


**Effluent Quality Assessment**

# Trademark Nitrogen Corporation

Hillsborough County

NPDES #FL0000647

Sampled March 17, 19 and 21, 2008

August 2008

**Biology Section**
**Bureau of Laboratories**
**Division of Resource Assessment and Management**

Quality Manual No. 870346G
NELAC Certification No. E31780

Trademark Nitrogen Corporation, 1216 Old Hopewell Road, Tampa, Hillsborough County, Florida, NPDES #FL0000647. Effluent samples for chemical, algal growth potential and bioassay analyses for this facility were collected on March 17, 2008. Additional effluent samples for bioassay analyses were collected on March 19 and 21, 2008.

## Introduction

Trademark Nitrogen Corporation is a fertilizer production plant. The facility consists of:

- a 125 ton/day nitric acid plant;
- a 150 ton/day ammonium nitrate plant;
- a fertilizer solutions plant;
- a magnesium nitrate plant;
- a nitrogen-phosphorus-potassium blend plant (liquid fertilizer);
- a small boiler plant.

The facility has a design flow of 0.09 million gallons per day (MGD) average daily flow (ADF). The ADF from March 2007 through February 2008 was 0.0882 MGD. Effluent from the facility consisting of non-process wastewater and stormwater is treated in an unlined vegetated detention ditch. The system provides for the uptake and settling of solids. Water can also be recycled through the ditch system via a manually controlled pump. Aeration and optional pH adjustment are performed prior to the effluent being discharge through Outfall D-001 to the ditch thence to the Class III freshwaters of the Palm River. The point of discharge for Outfall D-001 is located approximately at latitude 27º 57' 15.4" N, longitude 82º 21' 1.5" W.

The facility has no Administrative Orders but does have an active Consent Order (CO) concerning groundwater contamination. The CO has been amended under 91-1885B for the facility to proceed with site rehabilitation under Chapter 62-780 and 62-777 F.A.C. clean-up criteria. The facility has no mixing zones. A review of Discharge Monitoring Report Data for the past year revealed a dissolved oxygen violation (value 4.82 mg/L, permit limit ≥ 5.0 mg/L) in May 2007 (Facility information provided by Pete Wenner FDEP Southwest District.)

## Methods

Samples were collected on March 17, 2008, from the EFF-1 NPDES sampling point for Outfall D-001 by Pete Wenner and Chris Anastasiou (FDEP Southwest District). Effluent samples for bioassays were also collected on March 19 and March 21, 2008, by Pete Wenner. Samples were collected following DEP-SOP-001/01 FS 2400 Wastewater Sampling. All DEP SOPs are available on the web at: http://www.floridadep.org/labs/qa/sops.htm.

See Appendix A for the completed chain of custody forms.

The chronic screening toxicity tests and algal growth potential (AGP) test were performed following internal DEP SOPs TA07_05, TA07_09, and TA08_01 through TA08_09. All internal DEP SOPs are available on the web at: http://www.floridadep.org/labs/cgi-bin/sop/biosop.asp.

The chronic bioassay tests were performed in accordance with the facility's NPDES permit, with the exception that due to a composite sampler failure, the first bioassay sample was a grab sample rather than a composite sample.

Dunnett's procedure (available at Statistical Analysis for Biological Methods http://www.epa.gov/EERD/stat2.htm) and t-tests were used to analyze data from these tests.

## Toxicity Test Results

Tests were performed March 18 – 25*, 2008.

*Ceriodaphnia dubia* chronic screening bioassay – NOEC = 100%

*Pimephales promelas* chronic screening bioassay – NOEC = 100%

The bioassay samples were not chronically toxic to the test organisms.

See Appendix B for bioassay bench sheets. See Appendix C for results of the last 20 standard reference toxicant (SRT) tests and the bench sheets and statistical analyses of the SRT tests that correspond to these facility tests. See Appendix D for test organism receipts.

*NOTE: The *Ceriodaphnia dubia* bioassay was terminated on test day 6 (March 24, 2008) because termination criteria was met (≥ 60% of control organisms having 3rd brood).

## Algal Growth Potential

The effluent Algal Growth Potential (AGP) was 8.7 mg dry wt/L of the unicellular green alga, *Pseudokirchneriella subcapitata*, formerly known as *Selenastrum capricornutum*. Raschke and Shultz (1987) found that AGP values above 5.0 mg dry weight/L represent a “problem” threshold for fresh receiving waters, implying nutrient enrichment. The analytical chemistry suggests that the effluent is nitrogen-limited. The predicted AGP based on the inorganic nitrogen concentration was 2.5 (±20%) mg dry wt/L. The predicted AGP based on the total nitrogen concentration was 5.4 (±20%) mg dry wt/L (Table 1; Miller et al. 1978).

See Table 1 for AGP results.

## Chemistry Results

Total residual chlorine (TRC) and total ammonia were not detected in the laboratory in the any of the three bioassay samples. The total ammonia concentration in the sample collected and preserved for chemical analysis on March 17 was 0.035 mg/L. Based on the pH, salinity, and temperature of the effluent as collected, the calculated unionized ammonia concentration was < 0.02 mg/L.

Cadmium and iron were detected in the effluent between the laboratory method detection limits (MDL) and practical quantitation limits (PQL). Other substances of concern (62-302.530, F.A.C.) were not detected in the effluent.

See Table 2 for results of analytes detected in the effluent, and corresponding limits. See Appendix E for a complete list of chemical analyses performed. See Appendix F for laboratory instruments.

## Conclusion

The effluent samples collected from the EFF-1 NPDES sampling point for this facility on March 17, 19 and 21, 2008, were not chronically toxic to either test species during the chronic screening bioassays. The effluent AGP result exceeded the "problem” threshold for fresh receiving waters, suggesting nutrient enrichment may be an issue. Effluent water quality samples did not exceed any applicable water quality standards or violate any permit conditions.

## Literature Cited

Miller, W. E., T. E. Maloney, and J. C. Greene. 1978. The *Selenastrum capricornutum* Printz algal assay bottle test. EPA-600/9-78-018. U. S. EPA, Cincinnati, Ohio. 126 p.

Raschke, R. L. and D. A. Schultz. 1987. The use of the algal growth potential test for data assessment. J. Wat. Poll. Cont. Fed. 59(4): 222-227.

**Table 1. Measured and predicted algal growth potential (AGP; mg dry weight/L) for total soluble inorganic nitrogen (TSIN), orthophosphate (OP), total nitrogen (TN) and total phosphorus (TP) limitation of the freshwater species *Pseudokirchneriella subcapitata* and ratios of nitrogen to phosphorus for samples collected from the Trademark Nitrogen Corporation facility on March 17, 2008.**

| Trademark Nitrogen Corporation | | | | | | | |
|---|---|---|---|---|---|---|---|
| Location | AGP (measured) | Predicted AGP (TSIN) ± 20% | Predicted AGP (OP) ± 20% | Predicted AGP (TN) ± 20% | Predicted AGP (TP) ± 20% | Inorganic N:P ratio | Total N:P ratio |
| Effluent | 8.7 A | 2.5 ± 0.5 | 15.9 ± 3.2 | 5.4 ± 1.1 | 20.6 ± 4.1 | 1.8 | 3.0 |

**Table 2. Effluent limits, Class III Freshwater Criteria and chemical data for samples collected from the Trademark Nitrogen Corporation facility on March 17, 19 and 21, 2008.**

| Trademark Nitrogen Corporation NPDES #FL0000647 | Class III Criteria | Effluent Limits | Effluent | Bioassay 1 | Bioassay 2 | Bioassay 3 |
|---|---|---|---|---|---|---|
| **Organic Constituents (µg/L)** | | | | | | |
| None Detected | - | - | - | - | - | - |
| **Metals (µg/L unless otherwise noted)** | | | | | | |
| Aluminum | - | - | 60 U | - | - | - |
| Arsenic | ≤ 50 | - | 0.5 U | - | - | - |
| Cadmium | ≤ 0.53 b | - | 0.022 I | - | - | - |
| Calcium (mg/L) | - | - | 79.9 | - | - | - |
| Chromium-III | ≤ 180.40 b | - | 1 U | - | - | - |
| Copper | ≤ 20.16 b | - | 0.5 U | - | - | - |
| Iron | ≤ 1,000 | - | 31 I | - | - | - |
| Lead | ≤ 10.03 b | - | 0.2 U | - | - | - |
| Magnesium (mg/L) | - | - | 11.4 | - | - | - |
| Nickel | ≤ 111.89 b | - | 0.25 U | - | - | - |
| Selenium | ≤ 5.0 | - | 0.5 U | - | - | - |
| Silver | ≤ 0.07 | - | 0.025 U | - | - | - |
| Zinc | ≤ 257.30 b | - | 5 U | - | - | - |
| **Nutrients (mg/L)** | | | | | | |
| Ortho-phosphate | - | - | 0.037 | - | - | - |
| Total Phosphorus | - | - | 0.048 I | - | - | - |
| Total Ammonia | - | - | 0.035 | ≤ 0.017 U£ | ≤ 0.017 U£ | ≤ 0.017 U£ |
| Un-ionized Ammonia | ≤ 0.02 s | ≤ 0.02 s | ≤ 0.02 c | ≤ 0.02 c | ≤ 0.02 c | ≤ 0.02 c |
| Nitrate and Nitrite | - | Report | 0.032 p | - | - | - |
| Total Kjeldahl Nitrogen | - | Report | 0.11 Ip | - | - | - |
| Organic Nitrogen | - | - | 0.075 c | - | - | - |
| Total Nitrogen | - | ≤ 5.0 p | 0.142 cp | - | - | - |
| **General Physical and Chemical Parameters** | | | | | | |
| Dissolved Oxygen (mg/L) | ≥ 5.0 s | | 8.7 | 8.7 | 8.7 | 8.8 |
| pH (SU) | 6.0 - 8.5 s | 6.0 - 8.5 s | 7.9 | 7.9 | 7.8 | 7.8 |
| Conductivity (µmhos/cm) | ≤ 1,275 | ≤ 1,275 s | 506 | 506 | 478 | 511 |
| Temperature °C | - | Report | 21.0 | 21.0 | 21.0 | 18.9 |
| Sample Depth (m) | - | - | 0.2 | 0.2 | 0.2 | 0.2 |
| Total Residual Chlorine (mg/L) | ≤ 0.01 s | - | - | < 0.03 U£ | < 0.03 U£ | < 0.03 U£ |
| Chlorophyll a (µg/L) - Corrected | - | - | 2.6 | - | - | - |
| Phaeophytin (µg/L) | - | - | 1.6 | - | - | - |
| Total Suspended Solids (mg/L) | - | ≤ 12.0 s | 4 Up | - | - | - |
| CBOD, 5 day (mg/L), N - inhibited | - | - | 0.64 I | - | - | - |
| Flow (MGD) | - | Report | 0.0882 a | - | - | - |
| Hardness (mg $CaCO_3$) | - | - | 246.46 c | - | - | - |

Value exceeds the Class III Water Quality Criteria or permit limits

a - Annual average
b - Value is calculated based on hardness
c - Value is calculated
p - Limit based on 24-hour flow proportioned composite sample
s - Single sample
I - The reported value is between the laboratory method detection limit and the laboratory practical quantitation limit
U - Material analyzed for but not detected; value reported is the method detection limit
£ - Analysis performed in bioassay laboratory

# Florida Department of Environmental Protection

## Central Laboratory Sample Submittal Form

Event ID *

**Request Number: RQ-2008-03-03-36**
Trademark Nitrogen Corporation

Requester: Jennifer Paris
Field Report Prepared By:

**Customer: SW-DIST**
**Project ID: FYI3**
**PMAS: 1153**

Collected By: P. WENNER / C. ANASTASIOU
Sampling Agency: SW DISTRICT
Send Final Report To Tom Gucciardo

| Lab ID * | Location | Collection (begin) Eastern | Collection (end) Eastern | Bottle Group(s)** |
|---|---|---|---|---|
| | TRADEMARK NITROGEN D001 | ☐ Comp ☒ Grab Date 3/17/08 Time 1015 Central | Date Time Central | A |
| | Field ID: 36205 | Tot Res Chlorine (mg/L): — | Diss Oxygen (mg/L): 8.72 | Storet Station Number: — |
| | Matrix (Include type e.g. Salt, Fresh, etc): FRESH | Temp (C): 20.99 | pH: 7.89 | Sample Depth: 0.2 ☒ m ☐ ft |
| | Latitude: 27 ° 57 ' 15.4 " | Longitude: 82 ° 21 ' 01.3 " | Comments | 0.24 / 506 ☒ Salinity (PPTh) ☒ Sp Conductance (umho/cm) NPDES Number |

| Lab ID * | Location | Collection (begin) Eastern | Collection (end) Eastern | Bottle Group(s)** |
|---|---|---|---|---|
| | TRADEMARK NITROGEN D001 | ☐ Comp ☒ Grab Date 3/17/08 Time 1300 Central | Date Time Central | A |
| | Field ID: 36207 | Tot Res Chlorine (mg/L): — | Diss Oxygen (mg/L): 8.72 | Storet Station Number: — |
| | Matrix (Include type e.g. Salt, Fresh, etc): FRESH | Temp (C): 20.99 | pH: 7.89 | Sample Depth: 0.2 ☒ m ☐ ft |
| | Latitude: 27 ° 57 ' 15.4 " | Longitude: 82 ° 21 ' 01.3 " | Comments: TOXICITY SAMPLE COLLECTED AS GRAB SAMPLE NOT COMPOSITE | 0.24 / 506 ☒ Salinity (PPTh) ☒ Sp Conductance (umho/cm) NPDES Number |

| Lab ID * | Location | Collection (begin) Eastern | Collection (end) Eastern | Bottle Group(s)** |
|---|---|---|---|---|
| | FIELD BLANK | ☐ Comp ☒ Grab Date 3/17/08 Time 1043 Central | Date Time Central | B |
| | Field ID: 36206 | Tot Res Chlorine (mg/L): — | Diss Oxygen (mg/L): — | Storet Station Number: — |
| | Matrix (Include type e.g. Salt, Fresh, etc): FRESH | Temp (C): — | pH: — | Sample Depth: — ☐ m ☐ ft |
| | Latitude: o ' " | Longitude: o ' " | Comments | — ☐ Salinity (PPTh) ☐ Sp Conductance (umho/cm) NPDES Number: — |

| Lab ID * | Location | Collection (begin) Eastern | Collection (end) Eastern | Bottle Group(s)** |
|---|---|---|---|---|
| | | ☐ Comp ☐ Grab Date Time Central | Date Time Central | |
| | Field ID | Tot Res Chlorine (mg/L) | Diss Oxygen (mg/L) | Storet Station Number |
| | Matrix (Include type e.g. Salt, Fresh, etc) | Temp (C) | pH | Sample Depth ☐ m ☐ ft |
| | Latitude: o ' " | Longitude: o ' " | Comments | ☐ Salinity (PPTh) ☐ Sp Conductance (umho/cm) NPDES Number |

| Relinquished By: | Date/Time | Shipping Method: | Received By: | Date/Time | Relinquished By: | Date/Time | Received By: | Date/Time |
|---|---|---|---|---|---|---|---|---|
| PETE WENNER | 3/17/08 15:00 | DHL | Tghmaven Ebrighaus | 03/18/08 8:45 | 1 BSJ | BA 10:24 3-18-08 | | |

* Shaded Areas for Lab use only.

** Please see reverse side for Bottle Group information.

last revised October 1, 2003

# Florida Department of Environmental Protection

## Central Laboratory Sample Submittal Form

Event ID *

equest Number: RQ-2008-03-03-36
ademark Nitrogen Corporation

Requester: Jennifer Paris
Field Report Prepared By:

ustomer: SW-DIST
Collected By: PETER WENNER
Send Final Report To Tom Gucciardo

roject ID: FYI3
Sampling Agency: FDEP SWD

MAS: 1153

| Lab ID * | Location | Collection (begin) Eastern | Collection (end) Eastern | Bottle Group(s)** |
|---|---|---|---|---|
| | TRADEMARK NITROGEN D-001 | ☒ Comp ☐ Grab Date 3/18/08 Time 1050 Central | Date 3/19/08 Time 0950 Central | A |
| | Field ID: 36208 | Tot Res Chlorine (mg/L) | Diss Oxygen (mg/L): 8.70 | Storet Station Number: — |
| | Matrix (Include type e.g. Salt, Fresh, etc): FRESH | Temp (C): 21.05 | pH: 7.80 | Sample Depth ☒ m ☐ ft: 0.2 |
| | | | Salinity (PPTh): 0.25 ☐ Sp Conductance (umho/cm) | NPDES Number |
| | Latitude: 27 ° 57 ' 15.4 " | Longitude: 82 ° 21 ' 01.3 " | Comments: COMPOSITE SAMPLE: 24 SAMPLES @ 250mL/SAMPLE; 1SAMPLE/HOUR | |

| Lab ID * | Location | Collection (begin) Eastern | Collection (end) Eastern | Bottle Group(s)** |
|---|---|---|---|---|
| | | ☐ Comp ☐ Grab Date Time Central | Date Time Central | |
| | Field ID | Tot Res Chlorine (mg/L) | Diss Oxygen (mg/L) | Storet Station Number |
| | Matrix (Include type e.g. Salt, Fresh, etc) | Temp (C) | pH | Sample Depth ☐ m ☐ ft |
| | | | Salinity (PPTh) ☐ Sp Conductance (umho/cm) | NPDES Number |
| | Latitude ' " | Longitude ° ' " | Comments | |

| Lab ID * | Location | Collection (begin) Eastern | Collection (end) Eastern | Bottle Group(s)** |
|---|---|---|---|---|
| | | ☐ Comp ☐ Grab Date Time Central | Date Time Central | |
| | Field ID | Tot Res Chlorine (mg/L) | Diss Oxygen (mg/L) | Storet Station Number |
| | Matrix (Include type e.g. Salt, Fresh, etc) | Temp (C) | pH | Sample Depth ☐ m ☐ ft |
| | | | Salinity (PPTh) ☐ Sp Conductance (umho/cm) | NPDES Number |
| | Latitude ° ' " | Longitude ° ' " | Comments | |

| Lab ID * | Location | Collection (begin) Eastern | Collection (end) Eastern | Bottle Group(s)** |
|---|---|---|---|---|
| | | ☐ Comp ☐ Grab Date Time Central | Date Time Central | |
| | Field ID | Tot Res Chlorine (mg/L) | Diss Oxygen (mg/L) | Storet Station Number |
| | Matrix (Include type e.g. Salt, Fresh, etc) | Temp (C) | pH | Sample Depth ☐ m ☐ ft |
| | | | Salinity (PPTh) ☐ Sp Conductance (umho/cm) | NPDES Number |
| | Latitude ° ' " | Longitude ° ' " | Comments: 1 ✗ Biossay Rec'd in lab by J.D. 9.13.08 10:12 | |

| Relinquished By: | Date/Time | Shipping Method: | Received By: | Date/Time | Relinquished By: | Date/Time | Received By: | Date/Time |
|---|---|---|---|---|---|---|---|---|
| PETE WENNER | 3/19/08; 15:00 | DHL | | | | | SK | 3-20-08 9:00 |

* Shaded Areas for Lab use only.

** Please see reverse side for Bottle Group information.

last revised October 1, 2003

# Florida Department of Environmental Protection

## Central Laboratory Sample Submittal Form

Event ID *

Request Number: RQ-2008-03-03-36
rademark Nitrogen Corporation

Requester: Jennifer Paris
Field Report Prepared By:

Customer: SW-DIST
Collected By: PETER WENNER
Send Final Report To Tom Gucciardo

Project ID: FYI3
Sampling Agency: FDEP SWD
PMAS: 1153

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| Lab ID * | Location: TRADEMARK NITROGEN D-001 | | | ☒ Comp ☐ Grab | Collection (begin) Date 3/20/08 Time 0750 (Eastern) Central | Collection (end) Date 3/21/08 Time 0650 (Eastern) Central | | Bottle Group(s)** | |
| | Field ID: 36209 | | | Tot Res Chlorine (mg/L) | Diss Oxygen (mg/L): 8.77 DO%: 95.2 | Storet Station Number | | A | |
| | Matrix (Include type e.g. Salt, Fresh, etc): FRESH | Temp (C): 18.93 | pH: 7.82 | Sample Depth: ☒ m ☐ ft: 0.2 | 0.25 / 511 | ☒ Salinity (PPTh) ☒ Sp Conductance (umho/cm) | NPDES Number | | |
| | Latitude: 27 ° 57 15.4 | Longitude: 82 21 01.3 | | Comments: BOTH BOTTLES ARE 24-HOUR TIME COMPOSITE; 1-400mL SAMPLE/HR | | | | | |
| Lab ID * | Location | | | ☐ Comp ☐ Grab | Collection (begin) Date Time (Eastern) Central | Collection (end) Date Time Eastern Central | | Bottle Group(s)** | |
| | Field ID | | | Tot Res Chlorine (mg/L) | Diss Oxygen (mg/L) | Storet Station Number | | | |
| | Matrix (Include type e.g. Salt, Fresh, etc) | Temp (C) | pH | Sample Depth ☐ m ☐ ft | | ☐ Salinity (PPTh) ☐ Sp Conductance (umho/cm) | NPDES Number | | |
| | Latitude ' " | Longitude ° ' " | | Comments | | | | | |
| Lab ID * | Location | | | ☐ Comp ☐ Grab | Collection (begin) Date Time (Eastern) Central | Collection (end) Date Time Eastern Central | | Bottle Group(s)** | |
| | Field ID | | | Tot Res Chlorine (mg/L) | Diss Oxygen (mg/L) | Storet Station Number | | | |
| | Matrix (Include type e.g. Salt, Fresh, etc) | Temp (C) | pH | Sample Depth ☐ m ☐ ft | | ☐ Salinity (PPTh) ☐ Sp Conductance (umho/cm) | NPDES Number | | |
| | Latitude ° ' " | Longitude ° ' " | | Comments | | | | | |
| Lab ID * | Location | | | ☐ Comp ☐ Grab | Collection (begin) Date Time Eastern Central | Collection (end) Date Time Eastern Central | | Bottle Group(s)** | |
| | Field ID | | | Tot Res Chlorine (mg/L) | Diss Oxygen (mg/L) | Storet Station Number | | | |
| | Matrix (Include type e.g. Salt, Fresh, etc) | Temp (C) | pH | Sample Depth ☐ m ☐ ft | | ☐ Salinity (PPTh) ☐ Sp Conductance (umho/cm) | NPDES Number | | |
| | Latitude ° ' " | Longitude ° ' " | | Comments | | | | | |

| Relinquished By: | Date/Time | Shipping Method: | Received By: | Date/Time | Relinquished By: | Date/Time | Received By: | Date/Time |
|---|---|---|---|---|---|---|---|---|
| PETER WENNER | 3/21/08 | DHL | Marshall Hallett | 3-21-08 1030 | | | | 3-24-08 / 9:55a |

2 Biomass Jugs Temperature 1.0 °C
1 cooler

* Shaded Areas for Lab use only.

** Please see reverse side for Bottle Group information.

last revised October 1, 2003

## Appendix B. Bench Sheets

**FDEP Biology Laboratory - Chronic Screen Bench Sheet**
*Ceriodaphnia dubia*

Facility: Trademark Nitrogen Corp.
Address: 1216 Old Hopewell Rd
County: Hillsborough
NPDES Permit #: FL 0000647
Contact/District: Pete Wenner / SW - District

SOP#: TA07_05 EPA 821-R-02-013 Method 1002.0

Start Date: 3.18.08 Time: 13:00
End Date: 3.24.08 Time: 13:30

LIMS entry: 4.11.08 MF verified: 4.11.08 BA

Diluent/ Batch: 20% DMW / 3-10-08 Test Days Used: 0-3 (2 MF)
20% DMW / 3.17.08 Test Days Used: 4 - 6 (3 MF 5 MF)

Organism: *Ceriodaphnia dubia*
Organism Batch #: 11-08
Organism age : <24 hours
Feeding: Prior to Test and Daily
Food: YCT *P. subcapitata*
Food Batch: 7-26-07 1-31-08
Chamber size : 30 ml
Sample volume : 20 ml

| Sample Received | Lab Sample # | LIMS Job # | LIMS Sample # | Collection Date/Time | Received Date/Time |
|---|---|---|---|---|---|
| Test Day 0 | 1 | TLH-2008-03-18-03 | 1084393 | 3.17.08 / 13:00 | 3-18-08 / 10.25 |
| Test Day 2 | 2 | TLH-2008-03-20-05 | 1089864 | 3-18-08 / 10:50 | 3-20-08 / 10:15 |
| Test Day 4 | 3 | TLH-2008-03-24-03 | 1090352 | 3-21-08 / 0650 | 3-22-08 / 10 30 |

| Water Quality Parameters | Field Chlorine (mg/L) | | | Lab Chlorine (mg/L) | | | Alkalinity (mg/L as $CaCO_3$) | | |
|---|---|---|---|---|---|---|---|---|---|
| Lab Sample #: | 1 | 2 | 3 | 1 | 2 | 3 | MF 1 | MF 2 | MF 3 |
| Sample: | — | — | — | <0.03 | <0.03 | <0.03 | 97200 | ~~102203~~ | ~~106209~~ |
| Diluent/Control: | | | | <0.03 | <0.03 | <0.03 | 87 | 94 | ~~94~~ 94 BA |
| Method: | Not | test | test | Hach | Hach | Hach | Hach | Hach | Hach |
| Measured by: | — | — | — | JD | JD | MF | BA | BA | BA |
| Verified by: | BA | BA | BA | MF | MF | BA | MF | MF | MF |
| | Conductivity (circle one) (µmhos-cm) (mmhos-cm) | | | Ammonia (mg/L) | | | Hardness (mg/L as $CaCO_3$) | | |
| Lab Sample #: | 1 | 2 | 3 | 1 | 2 | 3 | MF 1 | MF 2 | 3 MF |
| Sample: | 527 | 528 | 524 | <0.017 | <0.017 | <0.017 | 20045 | 20645 | ~~14824~~ 3 |
| Diluent/Control: | 181 | 174 | 185 | <0.017 | <0.017 | <0.017 | 84 | 94 | 94 |
| Method: | Mettler | Mettler | Mettler | Denver | Denver | Denver | Hach | Hach | Hach |
| Measured by: | JD/MF | JD/MF | MF | MF | MF | MF | BA | BA | BA |
| Verified by: | MF BA | MF/BA | BA | BA | BA | BA | MF | MF | MF |

Temperature Shipped ≤6° C (all samples) (yes) no
Holding Time ≤36 hours (yes) no
Light Intensity : 50 - 100 ft. candles
Photoperiod : 16 light 8 dark
Temperature Range °C:
Incubator # 7 min. 25.7 max. 27.2 mean 26.5
Room B246 min. 24.2 max. 25.7 mean 25.0

| Test Replicate | Loaded from culture vial |
|---|---|
| 1 | A2 |
| 2 | A4 |
| 3 | A7 |
| 4 | A14 |
| 5 | A17 |
| 6 | A22 |
| 7 | A27 |
| 8 | A28 |
| 9 | B10 |
| 10 | B15 |

| Statistics | |
|---|---|
| Control % CV: | 13.9 |
| Dunnetts PMSD: | 10.1% |
| Reproduction NOEC: | 100% |
| Survival NOEC: | 100% |
| T-test value | 0,00000 |
| T- table value: | 1.94300 |
| df: | ~~18~~ 18 MF |

Investigators' signatures
James Daniel
Brad Ard
Marshall Faircloth

Loretta C. Wolfe
REVIEWER

Comments: TOST terminated on day 6 due to survival and reproduction criteria being met MF 3-24-08

## Appendix B. (continued)

**FDEP Biology Laboratory - Chronic Screen Bench Sheet**

*Ceriodaphnia dubia* Survival and Reproduction

Facility: Trade Mark Nitrogen

LIMS Sample # 1087373 for lab sample 1

Start Date: 3-18-08 Time: 13:00

End Date: 3-24-08 Time: 13:30

| Conc. | Day | Replicate Number 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | Total Neonates | Surviving adults | Neonates per adult |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Control | 0* | | | | | | | | | | | | 10 | |
| | 1 | | | | | | | | | | | | 10 | |
| | 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 10 | 0 |
| | 3 | 4 | 4 | 0 | 0 | 2 | 0 | 0 | 0 | 0 | 0 | 10 | 10 | 1 |
| | 4 | 7 | 8 | 0 | 4 | 7 | 0 | 4 | 2 | 4 | 4 | 34 | 10 | 3.4 |
| | 5 | 10 | 8 | 8 | 0 | 8 | 10 | 9 | 7 | 9 | 7 | 76 | 10 | 7.6 |
| | 6 | 0 | 14 | 16 | 12 | 10 | 12 | 11 | 12 | 9 | 9 | 105 | 10 | 10.5 |
| | 7 | | | | | | | | | | | | | |
| | 8 | | | | | | | | | | | | | |
| | Total | 21 | 26 | 28 | 19 | 20 | 26 | 24 | 23 | 20 | 20 | 225 | 10 | 22.5 |

| Conc. | Day | Replicate Number 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | Total Neonates | Surviving adults | Neonates per adult |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 100% sample | 0* | | | | | | | | | | | | 10 | |
| | 1 | | | | | | | | | | | | 10 | |
| | 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 10 | 0 |
| | 3 | 0 | 4 | 0 | 3 | 0 | 0 | 0 | 0 | 0 | 0 | 7 | 10 | 0.7 |
| | 4 | 3 | 0 | 5 | 0 | 2 | 6 | 4 | 4 | 2 | 3 | 29 | 10 | 2.9 |
| | 5 | 6 | 8 | 9 | 6 | 9 | 9 | 10 | 10 | 6 | 10 | 85 | 10 | 8.5 |
| | 6 | 9 | 10 | 9 | 10 | 11 | 12 | 9 | 13 | 14 | 7 | 104 | 10 | 10.4 |
| | 7 | | | | | | | | | | | | | |
| | 8 | | | | | | | | | | | | | |
| | Total | 20 | 22 | 23 | 19 | 22 | 27 | 23 | 27 | 22 | 20 | 225 | 10 | 22.5 |

| Organisms loaded by: BA | |
|---|---|
| Day | Counted/ verified by: |
| 0 | MF |
| 1 | MF |
| 2 | MF |
| 3 | MF |
| 4 | MF |
| 5 | BA |
| 6 | BA |
| 7 | — |
| 8 | — |

| Concentration | Mean neonates /adult | % survival |
|---|---|---|
| Control | 22.5 | 100 |
| 100% | 22.5 | 100 |

| Test acceptability | |
|---|---|
| Mean control reproduction 15/female | (yes) no |
| 3rd brood in 60% of surviving females | (yes) no |
| Control survival ≥80%: | (yes) no |

**Comments:**

*Day 0 is test initiation day

X = Dead (original organism)

Revised 09/21/06 MF/SP

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## Appendix B. (continued)

**FDEP Biology Laboratory - Chronic Screen Bench Sheet**

*Ceriodaphnia dubia* Parameters

Facility: Trademark Nitrogen

LIMS Sample #: 10853993 for lab sample 1

Circle µmhos-cm or mmhos-cm

| Test Day ↓ | | Concentration | New Sample: Test Replicate | pH (su) | Temp (°C) | DO (mg/L) | Conductivity (µmhos/cm) (mmhos/cm) | Old Sample: Test Replicate | pH (su) | Temp (°C) | DO (mg/L) | Conductivity (µmhos/cm) (mmhos/cm) |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Day 0 | sample: | Control | M | 8.2 | 25.1 | 7.86 | 181 | | | | | |
| | 1 | 100% | M | 8.3 | 24.0 | 8.3 | 526 | | | | | |
| | Measured by: | | | BA | BA | BA | BA | | | | | |
| | Recorded by: | | | BA | BA | BA | BA | | | | | |
| Day 1 | sample: | Control | M | 8.1 | 24.3 | 7.9 | 170 | M | 8.2 | 24.2 | 7.6 | 183 |
| | 1 | 100% | M | 8.1 | 24.8 | 8.4 | 518 | M | 8.5 | 24.2 | 7.4 | 510 |
| | Measured by: | | | MF | MF | MF | MF | | MF | MF | MF | MF |
| | Recorded by: | | | MF | MF | MF | MF | | MF | MF | MF | MF |
| Day 2 | sample: | Control | M | 8.3 | 25.0 | 7.6 | 174 | M | 8.1 | 24.5 | 7.3 | 175 |
| | 2 | 100% | M | 8.3 | 24.7 | 7.8 | 513 | M | 8.5 | 24.5 | 7.0 | 523 |
| | Measured by: | | | MF | MF | MF | MF | | MF | MF | MF | MF |
| | Recorded by: | | | MF | MF | MF | MF | | MF | MF | MF | MF |
| Day 3 | sample: | Control | M | 8.2 | 24.4 | 7.8 | 172 | M | 8.1 | 24.2 | 7.4 | 175 |
| | 2 | 100% | M | 8.2 | 25.1 | 8.3 | 523 | M | 8.4 | 24.3 | 7.7 | 520 |
| | Measured by: | | | MF | MF | MF | MF | | MF | MF | MF | MF |
| | Recorded by: | | | MF | MF | MF | MF | | MF | MF | MF | MF |
| Day 4 | sample: | Control | M | 8.2 | 24.8 | 7.7 | 185 | M | 8.1 | 24.9 | 7.2 | 184 |
| | 3 | 100% | M | 8.2 | 24.7 | 7.7 | 526 | M | 8.5 | 25.0 | 7.2 | 526 |
| | Measured by: | | | MF | MF | MF | MF | | MF | MF | MF | MF |
| | Recorded by: | | | MF | MF | MF | MF | | MF | MF | MF | MF |
| Day 5 | sample: | Control | M | 8.2 | 24.0 | 7.7 | 171 | M | 8.0 | 24.0 | 6.9 | 196 |
| | 3 | 100% | M | 8.3 | 24.0 | 8.2 | 512 | M | 8.1 | 24.0 | 7.1 | 515 |
| | Measured by: | | | BA | BA | BA | BA | | BA | BA | BA | BA |
| | Recorded by: | | | BA | BA | BA | BA | | BA | BA | BA | BA |
| Day 6 | sample: | Control | M | | | | | M | 8.1 | 24.0 | 7.6 | 184 |
| | 3 | 100% | M | | | | | M | 8.5 | 24.0 | 7.5 | 525 |
| | Measured by: | | | | | | | | BA | BA | BA | BA |
| | Recorded by: | | | | | | | | BA | BA | BA | BA |
| Day 7 | sample: | Control | M | Not renewed TJA 5-24-06 | | | | M | | | | |
| | 3 | | M | | | | | M | | | | |
| | Measured by: | | | | | | | | | | | |
| | Recorded by: | | | | | | | | | | | |
| Day 8 | sample: | Control | | | | | | M | | | | |
| | 3 | | | | | | | M | | | | |
| | Measured by: | | | | | | | | | | | |
| | Recorded by: | | | | | | | | | | | |

**Comments:**

Replicate M= "Metered" vial, a surrogate vial for parameters

Test Page 3 of 3 PAGE 000027 revised 08/24/06 MF/SP

## FDEP Biology Laboratory - Chronic Screen Bench Sheet

*Pimphales promelas or Menidia beryllina*

Facility: Trahanik Nitrogen Corp.
Address: 1216 Old Hopewell Rd
County: Hillsborough
NPDES Permit #: FL0000647
Contact/District: Pete Wenner / SW-Dist

Test Species (circle one)
(P. promelas) SOP#: TA07_09 EPA-821-R-02-013 Method 1000.0
M. beryllina SOP#: TA07_14 EPA-821-R-02-014 Method 1006.0

Start Date: 3.18.08 Time: 1230
End Date: 3-25-08 Time: 1030

Diluent/ Batch: MHW / 3.17.08 Test Days Used: 0-3
MHW / 3-21-08 Test Days Used: 3-Eff&Z
ASW / _____ Test Days Used: _____
ASW / _____ Test Days Used: _____

Organism age : <1 days
Feeding Prior to Test and Daily
Artemia Lot # : 651-536-90
Chamber size : 1000 ml
Test volume : 500 ml

| Artemia Batch # | | Day 3 | 52-08 |
|---|---|---|---|
| Day 0 | 49-08 | Day 4 | 53-08 |
| Day 1 | 50-08 | Day 5 | 54-08 |
| Day 2 | 51-08 | Day 6 | 55-08 |

| LIMS: | Date | Initials |
|---|---|---|
| Entry: | | |
| Verification: | 3-11-08 | BA |

| Sample Received | Lab Sample # | LIMS Job # | LIMS Sample # | Collection Date/Time | Received Date/Time |
|---|---|---|---|---|---|
| Test Day 0 | 1 | TLH-2008-03-18-03 | 08934 93 | 3-17-08/6:00 | 3-18-08/9:50 |
| Test Day 2 | 2 | TLH-2008-03-20-01 | 108 9869 | 3-18-08/10:30 | 3/20/08/10:15 |
| Test Day 4 | 3 | TLH-2008-03-21-03 | 109 0352 | 3-21-08/8:00 | 3/22/08/10:30 |

| Water Quality Parameters | Field Chlorine (mg/L) | | | Lab Chlorine (mg/L) | | | Alkalinity (mg/L as $CaCO_3$) | | | Hardness (mg/L as $CaCO_3$) | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Lab Sample #: | 1 | 2 | 3 | 1 | 2 | 3 | 1 | 2 | 3 | 1 | 2 | 3 |
| Sample: | 1523A | ~~BA~~ | ~~BA~~ | <0.03 | <0.03 | <0.03 | 200 | 203 | 200 | 245 | 245 | 243 |
| Diluent/Control: | | | | <0.03 | <0.03 | <0.03 | 67 | 81 | 81 | 106 | 147 | 147 |
| Method: | Not | tested | — | Hach | Hach | Hach | Hach | Hach | Hach | Hach | Hach | Hach |
| Measured by: | — | — | — | SJ | SJ | MF | BA | BA | BA | BA | BA | BA |
| Verified by: | BA | BA | BA | BA | BA | BA | MF | MF | MF | MF | MF | MF |

| | Conductivity (circle one) (µmhos-cm) mmhos-cm) | | | Ammonia (mg/L) | | | Salinity (ppth) | | |
|---|---|---|---|---|---|---|---|---|---|
| Lab Sample#: | 1 | 2 | 3 | 1 | 2 | 3 | 1 | 2 | 3 |
| Sample: | 527 | 528 | 524 | <0.017 | <0.017 | <0.017 | <1 | <1 | <1 |
| Diluent/Control: | 441 | 443 | 434 | <0.017 | <0.017 | <0.017 | <1 | <1 | <1 |
| Method: | Mettler | Mettler | Mettler | Denver | Denver | Denver | Mettler | Mettler | Mettler |
| Measured by: | BA/20 | BA/20 | MF | MF | MF | MF | BA/ | BA/20 | MF |
| Verified by: | MF | MF | BA | BA | BA | BA | MF | MF | BA |

Temperature Shipped ≤6° C (all samples) (yes) no
Holding Time ≤36 hours (yes) no
Light Intensity : 50 - 100 ft. candles
Photoperiod : 16 light 8 dark
Temperature Range °C:
Incubator # 2 min 25.7 max 27.2 mean 26.5
Water bath # ____ min _____ max _____ mean _____
Room B246 min 24.2 max 25.7 mean 25.0

Investigators' signatures
James Davis
Bill Lee
Marshall Faircloth

Reviewer: Dottie C. Wolfe

| Statistics | |
|---|---|
| Control %cv: | 1.76% |
| Dunnetts PMSD: | 4.4 |
| Survival NOEC: | 100% |
| Survival LOEC: | >100% |
| Growth NOEC: | 100% |
| Growth LOEC: | >100% |

Comments:

## Appendix B. (continued)

**FDEP Biology Laboratory - Chronic Screen Bench Sheet**

*Pimphales promelas or Menidia beryllina* Survival

Facility: ~~Lee County~~ Trademark Nitrogen Corporation J.D. 108393 BA

Organism: (*P. promelas*) *M.beryllina* (circle one)

LIMS Sample Number: 1040352 for lab sample #1

Start Date: 3.18.08 Time: 1230

End Date: 3-25-08 Time: 1030

| Concentration | Replicate | | TEST DAY 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 |
|---|---|---|---|---|---|---|---|---|---|---|
| Control | A | Z9 | 10 | 10 | 9 10 | 9 | 9 | 9 | 9 | 9 |
| | B | Z10 | 10 | 10 | 9 10 | 9 | 9 | 9 | 9 | 9 |
| | C | Z11 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 |
| | D | Z12 | 10 | 9 10 | 9 | 9 | 9 | 9 | 9 | 9 |
| % sample concentration | A | Z13 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 |
| | B | Z14 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 |
| | C | Z15 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 |
| | D | Z16 | 10 | 9 10 | 9 | 9 | 9 | 9 | 9 | 9 |
| Control | A | | 10 | | | | | | | |
| | B | | 10 | | | | | | | |
| | C | | 10 | | | | | | | |
| | D | | 10 | | | | | | | |
| % sample concentration | A | | 10 | | | | | | | |
| | B | | 10 | | | | | | | |
| | C | | 10 | | | | | | | |
| | D | | 10 | | | | | | | |
| Loaded by: JD | Checked by: | | | JD | JD | JD | BA | JD | JD | MF |
| Verified by: BA | | | | | | | | | | |

Comments:

Test Page 2 of 4

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revised 08/24/06 MF/SP

## Appendix B. (continued)

**Biology Laboratory Chronic Screening Bioassay Benchsheet**

Parameters

*P. promelas* SOP TA-07_09 *M. beryllina* SOP TA-07_14

Sample Source: Trademark Nitrogen Corporation

LIMS Sample #: ~~1090352~~ 1089393 (for Lab sample #1)

| Test Day ↓ | | Conc. | Old Sample: Test Rep. | pH (su) | Temp (°C) | DO (mg/L) | Cond (µmhos/cm) (mmhos/cm) | Salinity ppth | New Sample: Test Rep. | pH (su) | Temp (°C) | DO (mg/L) | Cond (µmhos/cm) (mmhos/cm) | Salinity ppth |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Day 0 | sample: | Control | | | | | | | A | 6.4 | 25.2 | 8.1 | 441 | <1 |
| | 1 | 100% | | | | | | | A | 8.3 | 24.3 | 8.4 | 534 | <1 |
| | Measured by: | | | | | | | | | JD | JD | JD | JD | JD |
| | Recorded by: | | | | | | | | | JD | JD | JD | JD | JD |
| Day 1 | sample: | Control | B | 6.8 | 24.5 | 6.4 | 445 | <1 | B | 6.9 | 24.0 | 7.2 | 440 | <1 |
| | 1 | 100% | B | 8.4 | 24.3 | 6.8 | 525 | <1 | B | 8.4 | 24.0 | 7.6 | 526 | <1 |
| | Measured by: | | | JD | JD | JD | JD | JD | | JD | JD | JD | JD | JD |
| | Recorded by: | | | JD | JD | JD | JD | JD | | JD | JD | JD | JD | JD |
| Day 2 | sample: | Control | C | 6.4 | 24.0 | 5.8 | 456 | <1 | C | 6.3 | 24.0 | 7.3 | 455 | <1 |
| | 2 | 100% | C | 8.4 | 24.0 | 6.0 | 542 | <1 | C | 8.4 | 24.0 | 7.3 | 529 | <1 |
| | Measured by: | | | JD | JD | JD | JD | JD | | JD | JD | JD | JD | JD |
| | Recorded by: | | | JD | JD | JD | JD | JD | | JD | JD | JD | JD | JD |
| Day 3 | sample: | Control | D | 6.4 | 24.0 | 6.2 | 449 ~~449~~ JD | <1 | D | 7.4 | 24.0 | 7.3 | 443 | <1 |
| | 2 | 100% | D | 8.1 | 24.0 | 6.2 | JD 538 | <1 | D | 8.3 | 24.2 | 7.2 | 540 | <1 |
| | Measured by: | | | JD | JD | JD | JD | JD | | JD | JD | JD | JD | JD |
| | Recorded by: | | | JD | JD | JD | JD | JD | | JD | JD | JD | JD | JD |
| Day 4 | sample: | Control | A | 6.8 | 24.0 | 5.4 | 481 | <1 | A | 7.3 | 24.0 | 7.5 | 395 | <1 |
| | 3 | 100% | A | 8.1 | 24.0 | 5.6 | 550 | <1 | A | 8.2 | 24.0 | 7.4 | 523 | <1 |
| | Measured by: | | | BA | BA | BA | BA | BA | | BA | BA | BA | BA | BA |
| | Recorded by: | | | BA | BA | BA | BA | BA | | BA | BA | BA | BA | BA |
| Day 5 | sample: | Control | B | 7.4 | 24.1 | 4.5 | 446 | <1 | B | 7.3 | 24.0 | 7.5 | 434 | <1 |
| | 3 | 100% | B | 8.1 | 24.1 | 4.7 | 543 | <1 | B | 8.2 | 24.0 | 7.3 | 530 | <1 |
| | Measured by: | | | JD | JD | JD | JD | JD | | JD | JD | JD | JD | JD |
| | Recorded by: | | | JD | JD | JD | JD | JD | | JD | JD | JD | JD | JD |
| Day 6 | sample: | Control | C | 7.6 | 24.2 | 5.7 | 407 | <1 | C | 7.9 | 24.0 | 7.4 | 345 | <1 |
| | 3 | 100% | C | 8.2 | 24.4 | 5.7 | 542 | <1 | C | 8.2 | 24.2 | 7.4 | 533 | <1 |
| | Measured by: | | | JD | JD | JD | JD | JD | | JD | JD | JD | JD | JD |
| | Recorded by: | | | JD | JD | JD | JD | JD | | JD | JD | JD | JD | JD |
| Day 7 | sample: | Control | D | 7.5 | 24.0 | 6.1 | 362 | <1 | | | | | | |
| | 3 | 100% | D | 8.2 | 24.0 | 6.2 | 546 | <1 | | | | | | |
| | Measured by: | | | BA | BA | BA | BA | BA | | | | | | |
| | Recorded by: | | | BA | BA | BA | BA | BA | | | | | | |

Test Rep. = Replicate | Record Salinity < 1 if freshwater | Circle µmhos-cm or mmhos-cm

Comments:

revised 6/24/05 mf

test page 3 of 4

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## Appendix B. (continued)

### FDEP Biology Laboratory - Chronic Screen Bench Sheet

Facility: Trademark Nitrogen

LIMS Sample Number: 089393 for lab sample #1

| Concentration | Replicate | Pan # | Pan wt. (g) | Pan + Fish (g) | Fish wt. (g) | Original # Fish | Mean wt. (mg) |
|---|---|---|---|---|---|---|---|
| | | | mean wt = fish wt divided by 10<br>convert g to mg: move decimal 3 places to right | | | | |
| Control | A | 9 | 0.01653 | 0.02204 | 0.00551 | 10 | 0.551 |
| | B | 10 | 0.01906 | 0.02473 | 0.00567 | 10 | 0.567 |
| | C | 11 | 0.01888 | 0.02445 | 0.00557 | 10 | 0.557 |
| | D | 12 | 0.01892 | 0.02436 | 0.00574 | 10 | 0.574 |
| 100 % sample | A | 13 | 0.01597 | 0.02177 | 0.00580 | 10 | 0.580 |
| | B | 14 | 0.01971 | 0.02565 | 0.00594 | 10 | 0.594 |
| | C | 15 | 0.01925 MF | 0.02484 | 0.00559 | 10 | 0.559 |
| | D | 16 | 0.01810 | 0.02351 | 0.00541 | 10 | 0.541 |
| Control | A | | | | | 10 | |
| | B | | | | | 10 | |
| | C | | | | | 10 | |
| | D | | | | | 10 | |
| % sample | A | | | | | 10 | |
| | B | | | | | 10 | |
| | C | | | | | 10 | |
| | D | | | | | 10 | |
| | Weighed by initials: | | MF | MF | | | |

| Growth | %CV | PMSD |
|---|---|---|
| Control | 1.75% | 4.14% |

| Growth | Table | Calculated | dF |
|---|---|---|---|
| T-test | 1.94310 | 0.56850 | 6 |
| Survival T-test | 1.94310 | -1.41421 | 6 |

| Drying 630f Sediment overnight | |
|---|---|
| Temperature °C: | 60°C |
| Hours: | 24 |

| Tare Weighing Date: 3-25-08 MF | Final Weighing Date: | | | Measured Value | | S-weight Value |
|---|---|---|---|---|---|---|
| Tare Pan 1 after Pan 5 | MF 0.01754 @ | 0.01757 | S-weights | 0.00998 | @ | 0.01000 |
| Tare Pan 1 after Pan 8 | 0.01755 @ | 0.01757 | at tare | 0.03000 | @ | 0.03000 |
| Final Pan 1 after Pan 5 | 0.02205 @ | 0.02204 | S-weights | 0.01000 | @ | 0.01000 |
| Final Pan 1 after Pan 8 | 0.02205 @ | 0.02204 | at final | 0.03000 | @ | 0.03000 |

Comments: 92.5% control survival 92.5% survival in 100% sample MF 3-27-08
mean tare weight = 0.552 mg dry wt/organism (0.555)
MF mean 100% sample weight = MF 0.569 mg MF dry wt/organism

Test Page 4 of 4 PAGE 000040 revised 08/24/06 MF/SP

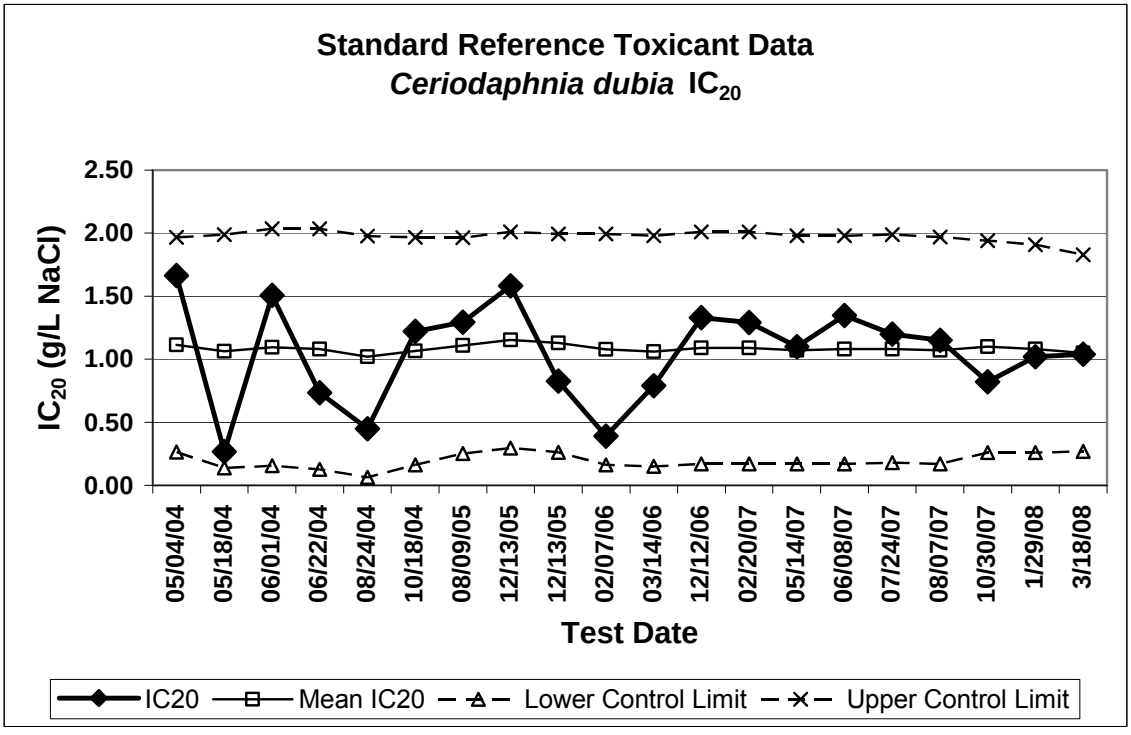
## Appendix C. SRT Bench Sheets and Statistical Analysis

Florida Department of Environmental Protection
Bureau of Laboratories

Standard Reference Toxicant (SRT) Test Data - 7-Day Chronic Toxicity
*Ceriodaphnia dubia* (water flea) - Sodium Chloride (NaCl)

| Date | IC20 | Mean IC20 | Std Dev | Lower Control Limit | Upper Control Limit |
|---|---|---|---|---|---|
| 05/04/04 | 1.66 | 1.12 | 0.43 | 0.26 | 1.97 |
| 05/18/04 | 0.27 | 1.06 | 0.46 | 0.14 | 1.99 |
| 06/01/04 | 1.51 | 1.10 | 0.47 | 0.15 | 2.04 |
| 06/22/04 | 0.73 | 1.08 | 0.48 | 0.13 | 2.03 |
| 08/24/04 | 0.45 | 1.02 | 0.48 | 0.06 | 1.98 |
| 10/18/04 | 1.22 | 1.07 | 0.45 | 0.16 | 1.97 |
| 11/16/04 | 1.29 | 1.11 | 0.43 | 0.25 | 1.96 |
| 08/09/05 | 1.58 | 1.15 | 0.43 | 0.30 | 2.01 |
| 12/13/05 | 0.83 | 1.13 | 0.43 | 0.26 | 1.99 |
| 02/07/06 | 0.39 | 1.08 | 0.46 | 0.16 | 1.99 |
| 03/14/06 | 0.79 | 1.06 | 0.46 | 0.15 | 1.98 |
| 12/12/06 | 1.33 | 1.09 | 0.46 | 0.17 | 2.01 |
| 02/20/07 | 1.29 | 1.09 | 0.46 | 0.17 | 2.01 |
| 05/14/07 | 1.10 | 1.07 | 0.45 | 0.17 | 1.98 |
| 06/08/07 | 1.35 | 1.08 | 0.45 | 0.17 | 1.98 |
| 07/24/07 | 1.20 | 1.08 | 0.45 | 0.18 | 1.99 |
| 08/07/07 | 1.15 | 1.07 | 0.45 | 0.17 | 1.97 |
| 10/30/07 | 0.82 | 1.10 | 0.42 | 0.26 | 1.94 |
| 1/29/08 | 1.02 | 1.08 | 0.41 | 0.26 | 1.91 |
| 3/18/08 | 1.04 | 1.05 | 0.39 | 0.27 | 1.83 |

Means are of the last 20 SRT tests.
Lower Confidence Limit = mean - 2 * Std Dev
Upper Confidence Limit = mean + 2 * Std Dev



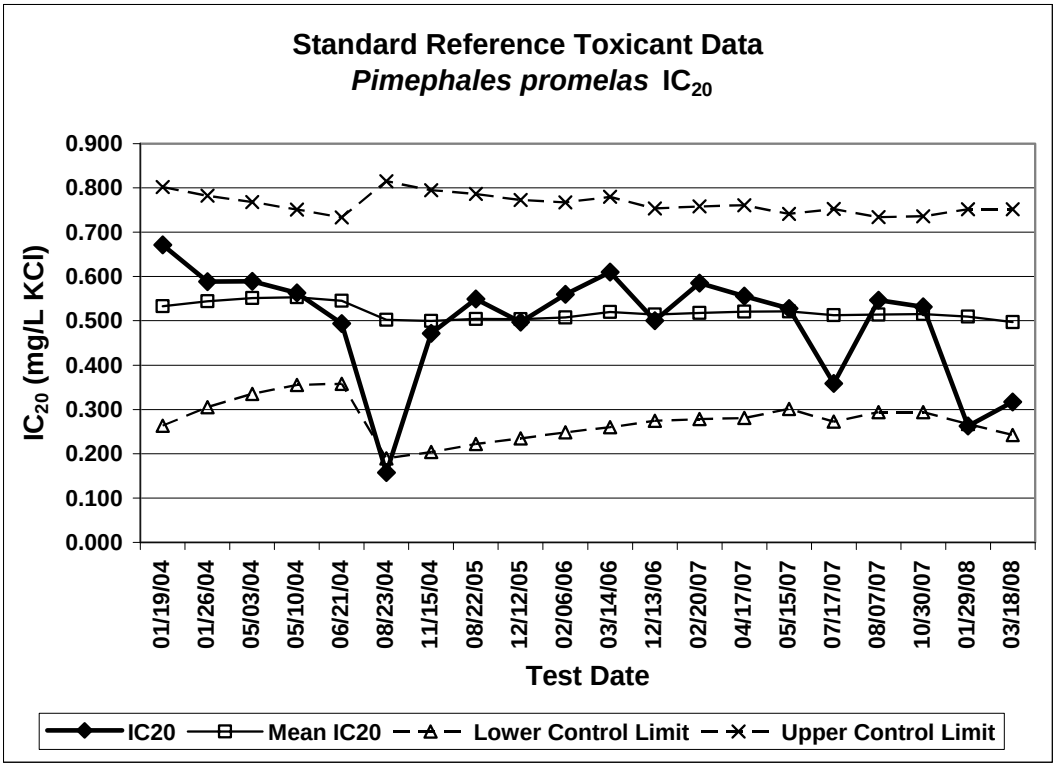
## Appendix C. (continued)

Florida Department of Environmental Protection
Bureau of Laboratories

Standard Reference Toxicant (SRT) Test Data - 7-Day Chronic Toxicity
*Pimephales promelas* (fathead minnow) - Potassium Chloride (KCl)

| Date | IC20 | Mean IC20 | Std Dev | Lower Control Limit | Upper Control Limit |
|---|---|---|---|---|---|
| 01/19/04 | 0.671 | 0.533 | 0.13 | 0.263 | 0.802 |
| 01/26/04 | 0.588 | 0.544 | 0.12 | 0.305 | 0.782 |
| 05/03/04 | 0.589 | 0.551 | 0.11 | 0.335 | 0.768 |
| 05/10/04 | 0.563 | 0.553 | 0.10 | 0.355 | 0.751 |
| 06/21/04 | 0.494 | 0.546 | 0.09 | 0.358 | 0.734 |
| 08/23/04 | 0.157 | 0.503 | 0.16 | 0.190 | 0.815 |
| 11/15/04 | 0.472 | 0.499 | 0.15 | 0.204 | 0.795 |
| 08/22/05 | 0.549 | 0.504 | 0.14 | 0.222 | 0.786 |
| 12/12/05 | 0.497 | 0.503 | 0.13 | 0.234 | 0.772 |
| 02/06/06 | 0.559 | 0.508 | 0.13 | 0.248 | 0.767 |
| 03/14/06 | 0.610 | 0.520 | 0.13 | 0.260 | 0.780 |
| 12/13/06 | 0.501 | 0.514 | 0.12 | 0.274 | 0.754 |
| 02/20/07 | 0.585 | 0.518 | 0.12 | 0.278 | 0.758 |
| 04/17/07 | 0.556 | 0.521 | 0.12 | 0.281 | 0.761 |
| 05/15/07 | 0.528 | 0.521 | 0.11 | 0.301 | 0.741 |
| 07/17/07 | 0.358 | 0.513 | 0.12 | 0.273 | 0.753 |
| 08/07/07 | 0.547 | 0.514 | 0.11 | 0.294 | 0.734 |
| 10/30/07 | 0.531 | 0.515 | 0.11 | 0.294 | 0.736 |
| 01/29/08 | 0.263 | 0.509 | 0.12 | 0.267 | 0.752 |
| 03/18/08 | 0.317 | 0.497 | 0.13 | 0.242 | 0.752 |

Means are from all SRT tests, up to 20 tests..
Lower Confidence Limit = mean - 2 * Std Dev
Upper Confidence Limit = mean + 2 * Std Dev



## FDEP Biology Laboratory - Chronic SRT Bench Sheet

*Ceriodaphnia dubia*

Diluent/ Batch: 20% DMW / 3-10-08 Test Days Used: 0-3
20% DMW / 3-12-08 Test Days Used: 4-5
Toxicant/ Batch: NaCl / 3-17-08 3ppt

SOP # : TA 07_05
EPA-821-R-02-013 Method 1002.0

Organism: *Ceriodaphnia dubia*
Organism Batch #/ Age: 11-08 / <24 hr.
Feeding: Prior to Test and Daily
Food: YCT / *P. subcapitata*
Food Batch: 7-26 Bot 1-31-08
Chamber size: 30 mL
Test Volume: 20 mL

Start Date: 3.18.08 Time: 1330 1345
End Date: 3-24-08 Time: 14:15

| Water Quality Parameters | 20% DMW 3-10-08 | Diluent (or other) | Method | Measured by: | Verified by: |
|---|---|---|---|---|---|
| Lab Total Residual $Cl_2$ (mg/L): | <0.03 | <0.03 | HACH | JD | MF |
| Alkalinity (mg/L as $CaCO_3$): | 87 | 94 | HACH | BA | MF |
| Hardness (mg/L as $CaCO_3$): | 84 | 94 | HACH | BA | MF |
| Total Ammonia (mg/L as N): | <0.017 | <0.017 | DENVER | MF | BA |
| Conductivity (umhos-cm) mmhos-cm : | 175 | 188 | YSI (Mettler) | MF | BA |

20% DMW 3-12-08

| Results | |
|---|---|
| Control % CV: | 15.7 |
| Dunnetts PMSD: | 16.9 |
| Reproduction NOEC: | 0.89 ppth |
| Reproduction LOEC: | 1.3 ppth |
| Survival NOEC: | 1.3 |
| Survival LOEC: | 2 |
| $ICP_{20}$: | 1.04 |
| $ICP_{25}$: | 1.11 |

| Day | Concentrations prepared by | Counted by |
|---|---|---|
| 0 | MF | MF/BA |
| 1 | MF | MF |
| 2 | MF | MF |
| 3 | MF | MF |
| 4 | MF | MF |
| 5 | MF | BA |
| 6 | — | BA |
| 7 | — | — |
| 8 | — | - |

| Test Replicate | Loaded from culture vial |
|---|---|
| 1 | A2 |
| 2 | A4 |
| 3 | A7 |
| 4 | A14 |
| 5 | A17 |
| 6 | A22 |
| 7 | A27 |
| 8 | A28 |
| 9 | B10 |
| 10 | B15 |

**Comments:**

Test terminated at Day 6 because control survival and reproduction criteria was met. MF 3-24-08

**Investigators' signatures**

Marshall Faircloth

Loretta C. Wolfe
REVIEWER

Light Intensity : 50 - 100 ft. candles
Photoperiod : 16 light 8 dark
Temperature Range °C:
Incubator # 2 min. 25.7 max. 27.2 mean 26.5
Incubator # ____ min. _____ max. _____ mean _____
Room B246 min. 24.2 max. 25.7 mean 25.0

## Appendix C. (continued)

*Ceriodaphnia dubia* Survival and Reproduction

Start Date: 3-18-08 Time: 13:30 Loaded by: BA
End Date: 3-24-08 Time: 14:15 Verified by: [illegible]

| Concentration | Day | Replicate Number 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | Total neonates | Live adults | Neonates per adult |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Control | 0 | | | | | | | | | | | | 10 | |
| | 1 | | | | | | | | | | | | 10 | |
| | 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | | 10 | |
| | 3 | 0 | 4 | 0 | 4 | 0 | 0 | 0 | 0 | 0 | 0 | 8 | 10 | 0.8 |
| | 4 | 4 | 0 | 2 | 0 | 0 | 4 | 5 | 5 | 3 | 4 | 27 | 10 | 2.7 |
| | 5 | 5 | 8 | 8 | 9 | 8 | 10 | 9 | 10 | 10 | 9 | 86 | 10 | 8.6 |
| | 6 | 12 | 11 | 13 | 9 | 10 | 14 | 13 | 16 | 10 | 13 | 121 | 10 | 12.1 |
| | 7 | | | | | | | | | | | | | |
| | 8 | | | | | | | | | | | | | |
| | Total | 21 | 23 | 23 | 22 | 18 | 28 | 27 | 31 | 23 | 26 | 242 | 10 | 24.2 |
| 0.59 ppth | 0 | | | | | | | | | | | | 10 | |
| | 1 | | | | | | | | | | | | 10 | |
| | 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 10 | 0 |
| | 3 | 0 | 3 | 0 | 2 | 0 | 0 | 0 | 0 | 0 | 0 | 5 | 10 | 0.5 |
| | 4 | 5 | 1 | 2 | 0 | 3 | 6 | 5 | 3 | 4 | 3 | 32 | 10 | 3.2 |
| | 5 | 11 | 9 | 8 | 9 | 9 | 9 | 9 | 8 | 9 | 8 | 89 | 10 | 8.9 |
| | 6 | 14 | 9 | 10 | 12 | 13 | 8 | 14 | 12 | 13 | 10 | 115 | 10 | 11.5 |
| | 7 | | | | | | | | | | | | | |
| | 8 | | | | | | | | | | | | | |
| | Total | 30 | 22 | 20 | 23 | 25 | 23 | 28 | 23 | 26 | 21 | 241 | 10 | 24.1 |
| 0.89 ppth | 0 | | | | | | | | | | | | 10 | |
| | 1 | | | | | | | | | | | | 10 | |
| | 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | — | 10 | — |
| | 3 | 0 | 0 | 0 | 4 | 0 | 0 | 0 | 0 | 0 | 0 | 4 | 10 | 0.4 |
| | 4 | 4 | 3 | 4 | 0 | 4 | 3 | 5 | 3 | 3 | 5 | 34 | 10 | 3.4 |
| | 5 | 8 | 7 | 8 | 8 | 7 | 11 | 8 | 5 | 7 | 7 | 76 | 10 | 7.6 |
| | 6 | 10 | 11 | 11 | 12 | 12 | 10 | 12 | 9 | 10 | 12 | 109 | 10 | 10.9 |
| | 7 | | | | | | | | | | | | | |
| | 8 | | | | | | | | | | | | | |
| | Total | 22 | 21 | 23 | 24 | 23 | 24 | 25 | 17 | 20 | 24 | 223 | 10 | 22.3 |

**Comments:**

## Appendix C. (continued)

**FDEP Biology Laboratory - Chronic SRT Bench Sheet**

*Ceriodaphnia dubia* Survival and Reproduction

| Concentration | Day | Replicate Number 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | Total neonates | Live adults | Neonates per adult |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 1.3 ppth | 0 | | | | | | | | | | | | 10 | |
| | 1 | | | | | | | | | | | | 10 | |
| | 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 10 | 0 |
| | 3 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 10 | 0 |
| | 4 | 3 | 2 | 3 | 0 | 2 | 4 | 3 | 3 | 4 | 4 | 28 | 10 | 2.8 |
| | 5 | 0 | 6 | 6 | D | 4 | 10 | 3 | 8 | 6 | 7 | 50 | 10 | 5.0 |
| | 6 | 0 | 10 | 10 | 0* | 8 | 1 | 9 | 11 | 9 | 9 | 67 | 10 | 6.7 |
| | 7 | | | | | | | | | | | | | |
| | 8 | | | | | | | | | | | | | |
| | Total | 3 | 18 | 19 | 0 | 14 | 15 | 15 | 22 | 19 | 20 | 145 | 10 | 14.5 |
| 2.0 ppth | 0 | | | | | | | | | | | | 10 | |
| | 1 | | | | | | | | | | | | 10 | |
| | 2 | 0 | X | 0 | 0 | X | 0 | X | 0 | 0 | 0 | 0 | 7 | 0 |
| | 3 | 0 | \| | X | 0 | \| | 0 | \| | 0 | 0 | 0 | 0 | 6 | 0 |
| | 4 | 1 | \| | \| | 0 | \| | 0 | \| | 0 | 0 | 0 | 1 | 6 | 0.16 |
| | 5 | 1 | \| | \| | 0 | \| | 0 | \| | 1 | 1 | 0 | 3 | 6 | 0.5 |
| | 6 | 0 | \| | \| | †♂BA | \| | †♂BA | \| | 0 | ♂ BA | 0 | 0 | 6 | 0 |
| | 7 | | \| | \| | \| | \| | \| | \| | \| | \| | \| | \| | \| | |
| | 8 | | \| | \| | \| | \| | \| | \| | \| | \| | \| | \| | \| | |
| | Total | 2 | \| | \| | 0 | \| | 0 | \| | 1 | 1 | 0 | 4 | 10 | 0.4 |
| 3.0 ppth | 0 | | | | | | | | | | | | 10 | |
| | 1 | | | | | | | | | | | | 10 | |
| | 2 | X | X | X | X | X | X | X | X | X | X | | 0 | |
| | 3 | | | | | | | | | | | | | |
| | 4 | | | | | | | | | | | | | |
| | 5 | | | | | | | | | | | | | |
| | 6 | | | | | | | | | | | | | |
| | 7 | | | | | | | | | | | | | |
| | 8 | | | | | | | | | | | | | |
| | Total | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |

**Comments:** 3-24-08 1.3 ppth rep. 4 aborted ME eggs - MF

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## Appendix C. (continued)

**FDEP Biology Laboratory - Chronic SRT Bench Sheet**

*Ceriodaphnia dubia* Parameters

| Test Day ↓ | Conc. | New Sample Replicate | pH (su) | Temp (°C) | DO (mg/L) | Conductivity (µmhos/cm) (mmhos/cm) | Old Sample Replicate | pH (su) | Temp (°C) | DO (mg/L) | Conductivity (µmhos/cm) (mmhos/cm) |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Day 0 | Control | M | 8.2 | 25.2 | 7.6 | 0.175 | | | | | |
| | 0.59 ppth | M | 8.1 | 25.2 | 7.8 | 1.34 | | | | | |
| | 0.89 ppth | M | 8.1 | 25.2 | 7.8 | 1.93 | | | | | |
| | 1.3ppth | M | 8.1 | 25.2 | 7.8 | 2.75 | | | | | |
| | 2.0 ppth | M | 8.1 | 24.9 | 7.8 | 3.98 | | | | | |
| | 3.0 ppth | M | 8.1 | 24.9 | 7.7 | 5.82 | | | | | |
| | Measured by: | | BA | BA | BA | BA | | | | | |
| | Recorded by: | | BA | BA | BA | BA | | | | | |
| Day 1 | Control | M | 8.1 | 24.3 | 7.9 | 0.173 | M | 8.2 | 24.8 | 7.2 | 0.195 |
| | 0.59 ppth | M | 8.1 | 24.2 | 7.7 | 1.290 | M | 8.1 | 24.2 | 7.2 | 1.39 |
| | 0.89 ppth | M | 8.1 | 24.4 | 7.7 | 1.87 | M | 8.1 | 24.1 | 7.2 | 1.97 |
| | 1.3ppth | M | 8.1 | 24.4 | 7.7 | 2.70 | M | 8.1 | 24.1 | 7.2 | 2.80 MF |
| | 2.0 ppth | M | 8.1 | 24.4 | 7.6 | 3.92 | M | 8.1 | 24.2 | 7.2 | 4.0 |
| | 3.0 ppth | M | 8.1 | 24.4 | 7.6 | 5.74 | M | 8.1 | 24.2 | 7.1 | 5.79 |
| | Measured by: | | MF | MF | MF | MF | | MF | MF | MF | MF |
| | Recorded by: | | MF | MF | MF | MF | | MF | MF | MF | MF |
| Day 2 | Control | M | 8.3 | 25.0 | 7.6 | 0.175 | M | 8.2 | 24.5 | 7.3 | 0.173 |
| | 0.59 ppth | M | 8.2 | 25.5 | 7.6 | 1.39 | M | 8.1 | 24.5 | 7.2 | 1.32 |
| | 0.89 ppth | M | 8.1 | 25.4 | 7.5 | 1.93 | M | 8.1 | 24.5 | 7.3 | 1.89 |
| | 1.3ppth | M | 8.1 | 25.4 | 7.5 | 2.73 | M | 8.1 | 24.3 | 7.3 | 2.81 |
| | 2.0 ppth | M | 8.1 | 25.3 | 7.5 | 3.94 | M | 8.0 | 24.3 | 7.3 | 3.95 |
| | 3.0 ppth | M | — | — | — | — | M | 8.0 | 24.4 | 7.2 | 5.81 |
| | Measured by: | | MF | MF | MF | MF | | MF | MF | MF | MF |
| | Recorded by: | | MF | MF | MF | MF | | MF | MF | MF | MF |
| Day 3 | Control | M | 8.2 | 7.8 (A) | 25.5 | 0.170 | M | 8.0 | 24.1 | 7.4 | 0.172 |
| | 0.59 ppth | M | 8.1 | 7.8 (A) | 24.4 | 1.33 | M | 7.9 | 24.3 | 7.4 | 1.39 |
| | 0.89 ppth | M | 8.1 | 7.9 (A) | 24.7 | 1.91 | M | 7.9 | 24.3 | 7.4 | 1.94 |
| | 1.3ppth | M | 8.1 | 7.8 (A) | 24.6 | 2.69 | M | 7.9 | 24.3 | 7.4 | 2.76 |
| | 2.0 ppth | M | 8.1 | 7.7 (A) | 24.7 | 3.86 | M | 7.9 | 24.4 | 7.4 | 3.91 |
| | 3.0 ppth | M | — | — | — | — | | — | — | — | — |
| | Measured by: | | MF | MF | MF | MF | | MF | MF | MF | MF |
| | Recorded by: | | MF | MF | MF | MF | | MF | MF | MF | MF |

**Comments:** (A) MF 3-21-08 Recorded DO interpolation and Temperature in DO spaces.

M= "Metered" vial, a surrogate vial for parameters

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## Appendix C. (continued)

**FDEP Biology Laboratory - Chronic SRT Bench Sheet**

*Ceriodaphnia dubia* Parameters

| Test Day ↓ | Conc. | New Sample Replicate | pH (su) | Temp (°C) | DO (mg/L) | Conductivity (µmhos/cm) (mmhos/cm) | Old Sample Replicate | pH (su) | Temp (°C) | DO (mg/L) | Conductivity (µmhos/cm) (mmhos/cm) |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Day 4 | Control | M | 8.2 | 24.8 | 7.6 | 0.188 | m | 8.1 | 25.0 | 7.4 | 0.190 |
| | 0.59 ppth | M | 8.1 | 25.2 | 7.6 | 1.33 | m | 8.1 | 24.8 | 7.3 | 1.36 |
| | 0.89 ppth | M | 8.1 | 25.2 | 7.3 | 1.90 | m | 8.1 | 24.8 | 7.1 | 1.92 |
| | 1.3ppth | M | 8.0 | 25.3 | 7.3 | 2.71 | m | 8.0 | 24.8 | 7.1 | 2.87 |
| | 2.0 ppth | M | 8.0 | 25.4 | 7.4 | 3.87 | m | 8.0 | 24.8 | 7.1 | 3.85 |
| | 3.0 ppth | M | — | — | — | — | — | — | — | — | — |
| | Measured by: | | MF | MF | MF | MF | | MF | MF | MF | MF |
| | Recorded by: | | MF | MF | MF | MF | | MF | MF | MF | MF |
| Day 5 | Control | M | 8.2 | 24.0 | 7.5 | 0.179 | A | 8.1 | 24.0 | 6.9 | 0.195 |
| | 0.59 ppth | M | 8.1 | 24.7 | 7.7 | 1.36 | A | 8.1 | 24.0 | 6.9 | 1.38 |
| | 0.89 ppth | M | 8.1 | 24.8 | 7.6 | 1.92 | A | 8.1 | 24.0 | 7.0 | 1.98 |
| | 1.3ppth | M | 8.1 | 24.8 | 7.6 | 2.71 | A | 8.1 | 24.0 | 7.1 | 2.9 |
| | 2.0 ppth | M | 8.1 | 24.9 | 7.6 | 3.91 | A | 8.1 | 24.0 | 7.1 | 3.82 |
| | 3.0 ppth | M | — | — | — | — | A | — | — | — | — |
| | Measured by: | | BA | BA | BA | BA | | BA | BA | BA | BA |
| | Recorded by: | | BA | BA | BA | BA | | BA | BA | BA | BA |
| Day 6 | Control | M | | | | | M | 8.1 | 24.0 | 7.3 | 0.182 |
| | 0.59 ppth | M | | | | | M | 8.1 | 24.0 | 7.3 | 1.42 |
| | 0.89 ppth | M | | | | | M | 8.1 | 24.0 | 7.2 | 01.95 |
| | 1.3ppth | M | | | | | M | 8.1 | 24.0 | 7.1 | 2.81 |
| | 2.0 ppth | M | | | | | M | 8.1 | 24.0 | 7.1 | 3.99 |
| | 3.0 ppth | M | | | | | M | — | — | — | — |
| | Measured by: | | | | | | | BA | BA | BA | BA |
| | Recorded by: | | | | | | | BA | BA | BA | BA |
| Day 7 | Control | M | | | | | | | | | |
| | 0.59 ppth | M | | | | | | | | | |
| | 0.89 ppth | M | | | | | | | | | |
| | 1.3ppth | M | | | | | | | | | |
| | 2.0 ppth | M | | | | | | | | | |
| | 3.0 ppth | M | | | | | | | | | |
| | Measured by: | | | | | | | | | | |
| | Recorded by: | | | | | | | | | | |
| Day 8 | Control | M | | | | | | | | | |
| | 0.59 ppth | M | | | | | | | | | |
| | 0.89 ppth | M | | | | | | | | | |
| | 1.3ppth | M | | | | | | | | | |
| | 2.0 ppth | M | | | | | | | | | |
| | 3.0 ppth | M | | | | | | | | | |
| | Measured by: | | | | | | | | | | |
| | Recorded by: | | | | | | | | | | |

Not renewed BA 3-25-06

Replicate M= "Metered" vial, a surrogate vial for parameters

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## Appendix C. (continued)

### FDEP Biology Laboratory - Chronic SRT Benchsheet

*Pimephales promelas* or *Menidia beryllina*

Test Species (circle one)

| | | | |
|---|---|---|---|
| (*P. promelas*) | SOP#: TA07_09 | EPA-821-R-02-013 | Method 1000.0 |
| *M. beryllina* | SOP#: TA07_14 | EPA-821-R-02-014 | Method 1006.0 |

Diluent/ Batch: Mod. Hard 3.17.08 Test Days Used: 0-3 Start Date: 3.18.08 Time: 1305

Diluent/ Batch: MHW 3-21-08 Test Days Used: 3-7 End Date: 3-25-08 Time: 1130

Toxicant/ Batch # : KCL / 3.17.08 Organism Batch #/ Age: 3-15-08 / <24hrs

Artemia Lot # : 384384256R Artemia lot # GSL-536-90 used Day 2,

| | | |
|---|---|---|
| Artemia Batch # | Day 2 51-08 | Day 5 54-08 |
| Day 0 49-08 | Day 3 52-08 | Day 6 55-08 |
| Day1 50-08 | Day 4 53-08 | |

Chamber size : 1000 ml

Test volume : 500 ml

Feeding: *P. promelas* 0.15 g wet wt. 2X day 6 hour interval

*M. beryllina* 0.10 g wet wt days 0-2 0.15g wet wt. days 3-7

| Water Quality Parameters | Diluent 3-17-08 | Diluent 3-21-08 (or other) | Method | Measured by | Verified by |
|---|---|---|---|---|---|
| Total Residual $Cl_2$ (mg/L): | <0.03 | <0.03 | HACH | SD | BA |
| Alkalinity (mg/L as $CaCO_3$): | 67 | 81 | HACH | BA | MF |
| Hardness (mg/L as $CaCO_3$): | 106 | 147 | HACH | BA | MF |
| Total Ammonia (mg/L as N): | <0.07 | <0.07 | DENVER | MF | BA |
| Conductivity µmhos-cm (mmhos-cm) | 0.440 | 0.406 | METTLER | SD | BA |

| Statistics | |
|---|---|
| Growth NOEC: | 0.25 g/L |
| Growth LOEC: | 0.50 g/L |
| Growth $IC_{20}$: | 0.317 g/L |
| Growth $IC_{25}$: | 0.336 g/L |
| Ctrl % CV: | 6.89% |
| Dunnetts PMSD: | 21.2% |
| Survival NOEC: | 0.25 g/L |
| Survival LOEC: | 0.5 g/L |

Light Intensity : 50 - 100 ft. candles

Photoperiod : 16 light 8 dark

Temperature Range °C:

Incubator # 3 min. 25.1 max. 26.7 mean 25.9

Water bath # ____ min. _____ max. _____ mean _____

Room B246 min. 24.2 max. 25.7 mean 25.0

*P. promelas* PMSD Bounds for Growth: Lower 12 Upper 30

*M. beryllina* PMSD Bounds for Growth: Lower 11 Upper 28

Investigators' signatures

James Daniel

Beau Absher

Marshall Faircloth

Loretta C. Wolfe

REVIEWER

**Comments:** Concurrent with Lee County and trademark Nitrogen MF 3-27-08

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## Appendix C. (continued)

**FDEP Biology Laboratory - Chronic SRT Benchsheet**

*Pimephales promelas or Menidia beryllina* Survival

Organism: ~~P. promelas~~ *M.beryllina* (circle one) D = dead M = missing

Concentration: µg/L g/L (circle one)

| Concentration | Replicate | Chamber | TEST DAY 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 |
|---|---|---|---|---|---|---|---|---|---|---|
| Control | A | F1 | 10 | 10 | 10 | 9 10 | 9 | 9 | 9 | 9 |
| | B | F2 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 |
| | C | F3 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 |
| | D | F4 | 10 | 10 | 9 10 | 9 | 9 | 9 | 9 | 9 |
| 0.0625 | A | F5 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 |
| | B | F6 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 |
| | C | F7 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 |
| | D | F8 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 |
| .125 | A | F9 | 10 | 8 2D | 7 10 | 7 | 7 | 7 | 7 | 7 |
| | B | F10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 |
| | C | F11 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 |
| | D | F12 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 |
| .25 | A | F13 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 |
| | B | F14 | 10 | 10 | 10 | 10 | 10 | 10 | 9 10 | 9 |
| | C | F15 | 10 | 7 3D | 7 | 7 | 7 | 7 | 7 | 7 |
| | D | F16 | 10 | 10 | 9 1D | 9 | 9 | 9 | 9 | 9 |
| .5 | A | F17 | 10 | 3 7D | 2 1D | 2 | 2 | 2 | 2 | 2 |
| | B | F18 | 10 | 6 4D | 5 1D | 5 | 5 | 5 | 5 | 5 |
| | C | F19 | 10 | 4 6D | 2 3D | 2 | 2 | 2 | 2 | 2 |
| | D | F20 | 10 | 5 5D | 0 5D | — | — | — | — | |
| 1.0 | A | F21 | 10 | 0 10D | | | | | | |
| | B | F22 | 10 | 0 10D | | | | | | |
| | C | F23 | 10 | 0 10D | | | | | | |
| | D | F24 | 10 | 0 10D | | | | | | |
| | A | | 10 | | | | | | | |
| | B | | 10 | | | | | | | |
| | C | | 10 | | | | | | | |
| | D | | 10 | | | | | | | |
| Loaded by (0 hr): JD | | Checked by*: | | JD | JD | JD | BA | JD | JD | MF |
| | | Concentrations prepared by: | | JD | JD | JD | BA | JD | JD | — |

**Comments:** Verified by BA 3-18-08

*verified by for 0 hour

Test Page 2 of 6 PAGE 000062 revised 8/24/06 MF/SP

## Appendix C. (continued)

**FDEP Biology Laboratory - Chronic SRT Benchsheet**

*Pimephales promelas* or *Menidia beryllina* Parameters

Start Date: 3.18.08 Time: 1305

End Date: 3-25-08 Time: 1130

Organism: (*P. promelas*) *M.beryllina* (circle one)

Concentration: µg/L (d/l) (circle one)

circle µmhos or mmhos

| Test Day | | Before Renewal | | | | | Test Start and After Renewal | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | Concentration | Test Replicate | pH (su) | Temp (°C) | DO (mg/L) | Conductivity µmhos mmhos | Test Replicate | pH (su) | Temp (°C) | DO (mg/L) | Conductivity µmhos (mmhos) |
| Day 0 | Control | | | | | | A | 6.3 | 24.9 | 8.0 | 0.440 |
| | .0625 | | | | | | A | 6.2 | 24.5 | 8.0 | 0.540 |
| | .125 | | | | | | A | 6.1 | 24.1 | 8.0 | 0.700 |
| | .25 | | | | | | A | 6.1 | 24.0 | 8.0 | 0.934 |
| | .5 | | | | | | A | 6.0 | 24.0 | 8.1 | 1.43 |
| | 1.0 | | | | | | A | 6.1 | 24.0 | 8.1 | 2.43 |
| | | | | | | | A | | | | |
| | Measured by: | | | | | | | JD | JD | JD | JD |
| | Recorded by: | | | | | | | JD | JD | JD | JD |
| | Concentration | Replicate | pH | Temp | DO | Conductivity | Replicate | pH | Temp | DO | Conductivity |
| Day 1 | Control | B | 6.8 | 25.4 | 6.8 | 0.452 | B | 7.0 | 24.1 | 7.6 | 0.435 |
| | .0625 | B | 6.7 | 25.0 | 7.0 | 0.549 | B | 6.2 | 24.0 | 7.5 | 0.511 |
| | .125 | B | 6.6 | 24.0 | 7.1 | 0.699 | B | 6.2 | 24.0 | 7.6 | 0.705 |
| | .25 | B | 6.5 | 24.2 | 7.3 | 0.933 | B | 6.1 | 24.0 | 7.7 | 0.950 |
| | .5 | B | 6.4 | 25.4 | 7.2 | 1.46 | B | 6.1 | 24.1 | 7.7 | 1.44 |
| | 1.0 | B | 6.3 | 24.4 | 6.9 | 2.42 | B | | | | |
| | | B | | | | | B | | | | |
| | Measured by: | | JD | JD | JD | JD | | JD | JD | JD | JD |
| | Recorded by: | | JD | JD | JD | JD | | JD | JD | JD | JD |
| | Concentration | Replicate | pH | Temp | DO | Conductivity | Replicate | pH | Temp | DO | Conductivity |
| Day 2 | Control | C | 6.3 | 24.2 | 6.8 | 0.448 | C | 6.1 | 24.0 | 7.5 | 0.438 |
| | .0625 | C | 6.0 | 24.0 | 6.7 | 0.509 | C | 6.0 | 24.0 | 7.5 | 0.529 |
| | .125 | C | 6.0 | 24.1 | 6.4 | 0.720 | C | 6.0 | 24.0 | 7.4 | 0.685 |
| | .25 | C | 6.0 | 25.4 | 6.3 | 0.983 | C | 6.0 | 24.0 | 7.4 | 0.954 |
| | .5 | C | 6.1 | 24.0 | 6.6 | 1.45 | C | 6.0 | 24.0 | 7.5 | 1.42 |
| | 1.0 | C | | | | | C | | | | |
| | | C | | | | | C | | | | |
| | Measured by: | | JD | JD | JD | JD | | JD | JD | JD | JD |
| | Recorded by: | | JD | JD | JD | JD | | JD | JD | JD | JD |
| | Concentration | Replicate | pH | Temp | DO | Conductivity | Replicate | pH | Temp | DO | Conductivity |
| Day 3 | Control | D | 6.0 | 24.0 | 6.4 | 0.445 | D | 7.3 | 24.0 | 7.7 | 0.406 |
| | .0625 | D | 6.0 | 24.0 | 6.1 | 0.541 | D | 7.8 | 24.0 | 7.6 | 0.562 |
| | .125 | D | 6.0 | 24.0 | 6.4 | 0.694 | D | 8.0 | 24.0 | 7.6 | 0.775 |
| | .25 | D | 6.0 | 24.0 | 6.5 | 0.972 | D | 8.2 | 24.0 | 7.7 | 0.999 |
| | .5 | D | 6.0 | 24.0 | 6.5 | 1.44 | D | 8.3 | 24.0 | 7.7 | 1.44 |
| | 1.0 | D | | | | | D | | | | |
| | | D | | | | | D | | | | |
| | Measured by: | | JD | JD | JD | JD | | JD | JD | JD | JD |
| | Recorded by: | | JD | JD | JD | JD | | JD | JD | JD | JD |

Comments:

Test Page 3 of 6 PAGE 000003 revised 8/24/06 MF/SP

## Appendix C. (continued)

**FDEP Biology Laboratory - Chronic SRT Benchsheet**

*Pimephales promelas* or *Menidia beryllina* Parameters

Organism: (*P. promelas*) *M.beryllina* (circle one)

Concentration: µg/L (mg/L) (circle one)

circle µmhos or mmhos

| Test Day | | Before Renewal | | | | | After Renewal | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | Concentration | Test Replicate | pH (su) | Temp (°C) | DO (mg/L) | Conductivity µmhos (mmhos) | Test Replicate | pH (su) | Temp (°C) | DO (mg/L) | Conductivity µmhos (mmhos) |
| Day 4 | Control | A | ~~12.9~~ 7.9 | 24.0 | 6.5 | 0.427 | A | 7.5 | 24.0 | 7.4 | 0.387 |
| | .0625 | A | ~~7.02~~ 7.8 | 24.0 | 6.2 | 0.577 | A | 7.9 | 24.0 | 7.4 | 0.487 |
| | .125 | A | 7.6 | 24.0 | 6.1 | 0.808 | A | 7.9 | 24.1 | 7.4 | 0.82 |
| | .25 | A | 8.0 | 24.0 | 6.0 | 1.16 | A | 7.9 | 24.0 | 7.4 | 0.911 |
| | .5 | A | 8.2 | 24.0 | 6.0 | 1.83 | A | 8.0 | 24.0 | 7.4 | 1.36 |
| | 1.0 | A | — | — | — | — | A | — | — | — | — |
| | | A | | | | | A | | | | |
| | Measured by: | | BA | BA | BA | BA | | BA | BA | BA | BA |
| | Recorded by: | | BA | BA | BA | BA | | BA | BA | BA | BA |
| | Concentration | Replicate | pH | Temp | DO | Conductivity | Replicate | pH | Temp | DO | Conductivity |
| Day 5 | Control | B | 7.4 | 24.0 | 6.2 | 0.396 | B | 7.7 | 24.0 | 7.6 | 0.380 |
| | .0625 | B | 7.4 | 24.0 | 6.3 | 0.501 | B | 7.8 | 24.0 | 7.5 | 0.598 |
| | .125 | B | 7.5 | 24.0 | 6.2 | 0.843 | B | 7.8 | 24.0 | 7.4 | 0.732 |
| | .25 | B | 7.6 | 24.0 | 6.3 | 0.927 | B | 7.8 | 24.0 | 7.4 | 0.941 |
| | .5 | B | 7.8 | 24.0 | 6.3 | 1.40 | B | 7.9 | 24.0 | 7.5 | 1.33 |
| | | B | | | | | B | | | | |
| | | B | | | | | B | | | | |
| | Measured by: | | DD | DD | DD | DD | | DD | DD | DD | DD |
| | Recorded by: | | DD | DD | DD | DD | | DD | DD | DD | DD |
| | Concentration | Replicate | pH | Temp | DO | Conductivity | Replicate | pH | Temp | DO | Conductivity |
| Day 6 | Control | C | 7.6 | 24.0 | 6.4 | 0.398 | C | 7.9 | 24.0 | 7.6 | 0.339 |
| | .06 | C | 7.6 | 24.0 | 6.4 | 0.617 | C | 7.9 | 24.0 | 7.4 | 0.606 |
| | .125 | C | 7.5 | 24.0 | ~~5.49~~ | 0.756 | C | 7.8 | 24.0 | 7.4 | 0.695 |
| | .25 | C | 7.6 | 24.0 | 5.8 | 0.973 | C | 7.8 | 24.0 | 7.4 | 0.933 |
| | .5 | C | 7.7 | 24.0 | 6.1 | 1.37 | C | 7.9 | 24.0 | 7.4 | 1.38 |
| | | C | | | | | C | | | | |
| | | C | | | | | C | | | | |
| | Measured by: | | DD | DD | DD | DD | | DD | DD | DD | DD |
| | Recorded by: | | DD | DD | DD | DD | | DD | DD | DD | DD |
| | Concentration | Replicate | pH | Temp | DO | Conductivity | Replicate | pH | Temp | DO | Conductivity |
| Day 7 | Control | D | 7.5 | 24.0 | 5.7 | 0.440 | D | | | | |
| | 0.06 | D | 7.5 | 24.0 | 6.0 | 0.622 | D | | | | |
| | 0.125 | D | 7.5 | 24.0 | 5.8 | 0.71 | D | | | | |
| | 0.25 | D | 7.4 | 25.9 | 5.4 | 0.945 | D | | | | |
| | 0.5 | ~~B~~ C | 7.4 | 24.4 | 5.4 | 1.37 | D | | | | |
| | | D | | | | | D | | | | |
| | | ~~C~~ D | | | | | D | | | | |
| | Measured by: | | BA | BA | BA | BA | | | | | |
| | Recorded by: | | BA | BA | BA | BA | | | | | |

Comments: D 5.4 J.D Day 6 .125 D.O.

## Appendix C. (continued)

**FDEP Biology Laboratory - Chronic SRT Benchsheet**

*Pimephales promelas* or *Menidia beryllina* Weights

SOP TA06_21

Organism: (*P. promelas*) *M.beryllina* (circle one)

Concentration: µg/L (mg/L) (circle one)

mean wt = fish wt divided by 10

convert g to mg: move decimal 3 places to right

| Concentration | Replicate | Pan # | Pan wt. (g) | Pan + Fish (g) | Fish wt. (g) | Original # of fish | Mean wt. (mg) |
|---|---|---|---|---|---|---|---|
| Control | A | 17 | 0.01712 | 0.02191 | 0.00479 | 10 | 0.479 |
| | B | 18 | 0.01746 | 0.02301 | 0.00555 | 10 | 0.555 |
| | C | 19 | 0.01702 | 0.02238 | 0.00536 | 10 | 0.536 |
| | D | 20 | 0.02068 | 0.02626 | 0.00558 | 10 | 0.558 |
| .0625 | A | 21 | 0.01864 | 0.02471 | 0.00607 | 10 | 0.607 |
| | B | 22 | 0.02068 | 0.02662 | 0.00594 | 10 | 0.594 |
| | C | 23 | 0.01809 | 0.02351 | 0.00542 | 10 | 0.542 |
| | D | 24 | 0.01868 | 0.02442 | 0.00574 | 10 | 0.574 |
| .125 | A | 25 | 0.01780 | 0.02292 | 0.00512 | 10 | 0.512 |
| | B | 26 | 0.01735 | 0.02299 | 0.00564 | 10 | 0.564 |
| | C | 27 | 0.01687 | 0.02371 | 0.00684 | 10 | 0.684 |
| | D | 28 | 0.01949 | 0.02387 | 0.00438 | 10 | 0.438 |
| .25 | A | 29 | 0.01896 | 0.02424 | 0.00528 | 10 | 0.528 |
| | B | 30 | 0.01970 | 0.02468 | 0.00498 | 10 | 0.498 |
| | C | 31 | 0.01889 | 0.02433 | 0.00544 | 10 | 0.544 |
| | D | 32 | 0.01946 | 0.02549 | 0.00603 | 10 | 0.603 |
| .5 | A | 33 | 0.01887 | 0.02040 | 0.00153 | 10 | 0.153 |
| | B | 34 | 0.02190 | 0.02557 | 0.00367 | 10 | 0.367 |
| | C | 35 | 0.01708 | 0.01883 | 0.00175 | 10 | 0.175 |
| | D | — | — | — | — | 10 | — |
| 1.0 | A | | | | | 10 | |
| | B | | | | | 10 | |
| | C | | | | | 10 | |
| | D | | | | | 10 | |
| | A | | | | | 10 | |
| | B | | | | | 10 | |
| | C | | | | | 10 | |
| | D | | | | | 10 | |
| Weighed by: | | | MF | MF | | | |

**Comments:**

Note: Pan number (#) refers to actual Pan number.

## Appendix C. (continued)

**FDEP Biology Laboratory - Chronic SRT Benchsheet**

*Pimephales promelas* or *Menidia beryllina* QC

SOPs TA07_09, TA07_14 and TA06_21

**Empty Pan Weights** Date: 3-25-08 MF

| | Pan 1 wt. | Pan 1 initial wt. |
|---|---|---|
| after Pan 5 ~~(20)~~ MF | 0.01757 @ | 0.01757 |
| after Pan 10 ~~(25)~~ MF | 0.01757 @ | 0.01757 |
| after Pan 15 ~~(30)~~ MF | 0.01758 @ | 0.01757 |
| after Pan 20 (35) MF | 0.01759 @ | 0.01757 |
| after Pan 25 | MF @ | 0.01757 |
| after Pan 30 | @ | |
| after Pan 35 | @ | |
| after Pan 40 | @ | |
| after Pan 45 | @ | |
| after Pan 48 | @ | |

| | Measured Value | S-weight Value |
|---|---|---|
| S-weights | 0.00998 @ | 0.01000 |
| | 0.03000 @ | 0.03000 |

~~(partial Pan #)~~ MF 3-25-08

**Fish + Pan Weights** Date: 3-26-08 MF

| | Pan 1 wt. | Pan 1 initial wt. |
|---|---|---|
| after Pan 5 | 0.02193 @ | 0.02191 |
| after Pan 10 | 0.02193 @ | 0.02191 |
| after Pan 15 | 0.02194 @ | 0.02191 |
| after Pan 20 | 0.02193 @ | 0.02191 |
| after Pan 25 | @ | |
| after Pan 30 | @ | |
| after Pan 35 | @ | |
| after Pan 40 | @ | |
| after Pan 45 | @ | |
| after Pan 48 | @ | |

| | Measured Value | S-weight Value |
|---|---|---|
| S-weights | 0.01000 @ | 0.01000 |
| | 0.03000 @ | 0.03000 |

Drying Oven ID: 630F Sediment oven #205

Temperature °C: 60°C

Drying Hours: 24 hours

Note: Pan number (5, 10, etc.) refers to sequence of weighing as actual Pan #s may differ.

**Comments:**

Test Page 6 of 6 PAGE 000006 revised 8/24/06 MF/SP

## Appendix C. (continued)

**Trademark Nitrogen Corporation *C. dubia* survival T-test**

| | | | |
|---|---|---|---|
| Sample: | Trademark Nitrogen Corporation | Date tested: | 3/18-24/2008 |
| Species: | Ceriodaphnia dubia | Test Type: | chronic screen |

| | Treatment A | Treatment B | # of Reps/Sample | 10 |
|---|---|---|---|---|
| | Control | 100% Effluent | | |
| | 1.00 | 1.00 | | p=0.05 |
| | 1.00 | 1.00 | df | Table t-value |
| | 1.00 | 1.00 | 1 | 6.31400 |
| | 1.00 | 1.00 | 2 | 2.92000 |
| | 1.00 | 1.00 | 3 | 2.35300 |
| | 1.00 | 1.00 | 4 | 2.13200 |
| | 1.00 | 1.00 | 5 | 2.01500 |
| | 1.00 | 1.00 | 6 | 1.94300 |
| | 1.00 | 1.00 | 7 | 1.89500 |
| | 1.00 | 1.00 | 8 | 1.86000 |
| Average | 1.000 | 1.000 | 9 | 1.83300 |
| | | | 10 | 1.81200 |
| | t-value= | 0.00000 | 18 | 1.73400 |
| | df= | 18.00000 | | |

**Trademark Nitrogen Corporation *C. dubia* Reproduction T-test**

| | | | |
|---|---|---|---|
| Sample: | Trademark Nitrogen Corporation | Dates tested: | 3/18/2008 - 3/24/2008 |
| Species: | *Ceriodaphnia dubia* | Test Type: | chronic screen |

| | Teatment A | Treatment B | #of Reps./sample | 10 |
|---|---|---|---|---|
| Replicate # | Control | 100% Effluent | | |
| 1 | 21.000 | 20.000 | | p=0.05 |
| 2 | 26.000 | 22.000 | df | Table t-value |
| 3 | 28.000 | 23.000 | 1 | 6.31400 |
| 4 | 19.000 | 19.000 | 2 | 2.92000 |
| 5 | 20.000 | 22.000 | 3 | 2.35300 |
| 6 | 26.000 | 27.000 | 4 | 2.13200 |
| 7 | 22.000 | 23.000 | 5 | 2.01500 |
| 8 | 23.000 | 27.000 | 6 | 1.94300 |
| 9 | 20.000 | 22.000 | 7 | 1.89500 |
| 10 | 20.000 | 20.000 | 8 | 1.86000 |
| | | | 9 | 1.83300 |
| Average | 22.50000 | 22.50000 | 10 | 1.81200 |
| | | | 18 | 1.734 |
| | t-value= | 0.00000 | | |
| | df= | 18 | | |

## Appendix C. (continued)

**Trademark Nitrogen Corporation *P. promelas* Survival T-test**

| | | | | | | |
|---|---|---|---|---|---|---|
| Sample: | Trademark Nitrogen Corporation | Dates tested: | 03/18/2008 - 03/25/2008 | | | |
| Species: | *Pimephales promelas* | Test Type: | Chronic Screen | | | |
| | | | | | | p=0.05 |
| | Treatment A | Treatment B | # of Reps/Sample | 4 | df | Table t-value |
| | Control | 100% Effluent | | | 1 | 6.31400 |
| | 0.90 | 1.00 | | | 2 | 2.92000 |
| | 0.90 | 1.00 | | | 3 | 2.35300 |
| | 1.00 | 1.00 | | | 4 | 2.13200 |
| | 0.90 | 0.90 | | | 5 | 2.01500 |
| | | | | | 6 | 1.94300 |
| Average | 0.925 | 0.975 | | | 7 | 1.89500 |
| | | | | | 8 | 1.86000 |
| t-value= | | -1.41421 | | | 9 | 1.83300 |
| df= | | 6.00000 | | | 10 | 1.81200 |
| | | | | | 18 | 1.73400 |

**Trademark Nitrogen Corporation *P. promelas* growth T-test**

| | | | |
|---|---|---|---|
| Sample: | Trademark Nitrogen Corporation | Date tested: | 03/18/2008-03/24/2008 |
| Species: | *Pimephales promelas* | Test Type: | Chronic screen |

| | Teatment A | Treatment B | #of Reps./sample | 4 |
|---|---|---|---|---|
| Replicate # | Control | 100% | | |
| 1 | 0.551 | 0.580 | | p=0.05 |
| 2 | 0.567 | 0.594 | df | Table t-value |
| 3 | 0.557 | 0.559 | 1 | 6.31400 |
| 4 | 0.574 | 0.541 | 2 | 2.92000 |
| 5 | | | 3 | 2.35300 |
| 6 | | | 4 | 2.13200 |
| 7 | | | 5 | 2.01500 |
| 8 | | | 6 | 1.94300 |
| 9 | | | 7 | 1.89500 |
| 10 | | | 8 | 1.86000 |
| | | | 9 | 1.83300 |
| Average | 0.56225 | 0.56850 | 10 | 1.81200 |
| | | | 18 | 1.734 |
| | t-value= | -0.49108 | | |
| | df= | 6 | | |

# Appendix C. (continued)

**Ceriodaphnia Survival and Reproduction Test-Reproduction**

| | | | | | |
|---|---|---|---|---|---|
| Start Date: | 3/18/2008 13:30 | Test ID: | CDcdsrt318 | Sample ID: | REF-Ref Toxicant |
| End Date: | 3/24/2008 14:15 | Lab ID: | DEP TOXLAB-FDEP Biology To | Sample Type: | NACL-Sodium chloride |
| Sample Date: | | Protocol: | EPAFW02-EPA/821/R-02-013 | Test Species: | CD-Ceriodaphnia dubia |
| Comments: | | | | | |

| Conc-ppt | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| D-Control | 21.000 | 23.000 | 23.000 | 22.000 | 18.000 | 28.000 | 27.000 | 31.000 | 23.000 | 26.000 |
| 0.59 | 30.000 | 22.000 | 20.000 | 23.000 | 25.000 | 23.000 | 28.000 | 23.000 | 26.000 | 21.000 |
| 0.89 | 22.000 | 21.000 | 23.000 | 24.000 | 23.000 | 24.000 | 25.000 | 17.000 | 20.000 | 24.000 |
| 1.3 | 3.000 | 18.000 | 19.000 | 0.000 | 14.000 | 15.000 | 15.000 | 22.000 | 19.000 | 20.000 |
| 2 | 2.000 | 0.000 | 0.000 | 0.000 | 0.000 | 0.000 | 0.000 | 1.000 | 1.000 | 0.000 |
| 3 | 0.000 | 0.000 | 0.000 | 0.000 | 0.000 | 0.000 | 0.000 | 0.000 | 0.000 | 0.000 |

| | | | Transform: Untransformed | | | | | Rank | 1-Tailed | Isotonic | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Conc-ppt | Mean | N-Mean | Mean | Min | Max | CV% | N | Sum | Critical | Mean | N-Mean |
| D-Control | 24.200 | 1.0000 | 24.200 | 18.000 | 31.000 | 15.681 | 10 | | | 24.200 | 1.0000 |
| 0.59 | 24.100 | 0.9959 | 24.100 | 20.000 | 30.000 | 13.041 | 10 | 103.50 | 77.00 | 24.100 | 0.9959 |
| 0.89 | 22.300 | 0.9215 | 22.300 | 17.000 | 25.000 | 10.789 | 10 | 93.00 | 77.00 | 22.300 | 0.9215 |
| *1.3 | 14.500 | 0.5992 | 14.500 | 0.000 | 22.000 | 50.496 | 10 | 61.00 | 77.00 | 14.500 | 0.5992 |
| 2 | 0.400 | 0.0165 | 0.400 | 0.000 | 2.000 | 174.801 | 10 | | | 0.400 | 0.0165 |
| 3 | 0.000 | 0.0000 | 0.000 | 0.000 | 0.000 | 0.000 | 10 | | | 0.000 | 0.0000 |

| Auxiliary Tests | Statistic | Critical | Skew | Kurt |
|---|---|---|---|---|
| Shapiro-Wilk's Test indicates normal distribution (p > 0.01) | 0.925709 | 0.919 | -1.14942 | 2.652279 |
| Bartlett's Test indicates unequal variances (p = 5.53E-03) | 12.62313 | 11.34487 | | |

| Hypothesis Test (1-tail, 0.05) | NOEC | LOEC | ChV | TU |
|---|---|---|---|---|
| Steel's Many-One Rank Test | 0.89 | 1.3 | 1.075639 | |
| Treatments vs D-Control | | | | |

**Linear Interpolation (200 Resamples)**

| Point | ppt | SD | 95% CL | | Skew |
|---|---|---|---|---|---|
| IC05 | 0.7750 | 0.1884 | 0.2735 | 0.9278 | -0.9974 |
| IC10 | 0.9173 | 0.1165 | 0.5470 | 1.0087 | -1.8016 |
| IC15 | 0.9809 | 0.0684 | 0.8383 | 1.1020 | -0.3823 |
| IC20 | 1.0445 | 0.0711 | 0.9318 | 1.2164 | 0.5838 |
| IC25 | 1.1081 | 0.0793 | 0.9950 | 1.3139 | 0.6328 |
| IC40 | 1.2989 | 0.0844 | 1.1486 | 1.4540 | -0.0887 |
| IC50 | 1.4191 | 0.0841 | 1.2492 | 1.5469 | -0.4285 |

# Appendix C. (continued)

**Larval Fish Growth and Survival Test-7 Day Growth**

| | | | | | |
|---|---|---|---|---|---|
| Start Date: | 3/18/2008 13:05 | Test ID: | ppcsrt318 | Sample ID: | REF-Ref Toxicant |
| End Date: | 3/25/2008 11:30 | Lab ID: | DEP TOXLAB-FDEP Biology To | Sample Type: | KCL-Potassium chloride |
| Sample Date: | | Protocol: | EPAF 94-EPA/600/4-91/002 | Test Species: | PP-Pimephales promelas |
| Comments: | Concurrent SRT for Facility Tests | | | | |

| Conc-gm/L | 1 | 2 | 3 | 4 |
|---|---|---|---|---|
| D-Control | 0.4790 | 0.5550 | 0.5360 | 0.5580 |
| 0.0625 | 0.6070 | 0.5940 | 0.5420 | 0.5740 |
| 0.125 | 0.5120 | 0.5640 | 0.6840 | 0.4380 |
| 0.25 | 0.5280 | 0.4980 | 0.5440 | 0.6030 |
| 0.5 | 0.1530 | 0.3670 | 0.1750 | 0.0000 |
| 1 | 0.0000 | 0.0000 | 0.0000 | 0.0000 |

| | | | Transform: Untransformed | | | | | Isotonic | |
|---|---|---|---|---|---|---|---|---|---|
| Conc-gm/L | Mean | N-Mean | Mean | Min | Max | CV% | N | Mean | N-Mean |
| D-Control | 0.5320 | 1.0000 | 0.5320 | 0.4790 | 0.5580 | 6.889 | 4 | 0.5556 | 1.0000 |
| 0.0625 | 0.5793 | 1.0888 | 0.5793 | 0.5420 | 0.6070 | 4.886 | 4 | 0.5556 | 1.0000 |
| 0.125 | 0.5495 | 1.0329 | 0.5495 | 0.4380 | 0.6840 | 18.836 | 4 | 0.5495 | 0.9890 |
| 0.25 | 0.5433 | 1.0211 | 0.5433 | 0.4980 | 0.6030 | 8.129 | 4 | 0.5433 | 0.9777 |
| 0.5 | 0.1738 | 0.3266 | 0.1738 | 0.0000 | 0.3670 | 86.629 | 4 | 0.1738 | 0.3127 |
| 1 | 0.0000 | 0.0000 | 0.0000 | 0.0000 | 0.0000 | 0.000 | 4 | 0.0000 | 0.0000 |

| Auxiliary Tests | Statistic | Critical | Skew | Kurt |
|---|---|---|---|---|
| Shapiro-Wilk's Test indicates normal distribution (p > 0.01) | 0.920948 | 0.868 | 0.376732 | 2.279355 |
| Bartlett's Test indicates equal variances (p = 0.04) | 10.15574 | 13.2767 | | |

**Linear Interpolation (200 Resamples)**

| Point | gm/L | SD | 95% CL(Exp) | | Skew |
|---|---|---|---|---|---|
| IC05 | 0.2604 | 0.0614 | 0.0000 | 0.2733 | -1.0280 |
| IC10 | 0.2792 | 0.0269 | 0.1927 | 0.2998 | -4.2498 |
| IC15 | 0.2980 | 0.0135 | 0.2455 | 0.3297 | 0.2755 |
| IC20 | 0.3168 | 0.0155 | 0.2675 | 0.3584 | 0.6656 |
| IC25 | 0.3356 | 0.0182 | 0.2849 | 0.3977 | 0.9029 |
| IC40 | 0.3920 | 0.0281 | 0.3277 | 0.5159 | 1.1155 |
| IC50 | 0.4296 | 0.0373 | 0.3576 | 0.6173 | 1.3805 |

## Appendix D. Test Organism Receipt

**MBL Aquaculture**
4569 Samuel Street
Sarasota, Florida 34233
TEL: (800) 889-0384
FAX: (941) 922-3874
EMAIL: sales@mblaquaculture.com
WEB: www.MBLAquaculture.com

**NELAC Accredited**

### Organism Shipment Record

State of Florida Aquaculture Certificate Number AQ0668007

Date: 3/17/08

Shipped to: FDEP

P.O. No: N/A

| Species | Quantity | Age | Brood Number | Temperature | Salinity |
|---|---|---|---|---|---|
| *Mysidopsis bahia* | | | | | |
| *Menidia beryllina* | | | | | |
| *Cyprinella leedsi* | 100 | 11 DAYS | CL080306 | 25°C | |
| *Pimephales promelas* | 450 | <24 HRS | FM080316-1550 | 25°C | |
| | | | | | |

Packed by:

Shipped Via: FedEx

Notes:

Thank you for your order.

**Appendix E. Chemical Analyses performed on the effluent from the Trademark Nitrogen Corporation Outfall D-001, sampled on March 17, 2008. Additional Field Data from Bioassay Samples collected on March 19 and 21, 2008.**

| Date Sampled | Field ID | Analysis | Component | Result | Remark | Units | MDL | PQL |
|---|---|---|---|---|---|---|---|---|
| 3/17/2008 10:15 | Effluent | AGP/LN | Algal Growth Potential | 8.72 | A | mg DryWt/L | 0.3 | 0.9 |
| 3/17/2008 10:15 | Effluent | Chlorophyll/Grain Size/BOD | Phaeophytin-a | 1.6 | | ug/L | 0.24 | 0.71 |
| 3/17/2008 10:15 | Effluent | Chlorophyll/Grain Size/BOD | Biochemical Oxygen Demand-5 Day,N-Inhib | 0.64 | I | mg/L | 0.2 | 2 |
| 3/17/2008 10:15 | Effluent | Chlorophyll/Grain Size/BOD | Chlorophyll-a, Corrected | 2.6 | | ug/L | 0.55 | 1.7 |
| 3/17/2008 10:15 | Effluent | Metals | Cadmium | 0.022 | I | ug/L | 0.02 | 0.08 |
| 3/17/2008 10:15 | Effluent | Metals | Calcium | 79.9 | | mg/L | 0.075 | 0.3 |
| 3/17/2008 10:15 | Effluent | Metals | Arsenic | 0.5 | U | ug/L | 0.5 | 2 |
| 3/17/2008 10:15 | Effluent | Metals | Copper | 0.5 | U | ug/L | 0.5 | 2 |
| 3/17/2008 10:15 | Effluent | Metals | Zinc | 5 | U | ug/L | 5 | 20 |
| 3/17/2008 10:15 | Effluent | Metals | Magnesium | 11.4 | | mg/L | 0.04 | 0.16 |
| 3/17/2008 10:15 | Effluent | Metals | Iron | 31 | I | ug/L | 30 | 120 |
| 3/17/2008 10:15 | Effluent | Metals | Nickel | 0.25 | U | ug/L | 0.25 | 1 |
| 3/17/2008 10:15 | Effluent | Metals | Lead | 0.2 | U | ug/L | 0.2 | 0.8 |
| 3/17/2008 10:15 | Effluent | Metals | Aluminum | 60 | U | ug/L | 60 | 240 |
| 3/17/2008 10:15 | Effluent | Metals | Chromium | 1 | U | ug/L | 1 | 4 |
| 3/17/2008 10:15 | Effluent | Metals | Selenium | 0.5 | U | ug/L | 0.5 | 2 |
| 3/17/2008 10:15 | Effluent | Metals | Silver | 0.025 | U | ug/L | 0.025 | 0.1 |
| 3/17/2008 10:15 | Effluent | Nutrients | Total-P | 0.048 | I | mg P/L | 0.02 | 0.06 |
| 3/17/2008 10:15 | Effluent | Nutrients | NO2NO3-N | 0.032 | | mg N/L | 0.004 | 0.01 |
| 3/17/2008 10:15 | Effluent | Nutrients | Nitrite-N | 0.01 | I | mg N/L | 0.004 | 0.01 |
| 3/17/2008 10:15 | Effluent | Nutrients | Ammonia-N | 0.035 | | mg N/L | 0.01 | 0.02 |
| 3/17/2008 10:15 | Effluent | Nutrients | Kjeldahl Nitrogen | 0.11 | I | mg N/L | 0.08 | 0.2 |
| 3/17/2008 10:15 | Effluent | Nutrients | TSS | 4 | U | mg/L | 4 | 16 |
| 3/17/2008 10:15 | Effluent | Nutrients | Organic Carbon | 1.9 | I | mg C/L | 1 | 5 |
| 3/17/2008 10:15 | Effluent | Nutrients | O-Phosphate-P | 0.037 | | mg P/L | 0.004 | 0.01 |
| 3/17/2008 10:15 | Effluent | Nutrients | Nitrate-N | 0.05 | U | mg N/L | 0.05 | 0.2 |
| 3/17/2008 10:15 | Effluent | Pesticides | Malathion | 0.14 | U | ug/L | 0.14 | 0.56 |
| 3/17/2008 10:15 | Effluent | Pesticides | Naled | 0.75 | U | ug/L | 0.75 | 3 |
| 3/17/2008 10:15 | Effluent | Pesticides | Phorate | 0.047 | U | ug/L | 0.047 | 0.19 |
| 3/17/2008 10:15 | Effluent | Pesticides | Ethion | 0.047 | U | ug/L | 0.047 | 0.19 |

## Appendix E. (continued)

| Date Sampled | Field ID | Analysis | Component | Result | Remark | Units | MDL | PQL |
|---|---|---|---|---|---|---|---|---|
| 3/17/2008 10:15 | Effluent | Pesticides | Parathion Ethyl | 0.14 | U | ug/L | 0.14 | 0.56 |
| 3/17/2008 10:15 | Effluent | Pesticides | Azinphos Methyl | 0.19 | U | ug/L | 0.19 | 0.76 |
| 3/17/2008 10:15 | Effluent | Pesticides | Butylate | 0.19 | UJ | ug/L | 0.19 | 0.76 |
| 3/17/2008 10:15 | Effluent | Pesticides | Fenamiphos | 0.19 | U | ug/L | 0.19 | 0.76 |
| 3/17/2008 10:15 | Effluent | Pesticides | Diazinon | 0.047 | U | ug/L | 0.047 | 0.19 |
| 3/17/2008 10:15 | Effluent | Pesticides | Chlorpyrifos Methyl | 0.094 | U | ug/L | 0.094 | 0.38 |
| 3/17/2008 10:15 | Effluent | Pesticides | Atrazine | 0.047 | U | ug/L | 0.047 | 0.19 |
| 3/17/2008 10:15 | Effluent | Pesticides | Metolachlor | 0.47 | U | ug/L | 0.47 | 1.9 |
| 3/17/2008 10:15 | Effluent | Pesticides | Ethoprop | 0.094 | U | ug/L | 0.094 | 0.38 |
| 3/17/2008 10:15 | Effluent | Pesticides | Chlorpyrifos Ethyl | 0.047 | U | ug/L | 0.047 | 0.19 |
| 3/17/2008 10:15 | Effluent | Pesticides | Alachlor | 0.57 | U | ug/L | 0.57 | 2.3 |
| 3/17/2008 10:15 | Effluent | Pesticides | Metribuzin | 0.094 | U | ug/L | 0.094 | 0.38 |
| 3/17/2008 10:15 | Effluent | Pesticides | Ametryn | 0.047 | U | ug/L | 0.047 | 0.19 |
| 3/17/2008 10:15 | Effluent | Pesticides | Fonofos | 0.094 | U | ug/L | 0.094 | 0.38 |
| 3/17/2008 10:15 | Effluent | Pesticides | Hexazinone | 0.094 | U | ug/L | 0.094 | 0.38 |
| 3/17/2008 10:15 | Effluent | Pesticides | Bromacil | 0.19 | U | ug/L | 0.19 | 0.76 |
| 3/17/2008 10:15 | Effluent | Pesticides | Parathion Methyl | 0.094 | U | ug/L | 0.094 | 0.38 |
| 3/17/2008 10:15 | Effluent | Pesticides | Norflurazon | 0.14 | U | ug/L | 0.14 | 0.56 |
| 3/17/2008 10:15 | Effluent | Pesticides | Simazine | 0.047 | U | ug/L | 0.047 | 0.19 |
| 3/17/2008 10:15 | Effluent | Pesticides | Prometryn | 0.14 | U | ug/L | 0.14 | 0.56 |
| 3/17/2008 10:15 | Effluent | Pesticides | Metalaxyl | 0.24 | U | ug/L | 0.24 | 0.96 |
| 3/17/2008 10:15 | Effluent | Pesticides | Mevinphos | 0.19 | U | ug/L | 0.19 | 0.76 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | 2,4-Dimethylphenol | 9.6 | U | ug/L | 9.6 | 38 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | 2,4-Dinitrophenol | 14 | U | ug/L | 14 | 58 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | 4,4'-DDT | 1.4 | U | ug/L | 1.4 | 5.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | 4-Nitrophenol | 14 | U | ug/L | 14 | 58 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Benzidine | 96 | U | ug/L | 96 | 380 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Benzo(a)anthracene | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Benzo(g,h,i)perylene | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Benzo(k)fluoranthene | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Butyl benzyl phthalate | 4.8 | U | ug/L | 4.8 | 19 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Dibenzo(a,h)anthracene | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Endosulfan sulfate | 1.4 | U | ug/L | 1.4 | 5.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Nitrobenzene | 1.9 | U | ug/L | 1.9 | 7.7 |

## Appendix E. (continued)

| Date Sampled | Field ID | Analysis | Component | Result | Remark | Units | MDL | PQL |
|---|---|---|---|---|---|---|---|---|
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Phenanthrene | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Bromoform | 0.5 | UJ | ug/L | 0.5 | 2 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | 2-Chloronaphthalene | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | 2-Methyl-4,6-dinitrophenol | 2.9 | U | ug/L | 2.9 | 12 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | 2-Nitrophenol | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | 4,4'-DDD | 1.4 | U | ug/L | 1.4 | 5.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Acenaphthene | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Acenaphthylene | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Bis(2-chloroethoxy)methane | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Bis(2-chloroisopropyl)ether | 2.9 | U | ug/L | 2.9 | 12 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Dieldrin | 1.4 | U | ug/L | 1.4 | 5.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Endosulfan I | 3.8 | U | ug/L | 3.8 | 15 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Endosulfan II | 3.8 | U | ug/L | 3.8 | 15 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Endrin | 1.4 | U | ug/L | 1.4 | 5.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Endrin aldehyde | 3.8 | UJ | ug/L | 3.8 | 15 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Heptachlor | 1.4 | U | ug/L | 1.4 | 5.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Indeno(1,2,3-cd)pyrene | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Naphthalene | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | 2,4-Dinitrotoluene | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Benzo(b)fluoranthene | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Dimethyl phthalate | 9.6 | U | ug/L | 9.6 | 38 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Fluoranthene | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Hexachloroethane | 2.9 | U | ug/L | 2.9 | 12 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Isophorone | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | N-Nitrosodiphenylamine/ Diphenylamine | 2.9 | U | ug/L | 2.9 | 12 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Chloroform | 0.2 | U | ug/L | 0.2 | 1 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | 1,2,4-Trichlorobenzene | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | 2,6-Dinitrotoluene | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | 4-Bromophenyl phenyl ether | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Bis(2-chloroethyl)ether | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Fluorene | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Hexachlorocyclopentadiene | 2.9 | U | ug/L | 2.9 | 12 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | N-Nitrosodi-n-propylamine | 1.9 | U | ug/L | 1.9 | 7.7 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Pentachlorophenol | 2.9 | U | ug/L | 2.9 | 12 |

## Appendix E. (continued)

| Date Sampled | Field ID | Analysis | Component | Result | Remark | Units | MDL | PQL |
|---|---|---|---|---|---|---|---|---|
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Dibromochloromethane | 0.2 | UJ | ug/L | 0.2 | 1 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | 4,4'-DDE | 1.4 | U | ug/L | 1.4 | 5.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Aldrin | 1.4 | U | ug/L | 1.4 | 5.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Benzo(a)pyrene | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Chrysene | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Hexachlorobenzene | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Phenol | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | alpha-BHC | 1.4 | U | ug/L | 1.4 | 5.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | delta-BHC | 1.4 | U | ug/L | 1.4 | 5.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | gamma-BHC | 1.4 | U | ug/L | 1.4 | 5.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | 2,4,6-Trichlorophenol | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | 2-Chlorophenol | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Bis(2-ethylhexyl)phthalate | 14 | U | ug/L | 14 | 58 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Di-n-octyl phthalate | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | beta-BHC | 1.4 | U | ug/L | 1.4 | 5.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | 2,4-Dichlorophenol | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | 4-Chloro-3-methylphenol | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Anthracene | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Di-n-butyl phthalate | 4.8 | U | ug/L | 4.8 | 19 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Hexachlorobutadiene | 2.9 | U | ug/L | 2.9 | 12 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Pyrene | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | 3,3'-Dichlorobenzidine | 38 | U | ug/L | 38 | 150 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | 4-Chlorophenyl phenyl ether | 1.9 | U | ug/L | 1.9 | 7.7 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Diethyl phthalate | 0.96 | U | ug/L | 0.96 | 3.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Heptachlor epoxide | 1.4 | U | ug/L | 1.4 | 5.8 |
| 3/17/2008 10:15 | Effluent | Priority Organic Pollutants | Bromodichloromethane | 0.2 | U | ug/L | 0.2 | 1 |
| 3/17/2008 10:15 | Effluent | Field Parameter | Sample Depth | 0.2 | | m | | |
| 3/17/2008 10:15 | Effluent | Field Parameter | Dissolved Oxygen | 8.72 | | mg/L | | |
| 3/17/2008 10:15 | Effluent | Field Parameter | Specific Conductance | 506 | | umhos/cm | | |
| 3/17/2008 10:15 | Effluent | Field Parameter | pH | 7.89 | | | | |
| 3/17/2008 10:15 | Effluent | Field Parameter | Temperature | 20.99 | | C | | |
| 3/17/2008 13:00 | Bioassay 1 | Toxicology | Bioassay-Chronic-Scrn-FW-Fish, NOEC | 100 | | NOEC | | |
| 3/17/2008 13:00 | Bioassay 1 | Toxicology | Bioassay-Chronic-Scrn-FW-C.dubia, NOEC | 100 | | NOEC | | |
| 3/17/2008 13:00 | Bioassay 1 | Field Parameter | Sample Depth | 0.2 | | m | | |

## Appendix E. (continued)

| Date Sampled | Field ID | Analysis | Component | Result | Remark | Units | MDL | PQL |
|---|---|---|---|---|---|---|---|---|
| 3/17/2008 13:00 | Bioassay 1 | Field Parameter | pH | 7.89 | | | | |
| 3/17/2008 13:00 | Bioassay 1 | Field Parameter | Specific Conductance | 506 | | umhos/cm | | |
| 3/17/2008 13:00 | Bioassay 1 | Field Parameter | Dissolved Oxygen | 8.72 | | mg/L | | |
| 3/17/2008 13:00 | Bioassay 1 | Field Parameter | Temperature | 20.99 | | C | | |
| 3/18/2008 10:50 | Bioassay 2 | Field Parameter | pH | 7.8 | | | | |
| 3/18/2008 10:50 | Bioassay 2 | Field Parameter | Specific Conductance | 0.25 | | umhos/cm | | |
| 3/18/2008 10:50 | Bioassay 2 | Field Parameter | Temperature | 21.05 | | C | | |
| 3/18/2008 10:50 | Bioassay 2 | Field Parameter | Dissolved Oxygen | 8.7 | | mg/L | | |
| 3/18/2008 10:50 | Bioassay 2 | Field Parameter | Sample Depth | 0.2 | | m | | |
| 3/20/2008 7:50 | Bioassay 3 | Field Parameter | pH | 7.82 | | | | |
| 3/20/2008 7:50 | Bioassay 3 | Field Parameter | Dissolved Oxygen | 8.77 | | mg/L | | |
| 3/20/2008 7:50 | Bioassay 3 | Field Parameter | Temperature | 18.93 | | C | | |
| 3/20/2008 7:50 | Bioassay 3 | Field Parameter | Sample Depth | 0.2 | | m | | |
| 3/20/2008 7:50 | Bioassay 3 | Field Parameter | Specific Conductance | 511 | | umhos/cm | | |
| 3/17/2008 10:45 | Field Blank | Chlorophyll/Grain Size/BOD | Biochemical Oxygen Demand-5 Day,N-Inhib | 0.2 | U | mg/L | 0.2 | 2 |
| 3/17/2008 10:45 | Field Blank | Metals | Nickel | 0.25 | U | ug/L | 0.25 | 1 |
| 3/17/2008 10:45 | Field Blank | Metals | Selenium | 0.5 | U | ug/L | 0.5 | 2 |
| 3/17/2008 10:45 | Field Blank | Metals | Chromium | 1 | U | ug/L | 1 | 4 |
| 3/17/2008 10:45 | Field Blank | Metals | Arsenic | 0.5 | U | ug/L | 0.5 | 2 |
| 3/17/2008 10:45 | Field Blank | Metals | Magnesium | 0.04 | U | mg/L | 0.04 | 0.16 |
| 3/17/2008 10:45 | Field Blank | Metals | Iron | 30 | U | ug/L | 30 | 120 |
| 3/17/2008 10:45 | Field Blank | Metals | Aluminum | 60 | U | ug/L | 60 | 240 |
| 3/17/2008 10:45 | Field Blank | Metals | Silver | 0.025 | U | ug/L | 0.025 | 0.1 |
| 3/17/2008 10:45 | Field Blank | Metals | Lead | 0.2 | U | ug/L | 0.2 | 0.8 |
| 3/17/2008 10:45 | Field Blank | Metals | Copper | 0.5 | U | ug/L | 0.5 | 2 |
| 3/17/2008 10:45 | Field Blank | Metals | Cadmium | 0.02 | U | ug/L | 0.02 | 0.08 |
| 3/17/2008 10:45 | Field Blank | Metals | Zinc | 5 | U | ug/L | 5 | 20 |
| 3/17/2008 10:45 | Field Blank | Metals | Calcium | 0.075 | U | mg/L | 0.075 | 0.3 |
| 3/17/2008 10:45 | Field Blank | Nutrients | Organic Carbon | 1 | U | mg C/L | 1 | 5 |
| 3/17/2008 10:45 | Field Blank | Nutrients | Kjeldahl Nitrogen | 0.08 | U | mg N/L | 0.08 | 0.2 |
| 3/17/2008 10:45 | Field Blank | Nutrients | Ammonia-N | 0.01 | U | mg N/L | 0.01 | 0.02 |
| 3/17/2008 10:45 | Field Blank | Nutrients | Organic Carbon | 1 | U | mg C/L | 1 | 5 |
| 3/17/2008 10:45 | Field Blank | Nutrients | Total-P | 0.02 | U | mg P/L | 0.02 | 0.06 |
| 3/17/2008 10:45 | Field Blank | Nutrients | NO2NO3-N | 0.004 | U | mg N/L | 0.004 | 0.01 |

## Appendix E. (continued)

| Date Sampled | Field ID | Analysis | Component | Result | Remark | Units | MDL | PQL |
|---|---|---|---|---|---|---|---|---|
| 3/17/2008 10:45 | Field Blank | Nutrients | TSS | 4 | U | mg/L | 4 | 16 |
| 3/17/2008 10:45 | Field Blank | Nutrients | Nitrate-N | 0.05 | U | mg N/L | 0.05 | 0.2 |
| 3/17/2008 10:45 | Field Blank | Nutrients | Nitrite-N | 0.004 | U | mg N/L | 0.004 | 0.01 |

## Appendix F. Bioassay Meter Identification

Facility ID: Trademark Nitrogen Corporation  Test Dates: March 18 -25, 2008

| METER / DEVICE | ID | SOP | Parameter | Method | √ used |
|---|---|---|---|---|---|
| YSI Model 58 Dissolved Oxygen Meter | 90H018262 | TA06_04 | Dissolved Oxygen | SM 4500 O G | √ |
| Accumet AR-10 pH Meter | 93312105 | TA06_23 | pH | SM 4500 $H^+$ | √ |
| Mettler-Toledo SevenMulti Conductivity Meter | SN 1225117049 Probe H04257 | TA06_25 | Conductivity<br>Salinity | SM 2510 B<br>SM 2520 B | √ |
| Denver Model 225 ISE with ammonia probe | SN K01377 | TA06_17 | Ammonia | SM 4500 NH3 F | √ |
| Thermometer (DO probe) | YSI 5750 SN 05J1720 | TA06_05 | Temperature | SM-2530B modified DEP SOP-001/01 FT1400 | √ |
| Hach Pocket Colorimeter II | 05050C026378 | TA06_22 | Total Residual Chlorine | EPA 330.5 SM 4500 CL G | √ |
| Alkalinity (Hach Kit) | No SN | TA06_08 | Alkalinity | SM 2320 B | √ |
| Hardness (Hach Kit) | No SN | TA06_09 | Hardness | SM 2340 C | √ |
| OHAUS Balance 250D | 1125481084 | TA06_21 TA06_27 | Weight | - | √ |
| S-Weights | BIO-S-WEIGHTS-01 | NA | Balance check | - | √ |
| | | | | | |

SM-2530B modified to allow use electronic and non-mercury filled thermometers.
Insert other device information in blank spaces(if used).  Test ID = Facility or SRTs (acute SRT--Chronic SRT)

V 1.1 6/12/08 MF

**The Bioassay of the Trademark Nitrogen Corporation effluent sampled on March 17, 19 and 21, 2008. NPDES #FL0000647.**

Fill Out This Section For All Surface Water Discharger Inspections(CEI, CSI, CBI, PAI, XSI-RI Optional)

| Transaction Code | NPDES NUMBER | YR/MO/DA | Insp Type | Inspector | Fac Type |
|---|---|---|---|---|---|
| 1 N | 2 5 | 3 F L 0 0 0 0 6 4 7 11 | 12 0 8 0 3 1 7 | 18 B | 19 S | 20 2 |

Remarks

66

**The Priority Pollutants Analysis of the Trademark Nitrogen Corporation effluent sampled on March 17, 19 and 21, 2008. NPDES #FL0000647.**

Fill Out This Section For All Surface Water Discharger Inspections(CEI, CSI, CBI, PAI, XSI-RI Optional)

| Transaction Code | NPDES NUMBER | YR/MO/DA | Insp Type | Inspector | Fac Type |
|---|---|---|---|---|---|
| 1 N | 2 5 | 3 F L 0 0 0 0 6 4 7 11 | 12 0 8 0 3 1 7 | 18 X | 19 S | 20 2 |

Remarks

66

**Biological Analyses of the Trademark Nitrogen Corporation effluent sampled on March 17, 19 and 21, 2008. NPDES #FL0000647.**

Fill Out This Section For All Surface Water Discharger Inspections(CEI, CSI, CBI, PAI, XSI-RI Optional)

| Transaction Code | NPDES NUMBER | YR/MO/DA | Insp Type | Inspector | Fac Type |
|---|---|---|---|---|---|
| 1 N | 2 5 | 3 F L 0 0 0 0 6 4 7 11 | 12 0 8 0 3 1 7 | 18 S | 19 S | 20 2 |

Remarks

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