

## Appendix D

D-1 Acute Lethality and Sublethal Toxicity Reports

D-2 EEM MMER Report

D-3 2003 MMER Report



**Stantec**

## Fathead minnow Test Report

Survival and Growth

1 of 6

Work Order : 204482

Sample Number : 7880

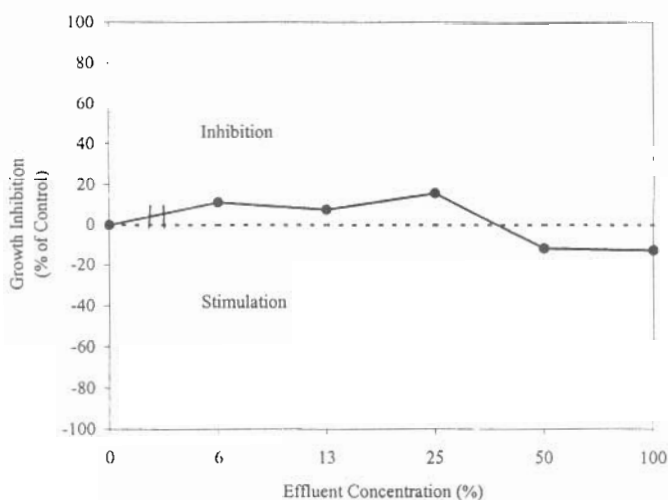
### Sample Identification

Company :	Nanisivik Mine (a Division of CanZinco Ltd.)	Date Collected :	2003-08-09
Location :	Nanisivik NU	Time Collected :	11:00
Substance :	159-4 August 8, 2003	Date Received :	2003-08-13
Sampling Method :	grab	Time Received :	10:30
Sampled By :	M. Markle	Date Tested :	2003-08-13
Shipped By:	FirstAir/air & Purolator/RD	Lab Storage:	4±2 °C
Temp. on arrival :	8.0°C		
Sample Description:	Clear, colourless, odourless.		

### Test Results

Effect	Value	95% Confidence Limits	Statistical Method
IC25 (Growth)	>100%	-	-
LC50 (Survival)	>100%	-	-

Fathead Minnow Growth Inhibition



Buildings  
Environment  
Industrial  
Transportation  
Urban Land

**Fathead minnow Test Report**

Survival and Growth

2 of 6

Work Order Number: 204482

Sample Number: 7880

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**Test Conditions**

Test Organism	<i>Pimephales promelas</i>	Test Volume Per Replicate	: 500 mL
Source	: ESG stock	Test Vessel	: 1 L polypropylene beaker
Life Stage	: Larval (<24 h old)	Depth of test solution	: 7.5 cm
pH Adjustment	None	# Replicates	: 3
Sample Filtration	: None	# Organisms per Replicate	: 10
Hardness Adjustment	None	Renewal Method <sup>d</sup>	: Syphon
Test Aeration	None	Renewal Period	: 24 h intervals
Photoperiod (h) light/dark	16/8	Feeding Rate	: 1500-2250 nauplii
Control/Dilution Water <sup>c</sup>	: Undiluted well water	Feeding Frequency	: 3 times daily
Test Type	Static renewal	Test Duration	: 7 days

<sup>c</sup> Control/Dilution Water: Well water with trace NaCl (29.6 mg/L).

<sup>d</sup> Renewal Method: Three subsamples were used for testing. Subsample 1 was used for day 0,1 and 2 renewals, subsample 2 was used for day 3 and 4 renewals, and subsample 3 was used for day 5 and 6 renewals. Approximately 85 - 90% of test solution removed by syphon. New solution added to achieve desired test volume.

Test Protocol: Biological Test Method: Test of Growth and Survival using fathead minnows. Environment Canada, Conservation and Protection. Ottawa, Ontario. Report EPS 1/RM/22 (including November 1997 amendments).

Test Organisms: No organisms exhibiting unusual appearance, behaviour, or undergoing unusual treatment were used in the test. All test organisms were from the same culture.

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**Comments**

Test conducted using three subsamples from a single sampling. The holding time was extended to four days as the sample was collected on 2003-08-09, but was not received until 2003-08-13. A power failure occurred on 2003-08-14 which resulted in the organisms being exposed to an extra 5 hours of darkness that day. There were no other unusual conditions or deviations from the test protocol. The results reported relate only to the sample tested.

## Fathead minnow Test Report

Survival and Growth

3 of 6

Work Order Number: 204482

Sample Number: 7880

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### Reference Toxicant Data

Substance	:	Potassium Chloride
Test Date	:	2003-08-18
IC25 Survival (g/L)	:	1.07
Lower 95% Confidence Limit	:	0.96
Upper 95% Confidence Limit	:	1.21
Test Duration	:	7 days
Historical Mean IC25 (g/L)	:	1.07
Upper Warning Limit (+2SD)	:	1.28
Lower Warning Limit (-2 SD)	:	0.89
Statistical Method	:	Linear Interpolation (Toxstat 3.5) <sup>b</sup>
Fathead minnow Batch Number	:	Fm03-08
Test Conducted By	:	D.Holtze/E.Williams

Reference toxicant test was conducted under conditions identical to the test.

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### References

<sup>a</sup> Stephan, C. E. 1977. Methods for calculating an LC50. P. 65-84 In: P.L. Mayer and J. L. Hamelink (eds.), Aquatic Toxicology and Hazard Evaluation. Amer. Soc. Testing and Materials, Philadelphia PA. ASTM STP 634.

<sup>b</sup> West, Inc. and D. Gulley. 1996. Toxstat Release 3.5. Western Ecosystems Technology. Cheyenne, WY, U.S.A.

Date: 2003-09-10

Approved By:

  
Keith Holtze, Director, Laboratory Operations

**Fathead minnow Test Report**  
Survival and Growth  
4 of 6

**Larval Fathead Minnow Survival**

Work Order Number : 204482

Sample Number : 7880

Industry : Nanisivik Mine (a Division of CanZinco Ltd.)

Time Start : 15:25

Fathead Batch # : Fm03-08

Previous 7d Stock Mort. : 0.0%

Start Date	Technician	Day 1		Day 2		Day 3		Day 4		Day 5		Day 6		Day 7		Mean Mortality (%)	Standard Deviation
		2003-08-13 DH(EW)	2003-08-14 DH(EW)	2003-08-15 TP	2003-08-16 KO(LM)	2003-08-17 KO(LM)	2003-08-18 AM(EW)	2003-08-19 TG	2003-08-20 JG(EW)	Completion Date							
Conc (%)	Rep	Mortality															
0	A	0	0	0	0	0	0	0	0	0	0	0	0	0	0	6.7	1.15
	B	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
	C	0	0	0	2	2	2	2	2	2	2	2	2	2	2		
6	A	0	0	0	0	0	0	0	0	0	0	0	0	0	0	6.7	1.15
	B	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
	C	0	0	0	2	2	2	2	2	2	2	2	2	2	2		
13	A	0	0	0	0	0	0	0	0	0	0	0	0	0	0	6.7	1.15
	B	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
	C	0	0	0	0	0	0	1	1	1	1	1	1	2	2		
25	A	0	0	0	0	0	0	0	0	0	0	0	0	0	0	3.3	0.58
	B	0	0	0	0	1	1	1	1	1	1	1	1	1	1		
	C	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
50	A	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.0	0.00
	B	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
	C	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
100	A	0	0	0	0	0	0	1	1	1	1	1	1	1	1	3.3	0.58
	B	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
	C	0	0	0	0	0	0	0	0	0	0	0	0	0	0		

Aberrant behaviour or swimming impairment noted during test: none

**Stantec**

Test Data Reviewed By: HR  
Date: 2003-09-06

**Fathead minnow Test Report**

Survival and Growth

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**Larval Fathead Minnow Weights**

Work Order Number : 204482

Sample Number : 7880

Concentration (%)	Replicate	# of Larvae Weighed	Mean Dry Wt. Of Larvae (mg)	Mean Dry Wt. (mg)	Standard Deviation
Control	A	10	0.778	0.757	0.049
	B	10	0.701		
	C	8	0.791		
6	A	10	0.682	0.671	0.064
	B	10	0.603		
	C	8	0.729		
13	A	10	0.626	0.699	0.073
	B	10	0.698		
	C	8	0.773		
25	A	10	0.614	0.637	0.077
	B	9	0.722		
	C	10	0.574		
50	A	10	0.811	0.846	0.031
	B	10	0.863		
	C	10	0.865		
100	A	9	0.921	0.854	0.067
	B	10	0.853		
	C	10	0.787		

**Larval Fathead Minnow Survival and Growth  
Water Chemistry Data**

Work Order Number : 204482  
Sample Number : 7880

Initial Parameters:		Temp. (°C)	D.O. (mg/L)	pH	Cond. ( $\mu$ s)	Hardness (mg/L as CaCO <sub>3</sub> )		
		25.0	9.9	7.6	2293	>1000		
		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Initial Temp. (°C):		25.0	25.0	24.0	25.0	25.0	25.5	25.0
Initial D.O. (mg/L):		9.9	9.3	9.8	9.5	9.8	9.1	9.3
% Sat. of 100% Effluent*		123	115	120	120	122	114	115
Preacteration (min)(≤100 bubbles/min):		20	20	20	20	20	20	20
		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Technician	New	DH(EW)	DH(EW)	EW	LM	JG(LM)	EW	DH(EW)
	Old	AM(EW)	TP	LM	JG(LM)	EW	DH(EW)	JG(EW)
Control		0%						
Temp. (°C)	Initial:	25.0	25.0	25.0	25.0	24.5	24.0	24.5
	Final:	25.0	25.5	25.5	25.0	25.5	25.0	25.5
D.O. (% Sat.*)	Initial:	100	100	101	99	100	97	101
	Final:	8.0	8.1	8.1	7.9	8.0	7.7	8.2
D.O. (mg/L)	Initial:	8.0	8.1	8.1	7.9	8.0	7.7	8.2
	Final:	7.5	7.1	7.2	7.3	7.0	7.4	6.7
pH	Initial:	8.3	8.4	8.4	8.5	8.3	8.3	8.4
	Final:	8.4	8.3	8.3	8.4	8.4	8.4	8.3
Conductivity	Initial:	535	491	575	513	545	508	545
Low		6%						
Temp. (°C)	Initial:	25.0	25.0	25.0	25.0	24.5	24.0	24.5
	Final:	25.0	25.5	25.5	25.0	25.5	25.0	25.5
D.O. (mg/L)	Initial:	8.1	8.1	8.1	7.9	8.0	7.8	8.2
	Final:	7.5	7.0	6.9	6.8	6.2	6.1	6.1
pH	Initial:	8.3	8.4	8.4	8.4	8.3	8.3	8.3
	Final:	8.3	8.2	8.2	8.3	8.1	8.1	8.1
Conductivity	Initial:	663	623	696	640	663	637	673
Middle		25%						
Temp. (°C)	Initial:	25.0	25.0	25.0	25.0	24.5	24.0	24.5
	Final:	25.0	25.5	25.5	25.0	25.5	25.0	25.5
D.O. (mg/L)	Initial:	8.2	8.3	8.3	7.9	8.1	7.9	8.3
	Final:	7.5	7.2	6.6	6.2	5.1	5.0	4.7
pH	Initial:	8.2	8.3	8.2	8.3	8.1	8.1	8.2
	Final:	8.2	8.0	8.0	7.9	7.7	7.6	7.6
Conductivity	Initial:	1038	1013	1064	1007	1015	1020	1021
High		100%						
Temp. (°C)	Initial:	25.0	25.0	25.0	25.0	24.5	24.0	24.5
	Final:	25.0	25.5	25.5	25.0	25.5	25.0	25.5
D.O. (mg/L)	Initial:	9.5	8.9	9.4	8.6	8.6	8.5	9.2
	Final:	7.6	6.8	6.2	6.1	5.5	5.7	4.8
pH	Initial:	7.8	7.8	7.6	7.7	7.8	7.6	7.6
	Final:	7.7	7.4	7.3	7.2	6.9	6.9	6.4
Conductivity	Initial:	2299	2300	2303	2291	2289	2307	2299

\* % saturation adjusted for actual temperature and barometric pressure



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## Ceriodaphnia dubia Test Report

Survival and Reproduction

1 of 6

Work Order : 204482

Sample Number : 7880

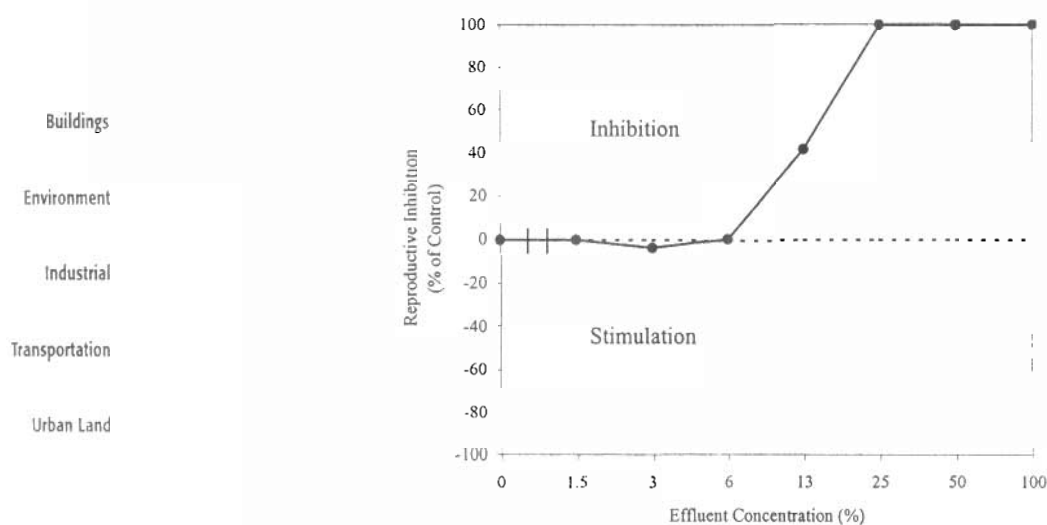
### Sample Identification

Company :	Nanisivik Mine (a Division of CanZinco Ltd.)	Date Collected :	2003-08-09
Location :	Nanisivik NU	Time Collected :	11:00
Substance :	159-4 August 8, 2003	Date Received :	2003-08-13
Sampling Method :	grab	Time Received :	10:30
Sampled By :	M. Markle	Date Tested :	2003-08-13
Shipped By:	FirstAir/air & Purolator/RD	Lab Storage:	4±2 °C
Temp. on arrival :	22.0°C		
Sample Description:	Clear, colourless, odourless.		

### Test Results

Effect	Value	95% Confidence Limits	Statistical Method
LC50 (Survival)	57.6%	40.5-94.6	Probit (Stephan) a
IC25 (Reproduction)	9.3%	7.4-11.5	Linear Interpolation (Toxstat 3.5) b

*Ceriodaphnia dubia* Reproductive Inhibition





**Ceriodaphnia dubia Test Report**

Survival and Reproduction

2 of 6

Work Order Number: 204482

Sample Number: 7880

**Test Conditions**

Test Organism	: <i>Ceriodaphnia dubia</i>	Control/Dilution Water <sup>c</sup>	: Lake Erie and Well water
Source	: ESG stock	Test Type	: Static renewal
Life Stage	: Neonate (<24 h)	Test Volume Per Replicate	: 15 mL
Mean Young Produced (previous 7 days)	: $\geq 15.0$	Test Vessel	: 15 mL polystyrene vial
# Young in Previous Brood	: $\geq 6.0$	Depth of test solution	: 4.5 cm
pH Adjustment	: None	# Replicates	: 10
Sample Filtration	: None	# Organisms per Replicate	: 1
Hardness Adjustment	: None	Renewal Method <sup>d</sup>	: transfer
Test Aeration	: None	Renewal Period	: 24 h intervals
Photoperiod (h) light/dark	: 16/8	Feeding Rate	: 0.2 mL YCT and algae
		Feeding Frequency	: once daily

<sup>c</sup> Control/Dilution Water: Mixture consisting of 25% Lake Erie water and 75% well water. The well water was supplemented with 29.6 mg/L of NaCl.

<sup>d</sup> Renewal Method: Three subsamples were used for testing. Subsample 1 was used for day 0,1 and 2 renewals, subsample 2 was used for day 3 and 4 renewals, and subsample 3 was used for the day 5 and 6 renewals. Organisms were transferred to new solution daily using a wide bore pipet.

Test Protocol: Biological Test Method: Test of Reproduction and Survival using the Cladoceran *Ceriodaphnia dubia*. Environment Canada, Conservation and Protection. Ottawa, Ontario. Report EPS 1/RM/21 (including November 1997 amendments).

Test Organisms: No organisms exhibiting unusual appearance, behaviour, or undergoing unusual treatment were used in the test. No ephippia were present in cultures. All organisms were within 12 hours of the same age. Test neonates were obtained from single broods from culture organisms. All test organisms were from the same culture.

**Comments**

Test conducted using three subsamples from a single sampling. The holding time was extended to four days as the sample was collected on 2003-08-09, but was not received until 2003-08-13. A power failure occurred on 2003-08-14 which resulted in 5 hours of inconsistent lighting. There were no other unusual conditions or deviations from the test protocol. The results reported relate only to the sample tested.

Work Order Number: 204482  
Sample Number: 7880

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**Reference Toxicant Data**

Substance	:	Sodium Chloride
Test Date	:	2003-08-22
IC25 Reproduction (g/L)	:	0.96
Lower 95% Confidence Limit	:	0.36
Upper 95% Confidence Limit	:	1.44
Test Duration	:	7 days
Historical Mean IC25 (g/L)	:	1.11
Upper Warning Limit (+2SD)	:	1.81
Lower Warning Limit (-2 SD)	:	0.70
Statistical Method	:	Linear Interpolation (Toxstat 3.5) <sup>b</sup>
<i>Ceriodaphnia</i> Batch Number	:	Cd03-08
Test Conducted By	:	T.Gardiner

Reference toxicant test was conducted under conditions identical to the test.

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**References**

<sup>a</sup> Stephan, C. E. 1977. Methods for calculating an LC50. P. 65-84 In: P.L. Mayer and J. L. Hamelink (eds.), Aquatic Toxicology and Hazard Evaluation. Amer. Soc. Testing and Materials, Philadelphia PA. ASTM STP 634.

<sup>b</sup> West, Inc. and D. Gulley. 1996. Toxstat Release 3.5. Western Ecosystems Technology. Cheyenne, WY, U.S.A.

Date: 2003-09-10

Approved By:

K. Holtze  
Keith Holtze, Director, Laboratory Operations

**Ceriodaphnia dubia Survival and Reproduction**

Work Order Number : 204482  
Sample Number : 7880  
Industry : Nanisivik Mine (a Division of CanZinco Ltd.)  
Time Start : 14:20  
Start Date : 2003-08-13  
Completion Date : 2003-08-20  
*C. dubia* Batch Number : Cd03-08  
Previous 7d culture mortality (%) : 11.7%

Concentration	Day	Replicate										% Mortality	Mean # Young	In1 JG(EW)
<b>Control</b>		1	2	3	4	5	6	7	8	9	10			
2003-08-14	1	0	0	0	0	0	0	0	0	0	0	0	0	LM
2003-08-15	2	0	0	0	0	0	0	0	0	0	0	0	0	LT
2003-08-16	3	0	0	0	6	6	1	0	0	0	0	0	1.3	JG(LM)
2003-08-17	4	6	7	0	0	0	5	4	7	5	6	0	4	JG(LM)
2003-08-18	5	13	13	5	13	12	10	9	8	12	13	0	10.8	EW/LM
2003-08-19	6	3	0	0	19	18	0	0	0	0	0	0	4	JG(EW)
2003-08-20	7	19	19	13	0	0	15	15	17	16	19	0	13.3	EW

Total neonates 41 39 18 38 36 31 28 32 33 38  
Mean # young: 33.4  
Total adult mortality: 0

Concentration	Day	Replicate										% Mortality	Mean # Young
		1	2	3	4	5	6	7	8	9	10		
<b>1.5%</b>	1	0	0	0	0	0	0	0	0	0	0	0	0
	2	0	0	0	0	0	0	0	0	0	0	0	0
	3	2	0	0	0	1	0	0	0	0	0	0	0.3
	4	2	6	0	4	5	4	6	5	6	7	0	4.5
	5	12	12	0	9	14	12	8	13	11	11	0	10.2
	6	21	0	4	0	19	0	0	0	0	0	0	4.4
	7	0	20	9	17	0	19	18	22	16	19	0	14

Total neonates 37 38 13 30 39 35 32 40 33 37  
Mean # young: 33.4  
Total adult mortality: 0

Concentration	Day	Replicate										% Mortality	Mean # Young
		1	2	3	4	5	6	7	8	9	10		
<b>3%</b>	1	0	0	0	0	0	0	0	0	0	0	0	0
	2	0	0	0	0	0	0	0	0	0	0	0	0
	3	1	0	0	3	5	0	0	0	0	0	0	0.9
	4	6	6	4	0	0	6	4	3	7	7	0	4.3
	5	17	14	3	12	12	12	11	7	10	13	0	11.1
	6	22	0	0	19	20	0	0	0	0	0	0	6.1
	7	0	20	16	0	0	21	14	14	19	18	0	12.2

Total neonates 46 40 23 34 37 39 29 24 36 38  
Mean # young: 34.6  
Total adult mortality: 0

Concentration	Day	Replicate										% Mortality	Mean # Young
		1	2	3	4	5	6	7	8	9	10		
<b>6%</b>	1	0	0	0	0	0	0	0	0	0	0	0	0
	2	0	0	0	0	0	0	0	0	0	0	0	0
	3	5	0	0	5	5	0	0	0	0	0	0	1.5
	4	0	6	4	0	0	6	5	5	6	7	0	3.9
	5	14	13	7	15	11	11	9	10	10	13	0	11.3
	6	19	21	0	11	17	0	0	0	0	0	0	6.8
	7	0	0	11	0	0	19	15	17	15	20	0	9.7

Total neonates 38 40 22 31 33 36 29 32 31 40  
Mean # young: 33.2  
Total adult mortality: 0

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x = mortality  
\* = accidental death

Test Data Reviewed By: *HL*  
Date: *2003-09-06*

**Ceriodaphnia dubia Test Report**  
Survival and Reproduction  
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Concentration	Day	Replicate										% Mortality	Mean # Young
		1	2	3	4	5	6	7	8	9	10		
13%	1	0	0	0	0	0	0	0	0	0	0	0	0
	2	0	0	0	0	0	0	0	0	0	0	0	0
	3	0	0	0	0	5	0	0	0	0	0	0	0.5
	4	3	2	0	0	0	0	3	5	0	2	0	1.5
	5	2	3	0	0	12	7	3	2	8	3	0	4
	6	0	0	0	0	19	0	0	0	0	0	0	1.9
	7	5	20	12	9 x	0	15	8	17	10	19	10	11.5
Total neonates		10	25	12	9 x	36	22	14	24	18	24		
Mean # young: 19.4													
Total adult mortality: 1													

Concentration	Day	Replicate										% Mortality	Mean # Young
		1	2	3	4	5	6	7	8	9	10		
25%	1	0	0	0	0	0	0	0	0	0	0	0	0
	2	0	0	0	0	0	0	0	0	0	0	0	0
	3	0	0	0	0	0	0	0	0	0	0	0	0
	4	0	0	0	0	0	0	0	0	0	0	0	0
	5	0	0	0	0	0	0	0	0	0	0	0	0
	6	0	0	0	0	0	0	0	0	0	0	0	0
	7	0	0	0	0	0	0	0	0	0	0	0	0
Total neonates		0	0	0	0	0	0	0	0	0	0		
Mean # young: 0													
Total adult mortality: 0													

Concentration	Day	Replicate										% Mortality	Mean # Young
		1	2	3	4	5	6	7	8	9	10		
50%	1	0	0	0	0	0	0	0	0	0	0	0	0
	2	0	0 x	0	0	0	0	0	0	0	0	10	0
	3	0	0	0	0	0	0	0	0	0	0	10	0
	4	0	0	0	0	0	0	0 x	0	0	0	20	0
	5	0	0	0 x	0	0	0	0	0	0	0	30	0
	6	0	0	0	0	0	0	0	0	0	0	30	0
	7	0	0	0	0	0	0	0	0	0	0	30	0
Total neonates		0	0 x	0 x	0	0	0	0 x	0	0	0		
Mean # young: 0													
Total adult mortality: 3													

Concentration	Day	Replicate										% Mortality	Mean # Young
		1	2	3	4	5	6	7	8	9	10		
100%	1	0	0	0	0	0	0	0	0	0	0	0	0
	2	0 x	0 x	0	0	0 x	0	0	0	0	0	30	0
	3	0	0	0	0	0	0	0	0	0	0	30	0
	4	0	0	0	0	0	0	0 x	0	0	0 x	50	0
	5	0	0	0 x	0 x	0	0 x	0	0	0 x	0	90	0
	6	0	0	0	0	0	0	0	0	0	0	90	0
	7	0	0	0	0	0	0	0	0	0	0	90	0
Total neonates		0 x	0 x	0 x	0 x	0 x	0 x	0 x	0	0 x	0 x		
Mean # young: 0													
Total adult mortality: 9													

**Stantec**

x = mortality  
\* = accidental death

Test Data Reviewed By: HL  
Date: 2003-09-06

***Ceriodaphnia dubia* Test Report**  
Survival and Reproduction  
5 of 6

Concentration	Day	Replicate										% Mortality	Mean # Young
		1	2	3	4	5	6	7	8	9	10		
13%	1	0	0	0	0	0	0	0	0	0	0	0	0
	2	0	0	0	0	0	0	0	0	0	0	0	0
	3	0	0	0	0	5	0	0	0	0	0	0	0.5
	4	3	2	0	0	0	0	3	5	0	2	0	1.5
	5	2	3	0	0	12	7	3	2	8	3	0	4
	6	0	0	0	0	19	0	0	0	0	0	0	1.9
	7	5	20	12	9	x	0	15	8	17	10	10	11.5
Total neonates		10	25	12	9	x	36	22	14	24	18	24	
Mean # young:													19.4
Total adult mortality:													1

Concentration	Day	Replicate										% Mortality	Mean # Young
		1	2	3	4	5	6	7	8	9	10		
25%	1	0	0	0	0	0	0	0	0	0	0	0	0
	2	0	0	0	0	0	0	0	0	0	0	0	0
	3	0	0	0	0	0	0	0	0	0	0	0	0
	4	0	0	0	0	0	0	0	0	0	0	0	0
	5	0	0	0	0	0	0	0	0	0	0	0	0
	6	0	0	0	0	0	0	0	0	0	0	0	0
	7	0	0	0	0	0	0	0	0	0	0	0	0
Total neonates		0	0	0	0	0	0	0	0	0	0	0	
Mean # young:													0
Total adult mortality:													0

Concentration	Day	Replicate										% Mortality	Mean # Young
		1	2	3	4	5	6	7	8	9	10		
50%	1	0	0	0	0	0	0	0	0	0	0	0	0
	2	0	0	x	0	0	0	0	0	0	0	10	0
	3	0	0	0	0	0	0	0	0	0	0	10	0
	4	0	0	0	0	0	0	0	x	0	0	20	0
	5	0	0	0	x	0	0	0	0	0	0	30	0
	6	0	0	0	0	0	0	0	0	0	0	30	0
	7	0	0	0	0	0	0	0	0	0	0	30	0
Total neonates		0	0	x	0	x	0	0	0	x	0	0	
Mean # young:													0
Total adult mortality:													3

Concentration	Day	Replicate										% Mortality	Mean # Young
		1	2	3	4	5	6	7	8	9	10		
100%	1	0	0	0	0	0	0	0	0	0	0	0	0
	2	0	x	0	x	0	0	0	0	0	0	30	0
	3	0	0	0	0	0	0	0	0	0	0	30	0
	4	0	0	0	0	0	0	0	x	0	0	50	0
	5	0	0	0	x	0	x	0	0	0	x	90	0
	6	0	0	0	0	0	0	0	0	0	0	90	0
	7	0	0	0	0	0	0	0	0	0	0	90	0
Total neonates		0	x	0	x	0	x	0	x	0	x	0	
Mean # young:													0
Total adult mortality:													9

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x = mortality  
\* = accidental death

Test Data Reviewed By: HL  
Date: 2003-09-06



**Stantec**

# **Lemna minor Test Report**

Growth Inhibition

1 of 5

Work Order : 204482  
 Sample Number : 7880

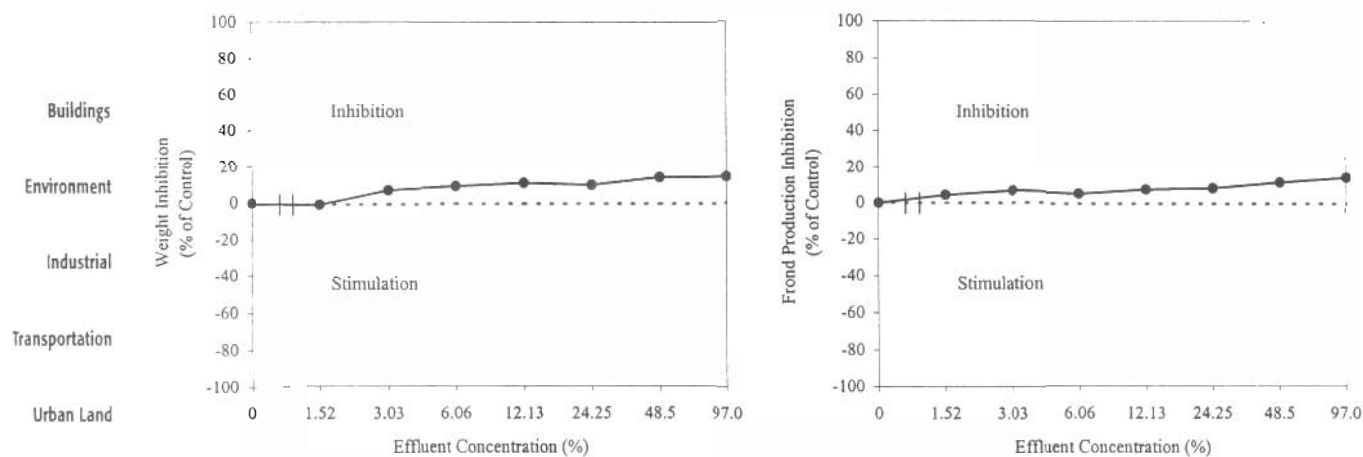
## **Sample Identification**

Company :	Nanisivik Mine (a Division of CanZinco Ltd.)	Date Collected :	2003-08-09
Location :	Nanisivik NU	Time Collected :	11:00
Substance :	159-4 August 8, 2003	Date Received :	2003-08-13
Sampling Method :	grab	Time Received :	10:30
Sampled By :	M. Markle	Date Tested :	2003-08-13
Shipped By:	FirstAir/air & Purolator/RD	Lab Storage:	4±2 °C
Temp. on arrival :	8.0°C		
Sample Description:	Clear, colourless, odourless.		

## **Test Results**

Effect	Value	95% Confidence Limits	Statistical Method
IC25 (Weight) <i>g dry wt</i>	>97.0%	-	-
IC25 (Frond Production)	>97.0%	-	-

## *Lemna minor* Growth Inhibition



**Lemna minor Test Report**

Growth Inhibition

2 of 5

Work Order Number: 204482

Sample Number: 7880

**Test Conditions**

Test Organism	: <i>Lemna minor</i> L., Strain 7730	Media Preparation Water <sup>b</sup>	: Millipore Super Q™
Organism Origin	: UTCC 492	Control/Dilution Water <sup>c</sup>	: Modified APHA
Source	: In-house culture	Test Type	: Static
Organism Age (days)	: 8	Test Volume Per Replicate	: 100 mL
Test Culture Frond # Increase	: >8	Test Vessel	: 250 mL Erlenmeyer flask
Initial Plant Inoculum	: 6 fronds (2 plants)	Depth of Test Solution	: 4.0 cm
Acclimation Time (h)	: 23:40	# Test Concentrations	: 6
Medium Sterilization	: No	# Control Replicates	: 3
Sample pH Adjustment	: None	# Test Replicates	: 3
Sample Hardness Adjustment	: None	Test Duration	: 7 days
Preaeration(≤100 bubbles/min)	: 20 min	Test Temperature	: 25 ± 2°C
Sample Filtration (µm)	: 1 (Whatman GF/C)	Lighting	: 24 hrs. 4200 – 4800 lux

<sup>b</sup> Media Preparation Water: Water used to prepare the liquid growth medium (Modified APHA). Obtained from the University of Guelph. No chemicals added.

<sup>c</sup> Control/Dilution Water: Millipore Super Q™ enriched with 10 mL/L nutrient stock solutions A, B, and C as described in EPS 1/RM/37.

Test Protocol: Environment Canada. 1999. Biological Test Method: Test for Measuring the Inhibition of Growth using the Freshwater Macrophyte, *Lemna minor*. Method Development and Application Section. Environmental Technology Centre. Environment Canada. Ottawa, ON. EPS 1/RM/37.

Test Organisms: No unusual appearance or treatment of culture prior to testing. Test culture was axenic.

**Comments**

Test sample was enriched with 10 mL/L nutrient stock solutions A, B, and C as described in EPS 1/RM/37.

Test dilutions were prepared and then allowed to acclimate for one hour before plants were added.

Control plants showed the required minimum growth of >8 times.

The holding time was extended to four days as the sample was collected on 2003-08-09, but was not received until 2003-08-13. A power failure occurred on 2003-08-14 which resulted in insufficient or variable lighting for 17 hours. There were no other unusual conditions or deviations from the test protocol. The results reported relate only to the sample tested.

**Lemna minor Test Report**

Growth Inhibition

3 of 5

Work Order Number: 204482  
Sample Number: 7880

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**Reference Toxicant Data**

Substance : Potassium Chloride  
Test Date : 2003-08-22  
IC25 Frond Production (g/L) : 2.64  
Lower 95% Confidence Limit : 0.43  
Upper 95% Confidence Limit : 3.43  
Test Duration : 7 days  
Historical Mean IC25 (g/L) : 2.25  
Upper Warning Limit (+2SD) : 3.58  
Lower Warning Limit (-2 SD) : 1.43  
Statistical Method : Linear Interpolation (Toxstat 3.5)<sup>a</sup>  
*Lemna minor* Batch Number : Lm03-08  
Test Conducted By : T.Patey/H.Roshon/K.Olaveson

Reference toxicant test was conducted under conditions identical to the test.

The first 5 reference toxicant tests were conducted using strain #8434, after which they were conducted using strain #7730.

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**References**

<sup>a</sup> West, Inc. and D. Gulley. 1996. Toxstat Release 3.5. Western Ecosystems Technology. Cheyenne, WY, U.S.A.

Date: 2003-09-10

Approved By: K Holtze  
Keith Holtze, Director, Laboratory Operations

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# Lemna minor Test Report

Growth Inhibition

4 of 5

## 7 Day Lemna minor Growth Inhibition Test

Work Order Number: 204482

Sample Number: 7880

Lemna Batch #: Lm03-08

Culture Age (days): 8

Culture Health Increase Factor (in APHA media): 11.2

Light Intensity: 4200-4800 lux

Sample Pre-aeration Time (min): 20

Sample Filtration (µm): 1

Acclimation of plants to APHA medium: Date: 2003-08-12 Time: 15:50

Start Date: 2003-08-13 Time: 15:30 Technician: T. Patey

Completion Date: 2003-08-20

Initial sample temperature: 25.0

Initial sample pH: 7.6

Daily Temperature			
Day	Date	Temp (°C)	Initials
0	2003-08-13	25.5	TP
1	2003-08-14	25.5	TP
2	2003-08-15	24.5	TP
3	2003-08-16	25.5	KO
4	2003-08-17	26.0	KO
5	2003-08-18	25.5	TP
6	2003-08-19	25.5	TP
7	2003-08-20	25.5	TP

pH		
Conc. (%)	Day 0	Day 7
Control	8.2	8.3
1.52	8.3	8.4
3.03	8.3	8.4
6.06	8.2	8.4
12.13	8.2	8.3
24.25	8.2	8.1
48.5	8.1	7.5
97.0	8.0	5.7

### FronD Counts at Day 7

Technician: K. Olaveson/ T. Patey/ H. Roshon

Conc. (%)	Rep A				Rep B				Rep C			
	Plant		FronD		Plant		FronD		Plant		FronD	
	# Live	# Dead	# Live	# Dead	# Live	# Dead	# Live	# Dead	# Live	# Dead	# Live	# Dead
Control	20	0	113	0	19	0	114	0	16	0	87	0
1.52	21	0	99	0	19	0	105	0	15	0	97	0
3.03	22	0	102	0	21	0	98	0	18	0	94	0
6.06	19	0	93	0	20	0	94	0	20	0	111	0
12.13	18	0	95	0	22	0	118	0	13	0	78	0
24.25	16	0	109	0	14	0	93	0	8	0	86	1
48.5	7	0	91	0	3	0	81	0	7	0	108	0
97.0	5	0	73	1	6	0	88	0	10	0	110	0

Summary (includes initial plant and frond numbers)

Conc. (%)	Average				Standard Deviation				Observations
	Plant		FronD		Plant		FronD		FronD/root appearance
	# Live	# Dead	# Live	# Dead	# Live	# Dead	# Live	# Dead	
Control	18.3	0.0	104.7	0.0	2.1	0.0	15.3	0.0	fronds healthy, appearance normal
1.52	18.3	0.0	100.3	0.0	3.1	0.0	4.2	0.0	
3.03	20.3	0.0	98.0	0.0	2.1	0.0	4.0	0.0	
6.06	19.7	0.0	99.3	0.0	0.6	0.0	10.1	0.0	
12.13	17.7	0.0	97.0	0.0	4.5	0.0	20.1	0.0	
24.25	12.7	0.0	96.0	0.3	4.2	0.0	11.8	0.6	
48.5	5.7	0.0	93.3	0.0	2.3	0.0	13.7	0.0	
97.0	7.0	0.0	90.3	0.3	2.6	0.0	18.6	0.6	

Notes: Large colonies in the 48.5% and 97.0% concentrations. Settled debris in the 97.0% concentration.

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Test Data Reviewed By: HL  
Date: 2003-09-03

**Lemna minor Test Report**

Growth Inhibition

5 of 5

**7 Day Lemna minor Growth Inhibition Test**

Work Order Number: 204482

Sample Number: 7880

**Increase in Frond Numbers**

Conc. (%)	Rep A	Rep B	Rep C	Average	Standard Deviation	%CV	% Inhibition
Control	107	108	81	98.7	15.3	15.5	0.0
1.52	93	99	91	94.3	4.2	4.4	4.4
3.03	96	92	88	92.0	4.0	4.3	6.8
6.06	87	88	105	93.3	10.1	10.8	5.4
12.13	89	112	72	91.0	20.1	22.1	7.8
24.25	103	87	81	90.3	11.4	12.6	8.4
48.5	85	75	102	87.3	13.7	15.6	11.5
97.0	68	82	104	84.7	18.1	21.4	14.2

Increase in control frond count: 17.4 fold

**Lemna minor Weights**

Concentration	Replicate	Total Replicate Biomass (mg)	Mean Treatment Biomass (mg)	Standard Deviation	%CV	% Inhibition
Control	A	10.10	9.14	1.45	15.9	0.0
	B	9.86				
	C	7.47				
1.52	A	9.57	9.17	0.49	5.4	-0.3
	B	9.31				
	C	8.62				
3.03	A	9.10	8.45	0.74	8.8	7.5
	B	8.62				
	C	7.64				
6.06	A	8.29	8.28	0.14	1.6	9.4
	B	8.41				
	C	8.14				
12.13	A	8.13	8.12	1.59	19.6	11.2
	B	9.71				
	C	6.53				
24.25	A	9.58	8.22	1.23	15.0	10.1
	B	7.90				
	C	7.18				
48.5	A	8.02	7.84	0.54	6.9	14.3
	B	7.23				
	C	8.26				
97.0	A	6.71	7.80	1.05	13.4	14.7
	B	7.88				
	C	8.80				

**Stantec**Test Data Reviewed By: HLDate: 2003-09-03



**Stantec**

*Selenastrum capricornutum*  
Growth Inhibition  
1 of 4

Work Order : 204482  
Sample Number : 7880

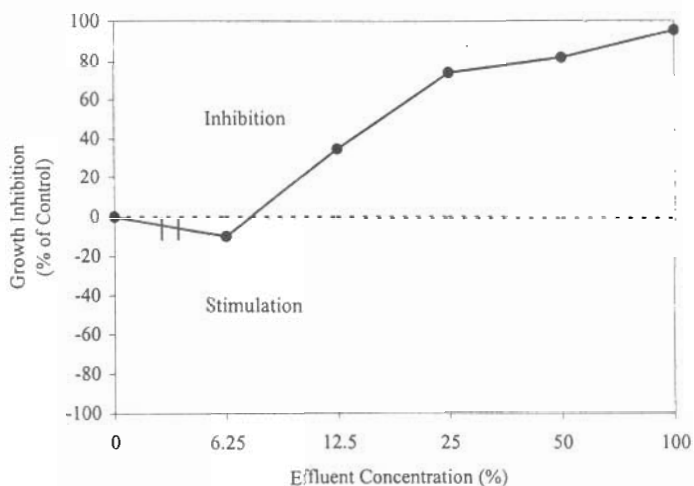
### Sample Identification

Company :	Nanisivik Mine (a Division of CanZinco Ltd.)	Date Collected :	2003-08-09
Location :	Nanisivik NU	Time Collected :	11:00
Substance :	159-4 August 8, 2003	Date Received :	2003-08-13
Sampling Method :	grab	Time Received :	10:30
Sampled By :	M. Markle	Date Tested :	2003-08-13
Shipped By:	FirstAir/air & Purolator/RD	Lab Storage:	4±2 °C
Temp. on arrival :	8.0°C		
Sample Description:	Clear, colourless, odourless.		

### Test Results\*

Effect	Value	95% Confidence Limits	Statistical Method
IC25 (Growth)	10.4%	9.4-10.8	Linear Interpolation (Toxstat 3.5) a

*Selenastrum capricornutum* Growth Inhibition



\*Note: The test results have not been adjusted for the 0.9091 dilution factor (see 'Concentrations' section on page 2).

Work Order Number: 204482  
Sample Number: 7880

### Test Conditions

Test Organism	: <i>Selenastrum capricornutum</i>	Control/Dilution Water <sup>c</sup>	: Millipore Milli Q™
Batch Number	: Sel03-08	Test Type	: Static
Organism Origin	: UTCC37	Test Volume Per Replicate	: 220 µL
Source	: In-house culture	Test Vessel	: U-shaped polystyrene microplate
Organism Age (days)	: 5 (in exponential growth)	# Test Concentrations <sup>d</sup>	: 10 + control
Initial Algal Inoculum	: 9448 cells/mL	# Control Replicates	: 10
pH Adjustment	: None	# Test Replicates	: 3
Sample Filtration <sup>b</sup>	: 0.45 µm	Test Duration	: 72 hours
Hardness Adjustment	: None	Lighting	: 24 hrs. 4000±10% lux
Sample Aeration	: None		

<sup>b</sup> Sample Filtration: 5-10 ml sub-sample was filtered through a preconditioned 0.45 µm filter prior to dilution.

<sup>c</sup> Control/Dilution Water: No chemicals added.

<sup>d</sup> Number of Concentrations: A total of ten concentrations were prepared and inoculated with algae. A minimum of five concentrations were enumerated at test completion.

Concentrations: Concentrations are prepared by making up a series of dilutions such as: 0.195, 0.39, 0.78, 1.56, 3.13, 6.25, 12.5, 25, 50, and 100%. The actual concentrations are slightly diluted by the addition of 10 µL of enrichment medium and 10 µL of algal inoculum. Therefore, the concentrations for the above series are actually 0.18, 0.35, 0.71, 1.42, 2.85, 5.68, 11.36, 22.73, 45.45, and 90.91% (a multiplication factor of 0.9091%). This multiplication factor has not been applied to the results.

Test Organisms: No unusual appearance or treatment of culture prior to testing.

Test Protocol: Biological Test Method. Growth Inhibition Test Using the Freshwater Alga *Selenastrum capricornutum*. Environment Canada, Conservation and Protection. Ottawa, Ontario. Report EPS1/RM/25 (including November 1997 amendments).