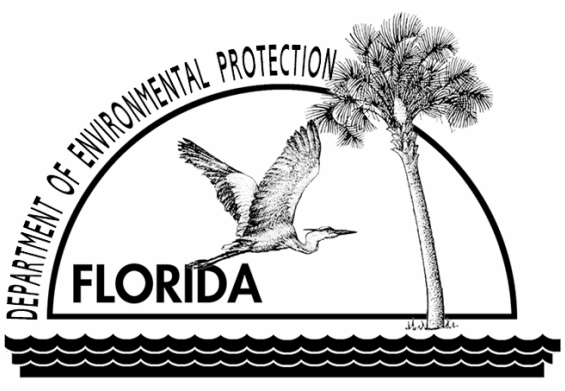


Effluent Quality Assessment

Paul Calcaterra Alligator Farm Wastewater Treatment and Disposal Facility

Okeechobee County

NPDES #FL0174696

Sampled August 18, 2008

January 2009

Biology Section
Bureau of Laboratories
Division of Environmental Assessment and Restoration

Quality Manual No. 870346G
NELAC Certification No. E31780

Paul Calcaterra Alligator Farm Wastewater Treatment and Disposal Facility, 33285 U. S. Highway 441 North, Okeechobee, Okeechobee County, Florida 34972, NPDES #FL0174696. Effluent samples for this facility were collected on August 18, 2008.

Introduction

Paul Calcaterra Alligator Farm Wastewater Treatment and Disposal Facility is an alligator farm in existence since 1986 under license by the Florida Fish and Wildlife Conservation Commission. The farm can reportedly rear up to 14,000 alligators in various growth stages, from eggs to hatchlings to market size, in enclosed pen structures, and another 700 adult breeding stock in the outdoor aquatic habitats. Water supplies to the raising pens and the processing house are provided by two separate on-site wells regulated by the St. Johns River Water Management District. The 8-inch main supply well that supplies the raising pens and the 4-inch supply well that serves the processing house have maximum daily (million gallons per day (MGD)) and annual (million gallons per year (MGY)) withdrawal allocations established, respectively, at 0.56 MGD and 25.3 MGY; and, 0.023 MGD and 0.32 MGY. Effluent is primarily discharged from the facility via land application. Discharge to surface water is permitted as a Wet-Weather Conditional Discharge (WWCD). There was no discharge to surface water during the past year. The facility has the following operations:

- eleven enclosed raising pens or grow-out buildings for hatchling alligators;
- two enclosed nursery buildings for egg incubation and hatching;
- eleven fenced outdoor “S” shape breeding ponds (some ponds are inter-connected); and,
- a processing house for harvesting the meat and hides.

The facility has two waste streams: The main waste stream is wastewater generated from the periodic flushing of the concrete floor slabs of the raising pens and nurseries. This wastewater consists mostly of animal wastes, residue of pelleted feed and feed additives, and cleaning chemicals that are used periodically when the pens are emptied. The smaller waste stream is wastewater from the processing house which consists mostly of fat, meat and blood contents residue left from harvesting of the alligator meat and hide products, and from normal building and equipment sanitation related activities.

The main waste stream is treated in a central facultative (aerobic-anaerobic) treatment pond, and from natural treatment in subsequent aerobic breeding ponds. The facility has a standby chlorinator (chlorine tablets) to disinfect the effluent from the breeding ponds in the event of a discharge to surface water. The smaller waste stream is treated in a conventional septic tank system.

Discharge to surface water consists of process wastewater and stormwater from Outfall D-001 at the north end of Breeding Pond No. 4. Discharge is authorized after rainfall events exceeding the 10-year/24-hour storm qualifying event. The threshold rainfall depth is established at 7.5 inches per day or 10 inches for any three consecutive days. Discharge from D-001 is to the perimeter spreader swale into an unnamed wetland which joins the SJRWMD jurisdictional wetland. The jurisdictional wetland drains into a roadside ditch and then flows into Boggy Branch, a tributary of Fort Drum Creek. All of the receiving waters are Class III freshwaters.

Land application of effluent consists of land application systems R-001 and R-002. Land application system R-001 has a maximum daily flow of 0.56 MGD. R-001 consists of the permeable areas at the Turtle Raising Pond facultative treatment system and the downstream breeding ponds, namely the six pond units that comprise Breeding Ponds No. 2, 3 and 4. R-001 also includes the other breeding ponds isolated from the facultative treatment pond stream, which receive normal outdoor breeding-herds waste. R-001 is located approximately at latitude 27º 33’ 01” N, longitude 80º 49’ 30” W.
Land application system R-002 has a maximum daily flow of 0.023 MGD. R-002 consists of the drain-field component for the septic tank system that serves the processing house. R-002 is located approximately at latitude 27º 32’ 59” N, longitude 80º 49’ 38” W.

The facility has no Administrative Orders, Consent Orders or mixing zones. A review of Discharge Monitoring Report (DMR) data for the past year, revealed no permit violations. (Facility information provided by Terry Davis of FDEP Southeast District).

Methods

Samples were collected from the EFF-01 NPDES sampling point for Outfall R-001 by Terry Davis (FDEP Southeast District) 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 form.

The 48-hour acute screening toxicity tests and algal growth potential test were performed following internal DEP SOPs TA07_01, TA07_02, 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.

Although the 48-hour acute screening toxicity test does not reflect the required permit conditions, passing test results provide confidence that the required permit conditions would likely be met.

Toxicity Test Results

Tests were performed August 19 - 21, 2008.

Ceriodaphnia dubia 48-hour acute screening bioassay –	LC_{50} > 100%, 5% mortality* in 100% sample at 48 hours
Cyprinella leedsi 48-hour acute screening bioassay –	LC_{50} > 100%, 0% mortality* in 100% sample at 48 hours

The bioassay samples were not acutely 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: In a 48-hour acute screening test of 100% effluent, mortality of <20% provides confidence that the effluent meets the acute toxicity criterion of Florida's Surface Water Quality Standards (Rule 62-302.500 (1)(a)4, F.A.C.). Mortality between 20-50% indicates low to moderate levels of toxicity, and further action may be required. Mortality of >50% indicates that the effluent fails to meet the minimum requirement to discharge to waters of the state (Rule 62-4.244(3)(a), F.A.C.).

Algal Growth Potential

The effluent Algal Growth Potential (AGP) was 27.7 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 result for total soluble inorganic nitrogen and ortho-phosphate suggests that the effluent is nitrogen-limited; while the chemical results for total nitrogen and total phosphorus suggest the effluent is co-limited by both nitrogen and phosphorus. The predicted AGP based on the inorganic nitrogen concentrations was 4.6 (±20%) mg dry wt/L. The predicted AGP based on the total nitrogen concentration was 91.2 (±20%) mg dry wt/L (Table 1; Miller et al. 1978).

Chemistry Results

Total residual chlorine and total ammonia were not detected in the bioassay sample in the laboratory. The total ammonia concentration in the sample collected and preserved for chemical analysis was 0.02 mg/L. Based on the pH, salinity, and temperature of the effluent as collected, the calculated unionized ammonia concentration was ≤ 0.02 mg/L.

The effluent dissolved oxygen (DO) as measured in the field (~4.5 mg/L) was less than the DO criterion for Class III fresh waters (≥ 5.0 mg/L; 62-302.530(30), F.A.C.); however, this was not a violation of permit conditions nor surface water criterion since there is not a DO limit in the permit and there was no discharge of effluent to surface waters.

No substances of concern were detected in the effluent at levels that did not comply with Class III fresh water quality criteria (62-302.530, F.A.C.). Aluminum was detected in the effluent. Chromium-III and lead were detected between the laboratory method detection limits (MDL) and practical quantitation limits (PQL).

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 bioassay lab equipment.

Conclusion

The effluent sample collected from the EFF-01 NPDES sampling point for Outfall R-001 for this facility on August 18, 2008, was not acutely toxic to either test species during the 48-hour acute screening bioassays. The effluent AGP result exceeded the "problem" threshold for fresh receiving waters. Effluent DO (~ 4.5 mg/L) was less than the Class III freshwater criterion for DO (≥ 5.0 mg/L; 62-302.530(30), F.A.C.); but was not in violation of the permit (no DO limit) and did not violate surface water criterion due to lack of discharge. Other 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 Paul Calcaterra Alligator Farm on August 18, 2008.

Paul Calcaterra Alligator Farm WWTDF							
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	27.7	4.6 ± 0.9	7.7 ± 1.5	91.2 ± 18.2	64.5 ± 12.9	6.7	16.0

Table 2. Effluent limits, Class III Freshwater Criteria and chemical and microbiological data for samples collected from the Paul Calcaterra Alligator Farm facility on August 18, 2008.

Paul Calcaterra Alligator Farm WWTDF NPDES# FL0174696	Class III Criteria	Effluent Limits	Effluent	
Metals (µg/L unless otherwise noted)				
Aluminum	-	-	280	
Arsenic	≤ 50	-	0.5	U
Cadmium	≤ 0.3 b	-	0.02	U
Calcium (mg/L)	-	-	18.5	
Chromium-III	≤ 89.5 b	-	0.94	I
Copper	≤ 9.7 b	-	0.5	U
Iron	≤ 1,000	-	30	U
Lead	≤ 3.4 b	-	0.3	I
Magnesium (mg/L)	-	-	14.2	
Nickel	≤ 54.2 b	-	0.25	U
Selenium	≤ 5.0	-	0.5	U
Silver	≤ 0.07	-	0.025	U
Zinc	≤ 124.5 b	-	5	U
Nutrients (mg/L)				
Ortho-phosphate	-	-	0.018	I
Total Phosphorus	-	Report	0.15	
Total Ammonia	-	Report	0.02	I
Un-ionized Ammonia	≤ 0.02 s	-	≤ 0.02	c
Nitrate and Nitrite	-	-	0.1	
Total Kjeldahl Nitrogen	-	Report	2.3	
Organic Nitrogen	-	-	2.28	c
Total Nitrogen	-	-	2.4	c
General Physical and Chemical Parameters				
Dissolved Oxygen (mg/L)	≥ 5.0	-	~ 4.5	c
pH (SU)	6.0 - 8.5	6.0 - 8.5 s	7.1	
Conductivity (µmhos/cm)	≤ 1,275	Report	359	
Temperature °C	-	-	28.4	
Sample Depth (m)	-	-	0.01	
Total Residual Chlorine (mg/L)	≤ 0.01	-	0.03	U£Q
Chlorophyll a (µg/L) - Corrected	-	-	78	
Phaeophytin (µg/L)	-	-	19	
Total Suspended Solids (mg/L)	-	Report	23	I
CBOD, 5 day (mg/L), N - inhibited	-	Report	3	V
Flow (MGD)	-	Report	No Discharge *	
Hardness (mg $CaCO_3$)	-	-	104.67	c
Microbiology (Number of colony forming units/100ml)				
Fecal Coliforms	≤ 400s #	800 s	5	

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

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

V - Analyte was detected in both sample and method blank

- If 10 or fewer samples are taken within a month, the single sample limit is 400 CFU (62-302.530(6), F.A.C.)

* - No discharge to surface waters during the past year

£ - Analyses performed in bioassay laboratory

Q - Sample held beyond normal holding time

Florida Department of Environmental Protection

Central Laboratory Sample Submittal Form

Request Number: RQ-2008-08-18-34
Paul Calcaterra Alligator Farm

Customer: SE-DIST

Project ID: FYI3

PMAS: 1143

Event ID *

Requester: Jennifer Paris

Collected By: Terry Davis

Sampling Agency: Southeast District

Field Report Prepared By: Terry Davis

Send Final Report To ~~Todd Brown~~ Terry Davis

Lab ID *	Location R-001			☐ Comp ☒ Grab	Collection (begin) Date 8/7/08 Time 1030 Eastern Central	Collection (end) Date Time Eastern Central	Bottle Group(s)** A
	Field ID SI-PC-1			Tot Res Chlorine (mg/L)	Diss Oxygen ~~(mg/L)~~ 5.8.20	Storet Station Number	
	Matrix (Include type e.g. Salt, Fresh, etc) water, fresh	Temp (C) 28.41	pH 7.12	Sample Depth ☒ m ☐ ft .01	359	☐ Salinity (PPTh) ☒ Sp Conductance (umho/cm)	NPDES Number
	Latitude o ' "	Longitude o ' "		Comments Pres as rqd. pH <2			
Lab ID *	Location Blank			☐ Comp ☒ Grab	Collection (begin) Date 8/18/08 Time 1050 Eastern Central	Collection (end) Date Time Eastern Central	Bottle Group(s)** B
	Field ID SI-PC-2			Tot Res Chlorine (mg/L)	Diss Oxygen (mg/L)	Storet Station Number	
	Matrix (Include type e.g. Salt, Fresh, etc) water, d.i.	Temp (C)	pH	Sample Depth ☒ m ☐ ft .01		☐ Salinity (PPTh) ☐ Sp Conductance (umho/cm)	NPDES Number
	Latitude o ' "	Longitude o ' "		Comments Pres as rqd pH <2			
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 o ' "	Longitude o ' "		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 o ' "	Longitude o ' "		Comments 2 bottles received in Tox by SD, 1005 8.19.08			

Relinquished By:	Date/Time	Shipping Method:	Received By:	Date/Time	Relinquished By:	Date/Time	Received By:	Date/Time
[illegible]	8/8/1330	DHL	Tahmoures Ghotbi	08/19/08 8:50				

* Shaded Areas for Lab use only.

** Please see reverse side for Bottle Group information.

last revised October 1, 2003

FDEP Biology Laboratory - Acute Screen Bioassay Bench Sheets

Facility: Paul Cafcaterra Alligator Farm WWTP & Disposal Facility
Address: 33285 US Hwy 441 N
City: Okeechobee **County:** Okeechobee
Contact/District: Terry Davis / SE-DIST
NPDES Permit #: FL 0174696
LIMS Job #: TLH-2008-08-19-45 **LIMS Sample #:** 112654 7
Lims Data Entry: MF 8-22-08 Data Entry Verification: SD 822-08

Sample Collection: Date: 8-18-08 Time: 10:30
Hold Time Start: Date: 8-18-08 Time: 10:30

Comments:

Instructions (for below): Circle appropriate wording. If yes is circled, complete blanks.

Test Type: Screen
(Static) / Static Renewal / Flow-through
Test Duration: (48) 96 Hours.

Test 1 validation: **Test 1:** SOP TA 07_01
Control survival ≥90%: (Yes) No
Temperature Range ≤3°C: (Yes) No

Test 2 validation: **Test 2:** SOP TA 07_02
Control survival ≥90%: (Yes) No
Temperature Range ≤3°C: (Yes) No

Initial sample handling:

PH adjustment : yes (no) Initial pH: ____ NaOH N HCl N ____ Drops mL Final pH: ____
Aeration : yes (no) Initial DO: ____mg/L Final DO: ____mg/L Duration: ____minutes Rate: ~ ____bubbles/min
Salinity adjusted (Test 1): yes (no) Initial Salinity: ____ Final Salinity: ____ Salts Hypersaline brine
Salinity adjusted (Test 2): yes (no) Initial Salinity: ____ Final Salinity: ____ Salts Hypersaline brine
Dechlorination: yes (no) ____mL of 0.025N Sodium Thiosulfate per liter of sample. Final TRC: ____

Sample Validation:
Temperature: Shipped ≤6°C (Yes) No Hand Delivered: Cooling (received °C < collected °C) (Yes) No
Holding Time: ≤36 Hours (Yes) No (Composite-end of collection; grab-when collected; 4 in 24 - time last sample collected)

Light Intensity : 50 - 100 ft. candles
Photoperiod : 16 light 8 dark
Temperature Range °C:
Incubator # 2 min 25.0 max 26.5 mean 26.0
Incubator # ____ min. ____ max. ____ mean ____
Room B246 min. 25.1 max. 25.7 mean 25.4

Test 1:% mortality in 100% sample: 5% LC50: 7100%
Test 2:% mortality in 100% sample: 0% LC50: 7100%

Investigators' Signatures
James Donat
Bruce Abel
Marshall Faircloth

REVIEWER

Water Quality Parameters	20% DMW	Moderately Hard Water/ (Well Water)	Salt Water ASW NSW Test 1	Salt Water ASW NSW Test 2	Other:	Original Sample	Method	Measured by	Verified by
Field Total Residual Cl_2 (mg/L):	N/A	N/A	N/A	N/A			Not Measured		BA
Lab Total Residual Cl_2 (mg/L):	<0.07	<0.03				<0.03	HACH	SD	BA
Alkalinity (mg/L as $CaCO_3$):	91	158				88	HACH	BA	SD
Hardness (mg/L as $CaCO_3$):	85	140				105	HACH	BA	SD
Total Ammonia (mg/L as N):	<0.017	<0.017				<0.017	DENVER	MF	BA
Salinity (ppth):	<1	<1				0.4	YSI / (Mettler)	SD	BA

V 1.4 9/12/05

Appendix B. (continued)

FDEP Biology Laboratory - Bioassay Survival Bench Sheet

LIMS Sample #: 1126547 Test #: 1/2 SOP#:TA 07 01 Test Started: Date 8.19.08 Time: 12:30

Organism/Batch/Age: C. dubia/30 l08 / ≤24hr Test Ended: Date 8-21-08 Time: 12:15

Chamber/Test Volume: 30 / 20 ml Diluent/ Batch: DMW / 8-5-08

Food/Batch: YCT 6-15-08 P.sub 7-18-08 Artemia 0hr — 24hr — 48hr — 72hr — Artemia Lot# —

Feeding: (Prior to Test) Prior to Renewal Daily

Concentration	Replicate	Chamber #	Test Hour: 0 hr	24 hr	48 hr BR	48 hr AR	72 hr	96 hr
Ctrl	A	A	5	5	5			
	B	B	5	5	5			
	C	C	5	5	5			
	D	D	5	5	5			
100%	A	A	5	5	5			
	B	B	5	5	5			
	C	C	5	5	4 1D			
	D	D	5	5	5			
Loaded/Verified by: [illegible]		Checked by:	BA	BA	BA			

m = missing d = dead BR/AR = Before/After Renewal

Comments:

Test Results:

Screening Tests: Report LC50 as >100%, =100%, or <100%.

% mortality in 100% sample: 5% LC50: >100%

If Calculated: 95% CI______ Method______

Substitue highest test concentration used if other than 100% (example: Ocean outfall tested at 30% concentration).

PAGE V 2.1 11/01/2005

000037

Appendix B. (continued)

FDEP Biology Laboratory - Bioassay Survival Bench Sheet

LIMS Sample #: 112654 7 Test #: 2/2 SOP#:TA 07 02 Test Started: Date 8.19.08 Time: 1330

Organism/Batch/Age: C.dubia/8.19.08/ 11 days Test Ended: Date 8.21.08 Time: 1300

Chamber/Test Volume: 1000 / 500 ml Diluent/ Batch: Well / 8.18.08

Food/Batch: YCT — P.sub — Artemia 0hr 129-08 24hr — 48hr — 72hr — Artemia Lot# 38438

Feeding: (Prior to Test) Prior to Renewal Daily

Concentration	Replicate	Chamber #	Test Hour 0 hr	24 hr	48 hr BR	48 hr AR	72 hr	96 hr
Ctrl	A	D1	5	5	5			
	B	D2	5	5	5			
	C	D3	5	5	5			
	D	D4	5	5	5			
100%	A	D5	5	5	5			
	B	D6	5	5	5			
	C	D7	5	5	5			
	D	D8	5	5	5			
Loaded/Verified by: JD / BA		Checked by:	√BA	JD	JD			

m = missing d = dead BR/AR = Before/After Renewal

Comments:

Test Results:

Screening Tests: Report LC50 as >100%, =100%, or <100%.

% mortality in 100% sample: 0 % LC50: > 100%

If Calculated: 95% CI ______ Method ______

Substitue highest test concentration used if other than 100% (example: Ocean outfall tested at 30% concentration).

PAGE V 2.1 11/01/2005

000038

FDEP Biology Laboratory - Bioassay Parameter Bench Sheet

LIMS Sample # : 1126547 **Test #:** 1 of 2

TEST SOP #: TA07_ D **Test Species:** (*Ceriodaphnia dubia*) *Cyprinella leedsi* *Pimephales promelas* *Americamysis bahia* *Menidia beryllina* Other:_____

Concentration Ctrl	0 Hr.	24 Hr.	48 Hr. before renewal	48 Hr. after renewal	72 Hr.	96 Hr.
Replicate	M		A			
pH (S.U.)	8.0		8.0			
Temperature °C	24.3		24.5			
Dissolved Oxygen mg/L	7.5		6.8			
Conductivity (µmhos) mmhos	176.4		BA 178			
(initials) Measured by:	BA		BA			
(initials) Recorded by:	BA		BA			

Comments:

Concentration 100%	0 Hr.	24 Hr.	48 Hr. before renewal	48 Hr. after renewal	72 Hr.	96 Hr.
Replicate	M		A			
pH (S.U.)	7.4		8.1			
Temperature °C	25.2		24.4			
Dissolved Oxygen mg/L	5.8		6.2			
Conductivity (µmhos) mmhos	367		376			
(initials) Measured by:	BA		BA			
(initials) Recorded by:	BA		BA			

Comments:

Concentration	0 Hr.	24 Hr.	48 Hr. before renewal	48 Hr. after renewal	72 Hr.	96 Hr.
Replicate						
pH (S.U.)						
Temperature °C						
Dissolved Oxygen mg/L						
Conductivity µmhos mmhos						
(initials) Measured by:						
(initials) Recorded by:						

Comments:

Concentration	0 Hr.	24 Hr.	48 Hr. before renewal	48 Hr. after renewal	72 Hr.	96 Hr.
Replicate						
pH (S.U.)						
Temperature °C						
Dissolved Oxygen mg/L						
Conductivity µmhos mmhos						
(initials) Measured by:						
(initials) Recorded by:						

Comments:

000039

V 2.0 2/24/04

FDEP Biology Laboratory - Bioassay Parameter Bench Sheet

LIMS Sample # : 1126547 **Test #:** 2 of 2

TEST SOP #: TA07_ 02 **Test Species:** *Ceriodaphnia dubia* (*Cyprinella leedsi*) *Pimephales promelas* *Americamysis bahia* *Menidia beryllina* Other:_____

Concentration Ctrl	0 Hr.	24 Hr.	48 Hr. before renewal	48 Hr. after renewal	72 Hr.	96 Hr.
Replicate	A	B	C			
pH (S.U.)	7.8	8.2	7.9			
Temperature °C	24.6	24.7	24.2			
Dissolved Oxygen mg/L	7.6	6.9	6.6			
Conductivity (µmhos) mmhos	276	275	272			
(initials) Measured by:	JD	JD	JD			
(initials) Recorded by:	JD	JD	JD			

Comments:

Concentration 100%	0 Hr.	24 Hr.	48 Hr. before renewal	48 Hr. after renewal	72 Hr.	96 Hr.
Replicate	A	B	C			
pH (S.U.)	7.4	7.8	7.6			
Temperature °C	25.9	24.7	24.2			
Dissolved Oxygen mg/L	5.8	6.6	6.8			
Conductivity (µmhos) mmhos	368	362	358			
(initials) Measured by:	JD	JD	JD			
(initials) Recorded by:	JD	JD	JD			

Comments:

Concentration	0 Hr.	24 Hr.	48 Hr. before renewal	48 Hr. after renewal	72 Hr.	96 Hr.
Replicate						
pH (S.U.)						
Temperature °C						
Dissolved Oxygen mg/L						
Conductivity µmhos mmhos						
(initials) Measured by:						
(initials) Recorded by:						

Comments:

Concentration	0 Hr.	24 Hr.	48 Hr. before renewal	48 Hr. after renewal	72 Hr.	96 Hr.
Replicate						
pH (S.U.)						
Temperature °C						
Dissolved Oxygen mg/L						
Conductivity µmhos mmhos						
(initials) Measured by:						
(initials) Recorded by:						

Comments:

V 2.0 2/24/04

Appendix C. SRT Bench Sheets and Statistical Analysis

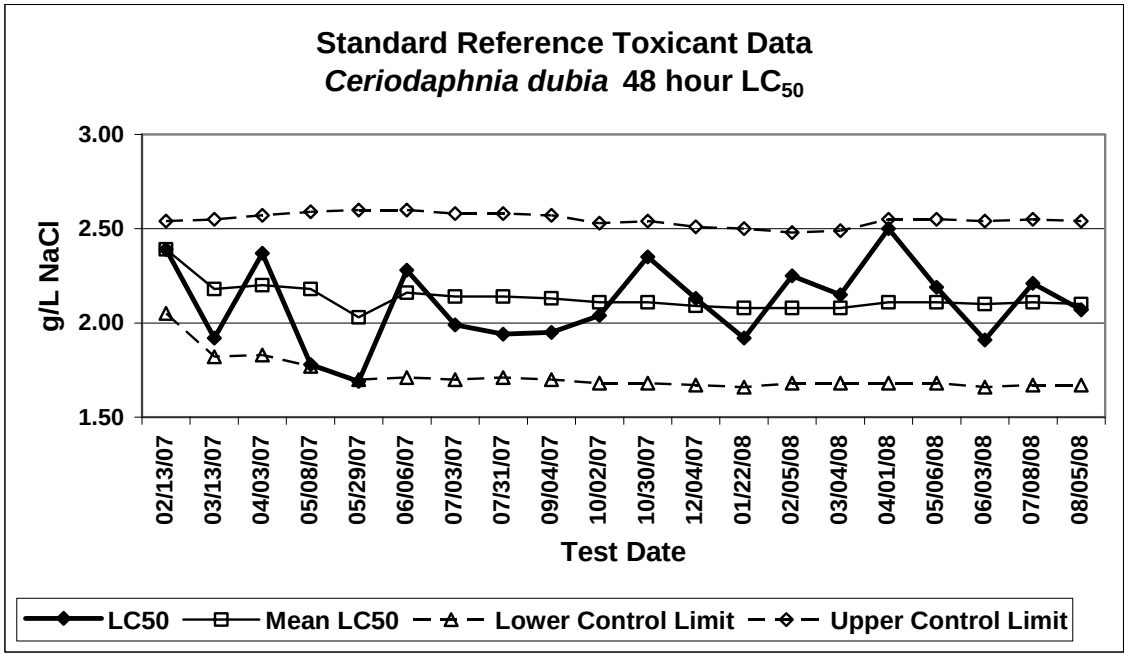
Florida Department of Environmental Protection
Bureau of Laboratories

Standard Reference Toxicant (SRT) Test Data - 48-hour Acute Toxicity
Ceriodaphnia dubia (water flea) - Sodium Chloride (NaCl)

Date	LC50	Mean LC50	Std Dev	Lower Control Limit	Upper Control Limit
02/13/07	2.39	2.39	0.17	2.05	2.54
03/13/07	1.92	2.18	0.18	1.82	2.55
04/03/07	2.37	2.20	0.18	1.83	2.57
05/08/07	1.78	2.18	0.21	1.77	2.59
05/29/07	1.69	2.03	0.23	1.70	2.60
06/06/07	2.28	2.16	0.22	1.71	2.60
07/03/07	1.99	2.14	0.22	1.70	2.58
07/31/07	1.94	2.14	0.22	1.71	2.58
09/04/07	1.95	2.13	0.22	1.70	2.57
10/02/07	2.04	2.11	0.21	1.68	2.53
10/30/07	2.35	2.11	0.21	1.68	2.54
12/04/07	2.13	2.09	0.20	1.67	2.51
01/22/08	1.92	2.08	0.20	1.66	2.50
02/05/08	2.25	2.08	0.20	1.68	2.48
03/04/08	2.15	2.08	0.20	1.68	2.49
04/01/08	2.50	2.11	0.22	1.68	2.55
05/06/08	2.19	2.11	0.22	1.68	2.55
06/03/08	1.91	2.10	0.22	1.66	2.54
07/08/08	2.21	2.11	0.22	1.67	2.55
08/05/08	2.07	2.10	0.22	1.67	2.54

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

CV = 10.32
EPA reference 75th quartile CV = 29%



Appendix C. (continued)

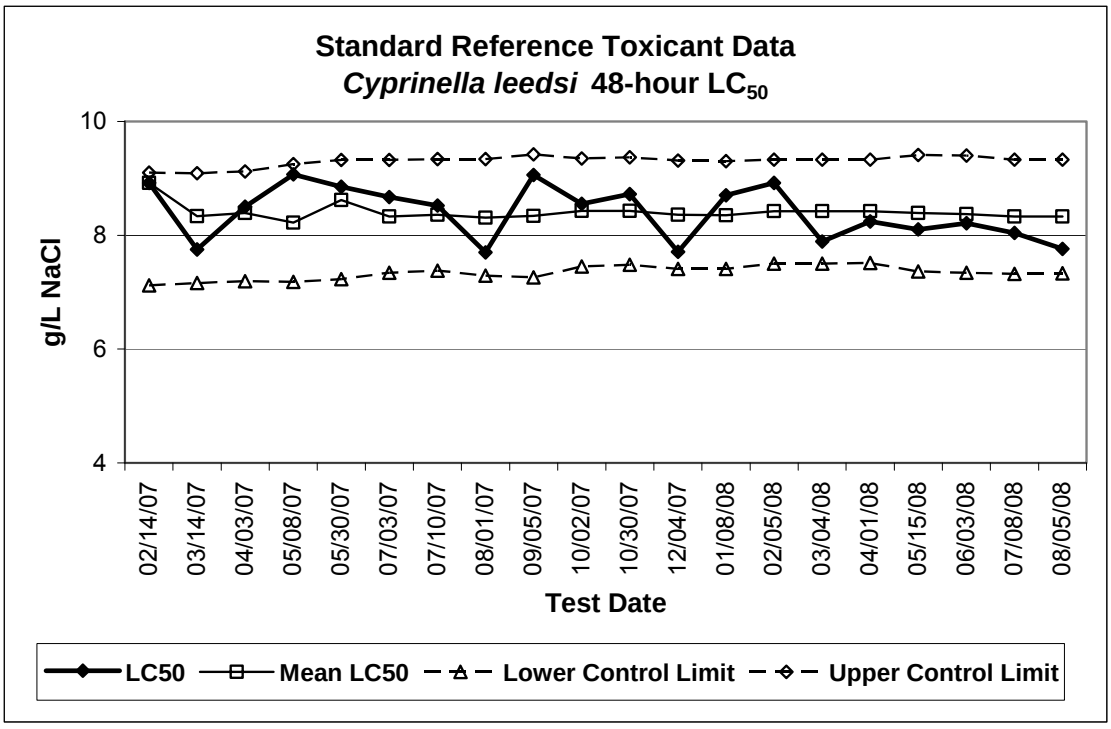
Florida Department of Environmental Protection
Bureau of Laboratories

Standard Reference Toxicant (SRT) Test Data - 48-hour Acute Toxicity
Cyprinella leedsi (bannerfin shiner) - Sodium Chloride (NaCl)

Date	LC50	Mean LC50	Std Dev	Lower Control Limit	Upper Control Limit
02/14/07	8.92	8.92	0.49	7.12	9.10
03/14/07	7.75	8.34	0.48	7.16	9.09
04/03/07	8.50	8.39	0.48	7.19	9.12
05/08/07	9.07	8.22	0.52	7.18	9.25
05/30/07	8.85	8.62	0.53	7.23	9.32
07/03/07	8.67	8.33	0.50	7.34	9.32
07/10/07	8.52	8.36	0.49	7.38	9.34
08/01/07	7.7	8.31	0.51	7.29	9.34
09/05/07	9.06	8.34	0.54	7.26	9.42
10/02/07	8.55	8.43	0.47	7.45	9.35
10/30/07	8.72	8.43	0.47	7.48	9.37
12/04/07	7.71	8.36	0.47	7.41	9.31
01/08/08	8.7	8.35	0.47	7.41	9.30
02/05/08	8.92	8.42	0.46	7.50	9.33
03/04/08	7.89	8.42	0.46	7.50	9.33
04/01/08	8.24	8.42	0.45	7.51	9.33
05/15/08	8.1	8.39	0.51	7.36	9.41
06/03/08	8.21	8.37	0.51	7.34	9.40
07/08/08	8.04	8.33	0.50	7.32	9.33
08/05/08	7.76	8.33	0.50	7.33	9.33

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

CV = 6.02
EPA reference 75th quartile CV = 19%



Appendix C. (continued)

FDEP Biology Laboratory - Acute SRT Bench Sheet

Test Organism: (check one) ☐ *Americamysis bahia* Method 2007.0 TA07.03 ☐ *Menidia Beryllina* Method 2006.0 TA07.04 ☐ *Hyalella azteca* Method 100.1 EPA-600-R-99-064 ☒ *Ceriodaphnia dubia* Method 2002.0 TA07.01 ☐ *Cyprinella leedsi* Method 2000.0 TA07.02 ☐ *Pimephales promelas* Method 2000.0 TA07.02

Method from EPA-821-R-02-012

Test Type: ☐ 96 hour non-renewal ☒ 96 hour renewal

Test Page 1 of 2

Diluent/ Batch #: DMW 18-508

Toxicant/ Batch#: NaCl 18-4-08

Organism Batch: 28-08 Age: <24 hr

Test Beginning: Date: 8-5-08 Time: 11:45

Test Ending: Date: 8-9-08 Time: 1305

Water Quality Parameters:	Diluent	SOP	Initials
Total Residual Cl_2 (mg/L):	<0.03	TA06_22	SD
Alkalinity (mg/L as $CaCO_3$):	89	TA06_08	BA
Hardness (mg.L as $CaCO_3$):	83	TA06_09	BA
Total Ammonia (mg/L as N):	<0.07	TA06_17	BA
Conductivity: µmhos (mmhos)	0.177	TA06_25	BA
Salinity (ppth):	<1	TA06_25	BA

Food: ☑ YCT Batch: 6-18-08; ☑ *P. subcapitata* 7-18-08; ☐ Artemia X

Artemia Lot #: —

Artemia Batch: 0 hr: — 24 hr: — 48 hr: — 72 hr: —

Chamber size: 30 mL

Test volume: 20 mL per replicate

Concentrations prepared by: BA

Concentration g/L MF

Temperature Range °C:

Incubator # 2 min 25.4 max 26.4 mean 25.9

Incubator # ___ min ___ max ___ mean ___

Room B246 min 25.2 max 26.0 mean 25.6

Light Intensity: 50-100 Ft. candles

Photoperiod: 16 hours Light / 8 hours dark

Feeding: √as needed

- ☑ Before test
- ☐ 0 hr
- ☐ 24 hr
- ☑ 48 hr
- ☐ 72 hr

all species 2 hrs prior to test

mysids daily

all species except *H. azteca* 2 hr prior to renewal

H. azteca 0 hr and 48 hr

Concentration µg mg g/L	Replicate	Chamber #	Test Hour 0 hr	24 hr	48 hr BR	48 hr AR	72 hr	96 hr
Control	A	A	5	5	5	5	5	5
	B	B	5	5	5	5	5	5
	C	C	5	5	5	5	5	5
	D	D	5	5	5	5	5	5
1.5ppth	A	A	5	5	5	5	5	5
	B	B	5	5	5	5	5	5
	C	C	5	5	5	5	3 2D	3
	D	D	5	5	4 1D	4	4	4
2.0ppth	A	A	5	5	4 1D	4	3 1D	3
	B	B	5	5	2 3D	2	2	2
	C	C	5	5	5	5	3 2D	2 1D
	D	D	5	4 1D	3 1D	3	1 2D	1
2.5ppth	A	A	5	4 1D	0 4D	—	—	
	B	B	5	2 3D	0 2D	—	—	
	C	C	5	3 2D	0 3D	—	—	
	D	D	5	5	0 5D	—	—	
3.0ppth	A	A	5	0 5D	—	—	—	
	B	B	5	0 4D	—	—	—	
	C	C	5	2 3D	0 2D	—	—	
	D	D	5	1 4D	0 1D	—	—	
4.0ppth	A	A	5	0 5D	—	—	—	
	B	B	5	0 5D	—	—	—	
	C	C	5	0 5D	—	—	—	
	D	A	5	0 5D	—	—	—	
Organisms loaded by: BA	Checked by:		BA	BA	BA	BA	MF	

Loading Verified by: JD

m = missing d = dead AR = after renewal BR = before renewal hr = hour

H. azteca		
0 hr	Alk	Hrd
Well		
100%		
96 hr	Alk	Hrd
Well		
100%		

Comments: Incubated in Incubator #2

Investigators' Signatures

Bruce Alexander

Marshall Faircloth

Reviewer: Kathleen Wolfe

Method: Spearman-Karber

Statistics: LC50	95% confidence intervals Lower	Upper
48 hour: 2.07	1.95	2.12
96 hour: 1.87	1.872	2.03

BA

PAGE 000001

Revised 7/15/2008 mf

Appendix C. (continued)

FDEP Biology Laboratory - Acute SRT Bench Sheet
0-96 hour parameters
Method from EPA-821-R-02-012

Test Page 2 of 2

Test Organism: (check one)
- ☐ *Americamysis bahia* Method 2007.0 TA07.03
- ☐ *Menidia Beryllina* Method 2006.0 TA07.04
- ☐ *Hyalella azteca* Method 100.1 EPA-600-R-99-064
- ☒ *Ceriodaphnia dubia* Method 2002.0 TA07.01
- ☐ *Cyprinella leedsi* Method 2000.0 TA07.02
- ☐ *Pimephales promelas* Method 2000.0 TA07.02

Test Beginning: Date: 8.5.08 Time: 1145
Test Ending: Date: 8.7.08 Time: 1305

Test Type: ☐ 96 hour non-renewal ☒ 96 hour renewal

hr = hour AR = after renewal BR = before renewal

Concentration µg mg (circle) mL = liter	Replicate	Chamber #	Test Time	pH (SU)	Temperature (°C)	Dissolved Oxygen (mg/L)	Conductivity ☐ µmhos ☒ mmhos
Control	M	—	0 hr	8.1	24.2	7.4	0.177
	—	—	24 hr	—	—	—	—
	A	—	48 hr BR	8.2	24.0	~~6.7~~ ~~7.5~~ ~~7.5~~	0.197
	M	—	48 hr AR	8.0	24.0	~~BR~~ 6.7 7.5	0.181
		—	72 hr				
	A	—	96 hr	8.0	24.7	7.6	1.91
1.5 ppt	M	—	0 hr	8.1	24.4	7.4	2.92
	—	—	24 hr	—	—	—	—
	A	—	48 hr BR	8.1	24.0	6.8	2.96
	M	—	48 hr AR	8.0	24.1	7.4	2.84
	—	—	72 hr				~~2.85~~ mF
	A	—	96 hr	8.0	24.7	7.4	2.85
2.0 ppt	M	—	0 hr	8.1	24.5	7.3	3.82
	—	—	24 hr	—	—	—	—
	A	—	48 hr BR	8.1	24.0	7.1	3.90
	M	—	48 hr AR	8.0	24.2	7.4	3.73
	—	—	72 hr				~~3.80~~ mF
	A	—	96 hr	8.0	24.6	7.5	3.80
2.5 ppt	M	—	0 hr	8.1	24.4	7.3	4.70
	—	—	24 hr	—	—	—	—
	A	—	48 hr BR	8.1	24.0	7.1	4.84
		—	48 hr AR				
		—	72 hr				
		—	96 hr				
3.0 ppt	M	—	0 hr	8.1	24.4	7.3	5.55
	—	—	24 hr	—	—	—	—
	A	—	48 hr BR	8.1	24.0	7.1	5.75
		—	48 hr AR				
		—	72 hr				
		—	96 hr				
4 ppt	M	—	0 hr	8.1	24.3	7.2	7.24
	A	—	24 hr	8.0	24.0	7.0	7.43
	~~A~~	—	48 hr BR				
		—	48 hr AR				
		—	72 hr				
		—	96 hr				

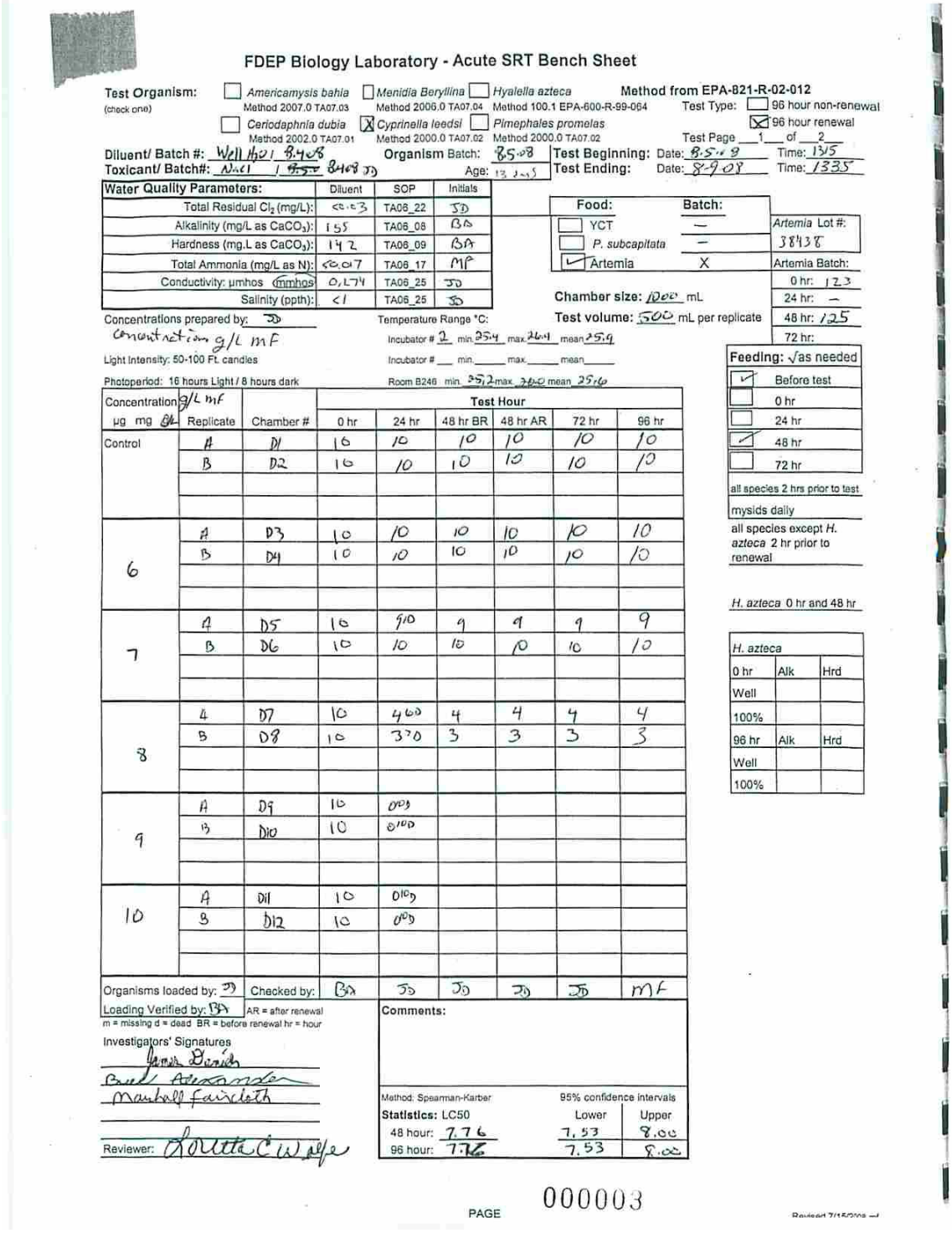
Investigators Signatures
Bruce Alexander
Marshall Brownlath
Martha C. Wolfe
Reviewed By:

Measured by:
0 hr: BA
24 hr: BA
48 hr BR: BA
48 hr AR: BA
72 hr: —
96 hr: mF

Comments: M = Motor Vial BA

PAGE 000002

Appendix C. (continued)



FDEP Biology Laboratory - Acute SRT Bench Sheet

Test Organism: (check one) ☐ *Americamysis bahia* Method 2007.0 TA07.03 ☐ *Menidia Beryllina* Method 2006.0 TA07.04 ☐ *Hyalella azteca* Method 100.1 EPA-600-R-99-064 ☐ *Ceriodaphnia dubia* Method 2002.0 TA07.01 ☒ *Cyprinella leedsi* Method 2000.0 TA07.02 ☐ *Pimephales promelas* Method 2000.0 TA07.02

Method from EPA-821-R-02-012

Test Type: ☐ 96 hour non-renewal ☒ 96 hour renewal

Test Page 1 of 2

Diluent/ Batch #: Well H2O / B.4.08 Organism Batch: 8.5.08 Test Beginning: Date: 8.5.08 Time: 1345

Toxicant/ Batch#: NaCl / ~~8.5.08~~ 8.4.08 JD Age: 13 days Test Ending: Date: 8.9.08 Time: 1335

Water Quality Parameters:	Diluent	SOP	Initials
Total Residual Cl_2 (mg/L):	<0.03	TA06_22	JD
Alkalinity (mg/L as $CaCO_3$):	155	TA06_08	BA
Hardness (mg.L as $CaCO_3$):	142	TA06_09	BA
Total Ammonia (mg/L as N):	<0.07	TA06_17	MF
Conductivity: µmhos (mmhos)	0,174	TA06_25	JD
Salinity (ppth):	<1	TA06_25	JD

Food:	Batch:
☐ YCT	—
☐ *P. subcapitata*	—
☑ Artemia	X

Artemia Lot #: 3843B

Artemia Batch:

0 hr: 123

24 hr: —

48 hr: 125

72 hr:

Chamber size: 1000 mL

Test volume: 500 mL per replicate

Concentrations prepared by: JD

Concentration g/L MF

Temperature Range °C:

Incubator # 2 min. 25.4 max. 26.1 mean 25.9

Incubator # ___ min. ___ max. ___ mean ___

Room B246 min. 25.2 max. 26.0 mean 25.6

Light Intensity: 50-100 Ft. candles

Photoperiod: 16 hours Light / 8 hours dark

Feeding: √as needed

- ☑ Before test
- ☐ 0 hr
- ☐ 24 hr
- ☑ 48 hr
- ☐ 72 hr

all species 2 hrs prior to test

mysids daily

all species except *H. azteca* 2 hr prior to renewal

H. azteca 0 hr and 48 hr

Concentration g/L MF µg mg %	Replicate	Chamber #	Test Hour 0 hr	24 hr	48 hr BR	48 hr AR	72 hr	96 hr
Control	A	D1	10	10	10	10	10	10
	B	D2	10	10	10	10	10	10
6	A	D3	10	10	10	10	10	10
	B	D4	10	10	10	10	10	10
7	A	D5	10	9/0	9	9	9	9
	B	D6	10	10	10	10	10	10
8	A	D7	10	4/0	4	4	4	4
	B	D8	10	3/0	3	3	3	3
9	A	D9	10	0/0				
	B	D10	10	0/0				
10	A	D11	10	0/0				
	B	D12	10	0/0				
Organisms loaded by: JD	Checked by:		BA	JD	JD	JD	JD	MF
Loading Verified by: BA	AR = after renewal							

m = missing d = dead BR = before renewal hr = hour

H. azteca		
0 hr	Alk	Hrd
Well		
100%		
96 hr	Alk	Hrd
Well		
100%		

Comments:

Investigators' Signatures

Jason Dierth

Brad Alexander

Marshall Faircloth

Reviewer: Collette C. Wolfe

Method: Spearman-Karber

Statistics: LC50	95% confidence intervals Lower	Upper
48 hour:	7.76	7.53	8.00
96 hour:	7.76	7.53	8.00

PAGE 000003

Revised 7/15/2008

Appendix C. (continued)

FDEP Biology Laboratory - Acute SRT Bench Sheet
0-96 hour parameters
Method from EPA-821-R-02-012

Test Page 2 of 2

Test Organism: (check one)
- ☐ *Americamysis bahia* Method 2007.0 TA07.03
- ☐ *Menidia Beryllina* Method 2006.0 TA07.04
- ☐ *Hyalella azteca* Method 100.1 EPA-600-R-99-064
- ☐ *Ceriodaphnia dubia* Method 2002.0 TA07.01
- ☒ *Cyprinella leedsi* Method 2000.0 TA07.02
- ☐ *Pimephales promelas* Method 2000.0 TA07.02

Test Beginning: Date: 8-5-08 Time: 1515
Test Ending: Date: 8-9-08 Time: 1335
Test Type: ☐ 96 hour non-renewal ☒ 96 hour renewal

concentration/L mL hr = hour AR = after renewal BR = before renewal

Concentration µg mg ⓪	Replicate	Chamber #	Test Time	pH (SU)	Temperature (° C)	Dissolved Oxygen (mg/L)	Conductivity ☐ µmhos ☒ mmhos
Control	A	D1	0 hr	7.9	24.0	7.5	0.274
	B	D2	24 hr	8.1	24.1	6.4	0.290
	A	D1	48 hr BR	8.0	24.4	6.6	0.333
	A	D1	48 hr AR	7.9	24.0	7.3	0.295
	B	D2	72 hr	8.0	24.8	6.3	0.279
	A	D1	96 hr	7.8	25.0	7.4	0.302
6	A	D3	0 hr	7.8	24.1	7.4	11.1
	B	D4	24 hr	8.0	24.3	6.1	10.9
	A	D3	48 hr BR	8.1	24.5	6.5	10.8
	A	D3	48 hr AR	7.9	24.0	7.1	10.8
	B	D4	72 hr	8.0	24.7	6.2	10.8
	A	D3	96 hr	8.0	25.1	6.6	10.9
7	A	D5	0 hr	7.9	24.1	7.4	12.8
	B	D6	24 hr	8.0	24.3	6.0	12.7
	A	D5	48 hr BR	8.2	24.4	6.4	12.6
	A	D5	48 hr AR	8.0	24.0	7.0	12.4
	B	D6	72 hr	8.0	24.7	6.3	13.1
	A	D5	96 hr	8.0	25.1	6.6	13.1
8	A	D7	0 hr	7.9	24.0	7.4	14.8
	B	D8	24 hr	8.0	24.3	6.0	14.7
	A	D7	48 hr BR	8.1	24.2	6.4	14.6
	A	D7	48 hr AR	8.0	24.0	7.0	14.7
	B	D8	72 hr	8.1	24.3	6.4	14.8
	A	D7	96 hr	8.1	25.2	6.5	14.8
9	A	D9	0 hr	7.9	24.0	7.5	16.2
	B	D10	24 hr	8.0	24.4	5.9	16.1
			48 hr BR				
			48 hr AR				
			72 hr				
			96 hr				
10	A	D11	0 hr	7.9	24.0	7.4	17.9
	B	D12	24 hr	7.9	24.4	5.6	17.7
			48 hr BR				
			48 hr AR				
			72 hr				
			96 hr				

Investigators Signatures
Jesse Daniels
Bruce Abel
Marshall Faircloth

Reviewed By: Kristie C. Wolfe

Measured by:
0 hr: JD
24 hr: JD
48 hr BR: JD
48 hr AR: JD
72 hr: JD
96 hr: MF

Comments:

PAGE 000004

Appendix C. (continued)

***Ceriodaphnia dubia* Acute Standard Reference Toxicant Test**

DATE: 8-5-08 DURATION: 48 hr TOXICANT : NaCl
SPECIES: *Ceriodaphnia dubia*

RAW DATA: Concentration (g/L)	Number Exposed	Mortalities
.00	20	0
1.50	20	1
2.00	20	6
2.50	20	20
3.00	20	20
4.00	20	20

SPEARMAN-KARBER TRIM: 5.00%

SPEARMAN-KARBER ESTIMATES: **LC50: 2.07**
95% LOWER CONFIDENCE: 1.95
95% UPPER CONFIDENCE: 2.20

***Cyprinella leedsi* Acute Standard Reference Toxicant Test**

DATE: 8-5-08 DURATION: 48 hr TOXICANT : NaCl
SPECIES: *Cyprinella leedsi*

RAW DATA: Concentration (g/L)	Number Exposed	Mortalities
.00	20	0
6.00	20	0
7.00	20	1
8.00	20	13
9.00	20	20
10.00	20	20

SPEARMAN-KARBER TRIM: .00%

SPEARMAN-KARBER ESTIMATES: **LC50: 7.76**
95% LOWER CONFIDENCE: 7.53
95% UPPER CONFIDENCE: 8.00

Appendix D. Test Organism Receipts

MARINCO BIOASSAY LABORATORY
The Aquatic Toxicology Specialists

NELAP Certification # E84191

Organism Shipment Record

State of Florida Aquaculture Certificate Number AQ0668007

Date: 8/4/08

Shipped to: FDEP

P.O. No: _____

Species	Quantity	Age	Brood Number	Temp. °C	pH S.U.	Salinity ‰
Mysidopsis bahia	150	4 DAYS	MS080731	25	7.8	20
Menidia beryllina	150	11 DAYS	SS080724	25	7.7	19.5
						Hardness
Cyprinella leedsi	150	12 DAYS	CL080723	25	7.8	90
Pimephales promelas	200	8 DAYS	FM080727 JU	25	7.9	90

Packed by: JJ

Shipped Via: Fed Ex

Notes: _____

Thank you for your order.

4569 Samuel Street · Sarasota, FL 34233 · Tel: 941-925-3594 · Fax: 941-922-3874 · Web: www.biologylab.com

Appendix D. (continued)

MARINCO BIOASSAY LABORATORY
The Aquatic Toxicology Specialists

NELAP Certification # E84191

Organism Shipment Record

State of Florida Aquaculture Certificate Number AQ0668007

Date: 8/18/08

Shipped to: FDEP

P.O. No: _____

Species	Quantity	Age	Brood Number	Temp.	pH	Salinity
Mysidopsis bahia						
Menidia beryllina						
						Hardness
Cyprinella leedsi	100	10 DAYS	CL080808	25°C	7.8	80 mg/L
Pimephales promelas						

Packed by: [signature]

Shipped Via: Fed Ex

Notes: _____

Thank you for your order.

4569 Samuel Street · Sarasota, FL 34233 · Tel: 941-925-3594 · Fax: 941-922-3874 · Web: www.biologylab.com

Appendix E. Chemical Analyses performed on the effluent from the EFF-01 NPDES sampling point for the Paul Calcaterra Alligator Farm facility Outfall R-001, sampled on August 18, 2008.

Date Sampled	Field ID	Analysis	Component	Result	Remark	Units	MDL	PQL
8/18/2008 10:30	Effluent R-001	AGP/LN	Algal Growth Potential	27.7		mg DryWt/L	0.3	0.9
8/18/2008 10:30	Effluent R-001	Chlorophyll/Grain Size/BOD	Biochemical Oxygen Demand-5 Day,N-Inhib	3	V	mg/L	0.2	2
8/18/2008 10:30	Effluent R-001	Chlorophyll/Grain Size/BOD	Phaeophytin-a	19		ug/L	0.8	2.4
8/18/2008 10:30	Effluent R-001	Chlorophyll/Grain Size/BOD	Chlorophyll-a, Corrected	78		ug/L	1.8	5.7
8/18/2008 10:30	Effluent R-001	Overflow	Fecal Coliforms-Membrane Filter	5		#/100 mL	1	
8/18/2008 10:30	Effluent R-001	Toxicology	Bioassay-Acute-Screen-FW-Fish, LC50	100	L	LC50		
8/18/2008 10:30	Effluent R-001	Toxicology	Bioassay-Acute-Screen-FW-C.dubia, LC50	100	L	LC50		
8/18/2008 10:30	Effluent R-001	Metals	Calcium	18.5		mg/L	0.075	0.3
8/18/2008 10:30	Effluent R-001	Metals	Cadmium	0.02	U	ug/L	0.02	0.08
8/18/2008 10:30	Effluent R-001	Metals	Arsenic	0.5	U	ug/L	0.5	2
8/18/2008 10:30	Effluent R-001	Metals	Chromium	0.94	I	ug/L	0.5	2
8/18/2008 10:30	Effluent R-001	Metals	Zinc	5	U	ug/L	5	20
8/18/2008 10:30	Effluent R-001	Metals	Silver	0.025	U	ug/L	0.025	0.1
8/18/2008 10:30	Effluent R-001	Metals	Nickel	0.25	U	ug/L	0.25	1
8/18/2008 10:30	Effluent R-001	Metals	Aluminum	280		ug/L	60	240
8/18/2008 10:30	Effluent R-001	Metals	Magnesium	14.2		mg/L	0.04	0.16
8/18/2008 10:30	Effluent R-001	Metals	Selenium	0.5	U	ug/L	0.5	2
8/18/2008 10:30	Effluent R-001	Metals	Copper	0.5	U	ug/L	0.5	2
8/18/2008 10:30	Effluent R-001	Metals	Iron	30	U	ug/L	30	120
8/18/2008 10:30	Effluent R-001	Metals	Lead	0.3	I	ug/L	0.2	0.8
8/18/2008 10:30	Effluent R-001	Nutrients	NO2NO3-N	0.1		mg N/L	0.004	0.01
8/18/2008 10:30	Effluent R-001	Nutrients	TSS	23	I	mg/L	7.2	29
8/18/2008 10:30	Effluent R-001	Nutrients	O-Phosphate-P	0.018	I	mg P/L	0.012	0.03
8/18/2008 10:30	Effluent R-001	Nutrients	Kjeldahl Nitrogen	2.3		mg N/L	0.08	0.2
8/18/2008 10:30	Effluent R-001	Nutrients	Ammonia-N	0.02	I	mg N/L	0.01	0.02
8/18/2008 10:30	Effluent R-001	Nutrients	Total-P	0.15		mg P/L	0.02	0.06
8/18/2008 10:30	Effluent R-001	Field Parameter	Temperature	28.41		C		
8/18/2008 10:30	Effluent R-001	Field Parameter	pH	7.12				
8/18/2008 10:30	Effluent R-001	Field Parameter	Specific Conductance	359		umhos/cm		
8/18/2008 10:30	Effluent R-001	Field Parameter	Sample Depth	0.01		m		
8/18/2008 10:50	Blank	Metals	Arsenic	0.5	U	ug/L	0.5	2
8/18/2008 10:50	Blank	Metals	Copper	0.5	U	ug/L	0.5	2
8/18/2008 10:50	Blank	Metals	Nickel	0.25	U	ug/L	0.25	1

Appendix E. (continued)

Date Sampled	Field ID	Analysis	Component	Result	Remark	Units	MDL	PQL
8/18/2008 10:50	Blank	Metals	Iron	30	U	ug/L	30	120
8/18/2008 10:50	Blank	Metals	Silver	0.025	U	ug/L	0.025	0.1
8/18/2008 10:50	Blank	Metals	Zinc	5	U	ug/L	5	20
8/18/2008 10:50	Blank	Metals	Calcium	0.075	U	mg/L	0.075	0.3
8/18/2008 10:50	Blank	Metals	Aluminum	60	U	ug/L	60	240
8/18/2008 10:50	Blank	Metals	Magnesium	0.04	U	mg/L	0.04	0.16
8/18/2008 10:50	Blank	Metals	Chromium	0.5	U	ug/L	0.5	2
8/18/2008 10:50	Blank	Metals	Lead	0.2	U	ug/L	0.2	0.8
8/18/2008 10:50	Blank	Metals	Cadmium	0.02	U	ug/L	0.02	0.08
8/18/2008 10:50	Blank	Metals	Selenium	0.5	U	ug/L	0.5	2
8/18/2008 10:50	Blank	Nutrients	Ammonia-N	0.01	U	mg N/L	0.01	0.02
8/18/2008 10:50	Blank	Nutrients	Total-P	0.02	U	mg P/L	0.02	0.06
8/18/2008 10:50	Blank	Nutrients	Kjeldahl Nitrogen	0.08	U	mg N/L	0.08	0.2
8/18/2008 10:50	Blank	Nutrients	O-Phosphate-P	0.004	U	mg P/L	0.004	0.01
8/18/2008 10:50	Blank	Nutrients	NO2NO3-N	0.12		mg N/L	0.004	0.01

Appendix F. Bioassay Instruments

Test Dates: August 19 - 21, 2008

Test ID: Paul Calcaterra Alligator Farm

SRT Dates: August 5 - 9, 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	SM 2510 B	√
			Salinity	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	-	√

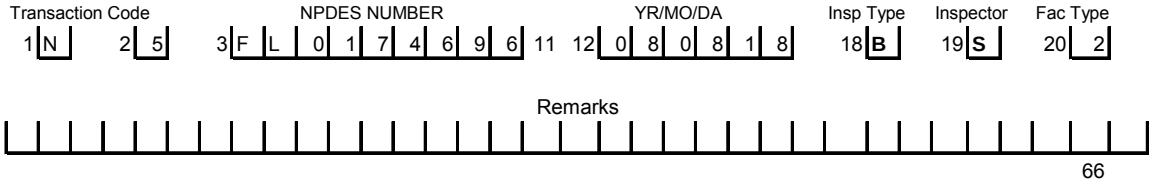
SM-2530B modified to allow use electronic and non-mercury filled thermometers.
Insert other device information in blank spaces(if used). Test ID = Facility or SRTs (acute SRT--Chronic SRT)

Comments:

V 1.4 8/21/08 BA

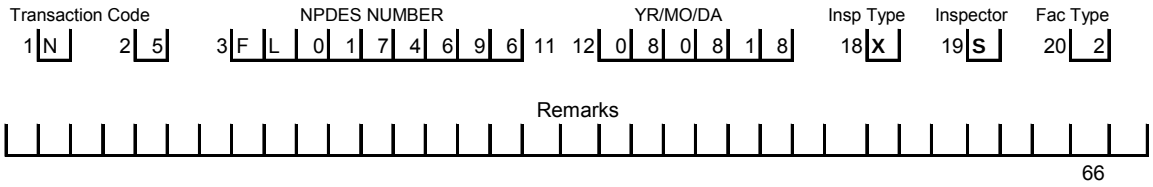
The Bioassay of the Paul Calcaterra Alligator Farm Wastewater Treatment and Disposal Facility effluent sampled on August 18, 2008. NPDES #FL0174696.

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



The Priority Pollutants Analysis of the Paul Calcaterra Alligator Farm Wastewater Treatment and Disposal Facility effluent sampled on August 18, 2008. NPDES #FL0174696.

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



Biological Analyses of the Paul Calcaterra Alligator Farm Wastewater Treatment and Disposal Facility effluent sampled on August 18, 2008. NPDES #FL0174696.

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

