APPENDIX D

Bioasssay Reports

ECOTOXICOLOGY LABORATORY
EP LABORATORIES
PRAIRIE & NORTHERN REGION
5320-122 STREET
EDMONTON, ALBERTA
T6H 3S5

REPORT OF TOXICITY TESTING USING DAPHNIA MAGNA

LAB SAMPLE NO. 02-01442

COMPANY/LOCATION: Echo Bay Mines, Lupin

RESULTS TO: **Anne Wilson**
Environment Canada
EPB / Northern Division
Suite 301
5204 - 50th (Franklin) Avenue
Yellowknife, Northwest Territories
X1A 1E2

PURPOSE: To examine the test material to determine its toxicity to daphnia.

RESULT: Not Acutely Lethal. No Mortality at 100% Concentration.

REPORT AUTHORIZATION: **Head, Ecotoxicology Laboratory**

SIGNATURE/DATE: Garth Elliott, August 02, 2002

Garth Elliott

cc. Gordon Manners, EPB, TSD.

REPORT OF TOXICITY TESTING USING DAPHNIA MAGNA

LAB SAMPLE NO. 02-01442

COMPANY/LOCATION: Echo Bay Mines, Lupin

SAMPLE COLLECTED BY:	**Mackenzie Sawyer**
DATE/TIME SAMPLED:	**July 08, 2002 07:00 h**
DATE/TIME RECEIVED:	**July 09, 2002 10:55 h**
DATE/TIME TEST START:	**July 09, 2002 14:55 h**
SAMPLE TYPE:	**liquid**
SAMPLE POINT:	**Pond 2, site 102 - Final Tailings Containment Site**
SAMPLING METHOD:	**Grab**
SAMPLE CONTAINER:	**6 X 10 L white plastic pails ~ 60 L total volume**
TRANSPORTATION INFORMATION:	**Sample arrived via courier.**
STORAGE INFORMATION:	**None Required**

METHOD: **EP Laboratories SOP# 820.0 Revision 2, for Daphnia Magna Testing in Compliance with May 1996 and December 2000 Amended Methods: Reference Method for Determining Acute Lethality of Effluents to *Daphnia magna*, July 1990, EPS 1/RM/14. Single Concentration Procedure.**

DEVIATION FROM "MUST" REQUIREMENTS: One - See QA/QC Conformance

RESULT: Not Acutely Lethal. No Mortality at 100% Concentration.

STATISTICS: **N/A**

TEST CONDITIONS:

SPECIES: *Daphnia magna <= 24 h old*

DAPHNIA PER TEST VESSEL: 10

TEST VESSEL: 300 mL beaker

TEST VOLUME (ml): 150

PHOTO PERIOD: 16h Light, 8h Dark **LUX: 714**

DILUTION WATER: Treated Edmonton City Water

No pH Adjustment and All Tests Initiated Are Reported.

PHYSICOCHEMICAL CONDITIONS:

SAMPLE APPEARANCE: Clear in color.

INITIAL PHYSICOCHEMICAL CONDITIONS: **Just Prior to Setup**

INITIAL	TEMPERATURE (° C)	DISSOLVED OXYGEN (%saturation)	CONDUCTIVITY (µmhos/cm)	pH	Hardness (CaCO3/L)
100% SAMPLE	20.3	102	846	7.2	220

PREAERATION: (Oil-Free Compressed) at a Rate of 25-50 ml/min/L for 30 Minutes.

TEST PHYSICOCHEMICAL CONDITIONS:

TEST	TEMPERATURE (°C)		DISSOLVED OXYGEN (mg/L)		CONDUCTIVITY (µmhos/cm)		pH		HARDNESS (CaCO3/L)
CONCENTRATIONS	0 h	48 h	0 h	48 h	0 h	48 h	0 h	48 h	0 h
CONTROL A	20.4	21.0	8.2	7.9	360	351	8.4	8.4	155
CONTROL B	20.6	20.5	8.3	7.9	357	359	8.4	8.4	155
CONTROL C	20.6	20.5	8.3	7.9	359	355	8.4	8.4	155
100% A	20.5	20.4	8.5	7.9	847	849	7.5	7.6	220
100% B	20.1	20.2	8.5	7.9	858	852	7.3	7.5	220
100% C	19.7	20.2	8.4	7.9	856	859	7.3	7.5	220

TEST DATA:

TEST START DATE/TIME: July 09, 2002 1455 h

REPLICATES = None

	CUMULATIVE MORTALITY/SUBLETHAL EFFECT			
TEST CONCENTRATION	24 h #immobile	48 h #dead/immobile	% MORTALITY	% IMMOBILE
CONTROL A	0	0/0	0.0	0.0
CONTROL B	0	0/0	0.0	0.0
CONTROL C	0	0/0	0.0	0.0
100% A	0	0/0	0.0	0.0
100% B	0	0/0	0.0	0.0
100% C	0	0/0	0.0	0.0

Mean % Mortality = 0/30 = 0.0 % Mean % Immobilized = 0/30 = 0.0 %

QUALITY ASSURANCE/QUALITY CONTROL:

TEST ORGANISM: (Daphnia magna)

Stock neonates used for Testing: H neonates - May 23, 2002

Most Recent Estimate of Time to First Brood: 9.2 Days

Average Neonates per Brood: 16.7

Frequency of ephippia from Adults of Neonates: None

Any special handling during test: None Required

CONFORMANCE: **Control Mortality within Acceptable Limits**
Ephippia must not be Present in the Culture
Less Than 25% of Brood Stock Mortality in the 7 days prior to a Test
Method "Must" Requirements Followed - Except One: The temperature in the maintenance beakers exceded 22.0 °C on June 25, 2002 and June 26, 2002. As a result water temperatures were not maintained at 20.0 °C plus or minus 2.0 °C for two weeks prior to testing. The results will be considered valid as no mortality was observed in the control.

QUALITY CONTROL:

Reference Toxicant: Sodium Chloride

Reference Toxicant Test # 02-0022-D

Date of Test: July 16, 2002

Reference Toxicant Analyst: W.A., R.B.

LC_{50}- 48 h = 6560 ppm

95% Confidence Limits: 6250 - 6960 ppm

HISTORIC GEOMETRIC MEAN:

LC_{50}- 48 h = 6120 ppm

Warning Limits(2SD): 5610 - 6670 ppm

CONFORMANCE: Control Mortality within Acceptable Limits
Reference Toxicant within Warning Limits
Less Than 25% Brood Stock Mortality in the 7 days Prior to Test
Method "Must" Requirements Followed

TEST ANALYST(S): **Lisa Mitchelmore** **Initial:** LM

Rene Beaulieu **Initial:** RB

DATA VALIDATION: Wendy Antonielli **Initial:** WA
(name)

Glossary of Terms

°C - degree(s) Celsius
cm - centimeter(s)
d - day(s)
DO - dissolved oxygen (concentration)
EC_{50} - median effective concentration
g - gram(s)
h - hour(s)
H_20 - water
IC_{50} - 50% inhibiting concentration
LC_{50} - median lethal concentration
L - litre(s)
mg - milligram(s)
min - minute(s)
mL - millilitre(s)
mm - millimetre(s)
mS - millisiemens
NaCl - sodium chloride
N/A - non applicable
OAS - Osmotic Adjustment Solution
ppm - parts per million
µg - microgram
µL - microlitre
> - greater than
< - less than
≥ - greater than or equal to
≤ - less than or equal to
± - plus or minus
% percentage or percent

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REPORT OF TOXICITY TESTING USING RAINBOW TROUT

LAB SAMPLE NO. 02-01441

COMPANY/LOCATION: Echo Bay Mines, Lupin

RESULTS TO: **Anne Wilson**
Environment Canada
EPB / Northern Division
Suite 301
5204 - 50th (Franklin) Avenue
Yellowknife, Northwest Territories
X1A 1E2

PURPOSE: To examine the test material to determine its toxicity to fish.

RESULT: Not Acutely Lethal. No Mortality at 100% Concentration

REPORT AUTHORIZATION: **Head, Ecotoxicology Laboratory**

SIGNATURE/DATE: Garth Elliott, August 02, 2002

Garth Elliott

cc. **Gordon Manners, EPB, TSD.**

REPORT OF TOXICITY TESTING USING RAINBOW TROUT

LAB SAMPLE NO. 02-01441

COMPANY/LOCATION: Echo Bay Mines, Lupin

SAMPLE COLLECTED BY:	**Mackenzie Sawyer**
DATE/TIME SAMPLED:	**July 08, 2002 07:00 h**
DATE/TIME RECEIVED:	**July 09, 2002 10:55 h**
DATE/TIME TEST START:	**July 09, 2002 14:15 h**
SAMPLE TYPE:	**liquid**
SAMPLE POINT:	**Pond 2, site 102 - Final Tailings Containment Site**
SAMPLING METHOD:	**Grab**
SAMPLE CONTAINER:	**6 X 10 L white plastic pails ~ 60 L total volume**
TRANSPORTATION INFORMATION:	**Sample arrived via courier**
STORAGE INFORMATION:	**None Required**

METHOD: **EP Laboratories SOP# 810.0 Revision 3, for Trout Testing in Compliance with May 1996 and December 2000 Amended Methods: Reference Method for Determining Acute Lethality of Effluents to Rainbow Trout, July 1990, EPS 1/RM/13. Single Concentration Procedure.**

DEVIATION FROM "MUST" REQUIREMENTS: One - see QA/QC

RESULT: Not Acutely Lethal. No Mortality at 100% Concentration

STATISTICS: **N/A**

TEST CONDITIONS:

SPECIES: *Oncorhynchus mykiss*

FISH PER TEST VESSEL: 10

TEST VESSEL: 83 L plastic containers

TEST VOLUME (L): 64 **SAMPLE DEPTH (cm): 35**

PHOTO PERIOD: 16h Light, 8h Dark

DILUTION WATER: Treated Edmonton City Water

Fish Not Fed 24 h Prior to Test

No pH Adjustment and All Tests Initiated Are Reported.

Aeration Continuous Throughout Test at : 6.5 ± 1.0 ml/min/L. Measured with a Rotometer.

PHYSICOCHEMICAL CONDITIONS:

SAMPLE APPEARANCE: Clear

INITIAL PHYSICOCHEMICAL CONDITIONS: **Just Prior to Setup**

INITIAL	TEMPERATURE (° C)	DISSOLVED OXYGEN (%saturation)	CONDUCTIVITY (µmhos/cm)	pH
100% SAMPLE	14.9	97.9	862	7.3

PREAERATION: (Oil-Free Compressed) at a Rate of 6.5 ± 1.0 ml/min/L for 30 Minutes.

TEST PHYSICOCHEMICAL CONDITIONS:

TEST	TEMPERATURE (°C)					DISSOLVED OXYGEN (mg/L)		CONDUCTIVITY (µmhos/cm)		pH		LUX
CONCENTRATIONS	0 h	24 h	48 h	72 h	96 h	0 h	96 h	0 h	96 h	0 h	96 h	0 h
CONTROL	14.8	14.9	14.9	14.9	14.6	9.2	9.0	352	346	8.2	8.4	214
100%	14.8	15.1	15.0	14.9	14.9	9.2	8.9	865	859	7.4	7.5	126

TEST DATA:

TEST START DATE/TIME: July 09, 2002 14:15 h

REPLICATES = None

	CUMULATIVE MORTALITY/SUBLETHAL EFFECT				
TEST CONCENTRATION	24 h	48 h	72 h	96 h	% MORTALITY
CONTROL	0	0	0	0	0.0
100%	0	0	0	0	0.0

TROUT LENGTHS AND WEIGHTS:

See attached table.

QUALITY ASSURANCE/QUALITY CONTROL:

TEST ORGANISM: (Oncorhynchus mykiss) **Rainbow Trout Fingerlings**

Source: Ackenberry Trout Farm

Lot# ABTF061202 Tank# 3 Arrival Date: June 12, 2002

Mortality of Trout Stock 7 Days Prior to Test: 0.0 %

Acclimation for >= 2 Weeks at Temp: 15°C ± 2°C

Approximate No. Of Trout Acclimated: 403

Flow of Water Through Tank (L/g of trout per day): 14.9

Litres in Tank per 10g of Trout: 10.8

CONFORMANCE: Control Mortality within Acceptable Limits
Less Than 2% Fish Stock Mortality in 7 Days Prior to Test
Method "Must" Requirements Followed Except One - the fish holding temperature on June 28, 2002 increased to 19.8°C, outside the range of 15.0°C +/- 2.0°C. Since no mortality was observed in the control, the test is considered valid.

QUALITY CONTROL:

Reference Toxicant: Phenol

Reference Toxicant Test # 02-0010-T

Date of Test: July 22, 2002

Reference Toxicant Analyst: W.A., R.B.

LC_{50}- 96 h = 10.4 ppm

95% Confidence Limits: 9.23 - 11.8 ppm

HISTORIC GEOMETRIC MEAN:

LC_{50}- 96 h = 11.4 ppm

Warning Limits(2SD): 10.0 - 12.9 ppm

CONFORMANCE: Control Mortality within Acceptable Limits
Reference Toxicant within Warning Limits
Less Than 2% Fish Stock Mortality in the 7 days Prior to Test
Method "Must" Requirements followed

TEST ANALYST(S): **Wendy Antoniolli** **Initial:** WA

Nancy Kruper **Initial:** for Nancy Kruper.

DATA VALIDATION: Lisa Mitchelmore **Initial:** LM
(name)

LAB SAMPLE NO: 02-01441

Trout Lengths and Weights

Single Concentration

	CONTROL LENGTH (cm)	CONTROL LENGTH (cm)	CONTROL WEIGHT (g)	CONTROL WEIGHT (g)	100% LENGTH (cm)	100% LENGTH (cm)	100% WEIGHT (g)	100% WEIGHT (g)
1	3.4		0.36		4.1		0.70	
2	3.2		0.27		3.6		0.41	
3	3.3		0.37		3.4		0.36	
4	3.5		0.39		3.5		0.40	
5	3.6		0.38		3.6		0.39	
6	3.2		0.28		3.3		0.28	
7	3.4		0.34		3.4		0.35	
8	3.4		0.37		3.5		0.38	
9	3.7		0.40		3.7		0.43	
10	3.5		0.41		3.0		0.21	
AVERAGE	3.4		0.36		3.5		0.39	
STD DEV.	0.2		0.05		0.3		0.13	
RANGE	3.2 - 3.7		0.27 - 0.41		3.0 - 4.1		0.21 - 0.70	
VOL (L)			64				64	
TOT WT (g)			3.57				3.91	
LOADING DENSITY (g/L)			0.06				0.06	

Attachment #1

Glossary of Terms

°C - degree(s) Celsius
cm - centimeter(s)
d - day(s)
DO - dissolved oxygen (concentration)
EC_{50} - median effective concentration
g - gram(s)
h - hour(s)
H_20 - water
IC_{50} - 50% inhibiting concentration
LC_{50} - median lethal concentration
L - litre(s)
mg - milligram(s)
min - minute(s)
mL - millilitre(s)
mm - millimetre(s)
mS - millisiemens
NaCl - sodium chloride
N/A - non applicable
OAS - Osmotic Adjustment Solution
ppm - parts per million
µg - microgram
µL - microlitre
> - greater than
< - less than
≥ - greater than or equal to
≤ - less than or equal to
± - plus or minus
% percentage or percent