National Oil and Hazardous Substances Pollution Contingency Plan:
3.0 Revised Dispersant Toxicity Tests Report
Menidia beryllina* & *Mysidopsis bahia
FR / Vol. 59, No. 178 / 47461 - 47464

prepared for

Natural Solutions Group Corporation

Services Requested By: Marcos Gonzalez

$VirO_2Syl$. EE USA Project No.: D-006-12

Sample Received: March 6, 2012

Results:	*M. bahia* Survival		*M. beryllina* Survival	
	48-hr LC50	95% Confidence Interval	96-hr LC50	95% Confidence Interval
$VirO_2Syl$	3.95 ppm	2.66 – 5.38 ppm	94.8 ppm	72.4 – 124 ppm
No. 2 Fuel Oil	6.43 ppm	5.68 – 7.28 ppm	40.5 ppm	38.0 – 43.2 ppm
10:1 No. 2 Fuel Oil / $VirO_2Syl$	7.45 ppm	6.43 – 8.77 ppm	10.1 ppm	9.23 – 11.0 ppm
Reference Toxicant Sodium Dodecyl Sulfate (SDS)	8.68 ppm	7.49 – 10.1 ppm	2.33 ppm	2.12 – 2.56 ppm

Report Date: March 26, 2012

by

ENVIRONMENTAL ENTERPRISES USA, INC.

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This report contains ten pages plus twelve appendices, A – L.
This report must not be reproduced in part, only in whole. The results and conclusions presented in this report apply only to the sample(s) tested.
All results included in this report are from valid tests.

Veronica McNew
Effluents Testing Supervisor

03/26/12
DATE

Jennifer Griffith
QA/QC Officer

03-26-12
DATE

David L. Daniel
Laboratory Director

3/26/12
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***Menidia beryllina* ACUTE, STATIC 96-hr REVISED STANDARD DISPERSANT TOXICITY TEST, FR / Vol. 59, No. 178 / 47461 – 47464.**

TEST OVERVIEW

Four, 96-hr static definitive LC50 tests were conducted by Environmental Enterprises USA, Inc. (EE USA) using a Miscellaneous Oil Spill Control Agent ($VirO_2Syl$ supplied by Natural Solutions Group Corp., Appendix K), No. 2 Fuel Oil (obtained from Resource Technology Corporation, Lab #: D-020-10, Appendix L), a 10:1 mixture of No. 2 Fuel Oil to $VirO_2Syl$, and Sodium Dodecyl Sulfate (SDS) (obtained from Sigma-Aldrich Chemical, Lot #: 019K0085, 92.5 – 100.5% based on total alkyl sulfate content, specific gravity of 0.37 g/ml). *Menidia beryllina* was cultured at EE USA and the test organisms were 7 days old when each test was initiated. Preliminary range-finding tests were set prior to the definitive LC50 tests (Appendix A). Synthetic seawater, hw-MARINEMIX + Bio-Elements and Crystal Sea Marinemix Bioassay Laboratory Formula sea salts (80:20), was used as the laboratory performance control solution and diluent. Each definitive test included three replicates of a laboratory performance control solution and either five or six test concentrations. This document presents methods, materials, and results of this testing. The definitive tests were conducted from March 14 – March 18, 2012, at the laboratory of EE USA.

MATERIALS AND METHODS

Materials and methods for the work performed are stated in FR / Vol. 59, No.178 / 47461 – 47464: Revised Standard Dispersant Toxicity Test. Actual materials and methods are detailed below. The tests were performed with strict adherence to the method as presented in the Federal Register with the following exception(s):

1) during this test, recorded temperatures fell outside the required range by not more than 0.3°C on at least one occasion. This was a minor excursion and did not affect the results of this test.

M. beryllina were cultured and maintained at 24±1°C and 25±1ppt salinity. Several clutches from different females comprised the embryo pool from which the test organism population hatched. Prior to test initiation the test organisms were acclimated to 20±1ppt salinity. Test organisms were fed 200 µl of a standardized suspension of less than 24-hr old *Artemia* nauplii once daily. The standardized suspension is equal to 0.05 grams wet weight strained *Artemia* nauplii per ml synthetic seawater.

On March 6, 2012 $VirO_2Syl$ was received at EE USA from Natural Solutions Group Corp., (Appendix K). A 40-ml glass vial with a Teflon septum was completely filled with this sample and sealed. The remaining sample in the original product container was immediately resealed. A gas-tight syringe was used to prepare stock solutions (SSOLs) as required. The product was stored at ambient laboratory temperatures.

USEPA – API Reference Oil, No. 2 Fuel Oil, was purchased from RTC and was received at EE USA on June 29, 2010 (Appendix L). The No. 2 Fuel Oil arrived in two approximately 600 ml containers that were sealed and filled to approximately 500 ml. Twenty 40-ml glass vials with Teflon septums were completely filled with No. 2 Fuel Oil, sealed, and stored in a dark refrigerator at 0.1 – 6°C. Only one vial of No.2 Fuel Oil was used to complete these tests. A gas-tight syringe was used to deliver aliquots of No. 2 Fuel Oil.

Range-finding Tests:

On March 9, 2012 exploratory range-finding tests were initiated to estimate the LC50s of $VirO_2Syl$, No. 2 Fuel Oil, a 10:1 mixture of No. 2 Fuel Oil to $VirO_2Syl$, and SDS (Appendix A). Test chambers were labeled with test treatment, concentration, organism, replicate identification, and EE USA's project number. Test concentrations were prepared using aliquots of stock solutions (SSOLs) prepared from each neat material as received. Each SSOL and each 1-liter test concentration was prepared and mixed for five minutes using a reciprocal shaker table (EBERBACH 6010, 280 excursions/minute, 3.8 cm stroke, ID# SH1 & A43). One replicate of a concurrent laboratory performance control and at least five dilutions each of $VirO_2Syl$, No. 2 Fuel Oil, a 10:1 mixture of No. 2 Fuel Oil to $VirO_2Syl$, and SDS were prepared. One liter of each test concentration was transferred to replicate test chambers as appropriate, ten *M. beryllina* were randomly loaded, and each test chamber was put into an environmental chamber at 25 +/- 1°C.

LC50s obtained from exploratory range-finding tests were used to select a series of test concentrations for each definitive test ($VirO_2Syl$, No. 2 Fuel Oil, a 10:1 mixture of No. 2 Fuel Oil to $VirO_2Syl$, and SDS) that was expected to bracket the LC50. The SSOL concentration and volumes for each test were prepared following the examples given in the test method.

Definitive Tests:

On March 14, 2012, three replicates of a concurrent laboratory performance control and five dilutions each of $VirO_2Syl$, No. 2 Fuel Oil, a 10:1 mixture of No. 2 Fuel Oil to $VirO_2Syl$, and SDS were prepared and put into an environmental chamber at 25 +/- 1°C. Test chambers were labeled with test treatment, concentration, organism, replicate identification, and EE USA's project number. Test concentrations were prepared using aliquots of SSOLs prepared from each neat material as received. Each SSOL and each 3-liter test concentration was prepared and mixed for five minutes using a reciprocal shaker table (EBERBACH 6010, 280 excursions/minute, 3.8 cm stroke, ID# SH1 & A43). One liter of each test concentration was transferred to three replicate test chambers per treatment as appropriate. Appendices B, C, D, and E contain copies of the raw data recorded for each test.

Appendix #	Toxicant
B	$VirO_2Syl$
C	No. 2 Fuel Oil
D	10:1 No.2 Fuel Oil to $VirO_2Syl$
E	SDS, Standard Reference Toxicant

$VirO_2Syl$:

An exploratory range-finding toxicity test indicated an estimated LC50 of 88.4 ppm $VirO_2Syl$ (Appendix A). The range-finding test resulted in an LC50 which requires a stock solution greater in volume than the 1000 ppm stock solution example given in the test method. The definitive test was prepared with an 1100 ml SSOL at 1,000 ppm: 1.1 ml of $VirO_2Syl$ plus 1098.9 ml synthetic seawater. The SSOL solution was mixed on a reciprocal shaker for five minutes. Test concentrations were prepared using aliquots of the SSOL and synthetic seawater and then mixed on the reciprocal shaker for five minutes (Appendix B, page 1).

No. 2 Fuel Oil:

An exploratory range-finding toxicity test indicated an estimated LC50 of 29.5 ppm No. 2 Fuel Oil (Appendix A). The definitive test was prepared with a 550 ml SSOL at 1000 ppm: 0.55 ml No. 2 Fuel Oil plus 549.45 ml synthetic seawater. The SSOL was mixed on a reciprocal shaker for five minutes. Test concentrations were prepared using aliquots of the SSOL and synthetic seawater and then mixed on the reciprocal shaker for five minutes (Appendix C, page 1).

10:1 No. 2 Fuel Oil / $VirO_2Syl$:

An exploratory range-finding toxicity test indicated an estimated LC50 of 70.7 ppm 10:1 No. 2 Fuel Oil to $VirO_2Syl$ (Appendix A). This LC50 result was greater than 50 ppm, the highest concentration tested. The definitive test concentrations were selected using the estimated LC50 from the range-finding test, and test concentrations were selected at 62.5 and 125 ppm in order to bracket the estimated LC50 from the range-finding test. Six concentrations rather than five were tested to ensure that a definitive LC50 result was obtained. The definitive test was prepared with a stock solution greater in volume than the 1000 ppm stock solution example given in the test method, 1100 ml SSOL at 1000 ppm: 1.0 ml No. 2 Fuel Oil plus 0.1 ml $VirO_2Syl$ plus 1098.90 ml synthetic seawater. The SSOL was mixed on a reciprocal shaker for five minutes. Test concentrations were prepared using aliquots of the SSOL and synthetic seawater and then mixed on the reciprocal shaker for five minutes (Appendix D, page 1).

Standard Reference Toxicant, SDS:

Sensitivity of test organisms to a known toxicant was determined by performing a concurrent Standard Reference Toxicant (SRT) test with SDS. An exploratory range-finding toxicity test indicated an estimated LC50 of 2.30 ppm SDS (Appendix A). The definitive test was prepared with a 500 ml SSOL at 2000 ppm: 1.00 g SDS plus 497.3 ml synthetic seawater to a total volume of 500 ml. The SSOL was mixed on a reciprocal shaker for five minutes. Test concentrations were prepared using aliquots of the SSOL and synthetic seawater and then mixed on the reciprocal shaker for five minutes (Appendix E, page 1).

The initial temperature, dissolved oxygen [DO], and salinity in each treatment was measured and recorded. At the end of each 24-hour exposure period, the ending DO, temperature, salinity, and pH in each treatment was measured and recorded (Appendix #, pages 3 - 4). *M. beryllina* from the same lot of test organisms, lot#: MN-067-12, was used in each test (Appendix #, page 3). The tests were initiated from 1747 to 1934 on March 14, 2012: ten *M. beryllina* larvae were randomly distributed to each test chamber. At 24-hr intervals, the number of survivors in each replicate of each treatment was recorded (Appendix #, page 2). After 96 hours, the final survival data were recorded and these tests were terminated.

Summary of Experimental Conditions	
Test Organisms	7-day-old *Menidia beryllina* larvae
Dilution Water	Synthetic seawater, 20±1 ppt
Temperature	25 +/- 1°C
Photoperiod	16 hours light; 8 hours dark
Test Chambers	Rectangular Pyrex dish, 21cm x 11cm x 7cm
Total Chamber Volume	1.45 liters
Test Solution Volume	1000 ml
Test Solution Renewal	No
Aeration	No, DO levels remained ≥4.0 mg/L

RESULTS AND CONCLUSION

The response used in statistical analysis of survival data was the number of surviving test organisms per concentration. The 96-hr survival data were used to estimate the 96-hr LC50: a point estimate of the concentration expected to result in 50% mortality to exposed *M. beryllina* larvae after 96 hours of exposure. Survival in the concurrent laboratory performance control was 100.0%.

$VirO_2Syl$:

Definitive test concentrations tested were 6.3, 12.5, 25, 50, and 100 ppm $VirO_2Syl$. The 96-hr LC50 was 94.8 ppm with a 95% confidence interval of 72.4 to 124 ppm as determined by the Trimmed Spearman-Karber method (Appendix B, page 5a).

No. 2 Fuel Oil:

Definitive test concentrations tested were 3.8, 7.5, 15.0, 30.0, and 60.0 ppm No. 2 Fuel Oil. The 96-hr LC50 was 40.5 ppm with a 95% confidence interval of 38.0 to 43.2 ppm as determined by the Trimmed Spearman-Karber method (Appendix C, page 5a).

10:1 No. 2 Fuel Oil / $VirO_2Syl$:

Definitive test concentrations were 3.9, 7.8, 15.6, 31.3, 62.5 and 125.0 ppm 10:1 No. 2 Fuel Oil to $VirO_2Syl$. The 96-hr LC50 was 10.1 ppm with a 95% confidence interval of 9.23 to 11.0 ppm as determined by the Trimmed Spearman-Karber method (Appendix D, page 5a).

Standard Reference Toxicant, SDS:

Definitive test concentrations were 1.0, 1.7, 2.9, 4.8, and 8.0 ppm SDS. The 96-hr LC50 was 2.33 ppm with a 95% confidence interval of 2.12 to 2.56 ppm as determined by the Trimmed Spearman-Karber method (Appendix E, page 5a).

***Mysidopsis bahia* ACUTE, STATIC 48-hr REVISED STANDARD DISPERSANT TOXICITY TEST, FR / Vol. 59, No. 178 / 47461 – 47464.**

TEST OVERVIEW

Four, 48-hr static definitive LC50 tests were conducted by Environmental Enterprises USA, Inc. (EE USA) using a Miscellaneous Oil Spill Control Agent ($VirO_2Syl$ supplied by Natural Solutions Group Corp., Appendix K), No. 2 Fuel Oil (obtained from Resource Technology Corporation, Lab #: D-020-10, Appendix L), a 10:1 mixture of No. 2 Fuel Oil to $VirO_2Syl$, and Sodium Dodecyl Sulfate (SDS) (obtained from Sigma-Aldrich Chemical, Lot #: 019K0085, 92.5 – 100.5% based on total alkyl sulfate content, density of 0.37 g/ml). *Mysidopsis bahia* was cultured at EE USA and the test organisms were 5 days old when each test was initiated. Preliminary range-finding tests were set prior to the definitive LC50 tests (Appendix F). Synthetic seawater, hw-MARINEMIX + Bio-Elements and Crystal Sea Marinemix Bioassay Laboratory Formula sea salts (80:20), was used as the laboratory performance control solution and diluent. Each definitive test included three replicates of a laboratory performance control solution and five test concentrations. This document presents methods, materials, and results of this testing. The definitive tests were conducted from March 13 – March 15, 2012, at the laboratory of EE USA.

MATERIALS AND METHODS

Materials and methods for the work performed are stated in FR / Vol. 59, No.178 / 47461 - 47464: Revised Standard Dispersant Toxicity Test. Actual materials and methods are detailed below. The tests were performed with strict adherence to the method as presented in the Federal Register.

M. bahia were cultured and maintained at 24±1°C and 25±1 ppt salinity. Four days before initiating this test 12- to 24-hr-old mysids were collected from breeding cultures, moved to a holding system, and acclimated to 25±1°C. Prior to test initiation the test organisms were acclimated to 20±1 ppt salinity. Test organisms were fed 200 ul of a standardized suspension of less than 24-hr old *Artemia* nauplii once daily. The standardized suspension is equal to 0.05 grams wet weight strained *Artemia* nauplii per ml synthetic seawater.

On March 6, 2012 $VirO_2Syl$ was received at EE USA from Natural Solutions Group Corp. (Appendix K). A 40-ml glass vial with a Teflon septum was completely filled with this sample and sealed. The remaining sample in the original product container was immediately resealed. A gas-tight syringe was used to prepare stock solutions (SSOLs) as required. The product was stored at ambient laboratory temperatures.

USEPA – API Reference Oil, No. 2 Fuel Oil, was purchased from RTC and was received at EE USA on June 29, 2010 (Appendix L). The No. 2 Fuel Oil arrived in two approximately 600 ml containers that were sealed and filled to approximately 500 ml. Twenty 40-ml glass vials with Teflon septums were completely filled with No. 2 Fuel Oil, sealed, and stored in a dark refrigerator at 0.1 – 6°C. Only one glass vial of No.2 Fuel Oil was used to complete these tests. A gas-tight syringe was used to deliver aliquots of No. 2 Fuel Oil.

Range-finding Tests:

On March 9, 2012 exploratory range-finding tests were initiated to estimate the LC50s of $VirO_2Syl$, No. 2 Fuel Oil, a 10:1 mixture of No. 2 Fuel Oil to $VirO_2Syl$, and SDS (Appendix F). Test chambers were labeled with test treatment, concentration, organism, replicate identification, and EE USA's project number. Test concentrations were prepared using aliquots of stock solutions (SSOLs) prepared from each neat material as received. Each SSOL was prepared and mixed for five seconds using a blender (OSTER, Model #: MG-W00, ID# A40) at approximately 7300 to 8300 rpm. One replicate of a concurrent laboratory performance control and at least five dilutions each of $VirO_2Syl$, No. 2 Fuel Oil, a 10:1 mixture of No. 2 Fuel Oil to $VirO_2Syl$, and SDS were prepared by dispensing aliquots of each SSOL by pipet into the appropriate test chambers and adding 800 ml of dilution water into each test chamber. Ten *M. bahia* and 200 ml of dilution water were randomly distributed to each test chamber to bring the total volume of each test chamber up to 1000 ml. Test chambers were put into an environmental chamber at 25 +/- 1°C.

LC50s obtained from exploratory range-finding tests were used to select a series of test concentrations for each definitive test ($VirO_2Syl$, No. 2 Fuel Oil, a 10:1 mixture of No. 2 Fuel Oil to $VirO_2Syl$, and SDS) that was expected to bracket the LC50. The SSOL concentration and volumes for each test were prepared following the examples given in the test method.

Definitive Tests:

On March 13, 2012, three replicates of a concurrent laboratory performance control and five dilutions of each product were prepared and put into an environmental chamber. Test chambers were labeled with test treatment, concentration, organism, replicate identification, and EE USA's project number. Test concentrations were prepared using aliquots of SSOLs prepared from each neat material as received. Each SSOL was prepared and mixed for five seconds using a blender (OSTER, Model #: MG-W00, ID# A40) at approximately 7300 to 8300 rpm. Aliquots of each SSOL were dispensed directly by pipet into the appropriate test chambers and then 800 ml dilution water were poured into each test chamber. Appendices G, H, I, and J contain copies of the raw data recorded for each test.

Appendix #	Toxicant
G	$VirO_2Syl$
H	No. 2 Fuel Oil
I	10:1 No.2 Fuel Oil to $VirO_2Syl$
J	SDS, Standard Reference Toxicant

$VirO_2Syl$:

An exploratory range-finding toxicity test indicated an estimated LC50 of 6.28 ppm $VirO_2Syl$ (Appendix F). The definitive test was prepared with a 550 ml SSOL at 1,000 ppm: 0.55 ml of $VirO_2Syl$ plus 549.45 ml synthetic seawater. The SSOL was mixed with a blender for five seconds. The test solutions were mixed with aliquots of the SSOL and synthetic seawater (Appendix G, page 1).

No. 2 Fuel Oil:

An exploratory range-finding toxicity test indicated an estimated LC50 of 3.30 ppm No. 2 Fuel Oil (Appendix F). The definitive test was prepared with a 550 ml SSOL at 1000 ppm: 0.55 ml No. 2 Fuel Oil plus 549.45 ml synthetic seawater. The SSOL was mixed with a blender for five seconds. The test solutions were mixed with aliquots of the SSOL and synthetic seawater (Appendix H, page 1).

10:1 No. 2 Fuel / $VirO_2Syl$:

An exploratory range-finding toxicity test indicated an estimated LC50 of 5.12 ppm 10:1 No. 2 Fuel Oil to $VirO_2Syl$ (Appendix F). The definitive test was prepared with a 550 ml SSOL at 1000 ppm: 0.50 ml No. 2 Fuel Oil plus 0.05 ml $VirO_2Syl$ plus 549.45 ml synthetic seawater. The SSOL was mixed with a blender for five seconds. The test solutions were mixed with aliquots of the SSOL and synthetic seawater (Appendix I, page 1).

Standard Reference Toxicant, SDS:

Sensitivity of test organisms to a known toxicant was determined by performing a concurrent Standard Reference Toxicant (SRT) test with SDS. An exploratory range-finding toxicity test indicated an estimated LC50 of 8.21 ppm SDS (Appendix F). The definitive test was prepared with a 500 ml SSOL at 2000 ppm: 1.00 g or 2.7 ml SDS plus 497.3 ml synthetic seawater. The SSOL was mixed with a blender for five seconds. The test solutions were mixed with aliquots of the SSOL and synthetic seawater (Appendix J, page 1).

The initial temperature, dissolved oxygen [DO], and salinity in each treatment was measured and recorded. At the end of each 24-hour exposure period, the ending DO, temperature, salinity, and pH in each treatment was measured and recorded (Appendix #, page 3). *M. bahia* from the same lot of test organisms, lot#: MB-151-12, was used in each test (Appendix #, page 2). The tests were initiated from 1737 to 1855 on March 13, 2012: ten *M. bahia* and 200 ml of dilution water were randomly distributed to each test chamber. The 200 ml of dilution water transferred with the mysids to each test chamber brought the total volume in each up to 1000 ml. At 24-hr intervals, the number of survivors in each replicate of each treatment was recorded (Appendix #, page 2). After 48 hours, the final survival data were recorded and these tests were terminated.

Summary of Experimental Conditions	
Test Organisms	5-day-old *Mysidopsis bahia*
Dilution Water	synthetic seawater, 20±1 ppt
Temperature	25 +/-1°C
Photoperiod	16 hours light; 8 hours dark
Test Chambers	Rectangular Pyrex dish, 21cm x 11cm x 7cm
Total Chamber Volume	1.45 liters
Test Solution Volume	1000 ml
Test Solution Renewal	No
Aeration	No, DO levels remained ≥4.0 mg/L

RESULTS AND CONCLUSION

The response used in statistical analysis of survival data was the number of surviving test organisms per concentration. The 48-hr survival data were used to estimate the 48-hr LC50: a point estimate of the concentration expected to result in 50% mortality to exposed *M. bahia* after 48 hours of exposure. Survival in the concurrent laboratory performance control was 100.0%.

VirO$_2$Syl:

Definitive test concentrations were 1.6, 3.1, 6.3, 12.5, and 25.0 ppm VirO$_2$Syl. The 48-hr LC50 was 3.95 ppm with a 95% confidence interval of 2.66 to 5.38 ppm as determined by the Probit method (Appendix G, page 4a).

No. 2 Fuel Oil:

Definitive test concentrations were 0.8, 1.5, 3.0, 6.0, and 12.0 ppm No. 2 Fuel Oil. The 48-hr LC50 was 6.43 ppm with a 95% confidence interval of 5.68 to 7.28 ppm as determined by the Trimmed Spearman-Karber method (Appendix H, page 4a).

10:1 No. 2 Fuel Oil / VirO$_2$Syl:

Definitive test concentrations were 0.6, 1.3, 2.5, 5.0, and 10.0 ppm 10:1 No. 2 Fuel Oil to VirO$_2$Syl. The 48-hr LC50 was 7.45 ppm with a 95% confidence interval of 6.43 to 8.77 ppm as determined by the Probit method (Appendix I, page 4a).

Standard Reference Toxicant, SDS:

Definitive test concentrations were 0.9, 1.8, 3.5, 7.0 and 14.0 ppm SDS. The 48-hr LC50 was 8.68 ppm with a 95% confidence interval of 7.49 to 10.1 ppm as determined by the Probit method (Appendix J, page 4a).

REFERENCES

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Environmental Enterprises USA, Inc.

APPENDIX A

Inland Silverside, *Menidia beryllina*
Acute Static 96–Hour Product Range-Finder Test (RFT)
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Natural Solutions Group Corporation – VirO$_2$Syl
Contact: Marcos Gonzalez

Test Concentrations, ppm VirO2Syl

Menidia beryllina	Total Volume/ Concentration, ml	Color Code	ml SSOL	ml DH_2O
500.0 ppm	1000.00	Black	500.00	500.00
250.0 ppm	"	Brown	250.00	750.00
125.0 ppm	"	Yellow	125.00	875.00
62.5 ppm	"	Green	62.50	937.50
31.3 ppm	"	Blue	31.30	968.70
0 ppm LPC	"	White	0.00	1000.00
Total Volume (ml) of product needed per day = 968.80				

1100 ml Stock Solution (SSOL) @ 1000 ppm: 1.10 ml VirO2Syl + 1098.90 ml DH_2O

Data Pages & Calculations by: Veronica M. Ybarra QA/QC Check by: Jennifer Griffith

M. beryllina = 1 Rep x 1000 ml

DH_2O = Dilution Water = **20 ppt Synthetic Seawater**

	LPC	Meter ID#
Date	03/09	//////
DH_2O Lot #	25R-061-12	//////
Alkalinity	120	//////
Salinity	20.2	A46
pH	8.0	A93
Temp.	23.5	A46
	VR	//////

Prep Date	03/09
Shaker Table ID#	SHKR A43
Initial	VR

LPC: Laboratory Performance Control, 20 ppt synthetic seawater
Alkalinity: mg/l as $CaCO_3$ Salinity: ppt pH: su Temp: °C

Comments: *Artemia* Lot # 030211-2; Feed *M. beryllina* once daily.

Inland Silverside, *Menidia beryllina*
Acute Static 96–Hour Product Range-Finder Test (RFT)
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Survival Data

Treatment, ppm VirO2Syl														
Time	REP	LPC 0 ppm White	REP	/////	REP	31.3 ppm Blue	REP	62.5 ppm Green	REP	125 ppm Yellow	REP	250 ppm Brown	REP	500 ppm Black	**Date & Initials**
0 HR 1738	1	10		/////	2	10	3	10	4	10	5	10	6	10	03/09/12
24 HR 1238	1	10		/////	2	10	3	10	4	10	5	9	6	6	03/10/12 VL
48 HR 1433	1	10		/////	2	10	3	10	4	8	5	7	6	1	03/11/12 VL
72 HR 1619	1	10		/////	2	10	3	10	4	3	5	4	6	1	03/12/12 VL
96 HR 1545	1	10		/////	2	10	3	10	4	0	5	0	6	0	03/13/12 VL
% Survival		100		/////		100		100		0		0		0	

Counted by: Penny Branch QC/QA by: Veronica McMenamin

Loaded by: Veronica McMenamin

Test Organisms Age: 7 days old Test Organisms Source: EE

Test Organisms Lot #: MN-062-12

Data Entry by: Veronica McMenamin

Double Data Entry by: Veronica McMenamin and/or

QC/QA by: Jennifer Griffith

Comments:

Product, RFT

D-006-12
LC50

M. beryllina Water Quality Data

LPC & All Treatments: Initial & Final Temp.: 23.5 to 26.4°C; Initial & Final DO: 4.0 to ≤ 7.4 mg/l
LPC: Initial Salinity: 18.5 to 21.4 ppt

0 to 24 HR		Treatment, ppm VirO2Syl							
03/09/12 – 03/10/12		LPC	/////	31.3 ppm	62.5 ppm	125 ppm	250 ppm	500 ppm	Meter ID#
DO	I	7.1	/////	7.4	7.5	7.6	7.9	8.7	S7
	F	7.1	/////	7.9	7.8	7.7	7.9	9.1	S7
Temp	I	23.5	/////	23.6	23.6	23.6	23.5	23.6	A46
	F	24.1	/////	23.9	23.8	24.0	23.9	23.9	A46
Salinity	I	20.2	/////	20.2	20.2	20.2	20.2	20.2	A46
	F	20.3	/////	20.3	20.2	20.2	20.2	20.3	A46
pH	F	7.9	/////	8.0	8.0	8.0	8.0	8.0	A93
Initials		Initial: SG VL				Final: SD VL			
Times		Initial Time: 1846				Final Time: 0804			

48 HR		Treatment, ppm VirO2Syl							
03/11/12		LPC	/////	31.3 ppm	62.5 ppm	125 ppm	250 ppm	500 ppm	Meter ID#
DO	F	6.8	/////	7.0	7.8	12.4	8.4	8.6	S7
Temp	F	25.0	/////	24.7	24.6	25.0	24.8	24.6	A46
Salinity	F	20.4	/////	20.5	20.4	20.4	20.3	20.3	A46
pH	F	8.0	/////	8.0	8.0	8.0	8.0	8.0	A93
Initials		Final: VM SD							
Times		Final Time: 0756							

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

Comments: _____

M. beryllina Water Quality Data

LPC & All Treatments: Initial & Final Temp.: 23.5 to 26.4°C; Initial & Final DO: 4.0 to ≤ 7.4 mg/l
LPC: Initial Salinity: 18.5 to 21.4 ppt

72 HR	Treatment, ppm VirO2Syl							
03/12/12	LPC	/////	31.3 ppm	62.5 ppm	125 ppm	250 ppm	500 ppm	Meter ID#
DO F	7.0	/////	7.0	7.0	7.1	12.6	8.7	S7
Temp F	24.5	/////	24.4	24.4	24.3	24.5	24.3	A46
Salinity F	20.6	/////	20.7	20.5	20.5	20.4	20.5	A46
pH F	8.0	/////	7.9	7.9	8.0	8.0	8.0	A93
Initials	Final: ME JG							
Times	Final Time: 1607							

96 HR	Treatment, ppm VirO2Syl							
03/13/12	LPC	/////	31.3 ppm	62.5 ppm	125 ppm	250 ppm	500 ppm	Meter ID#
DO F	6.7	/////	6.9	6.7	8.9	8.4	8.4	S7
Temp F	25.3	/////	24.8	24.8	24.8	25.1	24.8	A46
Salinity F	20.7	/////	20.8	20.5	20.5	20.7	20.5	A46
pH F	8.0	/////	8.0	7.9	7.9	8.0	8.0	A93
Initials	Final: MR SD							
Times	Final Time: 0916							

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

Comments: _____

Data Pages

- ☑ Calculations on mixing page are correct. (sign mixing page)
- ☑ Dates, dilutions, test method, # of replicates, replicate volume, product, acceptance limits, data analysis endpoint, and test organisms are correct throughout data pages.
- ☑ Format correct. (spaces for all entries, page numeration, no split pages, etc.)

VL Initials 03-09-12 Date

Chain-of-Custody

- ☑ Product on COC matches sample bottle/container.
- ☑ Product on COC matches test data pages.
- ☑ Lab # on COC matches sample bottle/container.
- ☑ Lab # on COC matches test data pages.
- ☑ Sample volume/amount is sufficient for test duration. (Sample volume in container(s) checked against sample volume on mixing page)

Sample volume/amount available: 8000 (ml) or g

Sample volume/amount needed: 0.55 (ml) or g

(Sample volume/amount insufficient if sample volume/amount available < sample volume/amount needed)

VL Initials 03/09/12 Date

Labels

- ☑ Lab # on labels matches test data pages.
- ☑ Test organism on data pages match labels (i.e. *Menidia beryllina* = MN, *Mysidopsis bahia* = MB)
- ☑ Test Concentrations on labels match Test Concentrations on test data pages.

VL Initials 03/09/12 Date

Raw Data QC/QA'd by: Veronica M. Nee 03/09/12

Acute Toxicity Test-96 Hr Survival

Start Date:	3/9/2012	Test ID:	mn00612RP	Sample ID:	NCP-National Contingency Plan
End Date:	3/13/2012	Lab ID:	EE-Environmental Enterprise	Sample Type:	PRD-Product
Sample Date:		Protocol:	EPAM 02-EPA Marine	Test Species:	MB-Menidia beryllina
Comments:					

Conc-ppm	1
PC-LP Control	1.0000
31.3	1.0000
62.5	1.0000
125	0.0000
250	0.0000
500	0.0000

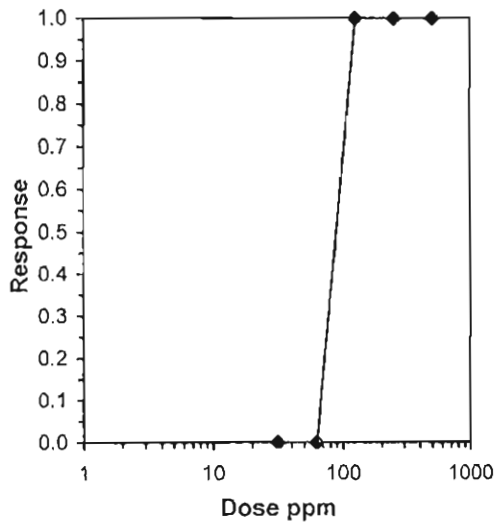
			Transform: Untransformed					Number	Total
Conc-ppm	Mean	N-Mean	Mean	Min	Max	CV%	N	Resp	Number
PC-LP Control	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
31.3	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
62.5	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
125	0.0000	0.0000	0.0000	0.0000	0.0000	0.000	1	10	10
250	0.0000	0.0000	0.0000	0.0000	0.0000	0.000	1	10	10
500	0.0000	0.0000	0.0000	0.0000	0.0000	0.000	1	10	10

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Normality of the data set cannot be confirmed				
Equality of variance cannot be confirmed				

Graphical Method

Trim Level	EC50
0.0%	88.388

88.388



Inland Silverside, *Menidia beryllina*
Acute Static 96–Hour No. 2 Fuel Oil Range-Finder Test (RFT)
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Test Concentrations, ppm No.2 Fuel Oil

Menidia beryllina	Total Volume/ Concentration, ml	Color Code	ml SSOL	ml DH_2O
50 ppm	1000.00	Black	50.00	950.00
25 ppm	"	Brown	25.00	975.00
12.5 ppm	"	Yellow	12.50	987.50
6.3 ppm	"	Green	6.30	993.70
3.2 ppm	"	Blue	3.20	996.80
0 ppm LPC	"	White	0.00	1000.00
		Total Volume (ml) of SSOL needed per day =		97.00

550 ml Stock Solution (SSOL) @ 1000 ppm = 0.55 ml No.2 Fuel Oil + 549.45 ml DH_2O

Data Pages & Calculations by: Veronica McMenamin QA/QC Check by: Jennifer Griffith

M. beryllina = 1 Rep x 1000 ml

DH_2O = Dilution Water = **20 ppt Synthetic Seawater**

	LPC	Meter ID#
Date	03/09	//////
DH_2O Lot #	25R-001-12	//////
Alkalinity	120	//////
Salinity	20.2	A462
pH	8.0	A93
Temp.	23.5	A412
	VR	//////

Prep Date	03/09
Shaker Table ID#	SH1 & A43
Initial	VR

LPC: Laboratory Performance Control, 20 ppt synthetic seawater
Alkalinity: mg/l as $CaCO_3$ Salinity: ppt pH: su Temp: °C

Comments: *Artemia* Lot # 030211-2; Feed *M. beryllina* once daily.

Inland Silverside, *Menidia beryllina*
Acute Static 96–Hour No. 2 Fuel Oil Range-Finder Test (RFT)
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Survival Data

Treatment, ppm No.2 Fuel Oil																		
Time	**REP**	**LPC 0 ppm White**	**REP**	/////	**REP**	**3.2 ppm Blue**	**REP**	**6.3 ppm Green**	**REP**	**12.5 ppm Yellow**	**REP**	**25.0 ppm Brown**	**REP**	**50.0 ppm Black**	**Date & Initials**
0 HR 1900	1	10		/////	2	10	3	10	4	10	5	10	6	10	03/09/12 VL
24 HR 1241	1	10		/////	2	10	3	10	4	10	5	10	6	1	03/10/12 JG
48 HR 1437	1	10		/////	2	10	3	10	4	9	5	9	6	0	03/11/12 VL
72 HR 1622	1	10		/////	2	10	3	10	4	9	5	9	6	0	03/12/12 JG
96 HR 1759	1	10		/////	2	10	3	10	4	9	5	9	6	0	03/13/12 VL
% Survival		100				100		100		90		90		0	

Counted by: Jenny Bunch QC/QA by: Veronica McNew

Loaded by: Veronica McNew

Test Organisms Age: 7 days old Test Organisms Source: EE

Test Organisms Lot #: MN-062-12

Data Entry by: Veronica McNew

Double Data Entry by: Veronica McNew and/or

QC/QA by: Jennifer Griffith

Comments:

M. beryllina Water Quality Data

LPC & All Treatments: Initial & Final Temp.: 23.5 to 26.4°C; Initial & Final DO: 4.0 to ≤ 7.4 mg/l
LPC: Initial Salinity: 18.5 to 21.4 ppt

0 to 24 HR	Treatment, ppm No.2 Fuel Oil							
03/09/12 – 03/10/12	LPC	/////	3.2 ppm	6.3 ppm	12.5 ppm	25.0 ppm	50.0 ppm	Meter ID#
DO I	7.1	/////	7.1	7.1	7.1	7.3	7.2	S7
F	7.1	/////	7.0	7.0	6.9	7.0	7.0	S7
Temp I	23.5	/////	23.6	23.9	24.0	23.7	23.7	~~A47~~ A46 (A)
F	24.1	/////	25.0	24.9	25.2	25.0	25.1	A46
Salinity I	20.2	/////	20.3	20.2	20.2	20.2	20.3	A46
F	20.3	/////	20.3	20.3	20.3	20.3	20.3	A46
pH F	7.9	/////	8.0	8.0	8.0	8.0	8.0	A93
Initials	Initial: JC				Final: SDVM			
Times	Initial Time: 1909				Final Time: 0808			

48 HR	Treatment, ppm No.2 Fuel Oil							
03/11/12	LPC	/////	3.2 ppm	6.3 ppm	12.5 ppm	25.0 ppm	50.0 ppm	Meter ID#
DO F	6.8	/////	6.7	6.8	6.8	6.7	6.5	S7
Temp F	25.0	/////	26.0	25.6	26.0	25.9	25.8	A46
Salinity F	20.4	/////	20.5	20.5	20.5	20.4	20.4	A46
pH F	8.0	/////	8.0	8.0	8.0	8.0	7.9	A93
Initials	Final: SDVM							
Times	Final Time: 0801							

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

Comments: (A) wrong data. JC 030912

M. beryllina Water Quality Data

LPC & All Treatments: Initial & Final Temp.: 23.5 to 26.4°C; Initial & Final DO: 4.0 to ≤ 7.4 mg/l
LPC: Initial Salinity: 18.5 to 21.4 ppt

72 HR	Treatment, ppm No.2 Fuel Oil							
03/12/12	LPC	/////	3.2 ppm	6.3 ppm	12.5 ppm	25.0 ppm	50.0 ppm	Meter ID#
DO F	7.0	/////	6.6	6.4	6.4	6.6		S7
Temp F	24.5	/////	24.4	24.1	24.4	24.2		A46
Salinity F	20.6	/////	20.7	20.6	20.7	20.6		A46
pH F	8.0	/////	7.9	7.9	7.9	7.9		A93
Initials	Final: MEJG							
Times	Final Time: 1612							

96 HR	Treatment, ppm No.2 Fuel Oil							
03/13/12	LPC	/////	3.2 ppm	6.3 ppm	12.5 ppm	25.0 ppm	50.0 ppm	Meter ID#
DO F	6.7	/////	6.6	6.5	6.4	6.5		S7
Temp F	25.3	/////	25.0	24.8	25.0	24.9		A46
Salinity F	20.7	/////	20.7	20.7	20.8	20.9		A46
pH F	8.0	/////	8.0	7.9	7.9	7.9		A93
Initials	Final: MR SD							
Times	Final Time: 0919							

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

Comments:

Data Pages

- ☑ Calculations on mixing page are correct. (sign mixing page)
- ☑ Dates, dilutions, test method, # of replicates, replicate volume, product, acceptance limits, data analysis endpoint, and test organisms are correct throughout data pages.
- ☑ Format correct. (spaces for all entries, page numeration, no split pages, etc.)

JC Initials 03-09-12 Date

Chain-of-Custody

- ☑ Product on COC matches sample bottle/container.
- ☑ Product on COC matches test data pages.
- ☑ Lab # on COC matches sample bottle/container.
- ☑ Lab # on COC matches test data pages.
- ☑ Sample volume/amount is sufficient for test duration. (Sample volume in container(s) checked against sample volume on mixing page)

Sample volume/amount available: 40 (ml) or g

Sample volume/amount needed: ~~0.50~~ 0.55 wrong data 03/09/12 VL ~~0.50~~ ml No.2 Fuel Oil ml or g

(Sample volume/amount insufficient if sample volume/amount available < sample volume/amount needed)

VL Initials 03/09/12 Date

Labels

- ☑ Lab # on labels matches test data pages.
- ☑ Test organism on data pages match labels (i.e. *Menidia beryllina* = MN, *Mysidopsis bahia* = MB)
- ☑ Test Concentrations on labels match Test Concentrations on test data pages.

VL Initials 03/09/12 Date

Raw Data QC/QA'd by: Veronica McManus 03/13/12

Acute Toxicity Test-96 Hr Survival

Start Date:	3/9/2012	Test ID:	mn00812RO	Sample ID:	NCP-National Contingency Plan
End Date:	3/13/2012	Lab ID:	EE-Environmental Enterprise	Sample Type:	2FO-No. 2 Fuel Oil
Sample Date:		Protocol:	EPAM 02-EPA Marine	Test Species:	MB-Menidia beryllina
Comments:					

Conc-ppm	1
PC-LP Control	1.0000
3.2	1.0000
6.3	1.0000
12.5	0.9000
25	0.9000
50	0.0000

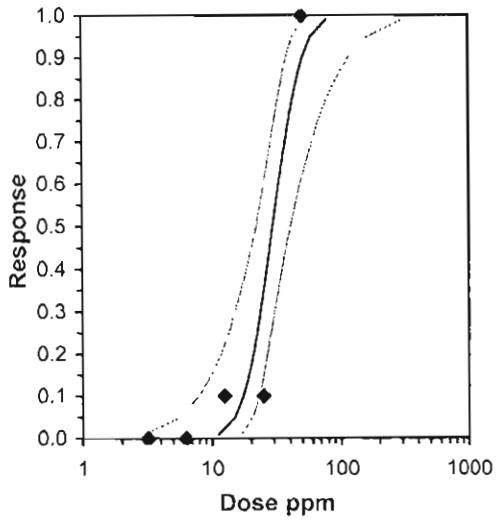
			Transform: Untransformed					Number	Total
Conc-ppm	Mean	N-Mean	Mean	Min	Max	CV%	N	Resp	Number
PC-LP Control	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
3.2	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
6.3	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
12.5	0.9000	0.9000	0.9000	0.9000	0.9000	0.000	1	1	10
25	0.9000	0.9000	0.9000	0.9000	0.9000	0.000	1	1	10
50	0.0000	0.0000	0.0000	0.0000	0.0000	0.000	1	10	10

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Normality of the data set cannot be confirmed				
Equality of variance cannot be confirmed				

Maximum Likelihood-Probit

Parameter	Value	SE	95% Fiducial Limits		Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	5.53743	1.61712	2.36787	8.70699	0	7.21681	7.81473	0.0653	1.47017	0.18059	8
Intercept	-3.141	2.38414	-7.8139	1.53195							
TSCR											

Point	Probits	ppm	95% Fiducial Limits	
EC01	2.674	11.2216	2.98145	16.7546
EC05	3.355	14.898	5.66303	20.4923
EC10	3.718	17.3275	7.91245	22.987
EC15	3.964	19.1867	9.86136	24.9763
EC20	4.158	20.8057	11.6901	26.8099
EC25	4.326	22.303	13.4612	28.629
EC40	4.747	26.5716	18.6219	34.841
EC50	**5.000**	**29.5236**	**21.9648**	**40.4091**
EC60	5.253	32.8036	25.2485	48.0907
EC75	5.674	39.0818	30.4034	67.2357
EC80	5.842	41.8945	32.3818	77.6245
EC85	6.036	45.4295	34.6819	92.2241
EC90	6.282	50.3041	37.6107	115.161
EC95	6.645	58.5076	42.115	161.189
EC99	7.326	77.6752	51.4258	306.669



3/9/2012 10:05 AM
Environmental Enterprises USA, Inc.

Inland Silverside, *Menidia beryllina*
Acute Static 96–Hour 10:1 No. 2 Fuel Oil/ Product Range-Finder Test (RFT)
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Natural Solutions Group Corporation – $VirO_2Syl$
Contact: Marcos Gonzales

Test Concentrations, ppm 10:1 No.2 Fuel Oil / $VirO_2Syl$

Menidia beryllina	Total Volume/ Concentration, ml	Color Code	ml SSOL	ml DH_2O
50.0 ppm	1000.00	Black	50.00	950.00
25.0 ppm	"	Brown	25.00	975.00
12.5 ppm	"	Yellow	12.50	987.50
6.3 ppm	"	Green	6.30	993.70
3.2 ppm	"	Blue	3.20	996.80
1.6 ppm	"	Wh/Blu	1.60	998.40
0 ppm LPC	"	White	0.00	1000.00
		Total Volume (ml) of SSOL needed per day =		98.60

550 ml Stock Solution (SSOL) @ 1000 ppm:
= 0.50 ml No.2 Fuel Oil + 0.05 ml $VirO_2Syl$ + 549.45 ml DH_2O

Data Pages & Calculations by: Veronica McKeon QA/QC Check by: Jennifer Griffith

M. beryllina = 1 Rep x 1000 ml

DH_2O = Dilution Water = **20 ppt Synthetic Seawater**

	LPC	Meter ID#
Date	03/09	//////
DH_2O Lot #	75R-061-12	//////
Alkalinity	120	//////
Salinity	20.2	A44
pH	8.0	A93
Temp.	23.5	A46
	VL	//////

Prep Date	03/09
Shaker Table ID#	S141 A43
Initial	VL

LPC: Laboratory Performance Control, 20 ppt synthetic seawater
Alkalinity: mg/l as $CaCO_3$ Salinity: ppt pH: su Temp: °C

Comments: *Artemia* Lot # 030211-2; Feed *M. beryllina* once daily.

Inland Silverside, *Menidia beryllina*

Acute Static 96–Hour 10:1 No. 2 Fuel Oil / Product Range-Finder Test (RFT)

3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Survival Data

Treatment ppm, 10:1 No. 2 Fuel Oil / VirO$_2$Syl														
Time	REP	LPC 0 ppm White	REP	1.6 ppm Wh/Blu	REP	3.2 ppm Blue	REP	6.3 ppm Green	REP	12.5 ppm Yellow	REP	25.0 ppm Brown	REP	50.0 ppm Black	Date & Initials
0 HR 1849	1	10	2	10	3	10	4	10	5	10	6	10	7	10	03/09/12 TB
24 HR 1240	1	10	2	10	3	10	4	10	5	10	6	10	7	10	03/10/12 JG
48 HR 1435	1	10	2	10	3	10	4	10	5	10	6	10	7	8	03/11/12 VL
72 HR 1621	1	10	2	10	3	10	4	10	5	10	6	10	7	7	03/12/12 JG
96 HR 1758	1	10	2	10	3	10	4	10	5	10	6	9	7	7	03/13/12 VL
% Survival		100		100		100		100		100		90		70	

Counted by: Jenny Bunch QC/QA by: Veronica McMenamin

Loaded by: Jenny Bunch

Test Organisms Age: 7 days old Test Organisms Source: CE

Test Organisms Lot #: MA-062-12

Data Entry by: Veronica McMenamin

Double Data Entry by: Veronica McMenamin and/or

QC/QA by: Jennifer Griffith

Comments:

M. beryllina Water Quality Data

LPC & All Treatments: Initial & Final Temp.: 23.5 to 26.4°C; Initial & Final DO: 4.0 to ≤ 7.4 mg/l
LPC: Initial Salinity: 18.5 to 21.4 ppt

0 to 24 HR	Treatment, ppm 10:1 No. 2 Fuel Oil / VirO$_2$Syl							
03/09/12-03/10/12	LPC	1.6 ppm	3.2 ppm	6.3 ppm	12.5 ppm	25.0 ppm	50.0 ppm	Meter ID#
DO I	7.1	7.3	7.2	7.3	7.3	7.3	7.3	57
F	7.1	7.2	7.2	7.2	7.1	7.1	7.3	57
Temp I	23.5	23.6	23.6	23.5	23.5	23.7	23.9	A46
F	24.1	24.1	23.9	23.8	24.2	24.1	23.9	A46
Salinity I	20.2	20.0	19.4	19.7	19.8	20.2	20.2	A46
F	20.3	20.0	19.5	19.8	19.9	20.2	20.2	A46
pH F	7.9	8.0	8.0	8.0	8.0	8.0	8.0	A93
Initials	Initial: SG				Final: SD VM			
Times	Initial Time: 1850				Final Time: 0806			

48 HR	Treatment, ppm 10:1 No. 2 Fuel Oil / VirO$_2$Syl							
03/11/12	LPC	1.6 ppm	3.2 ppm	6.3 ppm	12.5 ppm	25.0 ppm	50.0 ppm	Meter ID#
DO F	6.8	6.9	7.0	7.0	6.9	6.9	6.9	57
Temp F	25.0	25.0	24.7	24.5	24.6	24.8	24.7	A46
Salinity F	20.4	20.2	19.6	19.9	20.0	20.3	20.3	A46
pH F	8.0	8.0	8.0	8.0	8.0	8.0	8.0	A93
Initials	Final: SD VM							
Times	Final Time: 0759							

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

Comments: _____

M. beryllina Water Quality Data

LPC & All Treatments: Initial & Final Temp.: 23.5 to 26.4°C; Initial & Final DO: 4.0 to ≤ 7.4 mg/l
LPC: Initial Salinity: 18.5 to 21.4 ppt

72 HR	Treatment, ppm 10:1 No. 2 Fuel Oil / VirO$_2$Syl							
03/12/12	LPC	1.6 ppm	3.2 ppm	6.3 ppm	12.5 ppm	25.0 ppm	50.0 ppm	Meter ID#
DO F	7.0	7.2	6.9	6.9	6.8	6.8	6.8	57
Temp F	24.5	24.5	24.3	24.0	24.2	24.2	24.1	A46
Salinity F	20.6	20.7	19.8	20.2	20.2	20.2	20.5	A46
pH F	8.0	8.0	7.9	7.9	7.9	7.9	7.9	A93
Initials	Final: MEJG							
Times	Final Time: 1608							

96 HR	Treatment, ppm 10:1 No. 2 Fuel Oil / VirO$_2$Syl							
03/13/12	LPC	1.6 ppm	3.2 ppm	6.3 ppm	12.5 ppm	25.0 ppm	50.0 ppm	Meter ID#
DO F	6.7	6.6	6.8	6.8	6.7	6.7	6.7	57
Temp F	25.2	25.2	25.0	24.9	25.0	24.9	24.7	A46
Salinity F	20.7	20.5	19.7	20.2	20.2	20.5	20.5	A46
pH F	8.0	7.9	7.9	7.9	7.9	7.9	7.9	A93
Initials	Final: MJG SU							
Times	Final Time: 0918							

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

Comments:

Data Pages

- ☑ Calculations on mixing page are correct. (sign mixing page)
- ☑ Dates, dilutions, test method, # of replicates, replicate volume, product, acceptance limits, data analysis endpoint, and test organisms are correct throughout data pages.
- ☑ Format correct. (spaces for all entries, page numeration, no split pages, etc.)

JG Initials 03-09-12 Date

Chain-of-Custody

- ☑ Product on COC matches sample bottle/container.
- ☑ Product on COC matches test data pages.
- ☑ Lab # on COC matches sample bottle/container.
- ☑ Lab # on COC matches test data pages.
- ☑ Sample volume/amount is sufficient for test duration. (Sample volume in container(s) checked against sample volume on mixing page)

Sample volume/amount available: PRD 8000 Oil 40 (ml) or g

Sample volume/amount needed: 0.05 ml Product and 0.50 ml No.2 Fuel Oil (ml) or g

(Sample volume/amount insufficient if sample volume/amount available < sample volume/amount needed)

VL Initials 03/09/12 Date

Labels

- ☑ Lab # on labels matches test data pages.
- ☑ Test organism on data pages match labels (i.e. *Menidia beryllina* = MN, *Mysidopsis bahia* = MB)
- ☑ Test Concentrations on labels match Test Concentrations on test data pages.

VL Initials 03/09/12 Date

Raw Data QC/QA'd by: Veronica McMen 03/13/12

Acute Toxicity Test-96 Hr Survival

Start Date:	3/9/2012	Test ID:	mn00612RPO	Sample ID:	NCP-National Contingency Plan
End Date:	3/13/2012	Lab ID:	EE-Environmental Enterprise	Sample Type:	PIO-Product/No. 2 Fuel Oil Mixture
Sample Date:		Protocol:	EPAM 02-EPA Marine	Test Species:	MB-Menidia beryllina
Comments:					

Conc-ppm	1
PC-LP Control	1.0000
1.6	1.0000
3.2	1.0000
6.3	1.0000
12.5	1.0000
25	0.9000
50	0.7000

			Transform: Untransformed					Number	Total
Conc-ppm	Mean	N-Mean	Mean	Min	Max	CV%	N	Resp	Number
PC-LP Control	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
1.6	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
3.2	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
6.3	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
12.5	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
25	0.9000	0.9000	0.9000	0.9000	0.9000	0.000	1	1	10
50	0.7000	0.7000	0.7000	0.7000	0.7000	0.000	1	3	10

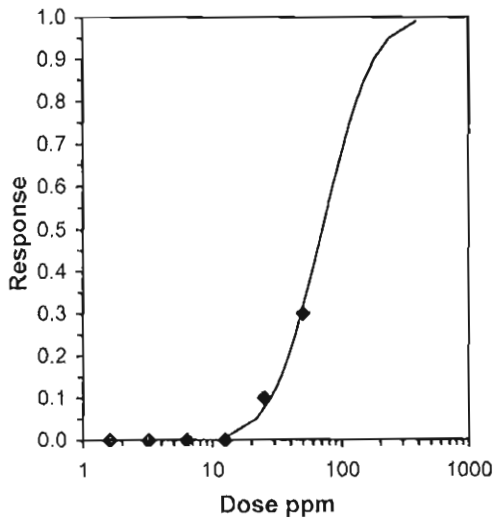
Auxiliary Tests	Statistic	Critical	Skew	Kurt
Normality of the data set cannot be confirmed				
Equality of variance cannot be confirmed				

Maximum Likelihood-Probit

Parameter	Value	SE	95% Fiducial Limits		Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	3.18169	1.6757	-0.1027	6.46607	0	0.18706	9.48773	0.99589	1.84969	0.3143	5
Intercept	-0.8852	2.63649	-6.0527	4.28237							

TSCR

Point	Probits	ppm	95% Fiducial Limits
EC01	2.674	13.138	
EC05	3.355	21.514	
EC10	3.718	27.9837	
EC15	3.964	33.4153	
EC20	4.158	38.4746	
EC25	4.326	43.4214	
EC40	4.747	58.8936	
EC50	**5.000**	**70.7448**	
EC60	5.253	84.9809	
EC75	5.674	115.262	
EC80	5.842	130.081	
EC85	6.036	149.777	
EC90	6.282	178.848	
EC95	6.645	232.632	
EC99	7.326	380.944	



Inland Silverside, *Menidia beryllina*
Acute Static 96–Hour Sodium Dodecyl Sulfate (SDS) Range-Finder Test (RFT)
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Test Concentrations, ppm SDS (Standard Reference Toxicant)

Menidia beryllina	Total Volume/ Concentration, ml	Color Code	ml SSOL	ml DH_2O
8.0 ppm	1000.00	Black	4.00	996.00
4.0 ppm	"	Brown	2.00	998.00
2.0 ppm	"	Yellow	1.00	999.00
1.0 ppm	"	Green	0.50	999.50
0.5 ppm	"	Blue	0.25	999.75
0 ppm LPC	"	White	0.0	1000.00
Total Volume (ml) of SSOL needed per day = 7.75				

500 ml Stock Solution (SSOL) @ 2,000 ppm = 1.00 g SDS + 497.3 ml DH_2O
Specific gravity of SDS = 0.37 g/ml; 1.00 g SDS = 2.7 ml

Weight of SDS = 1.0001 g Balance ID#: B2

Date & Time: 03/09/12 1649 Initials: VM

Data Pages & Calculations by: Veronica McNew QA/QC Check by: Jennifer Griffith

M. beryllina = 1 Rep x 1000 ml

DH_2O = Dilution Water = **20 ppt Synthetic Seawater**

	LPC	Meter ID#
Date	03/09	//////
DH_2O Lot #	25R-001-12	//////
Alkalinity	120	//////
Salinity	20.2	A416
pH	8.0	A193
Temp.	23.5	A416
	VM	//////

Prep Date	03/09
Shaker Table ID#	SH i-t A43
Initial	VM

LPC: Laboratory Performance Control, 20 ppt synthetic seawater
Alkalinity: mg/l as $CaCO_3$ Salinity: ppt pH: su Temp: °C

Comments: *Artemia* Lot # 030211-2; Feed *M. beryllina* once daily.

Inland Silverside, *Menidia beryllina*
Acute Static 96–Hour SDS Range-Finder Test

3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Survival Data

	Treatment, ppm SDS												
Time	REP	LPC 0 ppm White	REP	/////	REP	0.5 ppm Blue	REP	1.0 ppm Green	REP	2.0 ppm Yellow	REP	4.0 ppm Brown	REP	8.0 ppm Black	**Date & Initials**
0 HR 1800	1	10		/////	2	10	3	10	4	10	5	10	6	10	03/09/12 VL
24 HR 1233	1	10		/////	2	10	3	10	4	10	5	0	6	0	03/10/12 JG
48 HR 1429	1	10		/////	2	10	3	10	4	8	5	0	6	0	03/11/12 VL
72 HR 1615	1	10		/////	2	10	3	10	4	8	5	0	6	0	03/12/12 JG
96 HR 1752	1	10		/////	2	10	3	10	4	7	5	0	6	0	03/13/12 VL
% Survival		100				100		100		70		0		0	

Counted by: Jerry Bunch QC/QA by: Veronica McMinn

Loaded by: Veronica McMinn

Test Organisms Age: 7 days old Test Organisms Source: EE

Test Organisms Lot #: MN-0212-12

Data Entry by: Veronica McMinn

Double Data Entry by: Veronica McMinn and/or

QC/QA by: Jennifer Griffith

Comments:

Sodium Dodecyl Sulfate, RFT D-009-12 (For Product Test D-[illegible]-12)
LC50

M. beryllina Water Quality Data

LPC & All Treatments: Initial & Final Temp.: 23.5 to 26.4°C; Initial & Final DO: 4.0 to ≤ 7.4 mg/l
LPC: Initial Salinity: 18.5 to 21.4 ppt

0 to 24 HR		Treatment, ppm SDS							
03/09/12 – 03/10/12		LPC	/////	0.5 ppm	1.0 ppm	2.0 ppm	4.0 ppm	8.0 ppm	Meter ID#
DO	I	7.1	/////	7.2	7.2	7.2	7.2	7.2	S7
	F	7.1	/////	7.1	7.1	7.1	7.1	6.9	S7
Temp	I	23.5	/////	23.5	23.6	23.7	23.7	23.8	A46
	F	24.1	/////	23.8	23.6	23.8	23.6	23.6	A46
Salinity	I	20.2	/////	20.2	20.2	20.2	20.2	20.2	A46
	F	20.3	/////	20.3	20.3	20.3	20.3	20.3	A46
pH	F	7.9	/////	7.9	7.9	7.9	7.9	7.9	A93
Initials		Initial: VM SD				Final: VM SD			
Times		Initial Time: 1841				Final Time: 0759			

48 HR		Treatment, ppm SDS							
03/11/12		LPC	/////	0.5 ppm	1.0 ppm	2.0 ppm	4.0 ppm	8.0 ppm	Meter ID#
DO	F	6.8	/////	6.9	6.9	6.7			S7
Temp	F	25.0	/////	24.6	24.5	24.6			A46
Salinity	F	20.4	/////	20.5	20.4	20.4			A46
pH	F	8.0	/////	8.0	8.0	7.9			A93
Initials		Final: VM SD							
Times		Final Time: 0752							

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

Comments: _____

M. beryllina Water Quality Data

LPC & All Treatments: Initial & Final Temp.: 23.5 to 26.4°C; Initial & Final DO: 4.0 to ≤ 7.4 mg/l
LPC: Initial Salinity: 18.5 to 21.4 ppt

72 HR	Treatment, ppm SDS							
03/12/12	LPC	//////	0.5 ppm	1.0 ppm	2.0 ppm	4.0 ppm	8.0 ppm	Meter ID#
DO F	7.0	//////	6.9	6.9	6.8			S7
Temp F	24.5	//////	24.3	24.3	24.4			A46
Salinity F	20.6	//////	20.7	20.5	20.6			A46
pH F	8.0	//////	7.9	7.9	7.9			A93
Initials	Final: ME JG							
Times	Final Time: 1603							

96 HR	Treatment, ppm SDS							
03/13/12	LPC	//////	0.5 ppm	1.0 ppm	2.0 ppm	4.0 ppm	8.0 ppm	Meter ID#
DO F	6.7	//////	6.8	6.7	6.8			S7
Temp F	25.3	//////	25.0	24.9	24.9			A46
Salinity F	20.7	//////	20.7	20.6	20.9			A46
pH F	8.0	//////	7.9	7.9	7.9			A93
Initials	Final: MR JD							
Times	Final Time: 0913							

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

Comments: _____

Data Pages

- ☑ Calculations on mixing page are correct. (sign mixing page)
- ☑ Dates, dilutions, test method, # of replicates, replicate volume, product, acceptance limits, data analysis endpoint, and test organisms are correct throughout data pages.
- ☑ Format correct. (spaces for all entries, page numeration, no split pages, etc.)

JG Initials 03-09-12 Date

Chain-of-Custody

- ☑ Product on COC matches sample bottle/container.
- ☑ Product on COC matches test data pages.
- ☑ Lab # on COC matches sample bottle/container.
- ☑ Lab # on COC matches test data pages.
- ☑ Sample volume/amount is sufficient for test duration. (Sample volume in container(s) checked against sample volume on mixing page)

Sample volume/amount available: 95 ml or (g)

Sample volume/amount needed: 1.00 g SDS ml or (g)

(Sample volume/amount insufficient if sample volume/amount available < sample volume/amount needed)

VL Initials 03/09/12 Date

Labels

- ☑ Lab # on labels matches test data pages.
- ☑ Test organism on data pages match labels (i.e. *Menidia beryllina* = MN, *Mysidopsis bahia* = MB)
- ☑ Test Concentrations on labels match Test Concentrations on test data pages.

VL Initials 03/09/12 Date

Raw Data QC/QA'd by: Veronica M. Mena 03/13/12

Acute Toxicity Test-96 Hr Survival

Start Date:	3/9/2012	Test ID:	mn00912RS	Sample ID:	REF-Ref Toxicant
End Date:	3/13/2012	Lab ID:	EE-Environmental Enterprise	Sample Type:	SDS-Sodium dodecyl sulfate
Sample Date:		Protocol:	EPAM 02-EPA Marine	Test Species:	MB-Menidia beryllina
Comments:					

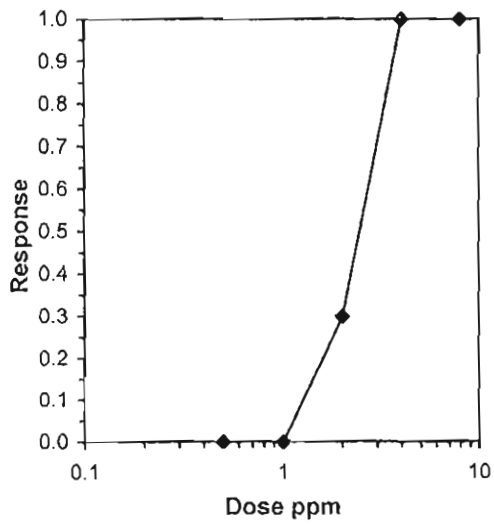
Conc-ppm	1
PC-LP Control	1.0000
0.5	1.0000
1	1.0000
2	0.7000
4	0.0000
8	0.0000

			Transform: Untransformed					Number	Total
Conc-ppm	Mean	N-Mean	Mean	Min	Max	CV%	N	Resp	Number
PC-LP Control	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
0.5	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
1	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
2	0.7000	0.7000	0.7000	0.7000	0.7000	0.000	1	3	10
4	0.0000	0.0000	0.0000	0.0000	0.0000	0.000	1	10	10
8	0.0000	0.0000	0.0000	0.0000	0.0000	0.000	1	10	10

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Normality of the data set cannot be confirmed				
Equality of variance cannot be confirmed				

Trimmed Spearman-Karber

Trim Level	EC50	95% CL	
0.0%	2.2974	1.8793	2.8086
5.0%	2.3288	1.8582	2.9186
10.0%	2.3589	1.8143	3.0670
20.0%	2.4114	1.6236	3.5814
Auto-0.0%	**2.2974**	**1.8793**	**2.8086**



Environmental Enterprises USA, Inc.

APPENDIX B

3/14/2012 6:00 PM
Environmental Enterprises USA, Inc.

Inland Silverside, *Menidia beryllina*
Acute Static 96–Hour Product Definitive Test (DFT)
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Natural Solutions Group Corporation – $VirO_2Syl$
Contact: Marcos Gonzalez

Test Concentrations, ppm $VirO_2Syl$

Menidia beryllina	Total Volume / Concentration, ml	Color Code	ml SSOL	ml DH_2O
100.0 ppm	3000.00	Black	300.00	2700.00
50.0 ppm	"	Brown	150.00	2850.00
25.0 ppm	"	Yellow	75.00	2925.00
12.5 ppm	"	Green	37.50	2962.50
6.3 ppm	"	Blue	18.90	2981.10
0.0 ppm LPC	"	White	0.00	3000.00
Total Volume (ml) of SSOL needed per day = 581.4				

1100 ml Stock Solution (SSOL) @ 1000 ppm: 1.10 ml $VirO_2Syl$ + 1098.9 ml DH_2O

Data Pages & Calculations by: Veronica McKeen QA/QC Check by: Jennifer Griffith

M. beryllina, 3000 ml total volume / treatment = 3 Rep x 1000 ml / replicate

DH_2O = Dilution Water = **20 ppt Synthetic Seawater**

	LPC	Meter ID#
Date	03/14	//////
DH_2O Lot #	25R-673-12	//////
Alkalinity	108	//////
Salinity	20.3	A46
pH	8.0	A93
Temp.	23.7	A46
	VR	//////

Prep Date	03/14
Shaker Table ID#	SH 1+ A43
Initial	VR

LPC: Laboratory Performance Control, 20 ppt synthetic seawater
Alkalinity: mg/l as $CaCO_3$ Salinity: ppt pH: su Temp: °C

Comments: *Artemia* Lot # 030211-2; Feed *M. beryllina* once daily.

Inland Silverside, *Menidia beryllina*
Acute Static 96–Hour Product Definitive Test
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Survival Data

Time	Treatment, ppm $VirO_2Syl$																		Date & Initials
	REP	LPC 0 ppm White	REP	6.3 ppm Blue	REP	12.5 ppm Green	REP	25.0 ppm Yellow	REP	50.0 ppm Brown	REP	100.0 ppm Black	REP						
0 HR	1	10	4	10	7	10	10	10	13	10	16	10	19						03/14/12
1850	2	10	5	10	8	10	11	10	14	10	17	10	20						TB
	3	10	6	10	9	10	12	10	15	10	18	10	21						
24 HR	1	10	4	10	7	10	10	10	13	10	16	10	19						03/15/12
1555	2	10	5	10	8	10	11	10	14	10	17	10	20						VL
	3	10	6	10	9	10	12	10	15	10	18	10	21						
48 HR	1	10	4	10	7	10	10	10	13	10	16	8	19						03/16/12
1536	2	10	5	10	8	10	11	10	14	10	17	8	20						VL
	3	10	6	10	9	10	12	10	15	10	18	7	21						
72 HR	1	10	4	10	7	9	10	10	13	10	16	8	19						03/17/12
1200	2	10	5	10	8	10	11	10	14	10	17	8	20						TK
	3	10	6	10	9	10	12	10	15	10	18	6	21						
96 HR	1	10	4	10	7	9	10	10	13	7	16	6	19						03/18/12
1816	2	10	5	9	8	10	11	10	14	10	17	4	20						JG
	3	10	6	10	9	10	12	10	15	10	18	4	21						
% Survival	100		96.7		96.7		100		90		46.7								

Product, DFT

D-006-12
LC50

Counted by: Terry Bunch QC/QA by: Veronica McNew

Loaded by: Terry Bunch

Test Organisms Age: 7 days old Test Organisms Source: EE

Test Organism Lot #: Mn-067-12

Data Entry by: Veronica McNew

Double Data Entry by: Veronica McNew and/or

QC/QA by: Jennifer Griffith

***M. beryllina* Water Quality Data**

LPC & All Treatments: Initial & Final Temp. – 23.5 – 26.4°C; Initial & Final DO – 4.0 < 7.4 mg/l
LPC: Initial Salinity 18.5 – 21.4 ppt

0 to 24 HR		Treatment, ppm $VirO_2Syl$							
03/14/12 – 03/15/12		LPC	6.3 ppm	12.5 ppm	25.0 ppm	50.0 ppm	100.0 ppm		Meter ID#
DO	I	7.3	7.3	7.3	7.4	7.6	7.8		S7
	F	7.0	7.2	7.2	7.2	7.5	7.6		S7
Temp	I	23.7	23.5	23.2	23.3	23.5	23.6		A46
	F	25.1	25.2	25.3	25.4	25.5	25.5		A46
Salinity	I	20.3	20.3	20.3	20.3	20.3	20.3		A46
	F	20.3	20.4	20.4	20.4	20.3	20.3		A46
pH	F	7.9	8.0	8.0	8.0	8.0	8.0		A93
Initials		Initial: TB VM				Final: SD MR			
Times		Initial Time: 1940				Final Time: 0922			

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

Comments:

M. beryllina Water Quality Data

LPC & All Treatments: Initial & Final Temp. – 23.5 – 26.4°C; Initial & Final DO – 4.0 < 7.4 mg/l
LPC: Initial Salinity 18.5 – 21.4 ppt

48 HR	Treatment, ppm $VirO_2Syl$							
03/16/12	LPC	6.3 ppm	12.5 ppm	25.0 ppm	50.0 ppm	100.0 ppm		Meter ID#
DO F	6.7	6.8	6.9	6.9	7.2	10.4		S7
Temp F	25.7	25.8	25.9	25.9	25.9	26.0		A46
Salinity F	20.5	20.6	20.5	20.5	20.5	20.5		A46
pH F	8.0	8.0	8.0	8.0	8.0	8.0		A93
Initials	Final: METK							
Times	Final Time: 0917							

72 HR	Treatment, ppm $VirO_2Syl$							
03/17/12	LPC	6.3 ppm	12.5 ppm	25.0 ppm	50.0 ppm	100.0 ppm		Meter ID#
DO F	6.4	6.6	6.8	6.6	6.7	6.7		S7
Temp F	25.7	25.8	25.7	25.7	25.8	25.6		A46
Salinity F	20.6	20.7	20.7	20.7	20.6	20.7		A46
pH F	8.0	8.0	8.0	8.0	8.0	8.0		A93
Initials	Final: MRTK							
Times	Final Time: 0810							

96 HR	Treatment, ppm $VirO_2Syl$							
03/18/12	LPC	6.3 ppm	12.5 ppm	25.0 ppm	50.0 ppm	100.0 ppm		Meter ID#
DO F	6.6	6.8	5.8	6.5	6.7	6.6		S7
Temp F	25.7	25.9	26.9	26.0	26.0	26.1		A46
Salinity F	20.7	20.9	20.9	20.8	20.7	20.8		A46
pH F	8.0	8.0	8.0	8.0	8.0	8.0		A93
Initials	Final: MEMR							
Times	Final Time: 0803							

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

Data Pages

- ☑ Calculations on mixing page are correct. (sign mixing page)
- ☑ Dates, dilutions, test method, # of replicates, replicate volume, product, acceptance limits, data analysis endpoint, and test organisms are correct throughout data pages.
- ☑ Format correct. (spaces for all entries, page numeration, no split pages, etc.)

JG Initials 03-14-12 Date

Chain-of-Custody

- ☑ Product on COC matches sample bottle/container.
- ☑ Product on COC matches test data pages.
- ☑ Lab # on COC matches sample bottle/container.
- ☑ Lab # on COC matches test data pages.
- ☑ Sample volume/amount is sufficient for test duration. (Sample volume in container(s) checked against sample volume on mixing page)

Sample volume/amount available: 8000 (ml) or g

Sample volume/amount needed: 1.1 ml or g

(Sample volume/amount insufficient if sample volume/amount available < sample volume/amount needed)

VM Initials 03/14/12 Date

Labels

- ☑ Lab # on labels matches test data pages.
- ☑ Test organism on data pages match labels (i.e. *Menidia beryllina* = MN, *Mysidopsis bahia* = MB)
- ☑ Test Concentrations on labels match Test Concentrations on test data pages.

VM Initials 03/14/12 Date

Raw Data QC/QA'd by: Veronica McMan 03/19/12

Acute Toxicity Test-96 Hr Survival

Start Date:	3/14/2012	Test ID:	mn00612DP	Sample ID:	NCP-National Contingency Plan
End Date:	3/18/2012	Lab ID:	EE-Environmental Enterprise	Sample Type:	PRD-Product
Sample Date:		Protocol:	EPAM 02-EPA Marine	Test Species:	MB-Menidia beryllina
Comments:					

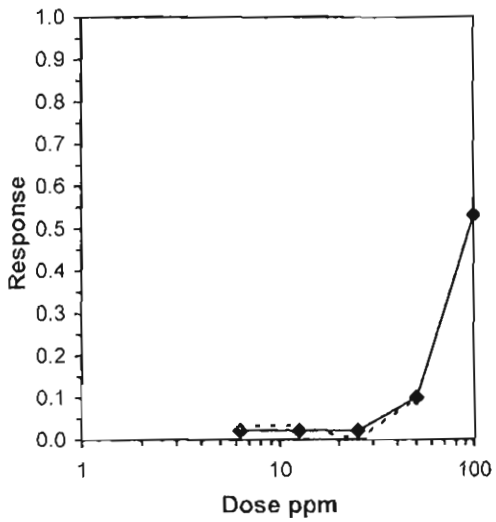
Conc-ppm	1	2	3
PC-LP Control	1.0000	1.0000	1.0000
6.3	1.0000	0.9000	1.0000
12.5	0.9000	1.0000	1.0000
25	1.0000	1.0000	1.0000
50	0.7000	1.0000	1.0000
100	0.6000	0.4000	0.4000

			Transform: Untransformed					Number	Total
Conc-ppm	Mean	N-Mean	Mean	Min	Max	CV%	N	Resp	Number
PC-LP Control	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	3	0	30
6.3	0.9667	0.9667	0.9667	0.9000	1.0000	5.973	3	1	30
12.5	0.9667	0.9667	0.9667	0.9000	1.0000	5.973	3	1	30
25	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	3	0	30
50	0.9000	0.9000	0.9000	0.7000	1.0000	19.245	3	3	30
100	0.4667	0.4667	0.4667	0.4000	0.6000	24.744	3	16	30

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.05)	0.91539	0.897	-0.6845	1.717
Equality of variance cannot be confirmed				

Trimmed Spearman-Karber

Trim Level	EC50	95% CL	
0.0%			
5.0%			
10.0%			
20.0%			
Auto-46.7%	94.808	72.424	124.110



Environmental Enterprises USA, Inc.

APPENDIX C

Inland Silverside, *Menidia beryllina*
Acute Static 96–Hour No. 2 Fuel Oil Definitive Test (DFT)
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Test Concentrations, ppm No. 2 Fuel Oil

Menidia beryllina	Total Volume / Concentration, ml	Color Code	ml SSOL	ml DH_2O
60.0 ppm	3000.00	Black	180.00	2820.00
30.0 ppm	"	Brown	90.00	2910.00
15.0 ppm	"	Yellow	45.00	2955.00
7.5 ppm	"	Green	22.50	2977.50
3.8 ppm	"	Blue	11.40	2988.60
0 ppm LPC	"	White	0.00	3000.00
Total Volume (ml) of SSOL needed per day = 348.90				

550 ml Stock Solution (SSOL) @ 1000 ppm: 0.55 ml No. 2 Fuel Oil + 549.45 ml DH_2O

Data Pages & Calculations by: Veronica Villa QA/QC Check by: Jennifer Griffith

M. beryllina, 3000 ml total volume / treatment = 3 Rep x 1000 ml / replicate

DH_2O = Dilution Water = **20 ppt Synthetic Seawater**

	LPC	Meter ID#
Date	03/14	//////
DH_2O Lot #	25R-573-12	//////
Alkalinity	108	//////
Salinity	20.3	A46
pH	8.0	A93
Temp.	(A) 20.2 3.7	A41
	VL	//////

Prep Date	03/14
Shaker Table ID#	5'11+ A43
Initial	VL

LPC: Laboratory Performance Control, 20 ppt synthetic seawater
Alkalinity: mg/l as $CaCO_3$ Salinity: ppt pH: su Temp: °C

Comments: *Artemia* Lot # 030211-2; Feed *M. beryllina* once daily.
(A) aurora data 03/14/12 VL

Inland Silverside, *Menidia beryllina*
Acute Static 96–Hour No. 2 Fuel Oil Definitive Test
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Survival Data

Time	REP	LPC 0 ppm White	REP	3.8 ppm Blue	REP	7.5 ppm Green	REP	15.0 ppm Yellow	REP	30.0 ppm Brown	REP	60.0 ppm Black	REP		Date & Initials
	Treatment, ppm No. 2 Fuel Oil														
0 HR	1	10	4	10	7	10	10	10	13	10	16	10	19		03/14/12
1934	2	10	5	10	8	10	11	10	14	10	17	10	20		TS
	3	10	6	10	9	10	12	10	15	10	18	10	21		
24 HR	1	10	4	10	7	10	10	10	13	10	16	0	19		03/15/12
1603	2	10	5	10	8	10	11	10	14	10	17	0	20		JG
	3	10	6	10	9	10	12	10	15	10	18	0	21		
48 HR	1	10	4	10	7	10	10	10	13	10	16	0	19		03/16/12
1544	2	10	5	10	8	10	11	10	14	10	17	0	20		VL
	3	10	6	10	9	10	12	10	15	10	18	0	21		
72 HR	1	10	4	10	7	10	10	10	13	10	16	0	19		03/17/12
1206	2	10	5	10	8	10	11	10	14	10	17	0	20		TK
	3	10	6	10	9	10	12	10	15	10	18	0	21		
96 HR	1	10	4	10	7	10	10	10	13	9	16	0	19		03/18/12
1821	2	10	5	10	8	10	11	10	14	9	17	0	20		JG
	3	10	6	10	9	10	12	10	15	10	18	0	21		
% Survival	100		100		100		100		93.3		0				

No. 2 Fuel Oil, DFT

D-008-12 (For Product Test D-006-12)
LC50

7/16/2010 7:52 PM
Environmental Enterprises USA, Inc.

Counted by: Jerry Bunch QC/QA by: Veronica McNew

Loaded by: Jerry Bunch

Test Organisms Age: 7 days old Test Organisms Source: EE

Test Organism Lot #: Mn-067-12

Data Entry by: Veronica McNew

Double Data Entry by: Veronica McNew and/or

QC/QA by: Jennifer Griffith

***M. beryllina* Water Quality Data**

LPC & All Treatments: Initial & Final Temp.: 23.5 to 26.4°C; Initial & Final DO: 4.0 to ≤ 7.4 mg/l
LPC: Initial Salinity: 18.5 to 21.4 ppt

0 to 24 HR	Treatment, ppm No. 2 Fuel Oil							
03/14/12-03/15/12	LPC	3.8 ppm	7.5 ppm	15.0 ppm	30.0 ppm	60.0 ppm		Meter ID#
DO I	7.3	7.3	7.3	7.4	7.4	7.3		57
F	7.0	7.2	7.1	7.0	7.1	7.2		57
Temp I	23.7	23.6	23.8	23.5	23.5	23.5		A42
F	25.1	24.4	24.4	24.5	24.5	24.6		A46
Salinity I	20.3	20.3	20.3	20.3	20.3	20.2		A46
F	20.3	20.3	20.3	20.3	20.4	20.3		A46
pH F	7.9	8.0	8.0	8.0	8.0	8.0		A93
Initials	Initial: TB VM				Final: 0925 DMR			
Times	Initial Time: 1946				Final Time: 0925			

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

Comments: A wrong Date SD 3-15-12

***M. beryllina* Water Quality Data**

LPC & All Treatments: Initial & Final Temp.: 23.5 to 26.4°C; Initial & Final DO: 4.0 to ≤ 7.4 mg/l

LPC: Initial Salinity: 18.5 to 21.4 ppt

48 HR	Treatment, ppm No. 2 Fuel Oil							
03/16/12	LPC	3.8 ppm	7.5 ppm	15.0 ppm	30.0 ppm	60.0 ppm		Meter ID#
DO F	6.7	6.9	6.8	6.7	6.7			S7
Temp F	25.7	24.9	24.9	25.0	25.2			A46
Salinity F	20.5	20.4	20.5	20.5	20.5			A46
pH F	8.0	8.0	8.0	7.9	7.9			A93
Initials	Final: MRTK							
Times	Final Time: 0919							

72 HR	Treatment, ppm No. 2 Fuel Oil							
03/17/12	LPC	3.8 ppm	7.5 ppm	15.0 ppm	30.0 ppm	60.0 ppm		Meter ID#
DO F	6.4	6.6	6.5	6.3	6.6			S7
Temp F	25.7	24.7	24.5	24.7	24.7			A46
Salinity F	20.6	20.6	20.7	20.6	20.6			A46
pH F	8.0	7.9	7.9	7.9	7.9			A93
Initials	Final: MRTK							
Times	Final Time: 0814							

96 HR	Treatment, ppm No. 2 Fuel Oil							
03/18/12	LPC	3.8 ppm	7.5 ppm	15.0 ppm	30.0 ppm	60.0 ppm		Meter ID#
DO F	6.6	6.9	6.9	6.5	6.8			S7
Temp F	25.7	25.0	24.9	25.0	25.2			A46
Salinity F	20.7	20.7	20.8	20.8	20.7			A46
pH F	8.0	8.0	8.0	8.0	8.0			A93
Initials	Final: MRMR							
Times	Final Time: 0815							

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

Data Pages

- ☑ Calculations on mixing page are correct. (sign mixing page)
- ☑ Dates, dilutions, test method, # of replicates, replicate volume, product, acceptance limits, data analysis endpoint, and test organisms are correct throughout data pages.
- ☑ Format correct. (spaces for all entries, page numeration, no split pages, etc.)

JC Initials 03-14-12 Date

Chain-of-Custody

- ☑ Product on COC matches sample bottle/container.
- ☑ Product on COC matches test data pages.
- ☑ Lab # on COC matches sample bottle/container.
- ☑ Lab # on COC matches test data pages.
- ☑ Sample volume/amount is sufficient for test duration. (Sample volume in container(s) checked against sample volume on mixing page)

Sample volume/amount available: 40 (ml) or g

Sample volume/amount needed: 0.55 (ml) or g

(Sample volume/amount insufficient if sample volume/amount available < sample volume/amount needed)

VL Initials 03/14/12 Date

Labels

- ☑ Lab # on labels matches test data pages.
- ☑ Test organism on data pages match labels (i.e. *Menidia beryllina* = MN, *Mysidopsis bahia* = MB)
- ☑ Test Concentrations on labels match Test Concentrations on test data pages.

VL Initials 03/14/12 Date

Raw Data QC/QA'd by: Veronica McNew 03/19/12

Acute Toxicity Test-96 Hr Survival

Start Date:	3/14/2012	Test ID:	mn00812DO	Sample ID:	NCP-National Contingency Plan
End Date:	3/18/2012	Lab ID:	EE-Environmental Enterprise	Sample Type:	2FO-No. 2 Fuel Oil
Sample Date:		Protocol:	EPAM 02-EPA Marine	Test Species:	MB-Menidia beryllina
Comments:					

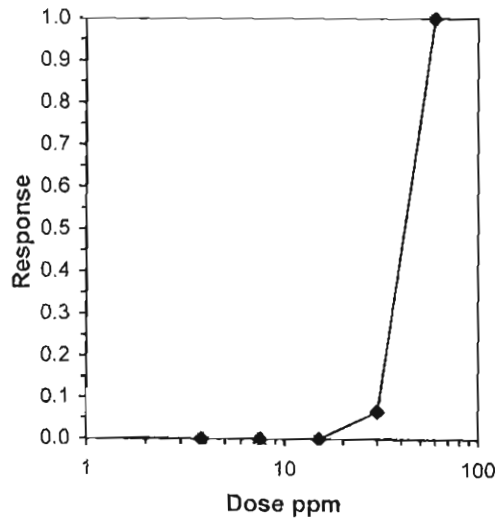
Conc-ppm	1	2	3
PC-LP Control	1.0000	1.0000	1.0000
3.8	1.0000	1.0000	1.0000
7.5	1.0000	1.0000	1.0000
15	1.0000	1.0000	1.0000
30	0.9000	0.9000	1.0000
60	0.0000	0.0000	0.0000

			Transform: Untransformed					Number	Total
Conc-ppm	Mean	N-Mean	Mean	Min	Max	CV%	N	Resp	Number
PC-LP Control	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	3	0	30
3.8	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	3	0	30
7.5	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	3	0	30
15	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	3	0	30
30	0.9333	0.9333	0.9333	0.9000	1.0000	6.186	3	2	30
60	0.0000	0.0000	0.0000	0.0000	0.0000	0.000	3	30	30

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates non-normal distribution (p <= 0.05)	0.58631	0.881	1.76253	7
Equality of variance cannot be confirmed				

Trimmed Spearman-Karber

Trim Level	EC50	95% CL	
0.0%	40.510	38.032	43.151
5.0%	41.327	37.779	45.209
10.0%	41.389	39.916	42.916
20.0%	41.389	39.916	42.916
Auto-0.0%	**40.510**	**38.032**	**43.151**



Environmental Enterprises USA, Inc.

APPENDIX D

Inland Silverside, *Menidia beryllina*

Acute Static 96–Hour 10:1 No. 2 Fuel Oil / Product Definitive Test (DFT)

3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461.- 47464

Natural Solutions Group Corporation – VirO$_2$Syl

Contact: Marcos Gonzalez

Test Concentrations, ppm 10:1 No. 2 Fuel Oil / VirO$_2$Syl

Menidia beryllina	Total Volume / Concentration, ml	Color Code	ml SSOL	ml DH_2O
125.0 ppm	3000.00	Black	375.00	2625.00
62.5 ppm	"	Brown	187.50	2812.50
31.3 ppm	"	Yellow	93.90	2906.10
15.6 ppm	"	Green	46.80	2953.20
7.8 ppm	"	Blue	23.40	2976.60
3.9 ppm	"	White/Blue	11.70	2988.30
0 ppm LPC	"	White	0.00	3000.00
Total Volume (ml) of SSOL needed per day = 738.30				

1100 ml Stock Solution (SSOL) @ 1000 ppm:
1.0 ml No.2 Fuel Oil + 0.10 ml VirO$_2$Syl + 1098.90 ml DH_2O

Data Pages & Calculations by: Veronica Ivette Nieves QA/QC Check by: Jennifer Griffith

M. beryllina, 3000 ml total volume / treatment = 3 Rep x 1000 ml / replicate

DH_2O = Dilution Water = **20 ppt Synthetic Seawater**

	LPC	Meter ID#
Date	03/14	//////
DH_2O Lot #	25R-073-12	//////
Alkalinity	108	//////
Salinity	20.3	A46
pH	8.0	A93
Temp.	23.7	A46
	VZ	//////

Prep Date	03/14
Shaker Table ID#	SH14 A43
Initial	VZ

LPC: Laboratory Performance Control, 20 ppt synthetic seawater
Alkalinity: mg/l as $CaCO_3$ Salinity: ppt pH: su Temp: °C

Comments: *Artemia* Lot # 030211-2; Feed *M. beryllina* once daily.

Inland Silverside, *Menidia beryllina*
Acute Static 96–Hour 10:1 No. 2 Fuel Oil / Product Definitive Test
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Survival Data

Time	Treatment, ppm 10:1 No. 2 Fuel Oil / VirO$_2$Syl																								Date & Initials
	REP	LPC 0 ppm White	REP	3.9 ppm Blue	REP	7.8 ppm Blue	REP	15.6 ppm Green	REP	31.3 ppm Yellow	REP	62.5 ppm Brown	REP	125.0 ppm Black	
0 HR	1	10	4	10	7	10	10	10	13	10	16	10	19	10	03/14/12
1913	2	10	5	10	8	10	11	10	14	10	17	10	20	10	TK
	3	10	6	10	9	10	12	10	15	10	18	10	21	10	
24 HR	1	10	4	10	7	10	10	2	13	2	16	0	19	0	03/15/12
1603	2	10	5	10	8	10	11	3	14	2	17	0	20	0	VL
	3	10	6	10	9	10	12	4	15	1	18	0	21	0	
48 HR	1	10	4	10	7	10	10	0	13	0	16	0	19	0	03/16/12
1541	2	10	5	10	8	9	11	0	14	0	17	0	20	0	VL
	3	10	6	10	9	10	12	0	15	0	18	0	21	0	
72 HR	1	10	4	10	7	9	10	C	13	C	16	C	19	0	03/17/12
	2	10	5	10	8	9	11	C	14	C	17	C	20	0	
1203	3	10	6	10	9	10	12	C	15	0	18	C	21	0	TK
96 HR	1	10	4	10	7	9	10	0	13	0	16	0	19	0	03/18/12
1818	2	10	5	10	8	8	11	0	14	0	17	0	20	0	JG
	3	10	6	10	9	9	12	0	15	0	18	0	21	0	
% Survival		100		100		84.7		0		0		0		0	

3/14/2012 7:44 PM
Environmental Enterprises USA, Inc.

Counted by: Terry Bunch QC/QA by: Veronica McMinn

Loaded by: Terry Bunch

Test Organisms Age: 7 days old Test Organisms Source: EE

Test Organism Lot #: MN-007-12

Data Entry by: Veronica McMinn

Double Data Entry by: Veronica McMinn and/or

QC/QA by: Jennifer Griffith

***M. beryllina* Water Quality Data**

LPC & All Treatments: Initial & Final Temp.: 23.5 to 26.4°C; Initial & Final DO: 4.0 to ≤ 7.4 mg/l
LPC: Initial Salinity: 18.5 to 21.4 ppt

0 to 24 HR		Treatment, ppm 10:1 No. 2 Fuel Oil / VirO$_2$Syl							
03/14/12 – 03/15/12		LPC	3.9 ppm	7.8 ppm	15.6 ppm	31.3 ppm	62.5 ppm	125 ppm	Meter ID#
DO	I	7.3	7.3	7.3	7.3	7.3	7.3	7.3	S7
	F	7.0	7.1	7.1	7.0	7.0	7.1	7.3	S7
Temp	I	23.7	23.6	23.7	23.6	23.5	23.5	23.5	A46
	F	25.1	24.7	24.7	24.7	24.7	24.6	24.5	A46
Salinity	I	20.3	20.3	20.3	20.3	20.3	20.3	20.3	A46
	F	20.3	20.3	20.2	20.3	20.3	20.3	20.3	A46
pH	F	7.9	8.0	8.0	8.0	8.0	8.0	8.0	A93
Initials		Initial: MR TB				Final: SD MR			
Times		Initial Time: 1949				Final Time: 0923			

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

Comments:

M. beryllina Water Quality Data

LPC & All Treatments: Initial & Final Temp.: 23.5 to 26.4°C; Initial & Final DO: 4.0 to ≤ 7.4 mg/l
LPC: Initial Salinity: 18.5 to 21.4 ppt

48 HR	Treatment, ppm 10:1 No. 2 Fuel Oil / $VirO_2Syl$							
03/16/12	LPC	3.9 ppm	7.8 ppm	15.6 ppm	31.3 ppm	62.5 ppm	125 ppm	Meter ID#
DO F	6.7	7.0	6.9	6.6	6.6			S7
Temp F	25.7	24.9	24.9	25.0	24.9			A46
Salinity F	20.5	20.4	20.3	20.4	20.4			A46
pH F	8.0	8.0	8.0	7.9	7.9			A93
Initials	Final: METK							
Times	Final Time: 0918							

72 HR	Treatment, ppm 10:1 No. 2 Fuel Oil / $VirO_2Syl$							
03/17/12	LPC	3.9 ppm	7.8 ppm	15.6 ppm	31.3 ppm	62.5 ppm	125 ppm	Meter ID#
DO F	6.4	6.4	6.6					S7
Temp F	25.7	24.7	24.7					A46
Salinity F	20.6	20.7	20.5					A46
pH F	8.0	7.9	7.9					A93
Initials	Final: MRTK							
Times	Final Time: 0812							

96 HR	Treatment, ppm 10:1 No. 2 Fuel Oil / $VirO_2Syl$							
03/18/12	LPC	3.9 ppm	7.8 ppm	15.6 ppm	31.3 ppm	62.5 ppm	125 ppm	Meter ID#
DO F	6.4	6.7	6.7					S7
Temp F	25.7	25.1	25.1					A46
Salinity F	20.7	20.7	20.6					A46
pH F	8.0	8.0	8.0					A93
Initials	Final: MBMR							
Times	Final Time: 0804							

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

10:1 No. 2 Fuel Oil / Product, DFT

D-006-12
LC50

Data Pages

- ☑ Calculations on mixing page are correct. (sign mixing page)
- ☑ Dates, dilutions, test method, # of replicates, replicate volume, product, acceptance limits, data analysis endpoint, and test organisms are correct throughout data pages.
- ☑ Format correct. (spaces for all entries, page numeration, no split pages, etc.)

JG Initials 03-14-12 Date

Chain-of-Custody

- ☑ Product on COC matches sample bottle/container.
- ☑ Product on COC matches test data pages.
- ☑ Lab # on COC matches sample bottle/container.
- ☑ Lab # on COC matches test data pages.
- ☑ Sample volume/amount is sufficient for test duration. (Sample volume in container(s) checked against sample volume on mixing page)

PRD 0.1
Sample volume/amount available: ~~1000~~ 40 (ml) or g

Sample volume/amount needed: 1.0 0.1 (ml) or g

(Sample volume/amount insufficient if sample volume/amount available < sample volume/amount needed)

VR Initials 03/14/12 Date

Labels

- ☑ Lab # on labels matches test data pages.
- ☑ Test organism on data pages match labels (i.e. *Menidia beryllina* = MN, *Mysidopsis bahia* = MB)
- ☑ Test Concentrations on labels match Test Concentrations on test data pages.

VR Initials 03/14/12 Date

Raw Data QC/QA'd by: Veronica McNew 03/19/12

Acute Toxicity Test-96 Hr Survival

Start Date:	3/14/2012	Test ID:	mn006DPO	Sample ID:	NCP-National Contingency Plan
End Date:	3/18/2012	Lab ID:	EE-Environmental Enterprise	Sample Type:	PIO-Product/No. 2 Fuel Oil Mixture
Sample Date:		Protocol:	EPAM 02-EPA Marine	Test Species:	MB-Menidia beryllina
Comments:					

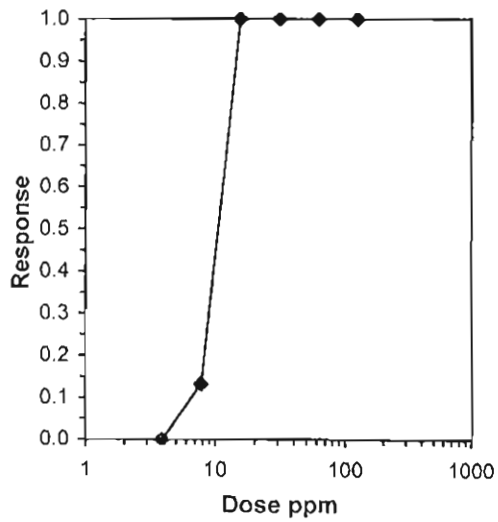
Conc-ppm	1	2	3
PC-LP Control	1.0000	1.0000	1.0000
3.9	1.0000	1.0000	1.0000
7.8	0.9000	0.8000	0.9000
15.6	0.0000	0.0000	0.0000
31.3	0.0000	0.0000	0.0000
62.5	0.0000	0.0000	0.0000
125	0.0000	0.0000	0.0000

			Transform: Untransformed					Number	Total
Conc-ppm	Mean	N-Mean	Mean	Min	Max	CV%	N	Resp	Number
PC-LP Control	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	3	0	30
3.9	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	3	0	30
7.8	0.8667	0.8667	0.8667	0.8000	0.9000	6.662	3	4	30
15.6	0.0000	0.0000	0.0000	0.0000	0.0000	0.000	3	30	30
31.3	0.0000	0.0000	0.0000	0.0000	0.0000	0.000	3	30	30
62.5	0.0000	0.0000	0.0000	0.0000	0.0000	0.000	3	30	30
125	0.0000	0.0000	0.0000	0.0000	0.0000	0.000	3	30	30

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates non-normal distribution (p <= 0.05)	0.72857	0.829	-1.4846	4
Equality of variance cannot be confirmed				

Trimmed Spearman-Karber

Trim Level	EC50	95% CL	
0.0%	10.057	9.228	10.961
5.0%	10.282	9.284	11.388
10.0%	10.426	9.091	11.958
20.0%	10.458	9.876	11.075
Auto-0.0%	**10.057**	**9.228**	**10.961**



APPENDIX E

3/14/2012 4:13 PM
Environmental Enterprises USA, Inc.

Inland Silverside, *Menidia beryllina*
Acute Static 96–Hour Sodium Dodecyl Sulfate Definitive Test (DFT)
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Test Concentrations, ppm Sodium Dodecyl Sulfate (SDS)
Standard Reference Toxicant

Menidia beryllina	Total Volume / Concentration, ml	Color Code	ml SSOL	ml DH_2O
8.0 ppm	3000.00	Black	12.00	2988.00
4.8 ppm	"	Brown	7.20	2992.80
2.9 ppm	"	Yellow	4.35	2995.65
1.7 ppm	"	Green	2.55	2997.45
1.0 ppm	"	Blue	1.50	2998.50
0 ppm LPC	"	White	0.00	3000.00
Total Volume (ml) of SSOL needed per day= 27.60				

500 ml Stock Solution (SSOL) @ 2000 ppm: 1.00 g (2.7 ml) SDS + 497.3 ml DH_2O
Specific Gravity of SDS = 0.37 g/ml

Weight of SDS = 1.0004 g Balance ID#: B2
Date & Time: 03-14-12 1658 Initials: JG

Data Pages & Calculations by: Veronica McMahon QA/QC Check by: Jennifer Duffy

M. beryllina, 3000 ml total volume / treatment = 3 Rep x 1000 ml / replicate

DH_2O = Dilution Water = **20 ppt Synthetic Seawater**

	LPC	Meter ID#
Date	03/14	//////
DH_2O Lot #	25R-073-12	//////
Alkalinity	108	//////
Salinity	20.3	A46
pH	8.0	A93
Temp.	23.7	A46
	VL	//////

Prep Date	03/14
Shaker Table ID#	3H1-4 A43
Initial	VL

LPC: Laboratory Performance Control, 20 ppt synthetic seawater
Alkalinity: mg/l as $CaCO_3$ Salinity: ppt pH: su Temp: °C

Comments: *Artemia* Lot # 030211-2; Feed *M. beryllina* once daily.

Inland Silverside, *Menidia beryllina*
Acute Static 96–Hour Sodium Dodecyl Sulfate (SDS) Definitive Test
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Survival Data

Time	Treatment, ppm SDS																		Date & Initials
	REP	LPC 0 ppm White	REP	1.0 ppm Blue	REP	1.7 ppm Green	REP	2.9 ppm Yellow	REP	4.8 ppm Brown	REP	8.0 ppm Black	REP						
0 HR	1	10	4	10	7	10	10	10	13	10	16	10	19						03/14/12
1747	2	10	5	10	8	10	11	10	14	10	17	10	20						TB
	3	10	6	10	9	10	12	10	15	10	18	10	21						
24 HR	1	10	4	10	7	8	10	3	13	0	16	0	19						03/15/12
1547	2	10	5	10	8	10	11	4	14	0	17	0	20						JG
	3	10	6	10	9	10	12	0	15	0	18	0	21						
48 HR	1	10	4	10	7	8	10	1	13	0	16	0	19						03/16/12
1312	2	10	5	10	8	10	11	4	14	0	17	0	20						VL
	3	10	6	10	9	10	12	0	15	0	18	0	21						
72 HR	1	10	4	10	7	8	10	1	13	0	16	0	19						03/17/12
1150	2	10	5	9	8	10	11	4	14	0	17	0	20						TK
	3	10	6	10	9	10	12	0	15	0	18	0	21						
96 HR	1	10	4	10	7	8	10	1	13	0	16	0	19						03/18/12
1808	2	10	5	9	8	10	11	4	14	0	17	0	20						JG
	3	10	6	10	9	10	12	0	15	0	18	0	21						
% Survival	100		96.7		93.3		16.7		0		0								

3/14/2012 12:31 PM
Environmental Enterprises USA, Inc.

Counted by: Terry Bunch QC/QA by: Veronica McNew

Loaded by: Terry Bunch

Test Organisms Age: 7 days old Test Organisms Source: EE

Test Organism Lot #: MN-067-12

Data Entry by: Veronica McNew

Double Data Entry by: Veronica McNew and/or

QC/QA by: Jennifer Griffith

***M. beryllina* Water Quality Data**

LPC & All Treatments: Initial & Final Temp.: 23.5 to 26.4°C; Initial & Final DO: 4.0 to ≤ 7.4 mg/l
LPC: Initial Salinity: 18.5 to 21.4 ppt

0 to 24 HR		Treatment, ppm SDS							
03/14/12 – 03/15/12		LPC	1.0 ppm	1.7 ppm	2.9 ppm	4.8 ppm	8.0 ppm		Meter ID#
DO	I	7.3	7.3	7.3	7.2	7.3	7.3		S7
	F	7.0	7.0	6.7	6.7	6.5	6.4		S7
Temp	I	23.7	23.9	24.0	23.9	23.9	24.0		A41b
	F	25.1	25.2	25.2	25.2	25.3	25.5		A46
Salinity	I	20.3	20.3	20.3	20.2	20.3	20.3		A41b
	F	20.3	20.3	20.4	20.4	20.4	20.3		A46
pH	F	7.9	8.0	8.0	8.0	7.9	7.8		A93
Initials		Initial: TB VM				Final: SD MR			
Times		Initial Time: 1936				Final Time: 0919			

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

Comments: _____

Sodium Dodecyl Sulfate, DFT D-009-12 (For Product Test D-006-12)
LC50

M. beryllina Water Quality Data

LPC & All Treatments: Initial & Final Temp.: 23.5 to 26.4°C; Initial & Final DO: 4.0 to ≤ 7.4 mg/l
LPC: Initial Salinity: 18.5 to 21.4 ppt

48 HR	Treatment, ppm SDS							
03/16/12	LPC	1.0 ppm	1.7 ppm	2.9 ppm	4.8 ppm	8.0 ppm		Meter ID#
DO F	6.7	6.7	6.5	6.2				S7
Temp F	25.7	25.8	26.0	26.0				A46
Salinity F	20.5	20.4	20.5	20.4				A46
pH F	8.0	8.0	7.9	7.9				A93
Initials	Final: METK							
Times	Final Time: 0913							

72 HR	Treatment, ppm SDS							
03/17/12	LPC	1.0 ppm	1.7 ppm	2.9 ppm	4.8 ppm	8.0 ppm		Meter ID#
DO F	6.4	6.4	6.3	6.3				S7
Temp F	25.7	25.1	25.9	25.8				A46
Salinity F	20.6	20.6	20.6	20.5				A46
pH F	8.0	7.9	7.9	7.9				A93
Initials	Final: MRTK							
Times	Final Time: 0805							

96 HR	Treatment, ppm SDS							
03/18/12	LPC	1.0 ppm	1.7 ppm	2.9 ppm	4.8 ppm	8.0 ppm		Meter ID#
DO F	6.6	6.7	6.6	6.6				S7
Temp F	25.7	25.8	25.9	25.8				A46
Salinity F	20.7	20.7	20.8	20.7				A46
pH F	8.0	8.0	8.0	8.0				A93
Initials	Final: MEMR							
Times	Final Time: 0800							

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

Data Pages

- ☑ Calculations on mixing page are correct. (sign mixing page)
- ☑ Dates, dilutions, test method, # of replicates, replicate volume, product, acceptance limits, data analysis endpoint, and test organisms are correct throughout data pages.
- ☑ Format correct. (spaces for all entries, page numeration, no split pages, etc.)

JG Initials 03-14-12 Date

Chain-of-Custody

- ☑ Product on COC matches sample bottle/container.
- ☑ Product on COC matches test data pages.
- ☑ Lab # on COC matches sample bottle/container.
- ☑ Lab # on COC matches test data pages.
- ☑ Sample volume/amount is sufficient for test duration. (Sample volume in container(s) checked against sample volume on mixing page)

Sample volume/amount available: 95 ml or (g)

Sample volume/amount needed: 1 ml or (g)

(Sample volume/amount insufficient if sample volume/amount available < sample volume/amount needed)

VL Initials 03/14/12 Date

Labels

- ☑ Lab # on labels matches test data pages.
- ☑ Test organism on data pages match labels (i.e. *Menidia beryllina* = MN, *Mysidopsis bahia* = MB)
- ☑ Test Concentrations on labels match Test Concentrations on test data pages.

VL Initials 03/14/12 Date

Raw Data QC/QA'd by: Veronica Lyle Vlen 03/19/12

Acute Toxicity Test-96 Hr Survival

Start Date:	3/14/2012	Test ID:	mn00912DS	Sample ID:	REF-Ref Toxicant
End Date:	3/18/2012	Lab ID:	EE-Environmental Enterprise	Sample Type:	SDS-Sodium dodecyl sulfate
Sample Date:		Protocol:	EPAM 02-EPA Marine	Test Species:	MB-Menidia beryllina
Comments:					

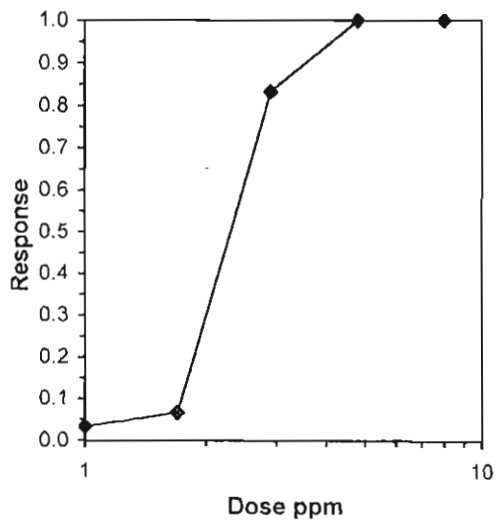
Conc-ppm	1	2	3
PC-LP Control	1.0000	1.0000	1.0000
1	1.0000	0.9000	1.0000
1.7	0.8000	1.0000	1.0000
2.9	0.1000	0.4000	0.0000
4.8	0.0000	0.0000	0.0000
8	0.0000	0.0000	0.0000

			Transform: Untransformed					Number	Total
Conc-ppm	Mean	N-Mean	Mean	Min	Max	CV%	N	Resp	Number
PC-LP Control	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	3	0	30
1	0.9667	0.9667	0.9667	0.9000	1.0000	5.973	3	1	30
1.7	0.9333	0.9333	0.9333	0.8000	1.0000	12.372	3	2	30
2.9	0.1667	0.1667	0.1667	0.0000	0.4000	124.900	3	25	30
4.8	0.0000	0.0000	0.0000	0.0000	0.0000	0.000	3	30	30
8	0.0000	0.0000	0.0000	0.0000	0.0000	0.000	3	30	30

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.05)	0.93927	0.859	0.55318	1.43951
Equality of variance cannot be confirmed				

Trimmed Spearman-Karber

Trim Level	EC50	95% CL	
0.0%			
5.0%	2.3344	2.1361	2.5511
10.0%	2.3140	2.1402	2.5018
20.0%	2.2991	2.1646	2.4419
Auto-3.3%	**2.3294**	**2.1236**	**2.5551**



APPENDIX F

3/9/2012 10:21 AM
Environmental Enterprises USA, Inc.

Mysid, *Mysidopsis bahia*
Acute Static 48–Hour Product Range-Finder Test (RFT)
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Natural Solutions Group Corporation – VirO$_2$Syl
Contact: Marcos Gonzalez

Test Concentrations, ppm VirO2Syl

Mysidopsis bahia	Total Volume/ Concentration, ml	Color Code	ml product	ml DH_2O
100.0 ppm	1000.00	Black	100.00	900.00
50.0 ppm	"	Brown	50.00	950.00
25.0 ppm	"	Yellow	25.00	975.00
12.5 ppm	"	Green	12.50	987.50
6.3 ppm	"	Blue	6.30	993.70
0 ppm LPC	"	White	0.00	1000.00
Total Volume (ml) of product needed per day = 193.80				

550 ml Stock Solution (SSOL) @ 1000 ppm: 0.55 ml VirO2Syl + 549.45 ml DH_2O

Data Pages & Calculations by: Veronica McAlear QA/QC Check by: Jennifer Griffith

M. bahia = 1 Rep x 1000 ml

DH_2O = Dilution Water = **20 ppt Synthetic Seawater**

	LPC	Meter ID#
Date	03/09	//////
DH_2O Lot #	25R-061-12	//////
Alkalinity	120	//////
Salinity	19.7	A46
pH	8.0	A93
Temp.	23.4	A46
	VL	//////

Prep Date	03/09
Blender ID#	A40
Initial	JG

LPC: Laboratory Performance Control, 20 ppt synthetic seawater
Alkalinity: mg/l as $CaCO_3$ Salinity: ppt pH: su Temp: °C

Comments: *Artemia* Lot # 030211-2; Feed *M. bahia* once daily.

Mysid, *Mysidopsis bahia*
Acute Static 48–Hour Product Range-Finder Test (RFT)
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Survival Data

Treatment, ppm VirO2Syl														
Time	REP	LPC 0 ppm White	REP	/////	REP	6.3 ppm Blue	REP	12.5 ppm Green	REP	25 ppm Yellow	REP	50 ppm Brown	REP	100 ppm Black	Date & Initials
0 HR 1744	1	10		/////	2	10	3	10	4	10	5	10	6	10	03/09/12 TB
24 HR 1226	1	10		/////	2	5	3	3	4	2	5	4	6	1	03/10/12 JG
48 HR 1629	1	10		/////	2	5	3	2	4	1	5	0	6	0	03/11/12 TB
% Survival		100		/////		50		20		10		0		0	

Counted by: Terry Bunch QC/QA by: Veronica C. McNew

Loaded by: Terry Bunch

Test Organisms Age: 5 days old Test Organisms Source: EE

Test Organisms Lot #: MB.042.12

Data Entry by: Veronica C. McNew

Double Data Entry by: Veronica C. McNew and/or

QC/QA by: Jennifer Griffith

Comments:

M. bahia Water Quality Data

LPC & All Treatments: Initial & Final Temp. – 23.5 – 26.4°C; Initial & Final DO – 4.0 < 7.4 mg/l
LPC: Initial Salinity 18.5 – 21.4 ppt

0 to 24 HR	Treatment, ppm VirO2Syl							
03/09/12 – 03/10/12	LPC	/////	6.3 ppm	12.5 ppm	25 ppm	50 ppm	100 ppm	Meter ID#
DO I	7.0	/////	7.3	7.3	7.6	7.7	8.0	S7
F	7.2	/////	(A) 23 7.4	7.7	7.9	8.2	98 (B) 8.6	S7
Temp I	23.6	/////	23.9	24.0	24.1	24.0	24.0	A46
F	24.3	/////	23.8	23.6	24.0	23.7	23.7	A46
Salinity I	19.7	/////	20.2	20.2	20.2	20.2	20.2	A46
F	19.8	/////	20.3	20.3	20.3	20.3	20.3	A46
pH F	8.0	/////	8.1	8.1	8.0	8.1	8.1	A93
Initials	Initial: JG				Final: SD VM			
Times	Initial Time: 1836				Final Time: 0812			

48 HR	Treatment, ppm VirO2Syl							
03/11/12	LPC	/////	6.3 ppm	12.5 ppm	25 ppm	50 ppm	100 ppm	Meter ID#
DO F	6.8	/////	7.0	7.0	7.1	7.7	8.9	S7
Temp F	25.3	/////	24.9	24.6	24.9	24.8	24.5	A46
Salinity F	20.0	/////	20.4	20.4	20.5	20.4	20.4	A46
pH F	7.9	/////	8.0	8.0	8.0	8.0	8.0	A93
Initials	Final: SD VM							
Times	Final Time: 0748							

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

Comments: (A) wrong date JG-030912 (B) wrong Date SD 3-10-12

Data Pages

- ☑ Calculations on mixing page are correct. (sign mixing page)
- ☑ Dates, dilutions, test method, # of replicates, replicate volume, product, acceptance limits, data analysis endpoint, and test organisms are correct throughout data pages.
- ☑ Format correct. (spaces for all entries, page numeration, no split pages, etc.)

JG Initials 03-09-12 Date

Chain-of-Custody

- ☑ Product on COC matches sample bottle/container.
- ☑ Product on COC matches test data pages.
- ☑ Lab # on COC matches sample bottle/container.
- ☑ Lab # on COC matches test data pages.
- ☑ Sample volume/amount is sufficient for test duration. (Sample volume in container(s) checked against sample volume on mixing page)

Sample volume/amount available: 8000 (ml) or g

Sample volume/amount needed: 0.55 (ml) or g

(Sample volume/amount insufficient if sample volume/amount available < sample volume/amount needed)

VM Initials 03/09/12 Date

Labels

- ☑ Lab # on labels matches test data pages.
- ☑ Test organism on data pages match labels (i.e. *Menidia beryllina* = MN, *Mysidopsis bahia* = MB)
- ☑ Test Concentrations on labels match Test Concentrations on test data pages.

VM Initials 03/09/12 Date

Raw Data QC/QA'd by: Veronica McVea 03/12/12

Acute Toxicity Test-48 Hr Survival

Start Date:	3/9/2012	Test ID:	mb00612RP	Sample ID:	NCP-National Contingency Plan
End Date:	3/11/2012	Lab ID:	EE-Environmental Enterprise	Sample Type:	PRD-Product
Sample Date:		Protocol:	EPAM 02-EPA Marine	Test Species:	MY-Mysidopsis bahia
Comments:					

Conc-ppm	1
PC-LP Control	1.0000
6.3	0.5000
12.5	0.2000
25	0.1000
50	0.0000
100	0.0000

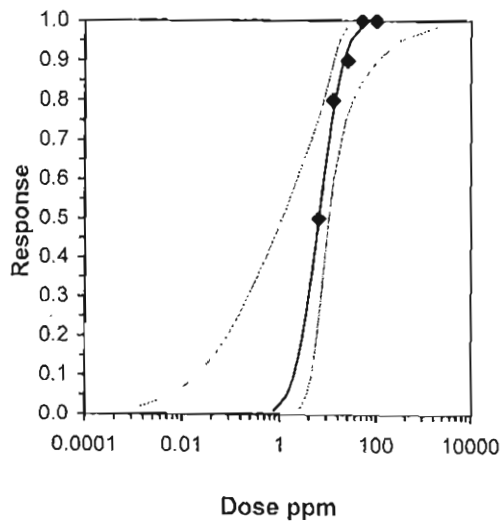
			Transform: Untransformed					Number	Total
Conc-ppm	Mean	N-Mean	Mean	Min	Max	CV%	N	Resp	Number
PC-LP Control	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
6.3	0.5000	0.5000	0.5000	0.5000	0.5000	0.000	1	5	10
12.5	0.2000	0.2000	0.2000	0.2000	0.2000	0.000	1	8	10
25	0.1000	0.1000	0.1000	0.1000	0.1000	0.000	1	9	10
50	0.0000	0.0000	0.0000	0.0000	0.0000	0.000	1	10	10
100	0.0000	0.0000	0.0000	0.0000	0.0000	0.000	1	10	10

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Normality of the data set cannot be confirmed				
Equality of variance cannot be confirmed				

Maximum Likelihood-Probit

Parameter	Value	SE	95% Fiducial Limits		Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	2.52972	0.92181	0.72298	4.33647	0	0.37	7.81473	0.94637	0.7979	0.3953	4
Intercept	2.98154	1.01325	0.99557	4.9675							
TSCR											

Point	Probits	ppm	95% Fiducial Limits	
EC01	2.674	0.7556	0.00078	2.48484
EC05	3.355	1.40504	0.00676	3.62408
EC10	3.718	1.9557	0.02123	4.45063
EC15	3.964	2.44454	0.04583	5.12699
EC20	4.158	2.91881	0.08426	5.75116
EC25	4.326	3.39839	0.14174	6.36228
EC40	4.747	4.98598	0.51672	8.34604
EC50	**5.000**	**6.27911**	**1.10008**	**10.0497**
EC60	5.253	7.90763	2.25496	12.5684
EC75	5.674	11.6018	6.038	22.4466
EC80	5.842	13.508	7.92958	31.8056
EC85	6.036	16.1287	10.0822	51.5916
EC90	6.282	20.1602	12.674	102.043
EC95	6.645	28.0614	16.574	300.989
EC99	7.326	52.1799	25.3019	2480.83



Mysid, *Mysidopsis bahia*
Acute Static 48–Hour No. 2 Fuel Oil Range-Finder Test (RFT)
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Test Concentrations, ppm No.2 Fuel Oil (Oil)

Mysidopsis bahia	Total Volume/ Concentration, ml	Color Code	ml SSOL	ml DH_2O
20 ppm	1000.00	Black	20.00	980.00
10 ppm	"	Brown	10.00	990.00
5.0 ppm	"	Yellow	5.00	995.00
2.5 ppm	"	Green	2.50	997.50
1.3 ppm	"	Blue	1.30	998.70
0 ppm LPC	"	White	0.00	1000.00
Total Volume (ml) of SSOL needed per day = 38.80				

550 ml Stock Solution (SSOL) @ 1000 ppm = 0.55 ml No.2 Fuel Oil + 549.45 ml DH_2O

Calculations by: Veronica McNew QA/QC Officer: Jennifer Griffith

M. bahia = 1 Rep x 1000 ml

DH_2O = Dilution Water = **20 ppt Synthetic Seawater**

	LPC	Meter ID#
Date	03/09	//////
DH_2O Lot #	25R-041-12	//////
Alkalinity	120	//////
Salinity	13.7	A41
pH	8.0	A93
Temp.	23.4	A41
	VL	//////

Prep Date	03/09
Blender ID#	A40
Initial	JG

LPC: Laboratory Performance Control, 20 ppt synthetic seawater
Alkalinity: mg/l as $CaCO_3$ Salinity: ppt pH: su Temp: °C

Comments: *Artemia* Lot # 030211-2; Feed *M. bahia* once daily.

Mysid, *Mysidopsis bahia*

Acute Static 48–Hour No.2 Fuel Oil Range-Finder Test

3.0 Revised Dispersant Toxicity Test
FR / Vol. 59, No. 178 / 47461 - 47464

Survival Data

Treatment ppm Oil														
Time	**REP**	**LPC 0 ppm White**	**REP**	**//////**	**REP**	**1.3 ppm Blue**	**REP**	**2.5 ppm Green**	**REP**	**5.0 ppm Yellow**	**REP**	**10.0 ppm Brown**	**REP**	**20.0 ppm Black**	**Date & Initials**
0 HR 1810	1	10		//////	2	10	3	10	4	10	5	10	6	10	03/09/12 TB
24 HR 1231	1	10		//////	2	10	3	10	4	8	5	0	6	0	03/10/12 JG
48 HR 1635	1	10		//////	2	10	3	8	4	1	5	0	6	0	03/11/12 TB
% Survival		100				100		80		10		0		0	

Counted by: Terry Brunk QC/QA by: Veronica McMeen

Loaded by: Terry Brunk

Test Organisms Age: 5 days old Test Organisms Source: EE

Test Organisms Lot #: MB-142-12

Data Entry by: Veronica McMeen

Double Data Entry by: Veronica McMeen and/or

QC/QA by: Jennifer Griffith

3/9/12, 9:15 AM
Environmental Enterprises USA, Inc.

M. bahia Water Quality Data

LPC & All Treatments: Initial & Final Temp.: 23.5 to 26.4°C; Initial & Final DO: 4.0 to ≤ 7.4 mg/l
LPC: Initial Salinity: 18.5 to 21.4 ppt

0 to 24 HR	Treatment ppm Oil							
03/09/12 – 03/10/12	LPC	//////	1.3 ppm	2.5 ppm	5.0 ppm	10.0 ppm	20.0 ppm	Meter ID#
DO I	7.0	//////	7.2	7.1	7.1	7.1	7.1	S7
F	7.2	//////	7.2	7.2	7.2	7.2	7.2	S7
Temp I	23.6	//////	23.7	24.2	24.3	24.1	24.2	A46
F	24.3	//////	24.8	24.9	25.0	25.0	25.0	A46
Salinity I	19.7	//////	20.3	20.2	20.2	20.2	20.2	A46
F	19.8	//////	20.4	20.3	20.3	20.3	20.3	A46
pH F	8.0	//////	8.0	8.0	8.0	8.0	8.0	A93
Initials	Initial: JG				Final: SD VM			
Times	Initial Time: 1840				Final Time: 0816			

48 HR	Treatment ppm Oil							
03/11/12	LPC	//////	1.3 ppm	2.5 ppm	5.0 ppm	10.0 ppm	20.0 ppm	Meter ID#
DO F	6.8	//////	7.0	6.9	6.9	6.9		S7
Temp F	25.3	//////	26.1	25.9	26.0	25.9		A46
Salinity F	20.0	//////	20.6	20.4	20.6	20.4		A46
pH F	7.9	//////	8.0	8.0	8.0	8.0		A93
Initials	Final: SD VM							
Times	Final Time: 0750							

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

Comments: _____

_____

Data Pages

- ☑ Calculations on mixing page are correct. (sign mixing page)
- ☑ Dates, dilutions, test method, # of replicates, replicate volume, product, acceptance limits, data analysis endpoint, and test organisms are correct throughout data pages.
- ☑ Format correct. (spaces for all entries, page numeration, no split pages, etc.)

JC Initials 03-09-12 Date

Chain-of-Custody

- ☑ Product on COC matches sample bottle/container.
- ☑ Product on COC matches test data pages.
- ☑ Lab # on COC matches sample bottle/container.
- ☑ Lab # on COC matches test data pages.
- ☑ Sample volume/amount is sufficient for test duration. (Sample volume in container(s) checked against sample volume on mixing page)

Sample volume/amount available: 40 (ml) or g

Sample volume/amount needed: 0.55 ml No.2 Fuel Oil (ml) or g

(Sample volume/amount insufficient if sample volume/amount available < sample volume/amount needed)

VM Initials 03/09/12 Date

Labels

- ☑ Lab # on labels matches test data pages.
- ☑ Test organism on data pages match labels (i.e. *Menidia beryllina* = MN, *Mysidopsis bahia* = MB)
- ☑ Test Concentrations on labels match Test Concentrations on test data pages.

VM Initials 03/09/12 Date

Raw Data QC/QA'd by: Veronica McMew 03/12/12

Acute Toxicity Test-48 Hr Survival

Start Date:	3/9/2012	Test ID:	mb00812RO	Sample ID:	NCP-National Contingency Plan
End Date:	3/11/2012	Lab ID:	EE-Environmental Enterprise	Sample Type:	2FO-No. 2 Fuel Oil
Sample Date:		Protocol:	EPAM 02-EPA Marine	Test Species:	MY-Mysidopsis bahia
Comments:					

Conc-ppm	1
PC-LP Control	1.0000
1.3	1.0000
2.5	0.8000
5	0.1000
10	0.0000
20	0.0000

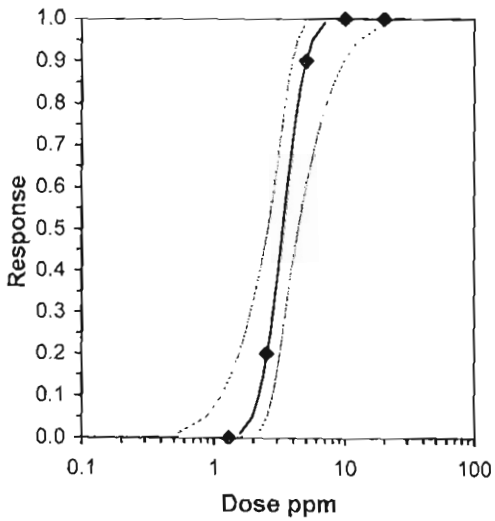
			Transform: Untransformed					Number	Total
Conc-ppm	Mean	N-Mean	Mean	Min	Max	CV%	N	Resp	Number
PC-LP Control	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
1.3	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
2.5	0.8000	0.8000	0.8000	0.8000	0.8000	0.000	1	2	10
5	0.1000	0.1000	0.1000	0.1000	0.1000	0.000	1	9	10
10	0.0000	0.0000	0.0000	0.0000	0.0000	0.000	1	10	10
20	0.0000	0.0000	0.0000	0.0000	0.0000	0.000	1	10	10

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Normality of the data set cannot be confirmed				
Equality of variance cannot be confirmed				

Maximum Likelihood-Probit

Parameter	Value	SE	95% Fiducial Limits		Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	7.20731	2.14499	3.00314	11.4115	0	0.02513	7.81473	0.99895	0.51816	0.13875	3
Intercept	1.26545	1.15621	-1.0007	3.53163							
TSCR											

Point	Probits	ppm	95% Fiducial Limits	
EC01	2.674	1.56814	0.53207	2.16618
EC05	3.355	1.94958	0.87994	2.53429
EC10	3.718	2.18952	1.14304	2.7737
EC15	3.964	2.36787	1.35735	2.96163
EC20	4.158	2.51993	1.54975	3.13258
EC25	4.326	2.65813	1.72967	3.2999
EC40	4.747	3.04095	2.22797	3.85205
EC50	**5.000**	**3.29732**	**2.53768**	**4.32242**
EC60	5.253	3.5753	2.83637	4.94268
EC75	5.674	4.09021	3.29481	6.39785
EC80	5.842	4.31454	3.46638	7.1497
EC85	6.036	4.5916	3.66236	8.17228
EC90	6.282	4.96562	3.9066	9.71418
EC95	6.645	5.57675	4.27163	12.6306
EC99	7.326	6.93326	4.99304	20.9073



3/9/2012 10:07 AM
Environmental Enterprises USA, Inc.

Mysid, *Mysidopsis bahia*

Acute Static 48–Hour 10:1 No. 2 Fuel Oil / Product Range-Finder Test (RFT)

3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Natural Solutions Group Corporation – $VirO_2Syl$

Contact: Marcos Gonzalez

Test Concentrations, ppm 10:1 No.2 Fuel Oil / $VirO_2Syl$

Mysidopsis bahia	Total Volume/ Concentration, ml	Color Code	ml SSOL	ml DH_2O
20.0 ppm	1000.00	Black	20.00	980.00
10.0 ppm	"	Brown	10.00	990.00
5.0 ppm	"	Yellow	5.00	995.00
2.5 ppm	"	Green	2.50	997.50
1.3 ppm	"	Blue	1.30	998.70
0.6 ppm	"	Wh/Blu	0.60	999.40
0 ppm LPC	"	White	0.00	1000.00
Total Volume (ml) of SSOL needed per day= 39.40				

550 ml Stock Solution (SSOL) @ 1000 ppm:
0.50 ml No.2 Fuel Oil + 0.05 ml $VirO_2Syl$ + 549.45 ml DH_2O

Data Pages & Calculations by: Veronica McKeon QA/QC Check by: Jeff Griffith

M. bahia = 1 Rep x 1000 ml
DH_2O = Dilution Water = **20 ppt Synthetic Seawater**

	LPC	Meter ID#
Date	03/09	//////
DH_2O Lot #	25R-Oil-12	//////
Alkalinity	120	//////
Salinity	19.7	A46
pH	8.0	A93
Temp.	23.6	A46
	VM	//////

Prep Date	03/09
Blender ID#	A40
Initial	JG

LPC: Laboratory Performance Control, 20 ppt synthetic seawater
Alkalinity: mg/l as $CaCO_3$ Salinity: ppt pH: su Temp: °C

Comments: *Artemia* Lot # 030211-2; Feed *M. bahia* once daily.

Mysid, *Mysidopsis bahia*
Acute Static 48–Hour 10:1 No. 2 Fuel Oil / Product Range-Finder Test (RFT)
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Survival Data

Treatment ppm, 10:1 No. 2 Fuel Oil / 0.50 ml No.2 Fuel Oil + 0.05 ml $VirO_2Syl$															
Time	**REP**	**LPC 0 ppm White**	**REP**	**0.6 ppm Wh/Blu**	**REP**	**1.3 ppm Blue**	**REP**	**2.5 ppm Green**	**REP**	**5.0 ppm Yellow**	**REP**	**10.0 ppm Brown**	**REP**	**20.0 ppm Black**	**Date & Initials**
0 HR 1803	1	10	2	10	3	10	4	10	5	10	6	10	7	10	03/09/12 TB
24 HR 1228	1	10	2	10	3	16	4	16	5	10	6	6	7	0	03/10/12 JG
48 HR 1632	1	10	2	10	3	10	4	10	5	4	6	1	7	0	03/11/12 TB
% Survival		100		100		100		100		40		10		0	

Counted by: Terry Bunch QC/QA by: Veronica McNew

Loaded by: Terry Bunch

Test Organisms Age: 5 days old Test Organisms Source: EE

Test Organisms Lot #: MB 142-12

Data Entry by: Veronica McNew

Double Data Entry by: Veronica McNew and/or

QC/QA by: Jennifer Griffith

Comments:

M. bahia Water Quality Data

LPC & All Treatments: Initial & Final Temp.: 23.5 to 26.4°C; Initial & Final DO: 4.0 to ≤ 7.4 mg/l
LPC: Initial Salinity: 18.5 to 21.4 ppt

0 to 24 HR	Treatment, ppm 10:1 No. 2 Fuel Oil / VirO$_2$Syl							
03/09/12 – 03/10/12	LPC	0.6 ppm	1.3 ppm	2.5 ppm	5.0 ppm	10.0 ppm	20.0 ppm	Meter ID#
DO I	7.0	7.2	7.1	7.3	7.3	7.2	7.3	S7
F	7.2	7.2	7.3	7.3	7.3	7.4	7.4	S7
Temp I	23.9 (A) 23.6	23.9	24.0	23.9	23.8	23.8	23.9	A46
F	24.3	23.7	23.5	23.6	23.8	23.6	23.6	A46
Salinity I	(A) 19.7 ~~20.3~~	20.3	20.2	20.2	20.2	20.2	20.2	A46
F	19.8	20.3	20.3	20.3	20.3	20.3	20.3	A46
pH F	8.0	8.1	8.0	8.0	8.0	8.0	8.0	A93
Initials	Initial: JG				Final: SD VM			
Times	Initial Time: 1837				Final Time: 0814			

48 HR	Treatment, ppm 10:1 No. 2 Fuel Oil / VirO$_2$Syl							
03/11/12	LPC	0.6 ppm	1.3 ppm	2.5 ppm	5.0 ppm	10.0 ppm	20.0 ppm	Meter ID#
DO F	6.8	7.0	7.1	7.2	7.1	7.1		S7
Temp F	25.3	24.9	24.5	24.3	24.6	24.5		A46
Salinity F	20.0	20.4	20.5	20.4	20.5	20.4		A46
pH F	7.9	8.0	8.0	8.0	8.0	8.0		A93
Initials	Final: SD VM							
Times	Final Time: 0749							

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

Comments: (A) wrong date JG-030912

Data Pages

- ☑ Calculations on mixing page are correct. (sign mixing page)
- ☑ Dates, dilutions, test method, # of replicates, replicate volume, product, acceptance limits, data analysis endpoint, and test organisms are correct throughout data pages.
- ☑ Format correct. (spaces for all entries, page numeration, no split pages, etc.)

JC Initials 03-09-12 Date

Chain-of-Custody

- ☑ Product on COC matches sample bottle/container.
- ☑ Product on COC matches test data pages.
- ☑ Lab # on COC matches sample bottle/container.
- ☑ Lab # on COC matches test data pages.
- ☑ Sample volume/amount is sufficient for test duration. (Sample volume in container(s) checked against sample volume on mixing page)

Sample volume/amount available: 40 oil 8000 PRD (ml) or g

Sample volume/amount needed: 0.50 ml No.2 Fuel Oil and 0.05 ml PRD (ml) or g

(Sample volume/amount insufficient if sample volume/amount available < sample volume/amount needed)

VL Initials 03/09/12 Date

Labels

- ☑ Lab # on labels matches test data pages.
- ☑ Test organism on data pages match labels (i.e. *Menidia beryllina* = MN, *Mysidopsis bahia* = MB)
- ☑ Test Concentrations on labels match Test Concentrations on test data pages.

VL Initials 03/09/12 Date

Raw Data QC/QA'd by: Veronica McMen 03/12/12

Acute Toxicity Test-48 Hr Survival

Start Date:	3/9/2012	Test ID:	mb00612RPO	Sample ID:	NCP-National Contingency Plan
End Date:	3/11/2012	Lab ID:	EE-Environmental Enterprise	Sample Type:	PIO-Product/No. 2 Fuel Oil Mixture
Sample Date:		Protocol:	EPAM 02-EPA Marine	Test Species:	MY-Mysidopsis bahia
Comments:					

Conc-ppm	1
PC-LP Control	1.0000
0.6	1.0000
1.3	1.0000
2.5	1.0000
5	0.4000
10	0.1000
20	0.0000

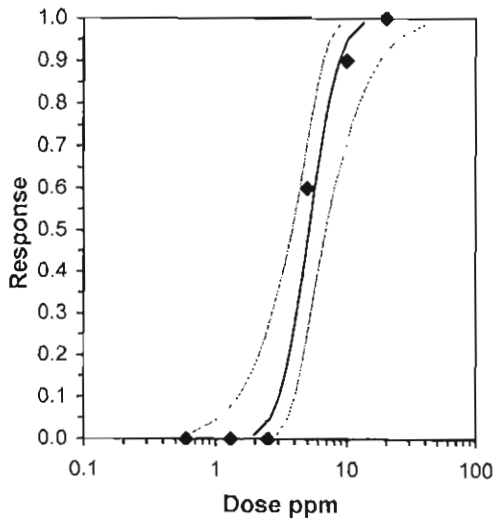
			Transform: Untransformed					Number	Total
Conc-ppm	Mean	N-Mean	Mean	Min	Max	CV%	N	Resp	Number
PC-LP Control	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
0.6	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
1.3	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
2.5	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
5	0.4000	0.4000	0.4000	0.4000	0.4000	0.000	1	6	10
10	0.1000	0.1000	0.1000	0.1000	0.1000	0.000	1	9	10
20	0.0000	0.0000	0.0000	0.0000	0.0000	0.000	1	10	10

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Normality of the data set cannot be confirmed				
Equality of variance cannot be confirmed				

Maximum Likelihood-Probit

Parameter	Value	SE	95% Fiducial Limits		Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	5.54959	1.51483	2.58052	8.51866	0	1.48411	9.48773	0.82945	0.70891	0.18019	7
Intercept	1.06584	1.11514	-1.1198	3.25152							
TSCR											

Point	Probits	ppm	95% Fiducial Limits	
EC01	2.674	1.94858	0.60728	2.88646
EC05	3.355	2.58534	1.09243	3.54365
EC10	3.718	3.00596	1.48349	3.98102
EC15	3.964	3.32775	1.81467	4.32754
EC20	4.158	3.60791	2.12082	4.64407
EC25	4.326	3.86697	2.41432	4.95442
EC40	4.747	4.6053	3.26367	5.98028
EC50	**5.000**	**5.11576**	**3.81999**	**6.85933**
EC60	5.253	5.68279	4.37893	8.03328
EC75	5.674	6.76781	5.28178	10.8672
EC80	5.842	7.25378	5.63368	12.3735
EC85	6.036	7.86445	6.04473	14.4634
EC90	6.282	8.70637	6.56991	17.695
EC95	6.645	10.1228	7.37976	24.0326
EC99	7.326	13.4308	9.05878	43.2381



Mysid, *Mysidopsis bahia*

Acute Static 48–Hour Sodium Dodecyl Sulfate (SDS) Range-Finder Test (RFT)

3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Test Concentrations, ppm SDS (Standard Reference Toxicant)

Mysidopsis bahia	Total Volume/ Concentration, ml	Color Code	ml SSOL	ml DH_2O
20.0 ppm	1000.00	Black	10.00	990.00
12.0 ppm	"	Brown	6.00	994.00
7.0 ppm	"	Yellow	3.50	996.50
4.0 ppm	"	Green	2.00	998.00
2.4 ppm	"	Blue	1.20	998.80
0 ppm LPC	"	White	0.00	1000.00
Total Volume (ml) of SSOL needed per day = 22.70				

500 ml Stock Solution (SSOL) @ 2,000 ppm = 1.00 g SDS + 497.3 ml DH_2O
Specific gravity of SDS = 0.37 g/ml; 1.00 g SDS = 2.7 ml

Weight of SDS = 1.0001 g Balance ID#: B2

Date & Time: 03/09/12 1651 Initials: VL

Data Pages & Calculations by: Veronica McMillan QA/QC Check by: Jennifer Griffith

M. bahia = 1 Rep x 1000 ml

DH_2O = Dilution Water = **20 ppt Synthetic Seawater**

	LPC	Meter ID#
Date	03/09	//////
DH_2O Lot #	25R-061-12	//////
Alkalinity	120	//////
Salinity	19.7	A46
pH	8.0	A93
Temp.	23.6	A46
	VL	//////

Prep Date	03/09
Blender ID#	A40
Initial	JG

LPC: Laboratory Performance Control, 20 ppt synthetic seawater
Alkalinity: mg/l as $CaCO_3$ Salinity: ppt pH: su Temp: °C

Comments: *Artemia* Lot # 030211-2; Feed *M. bahia* once daily.

Mysid, *Mysidopsis bahia*
Acute Static 48–Hour SDS Range-Finder Test
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Survival Data

Time	REP	LPC 0 ppm White	REP	/////	REP	2.4 ppm Blue	REP	4.0 ppm Green	REP	7.0 ppm Yellow	REP	12.0 ppm Brown	REP	20.0 ppm Black	Date & Initials
		Treatment, ppm SDS													
0 HR 1712	1	10		/////	2	10	3	10	4	10	5	10	6	10	03/09/12 TB
24 HR 1219	1	10		/////	2	10	3	10	4	10	5	0	6	0	03/10/12 JG
48 HR 1623	1	10		/////	2	10	3	10	4	8	5	0	6	0	03/11/12 TB
% Survival		100				100		100		80		0		0	

Counted by: Tony Bunch QC/QA by: Veronica McNew

Loaded by: Tony Bunch

Test Organisms Age: 5 days old Test Organisms Source: EE

Test Organisms Lot #: MB-142-12

Data Entry by: Veronica McNew

Double Data Entry by: Veronica McNew and/or

QC/QA by: Jennifer Griffith

Comments:

Sodium Dodecyl Sulfate, RFT

D-009-12 (for Product Test D-~~009~~ -12)
LC50

M. bahia Water Quality Data

LPC & All Treatments: Initial & Final Temp.: 23.5 to 26.4°C; Initial & Final DO: 4.0 to ≤ 7.4 mg/l
LPC: Initial Salinity: 18.5 to 21.4 ppt

0 to 24 HR		Treatment, ppm SDS							
03/09/12 – 03/10/12		LPC	//////	2.4 ppm	4.0 ppm	7.0 ppm	12.0 ppm	20.0 ppm	Meter ID#
DO	I	7.0	//////	7.0	7.1	7.0	7.1	7.1	57
	F	7.2	//////	7.2	7.1	7.1	7.1	7.1	57
Temp	I	23.6	//////	23.6	23.7	23.7	23.7	23.7	A46
	F	24.3	//////	24.4	24.3	(A) 24.3	24.3	24.3	A46
Salinity	I	19.7	//////	20.0	20.0	20.0	20.1	20.2	A46
	F	19.8	//////	20.0	20.1	20.3	20.2	20.2	A46
pH	F	8.0	//////	8.0	8.0	8.0	8.0	8.0	A93
Initials		Initial: JG				Final: SD VM			
Times		Initial Time: 1828				Final Time: 0809			

48 HR		Treatment, ppm SDS							
03/11/12		LPC	//////	2.4 ppm	4.0 ppm	7.0 ppm	12.0 ppm	20.0 ppm	Meter ID#
DO	F	6.8	//////	6.6	6.4	5.9			57
Temp	F	25.3	//////	25.2	25.1	25.2			A46
Salinity	F	20.0	//////	20.1	20.2	20.3			A46
pH	F	7.9	//////	7.9	7.8	7.8			A93
Initials		Final: SD VM							
Times		Final Time: 0745							

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

Comments: (A) wrong date SD 3-10-12

Data Pages

- ☑ Calculations on mixing page are correct. (sign mixing page)
- ☑ Dates, dilutions, test method, # of replicates, replicate volume, product, acceptance limits, data analysis endpoint, and test organisms are correct throughout data pages.
- ☑ Format correct. (spaces for all entries, page numeration, no split pages, etc.)

JG Initials 03-09-12 Date

Chain-of-Custody

- ☑ Product on COC matches sample bottle/container.
- ☑ Product on COC matches test data pages.
- ☑ Lab # on COC matches sample bottle/container.
- ☑ Lab # on COC matches test data pages.
- ☑ Sample volume/amount is sufficient for test duration. (Sample volume in container(s) checked against sample volume on mixing page)

Sample volume/amount available: 95 ml or (g)

Sample volume/amount needed: 1.00 g SDS ml or (g)

(Sample volume/amount insufficient if sample volume/amount available < sample volume/amount needed)

VL Initials 03/09/12 Date

Labels

- ☑ Lab # on labels matches test data pages.
- ☑ Test organism on data pages match labels (i.e. *Menidia beryllina* = MN, *Mysidopsis bahia* = MB)
- ☑ Test Concentrations on labels match Test Concentrations on test data pages.

VL Initials 03/09/12 Date

Raw Data QC/QA'd by: Veronica McMeen 03/12/12

Acute Toxicity Test-48 Hr Survival

Start Date:	3/9/2012	Test ID:	mb00912RS	Sample ID:	REF-Ref Toxicant
End Date:	3/11/2012	Lab ID:	EE-Environmental Enterprise	Sample Type:	SDS-Sodium dodecyl sulfate
Sample Date:		Protocol:	EPAM 02-EPA Marine	Test Species:	MY-Mysidopsis bahia
Comments:					

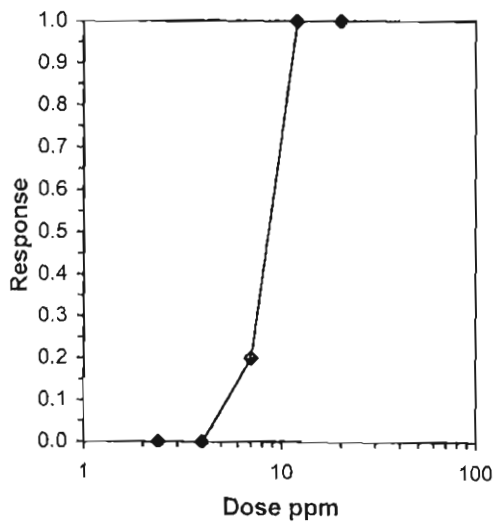
Conc-ppm	1
PC-LP Control	1.0000
2.4	1.0000
4	1.0000
7	0.8000
12	0.0000
20	0.0000

			Transform: Untransformed					Number	Total
Conc-ppm	Mean	N-Mean	Mean	Min	Max	CV%	N	Resp	Number
PC-LP Control	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
2.4	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
4	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	1	0	10
7	0.8000	0.8000	0.8000	0.8000	0.8000	0.000	1	2	10
12	0.0000	0.0000	0.0000	0.0000	0.0000	0.000	1	10	10
20	0.0000	0.0000	0.0000	0.0000	0.0000	0.000	1	10	10

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Normality of the data set cannot be confirmed				
Equality of variance cannot be confirmed				

Trimmed Spearman-Karber

Trim Level	EC50	95% CL	
0.0%	8.2116	7.1462	9.4358
5.0%	8.3435	7.1168	9.7816
10.0%	8.4550	6.9606	10.2703
20.0%	8.5680	7.7022	9.5311
Auto-0.0%	**8.2116**	**7.1462**	**9.4358**



APPENDIX G

3/13/2012 7:50 AM
Environmental Enterprises USA, Inc.

Mysid, *Mysidopsis bahia*
Acute Static 48–Hour Product Definitive Test (DFT)
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Natural Solutions Group Corporation – $VirO_2Syl$
Contact: Marcos Gonzalez

Test Concentrations, ppm $VirO_2Syl$

Mysidopsis bahia	Total Volume / Replicate / Concentration, ml	Color Code	ml SSOL / Replicate	ml DH_2O / Replicate
25.0 ppm	1000.00	Black	25.00	975.00
12.5 ppm	"	Brown	12.50	987.50
6.3 ppm	"	Yellow	6.30	993.70
3.1 ppm	"	Green	3.10	996.90
1.6 ppm	"	Blue	1.60	998.40
0 ppm LPC	"	White	0.00	1000.00
		Total Volume (ml) of SSOL needed per day =	48.5 ml / Replicate X 3 = 145.5 ml	

550 ml Stock Solution (SSOL) @ 1000 ppm: 0.55 ml $VirO_2Syl$+ 549.45 ml DH_2O

Data Pages & Calculations by: Veronica McMinn QA/QC Check by: Jennifer Griffith

M. bahia = 3 Rep x 3000 ml total volume / treatment

DH_2O = Dilution Water = **20 ppt Synthetic Seawater**

	LPC	Meter ID#
Date	03/13	//////
DH_2O Lot #	25R-072-12	//////
Alkalinity	112	//////
Salinity	19.8	A46
pH	8.0	A93
Temp.	23.8	A46
	VR	//////

Prep Date	03/13
Blender ID#	A44
Initial	JG

LPC: Laboratory Performance Control, 20 ppt synthetic seawater
Alkalinity: mg/l as $CaCO_3$ Salinity: ppt pH: su Temp: °C

Comments: *Artemia* Lot # 030211-2; Feed *M. bahia* once daily.

Mysid, *Mysidopsis bahia*
Acute Static 48–Hour Product Definitive Test (DFT)
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Survival Data

Time	REP	LPC 0.0 ppm White	REP	1.6 ppm Blue	REP	3.1 ppm Green	REP	6.3 ppm Yellow	REP	12.5 ppm Brown	REP	25.0 ppm Black	REP		Date & Initials
	Treatment, ppm $VirO_2Syl$														
0 HR	1	10	4	10	7	10	10	10	13	10	16	10	19		03/13/12
1800	2	10	5	10	8	10	11	10	14	10	17	10	20		TB
	3	10	6	10	9	10	12	10	15	10	18	10	21		
24 HR	1	10	4	8	7	4	10	2	13	5	16	7	19		03/14/12
1631	2	10	5	10	8	5	11	5	14	2	17	7	20		
	3	10	6	7	9	8	12	5	15	2	18	9	21		
48 HR	1	10	4	7	7	4	10	2	13	3	16	3	19		03/15/12
1729	2	10	5	10	8	5	11	4	14	2	17	0	20		
	3	10	6	7	9	7	12	3	15	1	18	1	21		
% Survival	100		80		53.3		30		20		13.3				

Counted by: Terry Bunch QC/QA by: Jennifer Griffith

Loaded by: Terry Bunch

Test Organisms Age: 5 days old Test Organisms Source: EE

Test Organism Lot #: Mb-151-12

Data Entry by: Veronica McNew

Double Data Entry by: Veronica McNew and/or

QC/QA by: Jennifer Griffith

Product, DFT

D-006-12
LC50

M. bahia Water Quality Data

LPC & All Treatments: Initial & Final Temp.: 23.5 to 26.4°C; Initial & Final DO: 4.0 to ≤ 7.4 mg/l
LPC: Initial Salinity: 18.5 to 21.4 ppt

0 to 24 HR		Treatment, ppm $VirO_2Syl$							
03/13/12 – 03/14/12		LPC	///	1.6 ppm	3.1 ppm	6.3 ppm	12.5 ppm	25.0 ppm	Meter ID#
DO	I	7.4	///	7.3	7.3	7.4	7.8	7.9	S7
	F	7.0	///	7.0	7.1	7.1	7.2	7.4	S7
Temp	I	23.8	///	24.4	24.4	24.4	24.4	24.5	A46
	F	25.1	///	25.0	25.0	25.0	25.0	25.1	A46
Salinity	I	19.8	///	19.8	19.8	19.8	19.8	19.8	A46
	F	19.9	///	19.8	19.9	19.8	19.8	19.8	A46
pH	F	7.9	///	8.0	8.0	8.0	8.0	8.0	A93
Initials		Initial: JG				Final: hmrsd			
Times		Initial Time: 1850				Final Time: 0918			

48 HR		Treatment, ppm $VirO_2Syl$							
03/15/12		LPC	///	1.6 ppm	3.1 ppm	6.3 ppm	12.5 ppm	25.0 ppm	Meter ID#
DO	F	6.9	///	6.9	7.1	7.0	7.1	7.3	S7
Temp	F	25.1	///	24.7	24.7	24.7	24.6	24.6	A46
Salinity	F	19.9	///	20.0	19.9	19.9	19.9	19.9	A46
pH	F	7.9	///	7.9	7.9	7.9	8.0	8.0	A93
Initials		Final: SD MR							
Times		Final Time: 0913							

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

Comments: _____

Data Pages

- ☑ Calculations on mixing page are correct. (sign mixing page)
- ☑ Dates, dilutions, test method, # of replicates, replicate volume, product, acceptance limits, data analysis endpoint, and test organisms are correct throughout data pages.
- ☑ Format correct. (spaces for all entries, page numeration, no split pages, etc.)

JG Initials 03-13-12 Date

Chain-of-Custody

- ☑ Product on COC matches sample bottle/container.
- ☑ Product on COC matches test data pages.
- ☑ Lab # on COC matches sample bottle/container.
- ☑ Lab # on COC matches test data pages.
- ☑ Sample volume/amount is sufficient for test duration. (Sample volume in container(s) checked against sample volume on mixing page)

Sample volume/amount available: 8000 (ml) or g

Sample volume/amount needed: 0.55 (ml) or g

(Sample volume/amount insufficient if sample volume/amount available < sample volume/amount needed)

VL Initials 03/13/12 Date

Labels

- ☑ Lab # on labels matches test data pages.
- ☑ Test organism on data pages match labels (i.e. *Menidia beryllina* = MN, *Mysidopsis bahia* = MB)
- ☑ Test Concentrations on labels match Test Concentrations on test data pages.

VL Initials 03/13/12 Date

Raw Data QC/QA'd by: Veronica McVea 03/19/12

Acute Toxicity Test-48 Hr Survival

Start Date:	3/13/2012	Test ID:	mb00612DP	Sample ID:	NCP-National Contingency Plan
End Date:	3/15/2012	Lab ID:	EE-Environmental Enterprise	Sample Type:	PRD-Product
Sample Date:		Protocol:	EPAM 02-EPA Marine	Test Species:	MY-Mysidopsis bahia
Comments:					

Conc-ppm	1	2	3
PC-LP Control	1.0000	1.0000	1.0000
1.6	0.7000	1.0000	0.7000
3.1	0.4000	0.5000	0.7000
6.3	0.2000	0.4000	0.3000
12.5	0.3000	0.2000	0.1000
25	0.3000	0.0000	0.1000

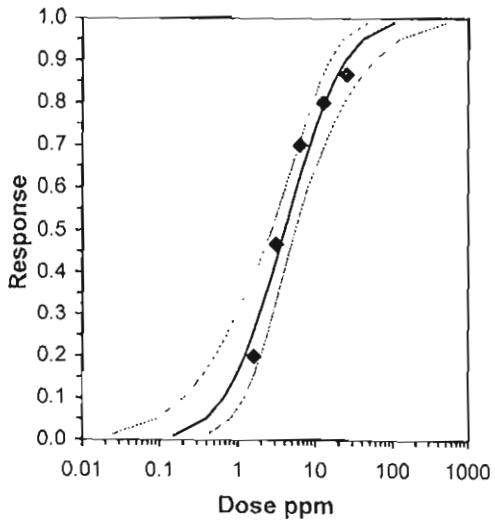
			Transform: Untransformed					Number	Total
Conc-ppm	Mean	N-Mean	Mean	Min	Max	CV%	N	Resp	Number
PC-LP Control	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	3	0	30
1.6	0.8000	0.8000	0.8000	0.7000	1.0000	21.651	3	6	30
3.1	0.5333	0.5333	0.5333	0.4000	0.7000	28.641	3	14	30
6.3	0.3000	0.3000	0.3000	0.2000	0.4000	33.333	3	21	30
12.5	0.2000	0.2000	0.2000	0.1000	0.3000	50.000	3	24	30
25	0.1333	0.1333	0.1333	0.0000	0.3000	114.564	3	26	30

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates normal distribution (p > 0.05)	0.90042	0.897	0.5699	-0.7752
Equality of variance cannot be confirmed				

Maximum Likelihood-Probit

Parameter	Value	SE	95% Fiducial Limits		Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	1.64022	0.28711	1.07748	2.20295	0	1.87675	7.81473	0.59838	0.59612	0.60968	3
Intercept	4.02223	0.23878	3.55423	4.49024							
TSCR											

Point	Probits	ppm	95% Fiducial Limits	
EC01	2.674	0.15059	0.02233	0.39071
EC05	3.355	0.39201	0.0945	0.80738
EC10	3.718	0.65281	0.20301	1.19429
EC15	3.964	0.92094	0.33892	1.56059
EC20	4.158	1.21061	0.50775	1.93628
EC25	4.326	1.53073	0.71588	2.33754
EC40	4.747	2.76478	1.65929	3.8522
EC50	**5.000**	**3.94567**	**2.65905**	**5.38317**
EC60	5.253	5.63093	4.06582	7.88404
EC75	5.674	10.1705	7.32836	16.7079
EC80	5.842	12.8599	9.00287	23.1487
EC85	6.036	16.9048	11.32	34.2214
EC90	6.282	23.848	14.9468	56.5399
EC95	6.645	39.7144	22.3024	120.405
EC99	7.326	103.381	46.427	505.908



APPENDIX H

3/13/2012 3:47 PM
Environmental Enterprises USA, Inc.

Mysid, *Mysidopsis bahia*
Acute Static 48–Hour No. 2 Fuel Oil Definitive Test (DFT)
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Test Concentrations, ppm No.2 Fuel Oil (Oil)

Mysidopsis bahia	Total Volume / Replicate / Concentration, ml	Color Code	ml SSOL / Replicate	ml DH_2O / Replicate
12.0 ppm	1000.00	Black	12.00	988.00
6.0 ppm	"	Brown	6.00	994.00
3.0 ppm	"	Yellow	3.00	997.00
1.5 ppm	"	Green	1.50	998.50
0.8 ppm	"	Blue	0.80	999.20
0 ppm LPC	"	White	0.00	1000.00
	Total Volume (ml) of SSOL needed per day =		23.3 ml / Replicate X 3 = 69.9 ml	

550 ml Stock Solution (SSOL) @ 1000 ppm: 0.55 ml No.2 Fuel Oil + 549.45 ml DH_2O

Data Pages & Calculations by: Veronica M. Hernandez QA/QC Check by: Jennifer Griffith

M. bahia = 3 Rep x 3000 ml total volume / treatment

DH_2O = Dilution Water = **20 ppt Synthetic Seawater**

	LPC	Meter ID#
Date	03/13	//////
DH_2O Lot #	25R-072-12	//////
Alkalinity	112	//////
Salinity	19.8	A416
pH	8.6	A93
Temp.	23.8	A416
	VH	//////

Prep. Date	03/13
Blender ID#	A44
Initial	JG

LPC: Laboratory Performance Control, 20 ppt synthetic seawater
Alkalinity: mg/l as $CaCO_3$ Salinity: ppt pH: su Temp: °C

Comments: *Artemia* Lot # 030211-2; Feed *M. bahia* once daily.

Mysid, *Mysidopsis bahia*
Acute Static 48–Hour No. 2 Fuel Oil Definitive Test (DFT)
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Survival Data

Time	Treatment, ppm Oil																		Date & Initials
	REP	LPC 0 ppm White	REP	0.8 ppm Blue	REP	1.5 ppm Green	REP	3.0 ppm Yellow	REP	6.0 ppm Brown	REP	12.0 ppm Black	REP						
0 HR	1	10	4	10	7	10	10	10	13	10	16	10	19						03/13/12
1855	2	10	5	10	8	10	11	10	14	10	17	10	20						TB
	3	10	6	10	9	10	12	10	15	10	18	10	21						
24 HR	1	10	4	10	7	10	10	10	13	9	16	0	19						03/14/12
1646	2	10	5	10	8	10	11	10	14	7	17	0	20						JG
	3	10	6	10	9	10	12	10	15	6	18	0	21						
48 HR	1	10	4	10	7	10	10	10	13	8	16	0	19						03/15/12
1741	2	10	5	10	8	10	11	10	14	6	17	0	20						VL
	3	10	6	10	9	10	12	10	15	4	18	0	21						
% Survival	100		100		100		100		60		0								

Counted by: Terry Brock QC/QA by: Jennifer Griffith

Loaded by: Terry Brock

Test Organisms Age: 5 days old Test Organisms Source: CE

Test Organism Lot #: MB-151-12

Data Entry by: Veronica McNew

Double Data Entry by: Veronica McNew and/or

QC/QA by: Jennifer Griffith

M. bahia Water Quality Data

LPC & All Treatments: Initial & Final Temp.: 23.5 to 26.4°C; Initial & Final DO: 4.0 to ≤ 7.4 mg/l
LPC: Initial Salinity: 18.5 to 21.4 ppt

0 to 24 HR		Treatment, ppm OIL							
03/13/12 – 03/14/12		LPC	///	0.8 ppm	1.5 ppm	3.0 ppm	6.0 ppm	12.0 ppm	Meter ID#
DO	I	7.4	///	7.7	7.8	7.8	7.8	7.8	57
	F	7.0	///	6.9	7.0	7.0	7.1	7.0	57
Temp	I	23.8	///	24.8	24.7	24.6	24.5	24.6	A46
	F	25.2	///	25.0	25.0	25.0	25.0	25.2	A46
Salinity	I	19.8	///	19.8	19.8	19.8	19.8	19.8	A46
	F	19.9	///	19.8	19.8	19.9	19.8	19.8	A46
pH	F	7.9	///	8.0	8.0	8.0	8.0	8.0	A93
Initials		Initial: JG				Final: JMR JD			
Times		Initial Time: 1857				Final Time: 0925			

48 HR		Treatment, ppm OIL							
03/15/12		LPC	///	0.8 ppm	1.5 ppm	3.0 ppm	6.0 ppm	12.0 ppm	Meter ID#
DO	F	6.9	///	6.9	7.0	7.0	7.0		57
Temp	F	25.1	///	24.7	24.7	24.7	24.7		A46
Salinity	F	19.9	///	19.9	19.9	20.0	19.9		A46
pH	F	7.9	///	7.9	7.9	7.9	8.0		A93
Initials		Final: SD MR							
Times		Final Time: 0918							

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

Comments:

Data Pages

- ☑ Calculations on mixing page are correct. (sign mixing page)
- ☑ Dates, dilutions, test method, # of replicates, replicate volume, product, acceptance limits, data analysis endpoint, and test organisms are correct throughout data pages.
- ☑ Format correct. (spaces for all entries, page numeration, no split pages, etc.)

JG Initials 03-13-12 Date

Chain-of-Custody

- ☑ Product on COC matches sample bottle/container.
- ☑ Product on COC matches test data pages.
- ☑ Lab # on COC matches sample bottle/container.
- ☑ Lab # on COC matches test data pages.
- ☑ Sample volume/amount is sufficient for test duration. (Sample volume in container(s) checked against sample volume on mixing page)

Sample volume/amount available: 40 (ml) or g

Sample volume/amount needed: 0.55 (ml) or g

(Sample volume/amount insufficient if sample volume/amount available < sample volume/amount needed)

VL Initials 03/13/12 Date

Labels

- ☑ Lab # on labels matches test data pages.
- ☑ Test organism on data pages match labels (i.e. *Menidia beryllina* = MN, *Mysidopsis bahia* = MB)
- ☑ Test Concentrations on labels match Test Concentrations on test data pages.

VL Initials 03/13/12 Date

Raw Data QC/QA'd by: Veronica McMeen 03/19/12

Acute Toxicity Test-48 Hr Survival

Start Date:	3/13/2012	Test ID:	mb00812DO	Sample ID:	NCP-National Contingency Plan
End Date:	3/15/2012	Lab ID:	EE-Environmental Enterprise	Sample Type:	2FO-No. 2 Fuel Oil
Sample Date:		Protocol:	EPAM 02-EPA Marine	Test Species:	MY-Mysidopsis bahia
Comments:					

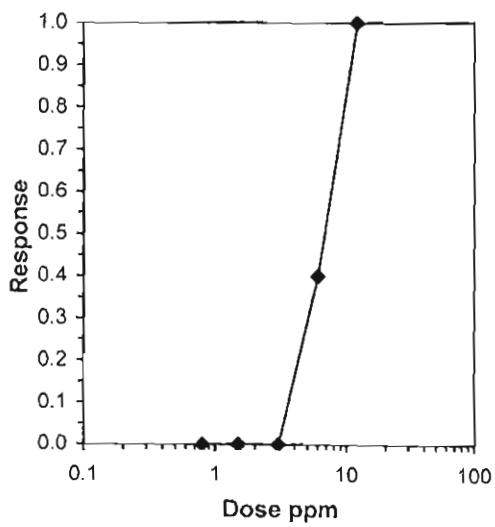
Conc-ppm	1	2	3
PC-LP Control	1.0000	1.0000	1.0000
0.8	1.0000	1.0000	1.0000
1.5	1.0000	1.0000	1.0000
3	1.0000	1.0000	1.0000
6	0.8000	0.6000	0.4000
12	0.0000	0.0000	0.0000

			Transform: Arcsin Square Root					Number	Total
Conc-ppm	Mean	N-Mean	Mean	Min	Max	CV%	N	Resp	Number
PC-LP Control	1.0000	1.0000	1.4120	1.4120	1.4120	0.000	3	0	30
0.8	1.0000	1.0000	1.4120	1.4120	1.4120	0.000	3	0	30
1.5	1.0000	1.0000	1.4120	1.4120	1.4120	0.000	3	0	30
3	1.0000	1.0000	1.4120	1.4120	1.4120	0.000	3	0	30
6	0.6000	0.6000	0.8926	0.6847	1.1071	23.670	3	12	30
12	0.0000	0.0000	0.1588	0.1588	0.1588	0.000	3	30	30

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates non-normal distribution (p <= 0.05)	0.5407	0.881	0.14228	7
Equality of variance cannot be confirmed				

Trimmed Spearman-Karber

Trim Level	EC50	95% CL	
0.0%	6.4306	5.6807	7.2795
5.0%	6.4752	5.6384	7.4361
10.0%	6.5195	5.5684	7.6330
20.0%	6.6063	5.2964	8.2403
Auto-0.0%	**6.4306**	**5.6807**	**7.2795**



APPENDIX I

Mysid, *Mysidopsis bahia*

Acute Static 48–Hour 10:1 No. 2 Fuel Oil / Product Definitive Test (DFT)

3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Natural Solutions Group Corporation – $VirO_2Syl$

Contact: Marcos Gonzalez

Test Concentrations, ppm 10:1 No. 2 Fuel Oil / $VirO_2Syl$

Mysidopsis bahia	Total Volume / Replicate / Concentration, ml	Color Code	ml SSOL / Replicate	ml DH_2O / Replicate
10.0 ppm	1000.00	Black	10.00	990.00
5.0 ppm	"	Brown	5.00	995.00
2.5 ppm	"	Yellow	2.50	997.50
1.3 ppm	"	Green	1.30	998.70
0.6 ppm	"	Blue	0.60	999.40
0 ppm LPC	"	White	0.00	1000.00
		Total Volume (ml) of SSOL needed per day =	19.4 ml / Replicate X 3 = 58.2 ml	

550 ml Stock Solution (SSOL) @ 1000 ppm: 0.50 ml No.2 Fuel Oil + 0.05 ml $VirO_2Syl$ + 549.45 ml DH_2O

Data Pages & Calculations by: Veronica M. Nu QA/QC Check by: Jennifer Griffith

M. bahia = 3 Rep x 3000 ml total volume / treatment

DH_2O = Dilution Water = **20 ppt Synthetic Seawater**

	LPC	Meter ID#
Date	03/13	//////
DH_2O Lot #	25R-072-12	//////
Alkalinity	112	//////
Salinity	19.8	A46
pH	8.0	A93
Temp.	23.8	A46
	VN	//////

Prep Date	03/13
Blender ID#	444
Initial	JG

LPC: Laboratory Performance Control, 20 ppt synthetic seawater
Alkalinity: mg/l as $CaCO_3$ Salinity: ppt pH: su Temp: °C

Comments: *Artemia* Lot # 030211-2; Feed *M. bahia* once daily.

Mysid, *Mysidopsis bahia*

Acute Static 48–Hour 10:1 No. 2 Fuel Oil / Product Definitive Test (DFT)

3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Survival Data

Treatment, ppm 10:1 No. 2 Fuel Oil / VirO$_2$Syl																								
Time	REP	LPC 0 ppm White	REP	0.6ppm Blue	REP	1.3 ppm Green	REP	2.5 ppm Yellow	REP	5.0 ppm Brown	REP	10.0 ppm Black	REP		Date & Initials									
0 HR	1	10	4	10	7	10	10	10	13	10	16	10	19		03/13/12									
1810	2	10	5	10	8	10	11	10	14	10	17	10	20		TB									
	3	10	6	10	9	10	12	10	15	10	18	10	21											
24 HR	1	10	4	10	7	10	10	10	13	10	16	3	19		03/14/12									
1610	2	10	5	10	8	10	11	10	14	10	17	5	20		JC									
	3	10	6	10	9	10	12	10	15	10	18	7	21											
48 HR	1	10	4	10	7	10	10	10	13	10	16	3	19		03/15/12									
1734	2	10	5	10	8	10	11	10	14	7	17	2	20		VL									
	3	10	6	10	9	10	12	10	15	8	18	2	21											
% Survival		100		100		100		100		83.3		23.3												

Counted by: Terry Branch QC/QA by: Jennifer Griffith

Loaded by: Terry Branch

Test Organisms Age: 5 days old Test Organisms Source: EE

Test Organism Lot #: MB-151-12

Data Entry by: Veronica McMeans

Double Data Entry by: Veronica McMeans and/or

QC/QA by: Jennifer Griffith

M. bahia Water Quality Data

LPC & All Treatments: Initial & Final Temp.: 23.5 to 26.4°C; Initial & Final DO: 4.0 to ≤ 7.4 mg/l
LPC: Initial Salinity: 18.5 to 21.4 ppt

0 to 24 HR	Treatment, ppm 10:1 No. 2 Fuel Oil / VirO$_2$Syl							
03/13/12 – 03/14/12	LPC	///	0.6 ppm	1.3 ppm	2.5 ppm	5.0 ppm	10.0 ppm	Meter ID#
DO I	7.4	///	7.5	7.5	7.6	7.6	7.4	57
F	7.6	///	6.9	7.6	7.0	7.6	6.9	57
Temp I	23.8	///	24.8	24.6	24.6	24.6	24.8	A46
F	25.1	///	25.0	25.9	25.9	25.8	25.9	A46
Salinity I	19.8	///	19.8	19.8	19.8	19.8	19.8	A46
F	19.9	///	19.9	19.9	19.8	19.8	19.8	A46
pH F	7.9	///	8.0	8.0	8.0	8.0	8.0	A93
Initials	Initial: JG				Final: MR SD			
Times	Initial Time: 1852				Final Time: 0919			

48 HR	Treatment, ppm 10:1 No. 2 Fuel Oil / VirO$_2$Syl							
03/15/12	LPC	///	0.6 ppm	1.3 ppm	2.5 ppm	5.0 ppm	10.0 ppm	Meter ID#
DO F	6.9	///	6.9	6.8	6.5	6.8	6.8	57
Temp F	25.1	///	25.7	25.8	25.8	25.7	25.7	A46
Salinity F	19.9	///	20.0	19.9	19.9	19.9	19.9	A46
pH F	7.9	///	7.9	8.0	8.0	8.0	8.0	A93
Initials	Final: SD MR							
Times	Final Time: 0914							

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

Comments: _____

Data Pages

- ☑ Calculations on mixing page are correct. (sign mixing page)
- ☑ Dates, dilutions, test method, # of replicates, replicate volume, product, acceptance limits, data analysis endpoint, and test organisms are correct throughout data pages.
- ☑ Format correct. (spaces for all entries, page numeration, no split pages, etc.)

JG Initials 03-13-12 Date

Chain-of-Custody

- ☑ Product on COC matches sample bottle/container.
- ☑ Product on COC matches test data pages.
- ☑ Lab # on COC matches sample bottle/container.
- ☑ Lab # on COC matches test data pages.
- ☑ Sample volume/amount is sufficient for test duration. (Sample volume in container(s) checked against sample volume on mixing page)

Sample volume/amount available: 40 (0.1) 8000 (PRD) (ml) or g

Sample volume/amount needed: 0.50 0.05 (ml) or g

(Sample volume/amount insufficient if sample volume/amount available < sample volume/amount needed)

VL Initials 03/13/12 Date

Labels

- ☑ Lab # on labels matches test data pages.
- ☑ Test organism on data pages match labels (i.e. *Menidia beryllina* = MN, *Mysidopsis bahia* = MB)
- ☑ Test Concentrations on labels match Test Concentrations on test data pages.

VL Initials 03/13/12 Date

Raw Data QC/QA'd by: Veronica McNew 03/19/12

Acute Toxicity Test-48 Hr Survival

Start Date:	3/13/2012	Test ID:	mb00612DPO	Sample ID:	NCP-National Contingency Plan
End Date:	3/15/2012	Lab ID:	EE-Environmental Enterprise	Sample Type:	PIO-Product/No. 2 Fuel Oil Mixture
Sample Date:		Protocol:	EPAM 02-EPA Marine	Test Species:	MY-Mysidopsis bahia
Comments:					

Conc-ppm	1	2	3
PC-LP Control	1.0000	1.0000	1.0000
0.6	1.0000	1.0000	1.0000
1.3	1.0000	1.0000	1.0000
2.5	1.0000	1.0000	1.0000
5	1.0000	0.7000	0.8000
10	0.3000	0.2000	0.2000

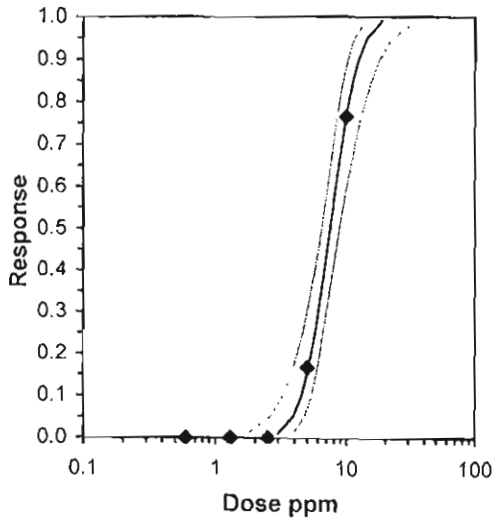
			Transform: Untransformed					Number	Total
Conc-ppm	Mean	N-Mean	Mean	Min	Max	CV%	N	Resp	Number
PC-LP Control	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	3	0	30
0.6	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	3	0	30
1.3	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	3	0	30
2.5	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	3	0	30
5	0.8333	0.8333	0.8333	0.7000	1.0000	18.330	3	5	30
10	0.2333	0.2333	0.2333	0.2000	0.3000	24.744	3	23	30

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates non-normal distribution (p <= 0.05)	0.70842	0.897	0.92057	5.85039
Equality of variance cannot be confirmed				

Maximum Likelihood-Probit

Parameter	Value	SE	95% Fiducial Limits		Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	5.82155	1.10633	3.65313	7.98997	0	0.11353	7.81473	0.99017	0.87203	0.17178	3
Intercept	-0.0766	0.95781	-1.9539	1.80076							
TSCR											

Point	Probits	ppm	95% Fiducial Limits	
EC01	2.674	2.96769	1.71941	3.87203
EC05	3.355	3.88582	2.61057	4.769
EC10	3.718	4.4863	3.24728	5.35257
EC15	3.964	4.94304	3.7505	5.80455
EC20	4.158	5.33898	4.19373	6.20822
EC25	4.326	5.70384	4.60281	6.59496
EC40	4.747	6.73766	5.72353	7.80841
EC50	**5.000**	**7.4478**	**6.42853**	**8.77352**
EC60	5.253	8.23279	7.13688	9.97323
EC75	5.674	9.72498	8.32685	12.5851
EC80	5.842	10.3896	8.81248	13.8645
EC85	6.036	11.2218	9.3948	15.5534
EC90	6.282	12.3642	10.1591	18.015
EC95	6.645	14.2749	11.3721	22.4683
EC99	7.326	18.6912	13.9716	34.1984



APPENDIX J

Mysid, *Mysidopsis bahia*
Acute Static 48–Hour Sodium Dodecyl Sulfate (SDS) Definitive Test (DFT)
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Test Concentrations, ppm SDS (Standard Reference Toxicant)

Mysidopsis bahia	Total Volume / Replicate / Concentration, ml	Color Code	ml SSOL / Replicate	ml DH_2O / Replicate
14.0 ppm	1000.00	Black	7.00	993.00
7.0 ppm	"	Brown	3.50	996.50
3.5 ppm	"	Yellow	1.75	998.25
1.8 ppm	"	Green	0.90	999.10
0.9 ppm	"	Blue	0.45	999.55
0 ppm LPC	"	White	0.00	1000.00
Total Volume (ml) of SSOL needed per day = 13.60 ml / Replicate X 3 = 40.8 ml				

500 ml Stock Solution (SSOL) @ 2,000 ppm = 1.00 g SDS (2.7 ml SDS) + 497.3 ml DH_2O
Specific gravity of SDS = 0.37 g/ml

Weight of SDS = 1.0000 g Balance ID#: B2

Date & Time: 03.13.12 1610 Initials: JG

Data Pages & Calculations by: Veronica McNew QA/QC Check by: Jennifer Griffith

M. bahia = 3 Rep x 3000 ml total volume / treatment
DH_2O = Dilution Water = **20 ppt Synthetic Seawater**

	LPC	Meter ID#
Date	03/13	//////
DH_2O Lot #	25R-0712-12	//////
Alkalinity	112.	//////
Salinity	19.8	A446
pH	8.0	A493
Temp.	23.8	A416
	VM	//////

Prep Date	03/13
Blender ID#	444
Initial	JG

LPC: Laboratory Performance Control, 20 ppt synthetic seawater
Alkalinity: mg/l as $CaCO_3$ Salinity: ppt pH: su Temp: °C

Comments: *Artemia* Lot # Lot # 030211-2; Feed *M. bahia* once daily.

Mysid, *Mysidopsis bahia*
Acute Static 48–Hour Sodium Dodecyl Sulfate (SDS) Definitive Test (DFT)
3.0 Revised Dispersant Toxicity Test: FR / Vol. 59, No. 178 / 47461 - 47464

Survival Data

Time	Treatment, ppm SDS																		Date & Initials
	REP	LPC 0 ppm White	REP	0.9 ppm Blue	REP	1.8 ppm Green	REP	3.5 ppm Yellow	REP	7.0 ppm Brown	REP	14.0 ppm Black	REP						
0 HR	1	10	4	10	7	10	10	10	13	10	16	10	19						03/13/12
1737	2	10	5	10	8	10	11	10	14	10	17	10	20						TB
	3	10	6	10	9	10	12	10	15	10	18	10	21						
24 HR	1	10	4	10	7	10	10	10	13	9	16	1	19						03/14/12
1623	2	10	5	10	8	10	11	10	14	10	17	2	20						JG
	3	10	6	10	9	10	12	10	15	9	18	3	21						
48 HR	1	10	4	10	7	10	10	10	13	7	16	0	19						03/15/12
1723	2	10	5	10	8	10	11	10	14	7	17	1	20						VL
	3	10	6	10	9	10	12	10	15	6	18	3	21						
% Survival	100		100		100		100		66.7		13.3								

Counted by: Jenny Bunch QC/QA by: Jennifer Griffith

Loaded by: Jenny Bunch

Test Organisms Age: 5 days old Test Organisms Source: EE

Test Organism Lot #: MB-151-12

Data Entry by: Veronica McNew

Double Data Entry by: Veronica McNew and/or

QC/QA by: Jennifer Griffith

Sodium Dodecyl Sulfate, DFT

D-009-12 (for Poduct Test D-0064-12)

LC50

M. bahia Water Quality Data

LPC & All Treatments: Initial & Final Temp.: 23.5 to 26.4°C; Initial & Final DO: 4.0 to ≤ 7.4 mg/l
LPC: Initial Salinity: 18.5 to 21.4 ppt

0 to 24 HR		Treatment, ppm SDS							
03/13/12 – 03/14/12		LPC	///	0.9 ppm	1.8 ppm	3.5 ppm	7.0 ppm	14.0 ppm	Meter ID#
DO	I	7.4	///	7.2	7.4	7.4	7.4	7.4	57
	F	7.0	///	7.6	6.9	6.9	6.9	6.9	57
Temp	I	23.8	///	24.1	24.2	24.3	24.4	24.5	A46
	F	25.1	///	25.0	25.0	25.0	24.9	25.0	A46
Salinity	I	19.8	///	19.8	19.8	19.8	19.8	19.8	A46
	F	19.9	///	19.9	19.9	19.9	19.9	19.9	A46
pH	F	7.9	///	8.0	8.0	8.0	8.0	8.0	A93
Initials		Initial: JG				Final: MR JD			
Times		Initial Time: 1849				Final Time: 0917			

48 HR		Treatment, ppm SDS							
03/15/12		LPC	///	0.9 ppm	1.8 ppm	3.5 ppm	7.0 ppm	14.0 ppm	Meter ID#
DO	F	4.9	///	7.0	6.8	6.4	5.9	6.1	57
Temp	F	25.1	///	24.9	24.8	24.8	24.7	24.6	A46
Salinity	F	19.9	///	19.9	20.0	19.9	19.9	20.0	A46
pH	F	7.9	///	7.9	7.9	7.9	7.8	7.8	A93
Initials		Final: SD MR							
Times		Final Time: 0911							

DO, Dissolved Oxygen: mg/l pH: SU Salinity: ppt Temp: °C

Comments:

Data Pages

- ☑ Calculations on mixing page are correct. (sign mixing page)
- ☑ Dates, dilutions, test method, # of replicates, replicate volume, product, acceptance limits, data analysis endpoint, and test organisms are correct throughout data pages.
- ☑ Format correct. (spaces for all entries, page numeration, no split pages, etc.)

JG Initials 03-13-12 Date

Chain-of-Custody

- ☑ Product on COC matches sample bottle/container.
- ☑ Product on COC matches test data pages.
- ☑ Lab # on COC matches sample bottle/container.
- ☑ Lab # on COC matches test data pages.
- ☑ Sample volume/amount is sufficient for test duration. (Sample volume in container(s) checked against sample volume on mixing page)

95 wrong data. 03/13/12 i

Sample volume/amount available: ~~8000~~ (ml) or (g)

Sample volume/amount needed: 1 ml or (g)

(Sample volume/amount insufficient if sample volume/amount available < sample volume/amount needed)

VL Initials 03/13/12 Date

Labels

- ☑ Lab # on labels matches test data pages.
- ☑ Test organism on data pages match labels (i.e. *Menidia beryllina* = MN, *Mysidopsis bahia* = MB)
- ☑ Test Concentrations on labels match Test Concentrations on test data pages.

VL Initials 03/13/12 Date

Raw Data QC/QA'd by: Veronica Mc Mew 03/19/12

Acute Toxicity Test-48 Hr Survival

Start Date:	3/13/2012	Test ID:	mb00912DS	Sample ID:	REF-Ref Toxicant
End Date:	3/15/2012	Lab ID:	EE-Environmental Enterprise	Sample Type:	SDS-Sodium dodecyl sulfate
Sample Date:		Protocol:	EPAM 02-EPA Marine	Test Species:	MY-Mysidopsis bahia
Comments:					

Conc-ppm	1	2	3
PC-LP Control	1.0000	1.0000	1.0000
0.9	1.0000	1.0000	1.0000
1.8	1.0000	1.0000	1.0000
3.5	1.0000	1.0000	1.0000
7	0.7000	0.7000	0.6000
14	0.0000	0.1000	0.3000

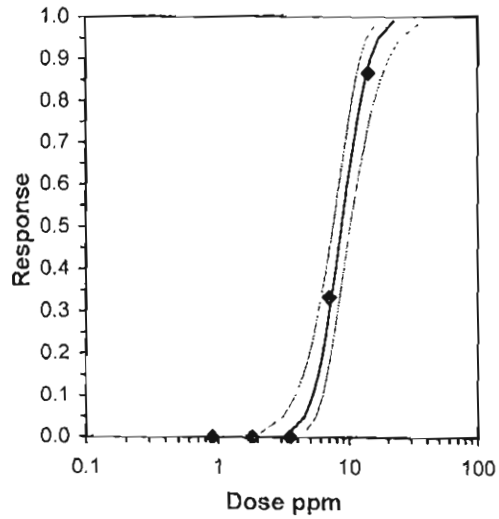
			Transform: Untransformed					Number	Total
Conc-ppm	Mean	N-Mean	Mean	Min	Max	CV%	N	Resp	Number
PC-LP Control	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	3	0	30
0.9	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	3	0	30
1.8	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	3	0	30
3.5	1.0000	1.0000	1.0000	1.0000	1.0000	0.000	3	0	30
7	0.6667	0.6667	0.6667	0.6000	0.7000	8.660	3	10	30
14	0.1333	0.1333	0.1333	0.0000	0.3000	114.564	3	26	30

Auxiliary Tests	Statistic	Critical	Skew	Kurt
Shapiro-Wilk's Test indicates non-normal distribution (p <= 0.05)	0.72126	0.897	0.7532	5.85039
Equality of variance cannot be confirmed				

Maximum Likelihood-Probit

Parameter	Value	SE	95% Fiducial Limits		Control	Chi-Sq	Critical	P-value	Mu	Sigma	Iter
Slope	5.74058	0.99036	3.79947	7.68168	0	0.6422	7.81473	0.88671	0.93866	0.1742	4
Intercept	-0.3885	0.93875	-2.2284	1.45148							
TSCR											

Point	Probits	ppm	95% Fiducial Limits	
EC01	2.674	3.41521	2.07576	4.438
EC05	3.355	4.48883	3.10129	5.50688
EC10	3.718	5.19302	3.82659	6.20225
EC15	3.964	5.72953	4.39762	6.73855
EC20	4.158	6.1952	4.90035	7.21426
EC25	4.326	6.62474	5.36538	7.6659
EC40	4.747	7.84389	6.65543	9.05012
EC50	**5.000**	**8.68289**	**7.48755**	**10.1193**
EC60	5.253	9.61163	8.34144	11.4264
EC75	5.674	11.3804	9.80189	14.2399
EC80	5.842	12.1695	10.4025	15.6107
EC85	6.036	13.1586	11.1246	17.4146
EC90	6.282	14.5181	12.0744	20.0334
EC95	6.645	16.7956	13.5861	24.7422
EC99	7.326	22.0755	16.8427	37.0002



APPENDIX K

ENVIRONMENTAL ENTERPRISES USA, INC.
58485 Pearl Acres Rd., Suite D
Slidell, Louisiana 70461
(985) 646-2787

Fed Ex # 258851425U

Kit No. BOX

CHAIN - OF - CUSTODY RECORD

Client: Natural Solutions Group Corp. Contact Person: _____

Address: 5120 NW 165 street
Suite 103
Miami Gardens, FL 33014

Phone#: _____

P.O. # _____

FAX #: _____

Project: _____

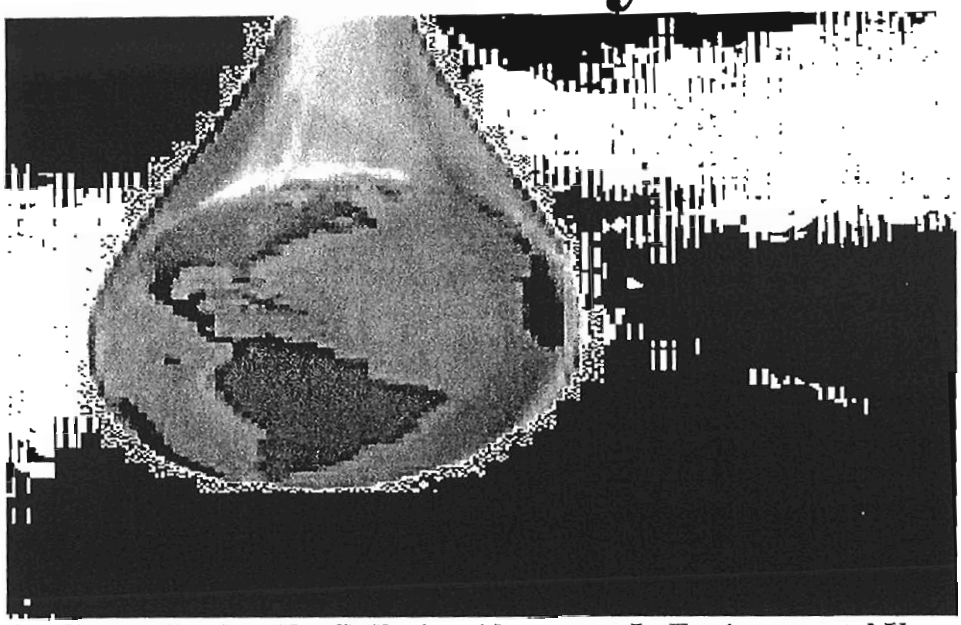
Special Handling Request

(X) RUSH
( ) VERBAL
( ) OTHER

Lab Sample Description	Date Collected	Time Collected	No. of Containers	Analysis Request	S/R No.	Lab No.
VirO2 Syl	N/A	N/A	1	NCP Toxicity	D-94107-12	D-006-12

Collected By:	Date	Time	Relinquished By:	Date	Time
Received By: Lana Powe EE	Date 3/6/12	Time 1125	Relinquished By: Lana Powe EE	Date 3/6/12	Time 1140
Received By: Veronica McNew	Date 03/06/12	Time 1143	Relinquished By:	Date	Time
Received By:	Date	Time	Relinquished By:	Date	Time
Received By:	Date	Time	Relinquished By:	Date	Time

VirO2Syl



Oxidizing Catalyst For Pollution Abatement In Environmental Use

MATERIAL SAFETY DATA SHEET

Hydrogen Peroxide (20 to 40%)
MSDS Ref. No.: 7722-84-1-3 Product VIRO2SYL – 72372-4- 87564
Date Approved: 04/05/2005
Revision No.: 9

This document has been prepared to meet the requirements of the U.S. OSHA Hazard Communication Standard, 29 CFR 1910.1200; the Canada's Workplace Hazardous Materials Information System (WHMIS) and the EC Directive, 2001/58/EC.

MANUFACTURER

Natural Solutions Group Corp.
Distributed by NSGC Global
5120 NW 165 Street, Suite 103, Miami Gardens, FL 33014

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1. PRODUCT AND COMPANY IDENTIFICATION

PRODUCT NAME: Hydrogen Peroxide (20 to 40%)

ALTERNATE PRODUCT NAME(S): VirO2Syl Standard 27.5 & 35%,

GENERAL USE: VIRO2SYL - Hydrogen Peroxide (20 to 40%) (7722-84-1-3)
Date: 04/05/2005
VirO2Syl –72372-4-87564

Standard 27.5 and 35% - most suitable grade for industrial bleaching, processing, pollution abatement and general oxidation reactions.

Semiconductor Reg, Seg, RGS, RGS 2, RGS 3, 31% - conform to ACS and Semi Specs. for wafer etching and cleaning, and applications requiring low residues.

Meets the Food Chemical Codex requirements for aseptic packaging and other food related applications. Certified by NSF to meet NSF/ANSI Standard 60 requirements for drinking water treatment.

MANUFACTURER
Natural Solutions Group Corp.
5120 NW 165 Street, Suite 103
Miami Gardens, FL 33014

2. HAZARDS IDENTIFICATION

EMERGENCY OVERVIEW:

- Clear, colorless, odorless liquid
- Oxidizer.
- Contact with combustibles may cause fire.
- Decomposes yielding oxygen that supports combustion of organic matters and can cause overpressure if confined.
- Corrosive to eyes, nose, throat, lungs and gastrointestinal tract.

POTENTIAL HEALTH EFFECTS: Corrosive to eyes, nose, throat and lungs. May cause irreversible tissue damage to the eyes including blindness. May cause skin irritation.

3. COMPOSITION / INFORMATION ON INGREDIENTS

Chemical Name CAS# Wt.% EC No. EC Class
Hydrogen Peroxide 7722-84-1 20 - 40 231-765-0 Xn, R22-37/38-41
Water 7732-18-5 60 - 80 231-791-2 Not classified
Hydrogen Peroxide (20 to 40%) (7722-84-1-3) Date: 04/05/2005

4. FIRST AID MEASURES

EYES: Immediately flush with water for at least 15 minutes, lifting the upper and lower eyelids intermittently. See a medical doctor or ophthalmologist immediately.

SKIN: Wash with plenty of soap and water. Get medical attention if irritation occurs and persists.

INGESTION: Rinse mouth with water. Dilute by giving 1 or 2 glasses of water. Do not induce vomiting. Never give anything by mouth to an unconscious person. See a medical doctor immediately.

INHALATION: Remove to fresh air. If breathing difficulty or discomfort occurs and persists, contact a medical doctor.

NOTES TO MEDICAL DOCTOR: Hydrogen peroxide at these concentrations is a strong oxidant. Direct contact with the eye is likely to cause corneal damage especially if not washed immediately. Careful ophthalmologic evaluation is recommended and the possibility of local corticosteroid therapy should be considered. Because of the likelihood of corrosive effects on the gastrointestinal tract after ingestion, and the unlikelihood of systemic effects, attempts at evacuating the stomach via emesis induction or gastric lavage should be avoided. There is a remote possibility, however, that a nasogastric or orogastric tube may be required for the reduction of severe distension due to gas formation.

5. FIRE FIGHTING MEASURES

EXTINGUISHING MEDIA: Flood with water.

FIRE / EXPLOSION HAZARDS: Product is non-combustible. On decomposition releases oxygen which may intensify fire.

FIRE FIGHTING PROCEDURES: Any tank or container surrounded by fire should be flooded with water for cooling. Wear full protective clothing and self-contained breathing apparatus.

FLAMMABLE LIMITS: Non-combustible

SENSITIVITY TO IMPACT: No data available

SENSITIVITY TO STATIC DISCHARGE: No data available

6. ACCIDENTAL RELEASE MEASURES

RELEASE NOTES: Dilute with a large volume of water and hold in a pond or diked area until hydrogen peroxide decomposes. Hydrogen peroxide may be decomposed by adding sodium metabisulfite or sodium sulfite after diluting to about 5%. Dispose according to methods outlined for waste disposal.
Hydrogen Peroxide (20 to 40%) (7722-84-1-3) Date: 04/05/2005
Combustible materials exposed to hydrogen peroxide should be immediately submerged in or rinsed with large amounts of water to ensure that all hydrogen peroxide is removed. Residual hydrogen peroxide that is allowed to dry (upon evaporation hydrogen peroxide can concentrate) on organic materials such as paper, fabrics, cotton, leather, wood or other combustibles can cause the material to ignite and result in a fire.

7. HANDLING AND STORAGE

HANDLING: Wear chemical splash-type monogoggles and full-face shield, impervious clothing, such as rubber, PVC, etc., and rubber or neoprene gloves and shoes. Avoid cotton, wool and leather. Avoid excessive heat and contamination. Contamination may cause decomposition and generation of oxygen gas which could result in high pressures and possible container rupture. Hydrogen peroxide should be stored only in vented containers and transferred only in a prescribed manner (see Technical Bulletins).
Never return unused hydrogen peroxide to original container, empty drums should be triple rinsed with water before discarding. Utensils used for handling hydrogen peroxide should only be made of glass, stainless steel, aluminum or plastic.

STORAGE: Store drums in cool areas out of direct sunlight and away from combustibles. For bulk storage refer to Technical Bulletins.

COMMENTS: VENTILATION: Provide mechanical general and/or local exhaust ventilation to prevent release of vapor or mist into the work environment.

8. EXPOSURE CONTROLS / PERSONAL PROTECTION

EXPOSURE LIMITS

Chemical Name ACGIH OSHA Supplier
Hydrogen Peroxide 1 ppm (TWA) 1 ppm (PEL)

ENGINEERING CONTROLS: Ventilation should be provided to minimize the release of hydrogen peroxide vapors and mists into the work environment. Spills should be minimized or confined immediately to prevent release into the work area. Remove contaminated clothing immediately and wash before reuse.

PERSONAL PROTECTIVE EQUIPMENT

EYES AND FACE: Use chemical splash-type monogoggles and a full-face shield made of polycarbonate, acetate, polycarbonate/acetate, PETG or thermoplastic.

RESPIRATORY: If concentrations in excess of 10 ppm are expected, use NIOSH/DHHS approved self-contained breathing apparatus (SCBA), or other approved atmospheric-supplied respirator (ASR) equipment (e.g., a full-face airline respirator (ALR)). DO NOT use any form of air-purifying respirator (APR) or filtering facepiece (AKA dust mask), especially those containing oxidizable sorbants such as activated carbon.
Hydrogen Peroxide (20 to 40%) (7722-84-1-3) Date: 04/05/2005

PROTECTIVE CLOTHING: For body protection wear impervious clothing such as an approved splash protective suit made of SBR Rubber, PVC (PVC Outershell w/Polyester Substrate), Gore-Tex (Polyester trilaminate w/Gore-Tex), or a specialized HAZMAT Splash or Protective Suite (Level A, B, or C). For foot protection, wear approved boots made of NBR, PVC, Polyurethane, or neoprene. Overboots made of Latex or PVC, as well as firefighter boots or specialized HAZMAT boots are also permitted. DO NOT wear any form of boot or overboots made of nylon or nylon blends. DO NOT use cotton, wool or leather, as these materials react RAPIDLY with higher concentrations of hydrogen peroxide. Completely submerge hydrogen peroxide contaminated clothing or other materials in water prior to drying. Residual hydrogen peroxide, if allowed to dry on materials such as paper, fabrics, cotton, leather, wood or other combustibles can cause the material to ignite and result in a fire.

GLOVES: For hand protection, wear approved gloves made of nitrile, PVC, or neoprene. DO NOT use cotton, wool or leather for these materials react RAPIDLY with higher concentrations of hydrogen peroxide. Thoroughly rinse the outside of gloves with water prior to removal. Inspect regularly for leaks.

9. PHYSICAL AND CHEMICAL PROPERTIES

ODOR: Odorless
APPEARANCE: Clear, colorless liquid
AUTOIGNITION TEMPERATURE: Non-combustible
BOILING POINT: 103°C/218°F (20%); 107°C/225°F (31%); 108°C/226°F (35%)
COEFFICIENT OF OIL / WATER: Not available
DENSITY / WEIGHT PER VOLUME: Not available
EVAPORATION RATE: Above 1 (Butyl Acetate = 1)
FLASH POINT: Non-combustible
FREEZING POINT: -15°C/6°F (20%); -26°C/-15°F (31%); -33°C/-27°F (35%)
ODOR THRESHOLD: Not available
OXIDIZING PROPERTIES: Strong oxidizer
PERCENT VOLATILE: 100%
pH: (as is) < / = 3.7
SOLUBILITY IN WATER: (in H_2O % by wt) 100%
SPECIFIC GRAVITY: 1.07 @ 20°C/4°C (20%); 1.11 @ 20°C/4°C (31%); 1.13 @ 20°C/4°C (35%)
VAPOR DENSITY: (Air = 1): Not available
VAPOR PRESSURE: 28 mmHg @ 30°C (20%); 24 mmHg @ 30°C (31%); 23mmHg @ 30°C (35%)

COMMENTS:

pH (1% solution) @ 25°C: 5.0 - 6.0
Hydrogen Peroxide (20 to 40%) (7722-84-1-3) Date: 04/05/2005

10. STABILITY AND REACTIVITY

CONDITIONS TO AVOID: Excessive heat or contamination could cause product to become unstable.

STABILITY: Stable (heat and contamination could cause decomposition)

POLYMERIZATION: Will not occur

INCOMPATIBLE MATERIALS: Reducing agents, wood, paper and other combustibles, iron and other heavy metals, copper alloys and caustic.

HAZARDOUS DECOMPOSITION PRODUCTS: Oxygen which supports combustion.

COMMENTS: Materials to Avoid : Dirt, organics, cyanides and combustibles such as wood, paper, oils, etc.

11. TOXICOLOGICAL INFORMATION

EYE EFFECTS: 35% hydrogen peroxide: Extremely irritating/corrosive (rabbit) [Study Number: I83-748]

SKIN EFFECTS: 35% hydrogen peroxide: Mildly irritating after 4-hour exposure (rabbit) Study Number: I83-747]

DERMAL LD_{50}: 35% hydrogen peroxide: > 2,000 mg/kg (rabbit) [Study Number: I83-746]

ORAL LD_{50}: 35% hydrogen peroxide: 1,193 mg/kg (rat) [Study Number: I83-745]

INHALATION LC_{50}: 50% hydrogen peroxide: > 0.17 mg/l (rat) [Study Number: I89-1080]

TARGET ORGANS: Eyes, nose, throat and lungs

ACUTE EFFECTS FROM OVEREXPOSURE: Extremely irritating/corrosive to eyes and gastrointestinal tract. May cause irreversible tissue damage to the eyes including blindness. Inhalation of mist or vapors may be severely irritating to nose, throat and lungs. May cause skin irritation.

CHRONIC EFFECTS FROM OVEREXPOSURE: The International Agency for Research on Cancer (IARC) has concluded that there is inadequate evidence for carcinogenicity of hydrogen peroxide in humans, but limited evidence in experimental animals (Group 3 - not classifiable as to its carcinogenicity to humans). The American Conference of Governmental Industrial Hygienists (ACGIH) has concluded that hydrogen peroxide is a 'Confirmed Animal Carcinogen with Unknown Relevance to Humans' (A3).
Hydrogen Peroxide (20 to 40%) (7722-84-1-3) Date: 04/05/2005

CARCINOGENICITY:

Chemical Name	IARC	NTP	OSHA	Other
Hydrogen Peroxide	Listed	Not listed	Not listed	(ACGIH) Listed (A3, Animal Carcinogen)

12. ECOLOGICAL INFORMATION

ECOTOXICOLOGICAL INFORMATION: Channel catfish 96-hour LC_{50} = 37.4 mg/L
Fathead minnow 96-hour LC_{50} = 16.4 mg/L
Daphnia magna 24-hour EC_{50} = 7.7 mg/L
Daphnia pulex 48-hour LC_{50} = 2.4 mg/L
Freshwater snail 96-hour LC_{50} = 17.7 mg/L
For more information refer to ECETOC "Joint Assessment of Commodity Chemicals No. 22, Hydrogen Peroxide." ISSN-0773-6339, January 1993

CHEMICAL FATE INFORMATION:

Hydrogen peroxide in the aquatic environment is subject to various reduction or oxidation processes and decomposes into water and oxygen. Hydrogen peroxide half-life in freshwater ranged from 8 hours to 20 days, in air from 10-20 hrs. and in soils from minutes to hours depending upon microbiological activity and metal contaminants.

13. DISPOSAL CONSIDERATIONS

DISPOSAL METHOD: An acceptable method of disposal is to dilute with a large amount of water and allow the hydrogen peroxide to decompose followed by discharge into a suitable treatment system in accordance with all regulatory agencies. The appropriate regulatory agencies should be contacted prior to disposal.

14. TRANSPORT INFORMATION

U.S. DEPARTMENT OF TRANSPORTATION (DOT)

PROPER SHIPPING NAME: Hydrogen peroxide, aqueous solutions with no less than 20% but not more than 40%hydrogen peroxide

PRIMARY HAZARD CLASS / DIVISION: 5.1 (Oxidizer)
UN/NA NUMBER: UN 2014
PACKING GROUP: II
LABEL(S): Oxidizer, Corrosive
PLACARD(S): 5.1 (Oxidizer)
Hydrogen Peroxide (20 to 40%) (7722-84-1-3) Date: 04/05/2005

ADDITIONAL INFORMATION: DOT Marking: Hydrogen Peroxide, aqueous solution with not less than 20%, but not more than40% Hydrogen Peroxide, UN 2014 Hazardous Substance/RQ: Not applicable
49 STCC Number: 4918775

DOT Spec: stainless steel/high purity aluminum cargo tanks and rail cars. UN Spec: HDPE drums. For Specific Details

INTERNATIONAL MARITIME DANGEROUS GOODS (IMDG)

PROPER SHIPPING NAME: Hydrogen peroxide, aqueous solutions with not less than 20%, but not more than 60%
hydrogen peroxide.

INTERNATIONAL CIVIL AVIATION ORGANIZATION (ICAO) / INTERNATIONAL AIR TRANSPORT ASSOCIATION (IATA)

PROPER SHIPPING NAME: Hydrogen peroxide, aqueous solutions with
not less than 20%, but not more than 40%
hydrogen peroxide (*).

OTHER INFORMATION:

(*) Air regulations permit shipment of Hydrogen Peroxide (20 - 40%) in non-vented containers for Air Cargo Only aircraft, as well as for Passenger and Cargo aircraft. HOWEVER, all Hydrogen Peroxide containers are vented and therefore, air shipments of H_2O_2 is not permitted. IATA air regulations state that venting of packages containing oxidizing substances is not permitted for air transport.
Protect from physical damage. Keep drums in upright position. Drums should not be stacked in transit. Do not store drum on wooden pallets.

15. REGULATORY INFORMATION

UNITED STATES

SARA TITLE III (SUPERFUND AMENDMENTS AND REAUTHORIZATION ACT)
SECTION 302 EXTREMELY HAZARDOUS SUBSTANCES (40 CFR 355, APPENDIX A):
Not listed
SECTION 311 HAZARD CATEGORIES (40 CFR 370):
Fire Hazard, Immediate (Acute) Health Hazard
Hydrogen Peroxide (20 to 40%) (7722-84-1-3) Date: 04/05/2005

SECTION 312 THRESHOLD PLANNING QUANTITY (40 CFR 370):
The Threshold Planning Quantity (TPQ) for this product, if treated as a mixture, is 10,000 lbs; however, this product contains the following ingredients with a TPQ of less than 10,000 lbs.:
None, (conc. <52%)

SECTION 313 REPORTABLE INGREDIENTS (40 CFR 372):
Not listed
CERCLA (COMPREHENSIVE ENVIRONMENTAL RESPONSE COMPENSATION AND LIABILITY ACT)
CERCLA DESIGNATION & REPORTABLE QUANTITIES (RQ) (40 CFR 302.4):
Unlisted (Hydrogen Peroxide 20-40%); RQ = 100 lbs.; Ignitability, Corrosivity
TSCA (TOXIC SUBSTANCE CONTROL ACT)
TSCA INVENTORY STATUS (40 CFR 710):
Listed
RESOURCE CONSERVATION AND RECOVERY ACT (RCRA)
RCRA IDENTIFICATION OF HAZARDOUS WASTE (40 CFR 261):
Waste Number: D001, D002

CANADA

WHMIS (WORKPLACE HAZARDOUS MATERIALS INFORMATION SYSTEM):
Product Identification Number: 2014
Hazard Classification / Division: Class C (Oxidizer), Class D, Div. 2, Subdiv. B. (Toxic), Class E (Corrosive) Ingredient Disclosure List: Listed

INTERNATIONAL LISTINGS

Hydrogen peroxide:
China: Listed
Japan (ENCS): (1)-419
Korea: KE-20204
Philippines (PICCS): Listed

HAZARD, RISK AND SAFETY PHRASE DESCRIPTIONS:

Hydrogen Peroxide, (Index #008-003-00-9):

EC Symbols: Xn (Harmful)

Hydrogen Peroxide (20 to 40%) (7722-84-1-3) Date: 04/05/2005

EC Risk Phrases: R22 (Harmful if swallowed.)

R37/38 (Irritating to respiratory system and to skin.)

R41 (Risk of serious damage to eyes.)

EC Safety Phrases: S1/2 (Keep locked up and out of reach of children.)

S3 (Keep in a cool place.)

S17 (Keep away from combustible material.)

S26 (In case of contact with eyes, rinse immediately with plenty of water and seek medical advice)

S28 (After contact with skin, wash immediately with plenty of water and soap.)

S36/37/39 (Wear suitable protective clothing, gloves and eye/face protection.)

S45 In case of accident or if you feel unwell, seek medical advice immediately –
(show the label where possible.)

16. OTHER INFORMATION

HMIS
Health 3
Flammability 0
Physical Hazard 1
Personal Protection (PPE) H
Protection = H (Safety goggles, gloves, apron, the use of a supplied air or SCBA respirator is required in lieu of a vapor cartridge respirator)
HMIS = Hazardous Materials Identification System
Degree of Hazard Code:
4 = Severe
3 = Serious
2 = Moderate
1 = Slight
0 = Minimal

NFPA
Health 3
Flammability 0
Reactivity 1
Special OX
SPECIAL = OX (Oxidizer)
NFPA = National Fire Protection Association
Degree of Hazard Code:
Hydrogen Peroxide (20 to 40%) (7722-84-1-3) Date: 04/05/2005

4 = Extreme
3 = High
2 = Moderate
1 = Slight
0 = Insignificant

REVISION SUMMARY:
This MSDS replaces Revision #8, dated November 04, 2004.
Changes in information are as follows:
Section 3 (Composition / Information on Ingredients)
Section 15 (Regulatory Information)
Section 16 (Other Information)
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APPENDIX L

@ EE on 6-29-10 @ 1030

ENVIRONMENTAL ENTERPRISES USA, INC.
58485 Pearl Acres Rd., Suite D
Slidell, Louisiana 70461
(985) 646-2787

Kit No. N/A

CHAIN - OF - CUSTODY RECORD

Client: EE USA INC. *Contact Person:* MARK A. O'NEIL
Address: 58485 Pearl Acres Rd. Slidell, LA 70461 *Phone#:* (985) 646-2787
P.O. #
FAX #:
Project:

Special Handling Request
( ) RUSH
( ) VERBAL
( ) OTHER

Lab Sample Description	Date Collected	Time Collected	No. of Containers	Analysis Request	S/R No.	Lab No.
U.S. EPA - API	N/A	N/A	2	NCP Toxicity Testing		D-020-10
Reference Oil						
No. 2 Fuel Oil						
Low Aromatic						
WF 681						
Purchased from RTC.						

Collected By:	Date	Time	Relinquished By:	Date	Time
Received By: UPS 1Z 7X9 262 03 5223 9857	Date	Time	Relinquished By: UPS	Date 6-29-10	Time 1035
Received By: T. O'Neill EE	Date 6-29-10	Time 1035	Relinquished By: T. O'Neill EE	Date 6-29-10	Time 1040
Received By: Mark A. O'Neil	Date 6/29/10	Time 1045	Relinquished By:	Date	Time
Received By:	Date	Time	Relinquished By:	Date	Time

D-020-10

U.S. Environmental Protection Agency
Environmental Monitoring and Support Laboratory - Cincinnati

American Petroleum Institute
Department of Environmental Affairs

STANDARD REFERENCE OIL SAMPLE

FUEL OIL NO. 2

This sample is made available for the sole purpose of providing a reference oil for research and laboratory testing purposes.

Storage and Handling

Store reference oil samples at a temperature of no more than 20°C, preferably in a dark area.

Ampuls, 20 mL - open the ampul by snapping off the top at the break area on the neck.

To retain a portion of the ampul contents, immediately transfer the oil to a clean, dry glass flask or vial, and seal. Do not use a plastic container. Non-glass stoppers must contain a Teflon insert to prevent oil contact with plastic or metal.

Bottles, one-pint - bottles of reference oils are closed with a plastic screw cap containing a Teflon insert. If bottle is used to store a portion of the oil contents after opening, be sure that the Teflon insert remains in the cap.

ASTM Standard Methods for Waterborne Oil Samples

Analyte	ASTM Method*
Specific and API gravity	D1298-80 (Part 23)
Nitrogen, sulfur, nickel and vanadium	D3327-79 (Part 31)
Sulfur compounds, profile	D3328-78 (Part 31)
Simulated distillation profile	D2887-73 (Part 24)
Infrared spectrum	D3414-79 (Part 31)
UV fluorescence spectrum	D3650-78 (Part 31)

*ASTM series available from: American Society for Testing and Materials, 1916 Race Street, Philadelphia, PA 19103.

REFERENCE VALUES

Fuel Oil No. 2

This oil has been analyzed by skilled oil testing and research laboratories to characterize it and to ensure that substantial compositional changes have not occurred during storage and sample preparation. Results for various selected parameters were as follows:

Analyte	Result
Specific gravity*	0.856 kg/L
API gravity*	33.7 degrees
Sulfur	0.12 weight %
Sulfur compounds, profile	See Fig. 1
Nitrogen	0.009 weight %
Vanadium	0.2 mg/L
Nickel	0.1 mg/L
Simulated distillation profile	See Fig. 2 and Table 1
Infrared spectrum	See Fig. 3
UV fluorescence spectrum	See Fig. 4
Pour point	-20°F
Viscosity,	
at 40°C	2.384
at 100°	1.034
Index	**
Saturates	57.2 weight %
Aromatics	41.8 weight %
Aromatics analyses, weight %:	
Alkylbenzenes	12.1
Naphthenebenzenes	12.1
Dinaphthenebenzene	2.6
Naphthalenes	8.2
Acenaphthenes	3.3
Fluorenes	1.6

REFERENCE VALUES

Fuel Oil No. 2
Continued

Analyte	Result
Phenanthrenes	1.5
Naphthenephenanthrenes	0
Pyrenes	0.1
Chrysenes	0
Perylenes	0
Dibenzanthracenes	0
Benzothiophenes	0
Dibenzothiophenes	0.1
Naphthabenzothiophenes	0.2
Class II-Class VII Unidentified	0

* At 15/15°C
** Not calculable when viscosity at 100°C is less than 2.0.

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