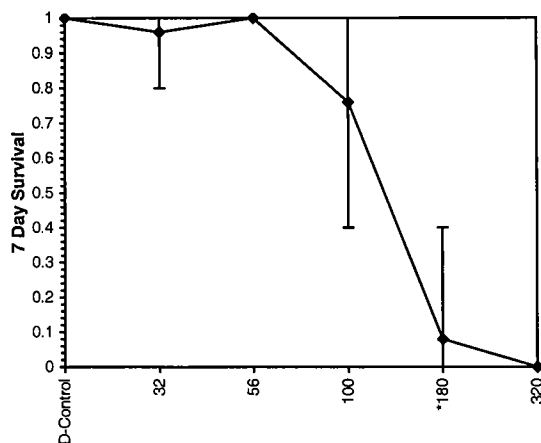
Transform: Arcsin Square Root								Rank	1-Tailed	Number	Total
Conc-ug/L	Mean	SD	Mean	Min	Max	CV%	N	Sum	Critical	Resp	Number
D-Control	1.0000	0.0000	1.3453	1.3453	1.3453	0.000	5			0	25
32	0.9600	0.0894	1.2977	1.1071	1.3453	8.207	5	25.00	17.00	1	25
56	1.0000	0.0000	1.3453	1.3453	1.3453	0.000	5	27.50	17.00	0	25
100	0.7600	0.2608	1.0737	0.6847	1.3453	26.959	5	20.00	17.00	6	25
*180	0.0800	0.1789	0.3174	0.2255	0.6847	64.711	5	15.00	17.00	23	25
320	0.0000	0.0000	0.2255	0.2255	0.2255	0.000	5			25	25

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates non-normal distribution ($p \leq 0.01$)	0.87543	0.888	0.19148	2.19139
Equality of variance cannot be confirmed				
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU
Steel's Many-One Rank Test	100	180	134.164	

Trimmed Spearman-Kärber			
Trim Level	EC50	95% CL	
0.0%			
5.0%	121.21	107.05	137.25
10.0%	122.51	106.69	140.68
20.0%	124.90	100.06	155.92
Auto-2.0%	121.16	107.14	137.03 ug/L Cu



Dose-Response Plot



Galpin
Oct 22/06

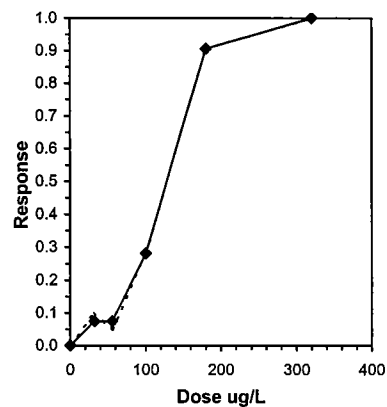
Larval Fish Growth and Survival Test-7 Day Growth (US)					
Start Date:	8/27/2004	Test ID:	RTAACu39	Sample ID:	REF-Ref Toxicant
End Date:	9/3/2004	Lab ID:	BCEVS-EVS Environment Cr	Sample Type:	CU-Copper
Sample Date:		Protocol:	EPAW 95-EPA West Coast	Test Species:	AA-Atherinops affinis
Comments:	09-0302-54, 0400384				
Conc-ug/L	1	2	3	4	5
D-Control	0.7120	0.9660	0.9200	1.0360	0.8980
32	0.5800	0.8580	0.9320	0.7520	0.9520
56	0.7800	0.7160	0.8120	1.0720	0.9340
100	0.6220	0.6920	0.9400	0.3300	0.6760
180	0.4280	0.0000	0.0000	0.0000	0.0000
320	0.0000	0.0000	0.0000	0.0000	0.0000

Conc-ug/L	Mean	SD	Transform: Untransformed					t-Stat	1-Tailed		Isotonic	
			Mean	Min	Max	CV%	N		Critical	MSD	Mean	N-Mean
D-Control	0.9064	0.1208	0.9064	0.7120	1.0360	13.330	5				0.9064	1.0000
32	0.8148	0.1529	0.8148	0.5800	0.9520	18.764	5	0.859	2.300	0.2451	0.8388	0.9254
56	0.8628	0.1413	0.8628	0.7160	1.0720	16.373	5	0.409	2.300	0.2451	0.8388	0.9254
*100	0.6520	0.2178	0.6520	0.3300	0.9400	33.401	5	2.387	2.300	0.2451	0.6520	0.7193
*180	0.0856	0.1914	0.0856	0.0000	0.4280	223.607	5	7.701	2.300	0.2451	0.0856	0.0944
320	0.0000	0.0000	0.0000	0.0000	0.0000	0.000	5				0.0000	0.0000

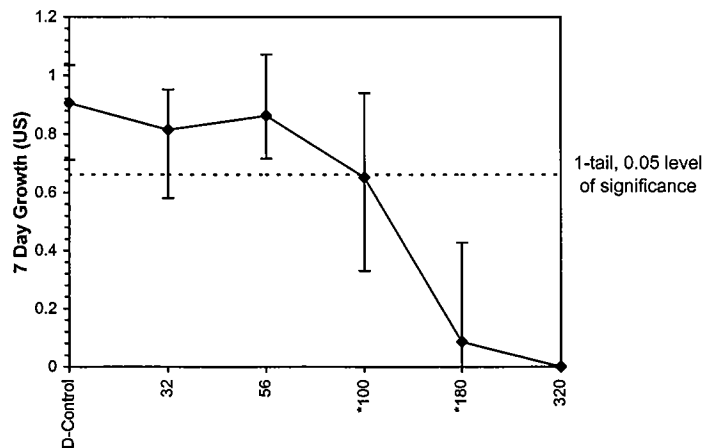
Auxiliary Tests						Statistic	Critical	Skew	Kurt			
Shapiro-Wilk's Test indicates normal distribution ($p > 0.01$)						0.97983	0.888	0.24836	0.34826			
Bartlett's Test indicates equal variances ($p = 0.81$)						1.6194	13.2767					
Hypothesis Test (1-tail, 0.05)			NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test			56	100	74.8331		0.24513	0.27045	0.56964	0.0284	9.0E-07	4, 20

Linear Interpolation (200 Resamples)					
Point	ug/L	SD	95% CL(Exp)	Skew	
IC05*	21.45	21.58	1.69	91.64	0.6918
IC10	61.43	22.13	0.00	103.56	-0.1239
IC15	72.10	20.41	1.19	118.52	-0.5135
IC20	82.78	15.91	41.12	121.23	-0.0104
IC25	93.45	13.98	53.16	124.15	0.0254
IC40	115.28	11.42	71.83	139.88	-0.3487
IC50	128.08	10.81	84.76	150.27	-0.4718

* indicates IC estimate less than the lowest concentration



Dose-Response Plot



APPENDIX B

Raw Data and Statistical Analyses:

Dendraster excentricus

**EVS ENVIRONMENT CONSULTANTS
ECHINOID FERTILIZATION TOXICITY TEST DATA SUMMARY**

Client AZIMUTH CONSULTING GROUP EVS Analysts KES / AKN
EVS Project No. 09-0302-J4/04-1424-044 Test Initiation Date 27 AUGUST 2004
EVS Work Order No. 0400385

SAMPLE

Identification G-CREEK-082404
Amount Received 5X20L
Date Collected 24 AUGUST 2004
Date Received 27 AUGUST 2004
Temperature (°C) 15.0
pH 8.0
Dissolved Oxygen (mg/L) 10.0 @ 8.5
Conductivity (µmhos/cm) 9240
Salinity (ppt) 5 @ 28
Ammonia (mg/L N) -
Chlorine (mg/L Cl) -
Other ① Salinity adjustment

TEST SPECIES

Organism Dendroaster eccentricus
Source Westwind Sealab Supply, 27 Aug. 2004
Date Received 27 August 2004
Reference Toxicant SDS
Current Reference Toxicant Result
Reference Toxicant Test Date 27 August 2004
IC50 (and 95% CL) 2.3 (2.1-2.4) mg/L SDS
Reference Toxicant Warning Limits (mean ± 2SD) and CV
3.9 ± 4.6 mg/L SDS; CV% = 60

DILUTION/CONTROL WATER (initial water quality)

Water Type UV Sterilized, 0.5µm filtered SW
Temperature (°C) 16.0
pH 8.0
Dissolved Oxygen (mg/L) 8.3
Salinity (ppt) 28
Other -

TEST CONDITIONS

Temperature Range (°C) 15.0 - 16.0
pH Range 8.0 - 8.1
Dissolved Oxygen Range (mg/L) 8.2 - 8.5
Salinity Range (ppt) 28
Sperm:Egg Ratio 2000:1
Test Duration 10:10
Other -

TEST RESULTS

IC50! 50.7 (48.1 - 53.6) % V/V
IC25! 17.5 (11.6 - 22.6) % V/V
NOEC! 4.7 % V/V
LOEC! 9.4 % V/V

Data Verified By Qualifly

Date Verified Oct. 26/04

EVS ENVIRONMENT CONSULTANTS
ECHINOID FERTILIZATION TOXICITY TEST INITIAL WATER QUALITY

Client AZIMUTH CONSULTING GROUP
 EVS Project No. 09-0302-54/04-1424-044
 EVS Work Order No. 0400385
 Logbook Echinoid #13 Pages 53-57

Test Initiation Date/Time 27 August 2004 @ 13:34
 Test Species Dendraster eccentricus
 Source/Date Received Westwind Sealab Supplies/27 August 2004
 Test Duration 10:10

Sample ID G-CREEK 082404	Conc/Rep % V/V	Temperature (°C)	pH	Salinity (ppt)	Dissolved Oxygen (mg/L)	Comments
CONTROL		16.0	8.0	28	8.3	
BRINE CTL		16.0	8.1	28	8.3	
	4.7	16.0	8.0	28	8.42	
	9.4	16.0	8.0	28	8.3	
	18.8	16.0	8.0	28	8.3	
	37.5	15.5 16.0	8.0	28	8.2	
	75.1	15.0 16.0	8.0	28	8.5	
Tech. Init.	Kef	Kef	Kef	Kef	Kef	

WQ Instruments Used: Temp. Calibrated Hg thermometer pH II-A-51 Salinity II-A-03030 DO II-A-011202
 Data Verified By Galt Date Verified Oct. 26/04

EVS ENVIRONMENT CONSULTANTS
ECHINOID FERTILIZATION TOXICITY TEST – EGG COUNT (SAMPLES)

Client AZIMUTH CONSULTING GROUP

Test Initiation Date/Time 27 August 2004 @ 13:34

EVS Project No. 09-0302-54/04-1424-044

Test Species Dendraster eccentricus

EVS Work Order No. 0400385

Source/Date Received Westwind Sealab Supplies/27 August 2004

Logbook Echinoid #13 Pages 53-57

Test Duration 10:10

Sperm:Egg Ratio 2000:1

Sample ID % V/V	Replicate	Number of Fertilized Eggs	Number of Unfertilized Eggs	Comments	Tech. Initials
4.7	A	78	22		Jaf ↓
	B	74	26		
	C	76	24		
	D	72	28		
9.4	A	61	39		Jaf ↓
	B	62	38		
	C	62	38		
	D	60	40		
18.8	A	56	44		Jaf ↓
	B	58	42		
	C	41 59	41		
	D	55	45		
37.5	A	49	51		Jaf ↓
	B	50	50		
	C	50	50		
	D	48	52		
75.1	A	25	75		Jaf ↓
	B	27	73		
	C	24	76		
	D	27	73		
CONTROL SEA WATER	A	77	4/29 23		Jaf ↓
	B	80	20		
	C	74	26		
	D	77	23		

Data Verified By Gal H

Date Verified Oct-26/04

EVS ENVIRONMENT CONSULTANTS
ECHINOID FERTILIZATION TOXICITY TEST – EGG COUNT (SAMPLES)

Client AZIMUTH CONSULTING GROUP.
 EVS Project No. 09-0302-54/04-1424-044
 EVS Work Order No. 0400385
 Logbook Echinoid #13 Pages 53-57

Test Initiation Date/Time 27 August 2004 @ 13:34
 Test Species Dendraster eccentricus
 Source/Date Received Westwind Sealab Supplies/27 August 2004
 Test Duration 10:10
 Sperm:Egg Ratio 2000:1

Sample ID	Replicate	Number of Fertilized Eggs	Number of Unfertilized Eggs	Comments	Tech. Initials
BRINE CONTROL	A	76	24		Jef ↓
	B	73	27		
	C	79	21		
	D	77	23		
	A				
	B				
	C				
	D				
	A				
	B				
	C				
	D				
	A				
	B				
	C				
	D				
	A				
	B				
	C				
	D				

Data Verified By Galfik

Date Verified Oct. 21/04

Test: SC-Sperm Cell Fertilization test				Test ID: 0400385			
Species: DE-Dendraster excentricus				Protocol: EPS1/RM/27-EC 92 (Sperm Cell)			
Sample ID: G CREEK-082404				Sample Type: EFF2-Industrial			
Start Date: 8/27/2004 10:10		End Date: 8/27/2004		Lab ID: BCEVS-EVS Environment Consultants			
Pos	ID	Rep	Group	Total Counted	Number Fertilized	Number Unfertilized	Notes
	1	1	D-Control	100	77	23	
	2	2	D-Control	100	80	20	
	3	3	D-Control	100	74	26	
	4	4	D-Control	100	77	23	
	5	1	B-Control	100	76	24	
	6	2	B-Control	100	73	27	
	7	3	B-Control	100	79	21	
	8	4	B-Control	100	77	23	
	9	1	4.700	100	78	22	
	10	2	4.700	100	74	26	
	11	3	4.700	100	76	24	
	12	4	4.700	100	72	28	
	13	1	9.400	100	61	39	
	14	2	9.400	100	62	38	
	15	3	9.400	100	62	38	
	16	4	9.400	100	60	40	
	17	1	18.800	100	56	44	
	18	2	18.800	100	58	42	
	19	3	18.800	100	59	41	
	20	4	18.800	100	55	45	
	21	1	37.500	100	49	51	
	22	2	37.500	100	50	50	
	23	3	37.500	100	50	50	
	24	4	37.500	100	48	52	
	25	1	75.100	100	25	75	
	26	2	75.100	100	27	73	
	27	3	75.100	100	24	76	
	28	4	75.100	100	27	73	

Comments: Azimuth Consulting Group (Polaris), 09-0302-54 (0400385)

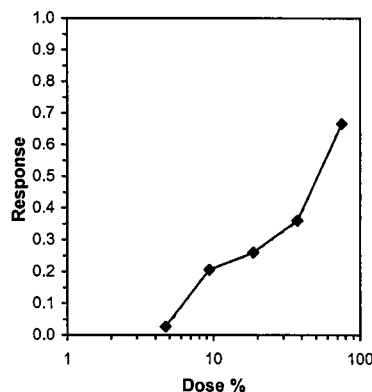
Sperm Cell Fertilization test-Proportion Fertilized					
Start Date:	8/27/2004 10:10	Test ID:	400385	Sample ID:	G CREEK-082404
End Date:	8/27/2004	Lab ID:	BCEVS-EVS Environment Cc	Sample Type:	EFF2-Industrial
Sample Date:	8/24/2004	Protocol:	EPS1/RM/27-EC 92 (Sperm (Test Species:	DE-Dendroaster excentricus
Comments:	Azimuth Consulting Group (Polaris), 09-0302-54 (0400385)				
Conc-%	1	2	3	4	
D-Control	0.7700	0.8000	0.7400	0.7700	
B-Control	0.7600	0.7300	0.7900	0.7700	
4.7	0.7800	0.7400	0.7600	0.7200	
9.4	0.6100	0.6200	0.6200	0.6000	
18.8	0.5600	0.5800	0.5900	0.5500	
37.5	0.4900	0.5000	0.5000	0.4800	
75.1	0.2500	0.2700	0.2400	0.2700	

Conc-%	Mean	SD	Transform: Untransformed					t-Stat	1-Tailed		Isotonic	
			Mean	Min	Max	CV%	N		Critical	MSD	Mean	N-Mean
D-Control	0.7700	0.0245	0.7700	0.7400	0.8000	3.181	4				0.7700	1.0000
B-Control	0.7625	0.0250	0.7625	0.7300	0.7900	3.279	4					
4.7	0.7500	0.0258	0.7500	0.7200	0.7800	3.443	4	1.546	2.410	0.0312	0.7500	0.9740
*9.4	0.6125	0.0096	0.6125	0.6000	0.6200	1.563	4	12.175	2.410	0.0312	0.6125	0.7955
*18.8	0.5700	0.0183	0.5700	0.5500	0.5900	3.203	4	15.460	2.410	0.0312	0.5700	0.7403
*37.5	0.4925	0.0096	0.4925	0.4800	0.5000	1.944	4	21.450	2.410	0.0312	0.4925	0.6396
*75.1	0.2575	0.0150	0.2575	0.2400	0.2700	5.825	4	39.616	2.410	0.0312	0.2575	0.3344

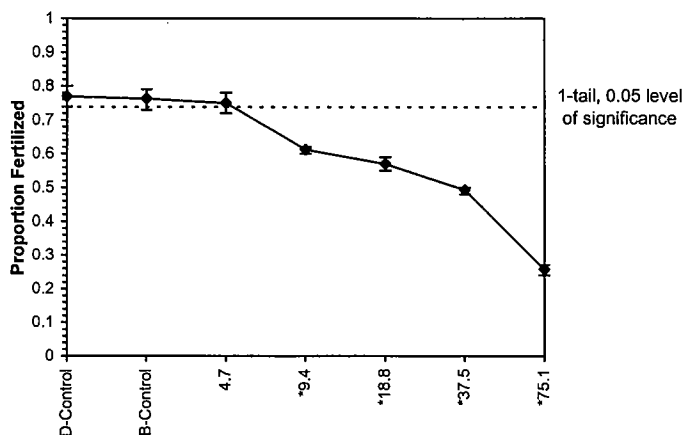
Auxiliary Tests					Statistic	Critical	Skew	Kurt		
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)					0.96771	0.884	-0.0461	-0.3022		
Bartlett's Test indicates equal variances (p = 0.47)					4.56528	15.0863				
The control means are not significantly different (p = 0.68)					0.42857	2.44691				
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test	4.7	9.4	6.6468	21.2766	0.03118	0.04049	0.14215	0.00033	5.1E-18	5, 18

Log-Linear Interpolation (200 Resamples)					
Point	%	SD	95% CL(Exp)	Skew	
IC05	5.180	0.661	1.565	6.056	-2.4799
IC10	6.314	0.363	5.072	7.317	-0.3761
IC15	7.655	0.380	6.468	8.804	-0.0227
IC20	9.242	0.946	8.159	13.672	1.6390
IC25	16.673	2.353	10.367	23.598	0.0429
IC40	41.060	0.897	38.450	44.063	-0.0075
IC50	51.581	0.976	49.063	54.697	0.0961

%v/v



Dose-Response Plot



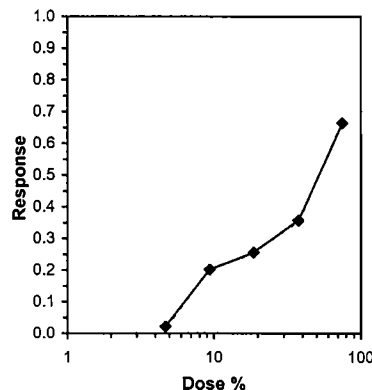
Statistical comparisons were against the D-control

Sperm Cell Fertilization test-Proportion Fertilized					
Start Date:	8/27/2004 10:10	Test ID:	400385	Sample ID:	G CREEK-082404
End Date:	8/27/2004	Lab ID:	BCEVS-EVS Environment Cc	Sample Type:	EFF2-Industrial
Sample Date:	8/24/2004	Protocol:	EPS1/RM/27-EC 92 (Sperm (Test Species:	DE-Dendroaster excentricus
Comments:	Azimuth Consulting Group (Polaris), 09-0302-54 (0400385)				
Conc-%	1	2	3	4	
D-Control	0.7700	0.8000	0.7400	0.7700	
B-Control	0.7600	0.7300	0.7900	0.7700	
4.7	0.7800	0.7400	0.7600	0.7200	
9.4	0.6100	0.6200	0.6200	0.6000	
18.8	0.5600	0.5800	0.5900	0.5500	
37.5	0.4900	0.5000	0.5000	0.4800	
75.1	0.2500	0.2700	0.2400	0.2700	

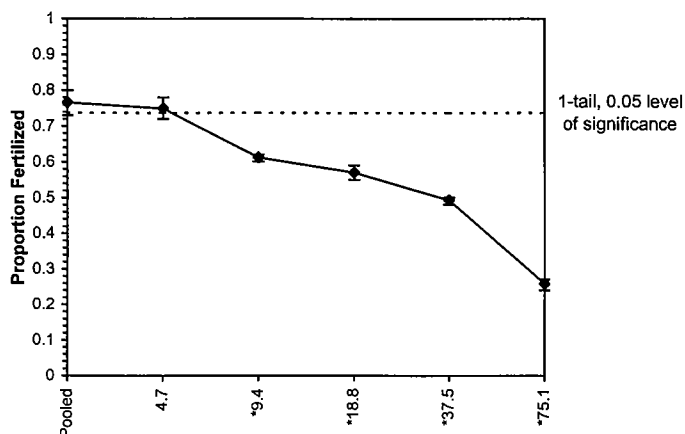
Conc-%	Mean	SD	Transform: Untransformed					t-Stat	1-Tailed Critical	MSD	Isotonic	
			Mean	Min	Max	CV%	N				Mean	N-Mean
Pooled	0.7663	0.0233	0.7663	0.7300	0.8000	3.036	8				0.7663	1.0000
4.7	0.7500	0.0258	0.7500	0.7200	0.7800	3.443	4	1.390	2.508	0.0293	0.7500	0.9788
*9.4	0.6125	0.0096	0.6125	0.6000	0.6200	1.563	4	13.156	2.508	0.0293	0.6125	0.7993
*18.8	0.5700	0.0183	0.5700	0.5500	0.5900	3.203	4	16.793	2.508	0.0293	0.5700	0.7439
*37.5	0.4925	0.0096	0.4925	0.4800	0.5000	1.944	4	23.424	2.508	0.0293	0.4925	0.6427
*75.1	0.2575	0.0150	0.2575	0.2400	0.2700	5.825	4	43.533	2.508	0.0293	0.2575	0.3361

Auxiliary Tests					Statistic	Critical	Skew	Kurt		
Shapiro-Wilk's Test indicates normal distribution (p > 0.01)					0.98075	0.896	-0.1412	-0.2089		
Bartlett's Test indicates equal variances (p = 0.44)					4.83474	15.0863				
The control means are not significantly different (p = 0.68)					0.42857	2.44691				
Hypothesis Test (1-tail, 0.05)	NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Bonferroni t Test	4.7	9.4	6.6468	21.2766	0.02931	0.03826	0.16613	0.00036	1.9E-21	5, 22

Log-Logit Interpolation (200 Resamples)						
Point	%	SD	95% CL(Exp)	Skew		
IC05	5.346	0.452	3.380	6.032	-1.8281	
IC10	6.555	0.307	5.439	7.178	-0.4707	
IC15	7.892	0.273	6.858	8.579	-0.3014	
IC20	9.379	0.715	8.461	12.502	1.9342	
IC25	17.460	2.031	11.554	22.636	0.0330	
IC40	40.999	0.628	38.998	42.863	0.1265	
IC50	50.738	0.809	48.086	53.622	0.1685	%v/v



Dose-Response Plot



Statistical comparisons were against the B-control

Pooled Control

Quelch
Oct. 26/04

**EVS ENVIRONMENT CONSULTANTS
ECHINOID FERTILIZATION TOXICITY TEST DATA SUMMARY**

Client Azimuth Consulting Group EVS Analysts KES, AKN
EVS Project No. 09-0302-54/04-1424-044 Test Initiation Date 27 AUGUST 2004
EVS Work Order No. 0400385

SAMPLE

Identification SDS Ref Tox Stock Sol
Amount ^{Prep'd} 1X1L
Date Collected ^{Prep'd} 26 April 2004
Date Received n/a
Temperature (°C) _____
pH _____
Dissolved Oxygen (mg/L) _____
Conductivity (µmhos/cm) _____
Salinity (ppt) _____
Ammonia (mg/L N) _____
Chlorine (mg/L Cl) _____
Other _____

TEST SPECIES

Organism Dendroaster eccentricus
Source Westwind Sealab Supplier
Date Received 27 August 2004
Reference Toxicant SDS
Current Reference Toxicant Result
Reference Toxicant Test Date 27 August 2004
IC50 (and 95% CL) 2.3 (2.1-2.4) mg/L SDS
Reference Toxicant Warning Limits (mean ± 2SD) and CV
3.9 ± 4.6 mg/L SDS, CV% = 60

DILUTION/CONTROL WATER (initial water quality)

Water Type UV Sterilized, 0.5µm filtered SW
Temperature (°C) 16.0
pH 8.0
Dissolved Oxygen (mg/L) 8.3
Salinity (ppt) 28
Other —

TEST CONDITIONS

Temperature Range (°C) 16.0
pH Range 8.0
Dissolved Oxygen Range (mg/L) 8.3
Salinity Range (ppt) 28
Sperm:Egg Ratio 2000:1
Test Duration 10:10
Other —

TEST RESULTS

IC 50: 2.3 (2.1-2.4) mg/L SDS
IC 25: 1.3 (1.2-1.4) mg/L SDS
NOEC: <1 mg/L SDS
LOEC: 1 mg/L SDS

Data Verified By Qualifit

Date Verified Oct. 26/04

EVS ENVIRONMENT CONSULTANTS
ECHINOID FERTILIZATION TOXICITY TEST INITIAL WATER QUALITY

Client AZIMOTH CONSULTING GROUP
 EVS Project No. 09-0302-54 / 04-1424-044
 EVS Work Order No. 0400385
 Logbook Echinoid #13 Pages 53-57

Test Initiation Date/Time 27 August 2004 @ 13:34
 Test Species Dendroaster excentricus
 Source/Date Received Westwind Seabed Supplies / 27 Aug 2004
 Test Duration 10:10

Sample ID	Conc/Rep	Temperature (°C)	pH	Salinity (ppt)	Dissolved Oxygen (mg/L)	Comments
SDS mg/L	CONTROL	16.0	8.0	28	8.3	
	1.0	16.0	8.0	28	8.3	
	1.8	16.0	8.0	28	8.3	
	3.2	16.0	8.0	28	8.3	
	5.6	16.0	8.0	28	8.3	
	10.0	16.0	8.0	28	8.3	
Tech. Init.	<i>del</i>	<i>del</i>	<i>del</i>	<i>del</i>	<i>del</i>	

WQ Instruments Used: Temp. Calibrated Hg thermometer pH II-A-51 Salinity II-A-020203 DO II-A-011202
 Data Verified By *Galpik* Date Verified Oct 21/04

EVS ENVIRONMENT CONSULTANTS
ECHINOID FERTILIZATION TOXICITY TEST – EGG COUNTS (CONTROLS)

Client AZIMUTH CONSULTING GROUP

Test Initiation Date/Time 27 August 2004 @ 13:34

EVS Project No. 09-0302-54/04-1424-044

Test Species Dendraster excentricus

EVS Work Order No. 0400385

Test Duration 10:10

Logbook Echinoid #13 Pages 53-57

Sperm:Egg Ratio 2000:1

Concentration SDS mg/L	Replicate	No. Fertilized Eggs	No. Unfertilized Eggs	Comments	Tech. Initials
Reference Toxicant					
1.0	A	70	30		ket
	B	72	28		↓
	C	69	31		↓
	D	68	32		↓
1.8	A	45	55		ket
	B	47	53		↓
	C	46	54		↓
	D	49	51		↓
3.2	A	25	75		ket
	B	26	74		↓
	C	24	76		↓
	D	27	73		↓
5.6	A	10	90		ket
	B	14	86		↓
	C	9	91		↓
	D	10	90		↓
10.0	A	2	98		ket
	B	2	98		↓
	C	1	99		↓
	D	2	98		↓
Control Seawater					
	A	81 77	23		ket
	B	74 80	20		↓
	C	74	26		↓
	D	77	23		↓

Data Verified By Galpin

Date Verified Oct. 21/04

Test: SC-Sperm Cell Fertilization test				Test ID: rtscsds9			
Species: DE-Dendraster excentricus				Protocol: EPS1/RM/27-EC 92 (Sperm Cell)			
Sample ID: REF-Ref Toxicant				Sample Type: SDS-Sodium dodecyl sulfate			
Start Date: 8/27/2004 10:10				End Date: 8/27/2004			
				Lab ID: BCEVS-EVS Environment Consultants			
Pos	ID	Rep	Group	Total Counted	Number Fertilized	Number Unfertilized	Notes
	1	1	D-Control	100	77	23	
	2	2	D-Control	100	80	20	
	3	3	D-Control	100	74	26	
	4	4	D-Control	100	77	23	
	5	1	1.0	100	70	30	
	6	2	1.0	100	72	28	
	7	3	1.0	100	69	31	
	8	4	1.0	100	68	32	
	9	1	1.8	100	45	55	
	10	2	1.8	100	47	53	
	11	3	1.8	100	46	54	
	12	4	1.8	100	49	51	
	13	1	3.2	100	25	75	
	14	2	3.2	100	26	74	
	15	3	3.2	100	24	76	
	16	4	3.2	100	27	73	
	17	1	5.6	100	10	90	
	18	2	5.6	100	14	86	
	19	3	5.6	100	9	91	
	20	4	5.6	100	10	90	
	21	1	10.0	100	2	98	
	22	2	10.0	100	2	98	
	23	3	10.0	100	1	99	
	24	4	10.0	100	2	98	

Comments: Azimuth Consulting Group 09-0302-54 (0400385)

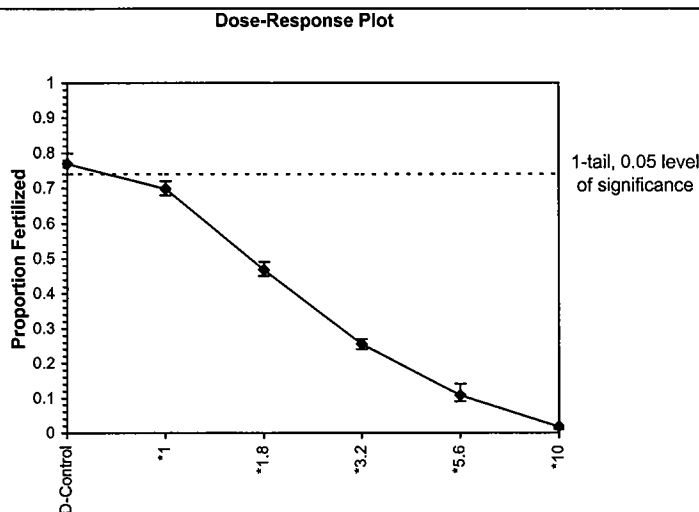
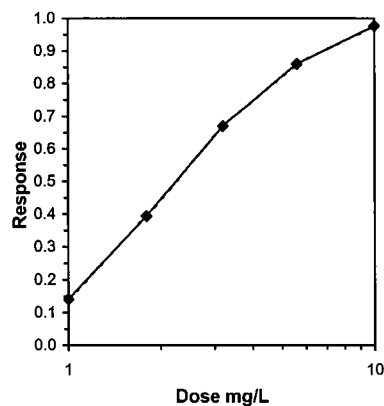
Sperm Cell Fertilization test-Proportion Fertilized					
Start Date:	8/27/2004 10:10	Test ID:	rtscsds9	Sample ID:	REF-Ref Toxicant
End Date:	8/27/2004	Lab ID:	BCEVS-EVS Environment Cr	Sample Type:	SDS-Sodium dodecyl sulfate
Sample Date:		Protocol:	EPS1/RM/27-EC 92 (Sperm t Test Species:	DE-Dendraster excentricus	
Comments:	Azimuth Consulting Group 09-0302-54 (0400385)				
Conc-mg/L	1	2	3	4	
D-Control	0.7700	0.8000	0.7400	0.7700	
1	0.7000	0.7200	0.6900	0.6800	
1.8	0.4500	0.4700	0.4600	0.4900	
3.2	0.2500	0.2600	0.2400	0.2700	
5.6	0.1000	0.1400	0.0900	0.1000	
10	0.0200	0.0200	0.0100	0.0200	

		Transform: Untransformed						1-Tailed		Isotonic	
Conc-mg/L	Mean	SD	Mean	Min	Max	CV%	N	t-Stat	Critical	MSD	Mean N-Mean
D-Control	0.7700	0.0245	0.7700	0.7400	0.8000	3.181	4				
*1	0.6975	0.0171	0.6975	0.6800	0.7200	2.448	4	5.813	2.410	0.0301	
*1.8	0.4675	0.0171	0.4675	0.4500	0.4900	3.653	4	24.254	2.410	0.0301	
*3.2	0.2550	0.0129	0.2550	0.2400	0.2700	5.063	4	41.292	2.410	0.0301	
*5.6	0.1075	0.0222	0.1075	0.0900	0.1400	20.627	4	53.118	2.410	0.0301	
*10	0.0175	0.0050	0.0175	0.0100	0.0200	28.571	4	60.334	2.410	0.0301	

Auxiliary Tests					Statistic	Critical	Skew	Kurt						
Shapiro-Wilk's Test indicates normal distribution ($p > 0.01$)					0.94353	0.884	0.48689	0.03974						
Bartlett's Test indicates equal variances ($p = 0.33$)					5.73659	15.0863								
Hypothesis Test (1-tail, 0.05)					NOEC	LOEC	ChV	TU	MSDu	MSDp	MSB	MSE	F-Prob	df
Dunnett's Test					<1	1			0.03006	0.03904	0.38532	0.00031	3.5E-22	5, 18

Log-Linear Interpolation (200 Resamples)					
Point	mg/L	SD	95% CL(Exp)	Skew	
IC05*	0.2818	0.0398	0.1777	0.4281	0.4398
IC10*	0.6429	0.1027	0.3830	1.0315	0.5219
IC15	1.0278	0.0806	0.6655	1.1534	-1.2547
IC20	1.1671	0.0441	1.0139	1.2853	-0.3348
IC25	1.3159	0.0419	1.1718	1.4292	-0.2285
IC40	1.8295	0.0507	1.6670	1.9678	-0.0985
IC50	2.2774	0.0469	2.1219	2.4035	-0.2200 mg/L SDS

* indicates IC estimate less than the lowest concentration



APPENDIX C

Chain-of-Custody Form

EVS environment
consultants

195 Pemberton Avenue
North Vancouver, BC
Canada V7P 2R4
Tel: 604-986-4331
Fax: 604-662-8548

Please see instructions for completion on back. Shaded areas to be completed by EVS Laboratory upon sample receipt.

Client Name: ALIMUTH CONSULTING, FDL

Client Contact Name: Cheryl Mackintosh/ Bruce Donald Ship to: EVS Environment Consultants

Address: 1001 W. 10th St.

Phone: 604-730-1220

195 Remberton Ave.

Address. 218-2402 West Broadway

Fax: 604-739-9070

North Vancouver, BC, V7P 2R4

VANCOUVER ISL, V61K 26

Sampled By: ~~Randy Baker~~ C. MacIntosh

Attn: Edmund Caravia

Shipping Date: Aug. 5, 2004[illegible]

PO/Reference No.: TC-03-03

Comments/Instructions: ① The sample ID was changed from G-Creek 081704 to G-Creek 082404 by the client's request.

Project Title: Polaris MMR

Results Needed By:

A) Released By: C. Mackintosh Date: Aug. 28/04
Company: Airnorth Time: 9:00am
Courier name: First air / FedEx
Shipping containers secured by: Tapex Straps Lock Other _____
(circle one)
Custody seals used? Yes No **METAL STRIPS**

B) Received by: **AXF** Date: **27 AUG 04**
Company: **EUS** Time: **0945**
Shipping containers received secure? ☒ Yes ☐ No
Custody seals intact? ☒ Yes ☐ No ☐ N/A

C) Released By: _____ Date: _____
 Company: _____ Time: _____
 Courier name: _____

Shipping containers secured by: Tape Straps Lock Other _____
 Custody seals used? Yes No (circle one)

D) Received by:	Date:
Company:	Time:
Shipping containers received secure?	Yes No
Custody seals intact?	Yes No N/A

1 Receiving Water (RW); Effluent (E); Elutriate (ELU); Sediment (SED); Chemical (CHEM); Stormwater (SW); Other (Please Specify)

2 Collapsible Carboy (CC); Glass Jar (GJ); Jerry Can (JC); Plastic HDPE (P); Plastic Bucket (PB); Other (Please Specify)

3 Please note any conditions the lab should be aware of for safety and storage concerns

4 Acceptable (A); Unacceptable (U). Please note specifics (e.g., broken, leaking, lid not on) under Comments/Instructions

Distribution of copies: White, yellow, pink - accompany the shipment
Orange - retained by consignor (e.g., shipper)
Yellow - retained by consignee (e.g., receiver)
Pink - for use as needed
White - returned to consignor by consignee

Revision Date: Sept. 25, 2000

APPENDIX F

**Letter from Environment Canada to Teck Cominco Metals, Ltd. re: Polaris 2004
Annual Report (dated August 9, 2005)**



Environment Canada
Environnement Canada

Prairie & Northern Region
Environment Canada
Rm 200, 4999 98th Ave.
Edmonton, AB T6B 2X3

Bruce Donald
Cominco Mining Partnership & Teck Cominco Metals Ltd.
Bag 2000
Kimberley, BC
V1A3E1

August 9, 2005

Dear Bruce Donald,

RE: Polaris 2004 Annual Report

In our review of the Polaris 2004 Annual Effluent and Water Quality Report, due March 31, 2005, it was noted that some Environmental Effects Monitoring (EEM) information does not appear to have been provided as required under Metal Mining Effluent Regulations (Schedule 5, Part 1). Please see the appended list of deficiencies relating to effluent characterization, water quality monitoring and sublethal toxicity testing for your facility.

If missing information was collected but not included in the above report, please submit this information to Environment Canada by September 30, 2005. Please ensure that all deficiencies are addressed in future reports. Failure to provide all information required under the Environmental Effects Monitoring Program will result in non-compliance.

Please be reminded that guidance is provided on the National EEM Website to assist you, at http://www.ec.gc.ca/eem/English/Publications/web_publication/ec_water/. If you have any questions, please do not hesitate to contact me [Phone (780) 951-8754, jenny.ferone@ec.gc.ca].

Sincerely,

Jenny Ferone, Regional EEM Coordinator

cc Peter Blackall
Chuck Brumwell
Ken Russell

Recycle Paper / Papier Recycle



Polaris (7834-3-37/C263-9)
2004 Annual Effluent and Water Quality Review

General:

- Methods, detection limits, and QA/QC need to be included in the future.
- Latitude/longitude and description of reference and exposure areas need to be included in hard copy.
- No units given in the hard copy for measured parameters in the Effluent or Water Quality tables.

Effluent Characterization:

- Collected and analyzed four effluent samples, but due to short discharge period (July and August only) only two were more than one month apart. Remaining two were only three days short of being one month apart.
- No methods described or detection limits, no QA/QC data or description.

Water Quality:

- Only two water sampling dates were more than one month apart due to short sampling period (July and August). Three samples were analyzed.
- No latitude or longitude of reference or exposure sampling areas in hard copy; No description of the ref or exp area – both described as Garrow Bay.
- No methods described or detection limits, no QA/QC data or description.

Sublethal Toxicity:

Please note that laboratories performing sublethal tests should provide all information required on the Environment Canada checklists (Annex C of the Metal Mining Guidance Document).

- A subset of the sublethal reports submitted by mines in the Prairie and Northern Region were sent for independent verification by Environment Canada toxicologists. This subset includes the test results submitted by your facility. We will inform you of any additional sublethal deficiencies identified by our reviewers once they have finished their review.
- For first sampling LC50 reported twice for fish species.
- First and second sampling: No LC50 done for sand dollar.
- All comments below are for both sampling dates (July 27 and August 24) unless otherwise stated

Atherinops affinis

- Information on labeling/coding of sample was not reported.
- Temperature of sample upon arrival at lab was not reported.
- The date that the sample was received at the lab was not reported.
- Dates or days during test when sub-samples or multiple samples were used, was not reported.
- The date for test completion was not reported.
- Temperature, DO, pH, and salinity of test solution and controls were not reported for each 24hr exposure period. They were only reported for the start of the test.
- Only give the final average mortality for each test concentration instead of the mortality as noted during each 24hr observation period

- Only give the average dry weight of each concentration instead of for each replicate of each concentration – also no indication if preserved in formalin/ethanol or not. If not preserved then 2 test concentration have weights <0.85mg (17.3% & 69%).
- No preparation procedure of salinity adjustment was reported.

Champia parvula

- Information on labeling and coding of sample was not reported.
- The date for sample receipt at the lab was not reported.
- The date for test completion was not reported.
- Procedure and preparation of the hypersaline brine was not reported.
- The type and quantity of chemicals (if any) added to the control/dilution water was not reported.
- Temperature, DO, pH and salinity of test solutions and controls were not reported for the start and 48hr of the exposure period or for the beginning and end of the recovery period.
- No note if there was anything unusual about the test organism prior to the test or if there was anything unusual about the test itself.
- Only the mean # of cystocarps per plant for each test concentration was reported. Needed to report the mean # of cystocarps per plant in each test vessel (in each replicate of each conc. and controls).
- No indication of quantitative statistic used to determine IC25 value.

Dendraster excentricus

- Information on labeling/coding of sample, the date it was received and its temperature upon arrival at lab were all not reported.
- There was no mention of the condition of the adult organisms prior to use.
- No note if there was anything unusual about the test organisms prior to use or anything unusual about the test itself.
- No procedure for salinity adjustment was reported – only statement that they followed EC guidance document.
- Only 100eggs/ vessel were used – should have been 2000 for the 10mL test volume.
- No indication of quantitative statistic used to determine the IC25 value.

APPENDIX G

**Letter from Azimuth Consulting Group, Inc. to Environment Canada re:
Clarification of reporting issues outlined in August 9, 2005 letter (dated September
14, 2005)**



**Azimuth Consulting
Group Inc.**
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Vancouver, BC
Canada V6K 2G8

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Fax: 604-739-8511
www.azimuthgroup.ca

Jenny Ferone
Regional EEM Coordinator
Prairie & Northern Region
Environment Canada
Rm 200, 4999 98th Ave.
Edmonton, AB, T6B 2X3

September 14, 2005

Dear Ms. Ferone,

**Re: Clarification of reporting issues outlined in Environment
Canada's letter to Teck Cominco regarding the Polaris 2004 EEM
Annual Report**

This letter is being written on behalf of Teck Cominco Metals Ltd. in response to Environment Canada's letter to Teck Cominco, dated August 9, 2005, regarding purported deficiencies in the Polaris 2004 Annual Effluent and Water Quality Report. Azimuth staff prepared the report and will be addressing the issues outlined in your letter by providing a revised 2004 EEM Annual Report. However, there are a few questions and clarifications that we would like to draw to your attention, to ensure that we include all required information in the revised report and that there are no misunderstandings or further deficiencies. Our specific responses to each of the points raised in the review letter are as follows:

General:

- Methods, detection limits and QA/QC will be included in all future reports
- Latitude and longitude and a description of the reference and exposure areas will be provided in the hard copy
- Units for water quality will be provided

Effluent Characterization:

- Effluent was collected and tested within allowable time periods, given the short duration of effluent discharge at Polaris.
- Methods, detection limits and QA/QC will be provided in the hard copy

Water Quality:

- Sampling of water quality was optimized to collect as many samples as possible, given short duration of discharge
- Latitude and longitude of exposure and reference areas were provided in the electronic copy; these will also be provided in the hard copy
- Methods, detection limits and QA/QC will be provided in the hard copy

Sublethal Toxicity:

- *“The first sampling LC50 reported twice for fish species.”* We presume the reviewers referring to the topsmelt tests. If so, we conducted 3 acute lethality tests on topsmelt in 2004. Results of all three tests are reported in the EEM under point *iii* of the Results. These values are listed as >72.6%, >69.0% and >71.0%.
- *“First and second sampling: No LC50 done for sand dollar.”* The sand dollar test is strictly a sublethal fertilization test, and no LC50 is measured.

Atherinops affinis

- Information on labeling and coding of samples will be reported
- Temperature on sample receipt at the lab will be reported.
- Date of sample receipt at the lab will be reported
- Dates or days when sub- or multiple samples were used will be reported
- Date for test completion will be reported
- *“Temperature, DO, pH, and salinity of test solution and controls were not reported for each 24hr exposure period...”* The laboratory (EVS) hand writes temperature, DO and pH on hard copy forms and does not enter these electronically. PDF copies of these forms will be provided with our hard copy report.
- *“Only give the final average mortality for each test concentration instead of the mortality as noted during each 24hr observation period.”* As above, these are written by hand in hard copy by the lab. PDF copies will be provided with the final report.
- *Comment regarding the dry weights of each replicate for each test concentration... “If not preserved then 2 test concentration have weights <0.85mg (17.3%, and 69%).”* Dry weights will be reported for each replicate of each concentration. Note that the 0.85mg test validity benchmark only refers to the **controls**. If there is a sublethal effect, such as slower growth, exposed animals will necessarily weigh less than the control and may weigh less than 0.85 mg.
- *“No preparation procedure of salinity adjustment was reported.”* Information regarding salinity adjustment was presented under point *ix* under Test Facilities and Conditions. The following text was included:
 - ix. *Indication that EC guidance document for salinity adjustment was followed*
 - *The following was done for all 3 tests:*
 - *No deviations from EC guidance document on preparation of hypersaline brine (HSB)*

- *HSB prepared from natural seawater concentrated to 90ppt (by freezing/refreezing to remove frozen layer and concentrate salts)*
- *No deviations from EC guidance document for salinity adjustment of sample*
- *HSB was added to samples to salinity adjust them to 30ppt*

We presume that this level of information is sufficient. If not, please let us know what additional information is required. Note that for the topmelt and echinoderm tests, the lab reports state: *“Salinity: 30±2 (sample adjusted with hypersaline brine [HSB]. Preparation of HSB and salinity adjustment as per EC guidance document on salinity adjustment – July 1997)”*

Champia parvula

- Information on labeling and coding will be provided in the revised final report
- Date for sample receipt at the lab will be reported
- Date for test completion will be reported
- *“Procedure and preparation of the hypersaline brine was not reported.”* Under point vii. of Test Conditions and Facilities we indicate:
 - *No deviations from EC guidance document on preparation of hypersaline brine*
 - *HSB prepared from natural seawater at 90ppt*
 - *No deviations from EC guidance document for salinity adjustment of sample*
 - *Salinity adjustment: 600mL effluent + 250mL HSB + 8.5mL test nutrient solution*
 - *Salinity of samples adjusted from 4ppt to 30ppt*

The lab report indicated for hypersaline brine that it was *“Prepared from natural seawater, at 90 ppt salinity.* As per EC guidance document on salinity adjustment”*. We presume that this is a sufficient level of detail. If not, please advise us what additional information the laboratory can provide to satisfy this comment.

- *“No note if there was anything unusual about the test organism prior to the test or if there was anything unusual about the test itself”*. We will include appropriate observations about the test in future reports. With regards to the organisms, under point iii of Test Organisms we indicated that *“[test species]...*
 - *Sexually mature male and female branches*
 - *Obtained from USEPA, Hatfield Marine Science Center, Newport Oregon, 1995*
 - *Appear in good health*
 - *Females have trichogynes, males have sori with spermatia*

We presume this is a sufficient level of detail. If not, please advise us on what additional information the laboratory can provide to satisfy this comment.

- *“No indication of quantitative statistic used to determine the IC25 value.”* Under point iii. of Results, we stated *“Quantal statistic method was linear interpolation”*. We presumed this was sufficient. Please advise if the reviewer wishes us to elaborate on this method.

Dendraster excentricus

- Information on labeling/coding, date of receipt, temperature at arrival at laboratory will be provided
- *“There was no mention of the condition of the adult organisms prior to use.”*
Under point xi (Test Organisms) it is stated that *“organisms appear healthy”*. We presumed this was sufficient information. If not, please elaborate on what additional level of detail is required.
- Future reports will note if there is anything unusual observed about the organisms or the test.
- *“No procedure for salinity adjustment was reported – only statement that they followed EC guidance document”* Under point ix (Test Facilities and Conditions) we make the following comments on hypersaline brine and salinity adjustment.
 - x. *Procedure for preparation of hypersaline brine (HSB) as per EC guidance document on salinity adjustment – July 1997*
 - *Test 1: na*
 - *Test 2: 30 ppt adjusted with hypersaline brine (HSB). Preparation of HSB and salinity adjustment as per EC guidance document on Salinity adjustment – July 1997*
 - *Test 2: 30 ppt adjusted with hypersaline brine (HSB). Preparation of HSB and salinity adjustment as per EC guidance document on Salinity adjustment – July 1997*
 - xi. *Procedure for salinity adjustment as per EC guidance document on salinity adjustment – July 1997*
 - *No deviations from EC guidance for salinity adjustment*
 - *Test 1: na*
 - *Test 2: salinity adjusted from 2.8 to 28 ppt*
 - *Test 3: salinity adjusted from 5 to 28 ppt*

We presume that this is a sufficient level of detail and was according to EC guidance. For both topsmelt and echinoderm tests the original lab report methods for salinity state *“30±2 (sample adjusted with hypersaline brine [HSB]. Preparation of HSB and salinity adjustment as per EC guidance document on salinity adjustment – July 1997)”* Please advise if your reviewers would like us to obtain more information from the laboratory.

- The laboratory does use 2000 eggs/10 mL, although a subset of 100 eggs are evaluated to determine the overall fertilization rate. We will clarify this.
- Quantitative statistic for determining IC25 will be provided.

We have raised a few issues where further guidance from your office is required. These are stated above as assumptions where clarification is requested from your office. We have incorporated all other appropriate additional information requested by the reviewers into a revised 2004 Polaris Annual Report. Once we have received a response from your office we will finalize this revised report. If we do not have a response from your office by Monday September 26, 2005, we will finalize the report

following our assumptions as stated in this letter, and submit the revised report by the September 30, 2005 deadline.

We would also like to communicate to Environment Canada that the electronic and hardcopy versions of the EEM report are not harmonized, as different information is requested for the two reports. As it is currently, the addition of the electronic report results in a duplication of effort and confuses the reporting requirements, rather than resulting in a more streamlined reporting process.

Sincerely,

Cheryl Mackintosh, M.R.M., R.P.Bio.

Azimuth Consulting Group Inc.

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APPENDIX H

Polaris Mine – Revised 2004 3rd Quarter Metal Mining Effluent Regulations Report (dated March 22, 2005)



March 22, 2005

Prairie & Northern Region
Environment Canada
Room 200, 4999 98th Ave.
Edmonton, AB T6B 2X3

Attention: Peter Blackall, Regional Director of Environmental Protection

Dear Sir;

Re: Polaris Mine – Revised 2004 3rd Quarter Metal Mining Effluent Regulations Report

Please find attached a **revised** Polaris Mine 3rd Quarter MMR regulatory data tables. The reason for the revision is to ensure consistency between the data entered electronically online through the RISS system, and the hard copy Annual and 3rd Quarter reports provided. As explained in the original 3rd Quarter report cover letter, effluent from Garrow Lake ceased flowing by mid-August but surface run off from adjacent slopes maintained minimal water flow at the designated final discharge point into August. By August 17, there was water at the final discharge point in a pool, but flow was too low to be measured. As a result, regulatory concentration data in the 3rd Quarter had originally been submitted with average daily flows of 0 m³ per day for the weeks of Aug. 17, Aug. 24, and Aug. 31, 2004. However, we continued to collect effluent, water quality and acute and sublethal toxicity samples on these latter three dates. Upon submitting the regulatory data into the online RISS system for the annual report, these August concentration data were not accepted with a flow rate of zero. Thus a nominal flow rate of 1 m³ per day was entered. As a result the loadings data set is slightly different from the data submitted in the original 3rd Quarter report. Note that these low flow conditions during August, as well as their implications for effluent sampling, were discussed with Environment Canada representatives early in August.

If you have any questions regarding the annual report or aspects of the application of the MMER to the Polaris Mine, please feel free to contact me at any time.

Yours truly,

Original signed by B. Donald

Bruce Donald

Attachments: Revised 2003 3rd Quarter Regulatory Data

cc: Walter Kuit (Teck Cominco Limited)

Randy Baker (Azimuth Consulting Group)

2004 3rd QUARTER MMER REPORT - REVISED

LOCATION - FINAL DISCHARGE POINT FROM GARROW LAKE (GARROW LAKE DAM SIPHONS)

CONCENTRATIONS OF EFFLUENT FOR MMER SCHEDULE 4 SAMPLED WEEKLY

Sample Taken		DELETERIOUS SUBSTANCE (mg/L) ¹									Collection Method
During The Week of	Date Sample Taken	Arsenic	Copper	Cyanide	Lead	Nickel	Zinc	TSS	Radium 226 ¹	pH ¹	
5-Jul-04	7-Jul-04	0.00100	0.00265	0.0050	0.00269	0.00442	0.1980	117	0.0200	8.05	Grab
12-Jul-04	13-Jul-04	0.00200	0.00070	0.0050	0.00032	0.00204	0.1060	5.7	0.0070	7.90	Grab
19-Jul-04	20-Jul-04	0.00200	0.00043	0.0050	0.00084	0.00088	0.0435	3.0	0.0050	7.86	Grab
26-Jul-04	27-Jul-04	0.00100	0.00052	0.0050	0.00157	0.00207	0.0429	3.0	0.0050	7.87	Grab
2-Aug-04	3-Aug-04	0.00020	0.00082	0.0050	0.00280	0.00338	0.0349	3.3	0.0060	8.00	Grab
9-Aug-04	10-Aug-04	0.00100	0.00100	0.0055	0.00120	0.00748	0.0482	3.0	0.0050	8.04	Grab
16-Aug-04	17-Aug-08	0.00020	0.00121	0.0050	0.00177	0.00644	0.0418	5.3	0.0100	7.95	Grab
23-Aug-04	24-Aug-04	0.00020	0.00134	0.0050	0.00119	0.00967	0.0498	4.4	0.0080	7.84	Grab
30-Aug-04	31-Aug-04	0.00020	0.00137	0.0050	0.00261	0.01340	0.0794	14.5	0.0080	7.90	Grab
6-Sep-04	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²
13-Sep-04	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²
20-Sep-04	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²
27-Sep-04	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²

Note¹ - All concentrations are in mg/L except Radium 226 which is Bq/L and pH which is in pH units

Note² - "nd" refers to no effluent discharge to sample

MONTHLY MEAN CONCENTRATIONS OF EFFLUENT FOR MMER SCHEDULE 4

MONTH OF	MONTHLY MEAN CONCENTRATION ¹ OF DELETERIOUS SUBSTANCE ³							
	Arsenic	Copper	Cyanide	Lead	Nickel	Zinc	TSS	Radium 226
July/04	0.0015	0.00107	0.0050	0.00135	0.00235	0.0976	32	0.0093
August/04	0.0004	0.00115	0.00510	0.00191	0.00807	0.0508	6.1	0.0074
September/04	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²	nd ²

Note¹ - All concentrations are in mg/L except Radium 226 which is Bq/L

Note² - "nd" refers to no effluent discharge to sample

Note³ - Monthly Mean Concentrations - the **MEAN** value of the concentrations measured in all water samples collected during each month when a deleterious substance is deposited.

MASS LOADING OF DELETERIOUS SUBSTANCE FOR EACH DAY SAMPLED

Sample Taken		DAILY MASS LOADING OF DELETERIOUS SUBSTANCE (kg/day) ¹								Average Daily
During The	Date									Flow Rate
Week of	Sample Taken	Arsenic	Copper	Cyanide	Lead	Nickel	Zinc	TSS	Radium 226 ¹	(m ³ /day)
5-Jul-04	7-Jul-04	0.018	0.048	0.091	0.049	0.080	3.588	2120	362,440	18,122
12-Jul-04	13-Jul-04	0.752	0.264	1.880	0.120	0.767	39.850	2143	2,631,608	375,944
19-Jul-04	20-Jul-04	0.205	0.044	0.512	0.086	0.090	4.458	307	512,395	102,479
26-Jul-04	27-Jul-04	0.028	0.015	0.141	0.044	0.058	1.211	85	141,090	28,218
2-Aug-04	3-Aug-04	0.005	0.022	0.132	0.074	0.089	0.923	87	158,700	26,450
9-Aug-04	10-Aug-04	0.026	0.026	0.142	0.031	0.194	1.248	78	129,470	25,894
16-Aug-04	17-Aug-08	0.000	0.000	0.000	0.000	0.000	0.000	0	0	1
23-Aug-04	24-Aug-04	0.000	0.000	0.000	0.000	0.000	0.000	0	0	1
30-Aug-04	31-Aug-04	0.000	0.000	0.000	0.000	0.000	0.000	0	0	1
6-Sep-04	nd ²	0	0	0	0	0	0	0	0	0
13-Sep-04	nd ²	0	0	0	0	0	0	0	0	0
20-Sep-04	nd ²	0	0	0	0	0	0	0	0	0
27-Sep-04	nd ²	0	0	0	0	0	0	0	0	0

Note¹ - Mass Loading is in kilograms per day of the deleterious substance deposited except Radium 226 which is in Bq per day

Note² - "nd" refers to no effluent discharge to sample

MASS LOADING PER CALENDAR MONTH FOR EACH DELETERIOUS SUBSTANCE

CALENDAR MONTH OF	MASS LOADING ¹ FOR DELETERIOUS SUBSTANCE (kg/month) ²								Average Weekly Flow Rate ³ (m ³ /week)	Total Monthly Volume ⁴ (m ³ /month)
	Arsenic	Copper	Cyanide	Lead	Nickel	Zinc	TSS	Radium 226 ²		
July/04	7.77	2.87	20.33	2.31	7.71	380.58	36,078.15	28,268,381	131,191	4,066,913
August/04	0.19	0.29	1.70	0.65	1.76	13.46	1,022.95	1,786,654	10,469	324,551
September/04	0	0	0	0	0	0	0	0	0	0

Note¹ - Total Mass Loading for Calendar month calculated by multiplying the Average Daily Mass Loading for the Month x # days in the month

Note² - Mass loading units are in kg per month except Radium 226, which is in Bq permonth

Note³ - Average Weekly Flow Rate calculated by multiplying Average Daily Flow Rate x 7 days per week

Note⁴ - Total Monthly Volume calculated by multiplying Average Daily Flow Rate for the month x days in month