

Bioassays of

Orange Park Wastewater Treatment Plant

Clay County

NPDES #FL0023922

Sampled January 29 and 30, 2006

April 2006

Biology Section
Bureau of Laboratories
Division of Resource Assessment and Management

Quality Manual No. 870346G
NELAC Certification No. E31780

Orange Park Wastewater Treatment Plant, 700 Ash Street, Orange Park, Clay County, Florida, NPDES #FL0023922. Effluent samples for this facility were collected on January 29 and 30, 2006.

Introduction

The Orange Park Wastewater Treatment Plant uses the contact-stabilization method to treat domestic wastewater for the Town of Orange Park. The facility has a design flow of 2.5 million gallons per day (MGD) and had an average annual daily flow (AADF) of 1.02 MGD from January 2005 through December 2005. The facility consists of three separate plants: Plant #1, consisting of a contact tank, a clarifier, a re-aeration tank, a chlorine contact chamber and an aerobic digester, has a capacity of 0.5 MGD; Plant #2, consisting of a contact tank, a clarifier, a re-aeration tank, a chlorine contact chamber and an aerobic digester, has a capacity of 1.0 MGD; Plant #3, consisting of a contact tank, a clarifier, a re-aeration tank, a chlorine contact chamber and an aerobic digester, also has a capacity of 1.0 MGD. Hypochlorite is used for chlorination and sodium bisulfite is used for dechlorination. Treated effluent from the three plants is combined prior to discharge into the Class III freshwaters of the St. Johns River through Outfall D-001. The D-001 Outfall, approximately 183 m (600 feet) in length, discharging at a depth of 2.43 m (8 feet), is located approximately at latitude 30º 10' 49" N, longitude -81º 41' 55" W. Wastewater residuals are hauled to a land application site for final disposal.

The facility is currently under enforcement action (Consent Order OGC number 04-0739) which establishes total nitrogen and total unionized ammonia interim limits until January 31, 2007. The corrective action of the Consent Order requires construction of new biological treatment plants to replace the existing ones. A review of Discharge Monitoring Report data showed no violations during the past year (facility information provided by Chet McGhee of FDEP Northeast District).

Methods

Samples were collected from the EFD-1 NPDES sampling point for Outfall D-001 by Chet McGhee, Joe Emery, Pat O'Connor, and Tom Kallemeyn (FDEP Northeast 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 1 for the completed chain of custody form.

The 96-hour acute screening toxicity tests and algal growth potential test were performed following internal DEP SOP's TA07_01, TA07_02, and TA08_01 through TA08_09. All internal DEP SOP's are available on the web at: http://www.floridadep.org/labs/cgi-bin/sop/biosop.asp. Bioassays were performed on each of four samples which were collected over a 24-hour period (one sample every six hours).

Toxicity Test Results

Tests were performed January 31 – February 4, 2006.

Ceriodaphnia dubia 96-hour acute screening bioassay 1 –	LC_{50} > 100%, 40% mortality in 100% sample at 96 hours
Cyprinella leedsi 96-hour acute screening bioassay 1 –	LC_{50} > 100%, 0% mortality in 100% sample at 96 hours
Ceriodaphnia dubia 96-hour acute screening bioassay 2 –	LC_{50} > 100%, 40% mortality in 100% sample at 96 hours

Cyprinella leedsi 96-hour acute screening bioassay 2 –	LC_{50} > 100%, 15% mortality in 100% sample at 96 hours
Ceriodaphnia dubia 96-hour acute screening bioassay 3 –	LC_{50} > 100%, 10% mortality in 100% sample at 96 hours
Cyprinella leedsi 96-hour acute screening bioassay 3 –	LC_{50} > 100%, 0% mortality in 100% sample at 96 hours
Ceriodaphnia dubia 96-hour acute screening bioassay 4 –	LC_{50} > 100%, 10% mortality in 100% sample at 96 hours
Cyprinella leedsi 96-hour acute screening bioassay 4 –	LC_{50} > 100%, 0% mortality in 100% sample at 96 hours

Bioassay samples 1 and 2 showed unacceptable* acute toxicity to the *Ceriodaphnia dubia* test organisms but not the *Cyprinella leedsi* test organisms. Bioassay samples 3 and 4 did not show unacceptable acute toxicity to either test species. Since mortality in all of the tests was less than 50%, the test results did not constitute a violation** of the permit conditions. The cause of toxicity was not determined in these bioassays.

* Unacceptable acute toxicity is defined in the permit as greater than 20% mortality of either test species in any grab sample test. Demonstration of unacceptable acute toxicity serves as a trigger for additional toxicity testing.

** Violation of permit condition is defined as mortalities greater than 50% in 100% effluent in any routine sample test (screening test with control and 100% effluent) or an LC_{50} of less than 100% effluent in any additional definitive test.

See Appendix 2 for bioassay bench sheets.

See Appendix 3 for results of the last 20 standard reference toxicant (SRT) tests, and bench sheets and statistical analyses of the SRT test that corresponds to this facility test. See Appendix 4 for test organism receipts.

Algal Growth Potential

The effluent Algal Growth Potential (AGP) was 221 mg dry wt/L of the freshwater 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 inorganic nitrogen concentrations was 604 (±20%) mg dry wt/L (Table 1; Miller et al. 1978).

See Table 1 for AGP results.

Chemistry Results

Total residual chlorine was not detected in the bioassay sample in the laboratory. Total ammonia was detected in the laboratory in bioassay samples 1, 2, 3, and 4 at concentrations of 8.33 mg/L, 6.69 mg/L, 3.30 mg/L and 3.62 mg/L, respectively. The total ammonia concentration in the sample collected and preserved for chemical analysis was 4.9 mg/L. Based on the pH, salinity, and temperature of the effluent as collected, the calculated unionized ammonia concentration in the preserved sample was 0.03 mg/L, which exceeded the Class III fresh water quality criterion and permit limit of 0.02 mg/L.

Aluminum, copper, iron, lead, and zinc were detected in the effluent at levels that comply with Class III fresh water quality criteria. Nickel was detected between the laboratory method detection limit (MDL) and practical quantitation limit (PQL). The effluent concentrations of silver (0.34 µg/L) and unionized ammonia (0.03 mg/L) exceeded the Class III fresh water criteria of ≤ 0.07 mg/L (62-302.530(60), F.A.C.) and ≤ 0.02 mg/L (62-302.530(3), F.A.C.), respectively.

See Table 2 for results of analytes detected in the effluent, and corresponding limits.

See Appendix 5 for a complete list of chemical analyses performed.

Conclusion

While effluent samples one (1) and two (2) collected from Outfall D-001 for this facility on January 29 and 30 showed unacceptable toxicity to the *Ceriodaphnia dubia* (waterflea) test species, none of the test results for either species constituted a violation of permit conditions during the acute screen 96-hour bioassays. The cause of toxicity was not determined. The effluent AGP result exceeded the "problem" threshold for fresh receiving waters. The effluent concentrations of silver (0.34 µg/L) and unionized ammonia (0.03 mg/L) exceeded the Class III fresh water criteria.

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) of the freshwater species Psuedokirchneriella subcapitata *for total soluble nitrogen (TSIN) and total nitrogen (TN) limitation and ratios of nitrogen to phosphorus for samples collected from Orange Park Wastewater Treatment Plant on January 29-30, 2006.*

Location	AGP (measured)	Predicted AGP (TSIN) ± 20%	Predicted AGP (TN) ± 20%	Inorganic N:P ratio	Total N:P ratio
Effluent Samples	221	604.2 ± 120.84	668.8 ± 133.76	4.82	4.76

Table 2. Effluent limits, Class III Freshwater Criteria, and chemical and microbiological data for samples collected from Orange Park Wastewater Treatment Plant on January 30, 2006.

Orange Park WWTP NPDES# FL0023922	Class III Freshwater Stds	Effluent Limits	Effluent Samples
Organic Constituents (µg/L)			
None Detected	-	-	-
Metals (µg/L unless otherwise noted)			
Aluminum	-	-	30
Arsenic	≤ 50	-	4 U
Cadmium	≤ 1.9 b	-	0.05 U
Calcium (mg/L)	-	-	43.7
Chromium-III	≤ 145.9 b	-	2 U
Copper	≤ 16.2 b	≤ 16.2 b	3.05
Iron	≤ 1,000	-	93
Lead	≤ 7.2 b	-	0.35
Magnesium (mg/L)	-	-	19.7
Nickel	≤ 89.9 b	-	1.5 I
Selenium	≤ 5	-	0.5 U
Silver	≤ 0.07	≤ 0.07	0.34
Zinc	≤ 206.6 b	-	42
Nutrients (mg/L)			
Ortho-phosphate	-	-	3.3
Total Phosphorus	-	-	3.7
Ammonia	-	Report	4.9
Unionized Ammonia	≤ 0.02	≤ 0.02	0.03 c
Nitrate+Nitrite	-	-	11 f
Total Kjeldahl Nitrogen	-	-	6.6
Organic Nitrogen	-	-	1.7 c
Total Nitrogen	-	Report	17.6 c
General Physical and Chemical Parameters			
Dissolved Oxygen (mg/L)	≥ 5	-	8.2
pH (S.U.)	6.0-8.5	6.0-8.5	7.1
Conductivity (umhos/cm)	≤ 1,275	-	815
Temperature (C)	-	Report	21.9
CBOD (mg/L)	-	60 s	2.7 Q
Chlorophyll *a* (µg/L)	-	-	0.96 U
Phaeophytin *a* (µg/L)	-	-	1.7 U
Flow (MGD)	-	2.5 a	1.02 a
TSS (mg/L)	-	60 s	4 U
Total residual chlorine (mg/L)	-	≤ 0.01	≤ 0.01
Alkalinity (mg/L)	≥ 20	-	113
Hardness (mg/L)	-	Report	190.2 c
Microbiology (# counts/100ml)			
Fecal Coliforms-Membrane Filter	≤800 s	≤800 s	-

Value exceeds the Class III Fresh Water Quality Criteria (62-302, F.A.C.) and/or Effluent Limits

a - Annual average

b - Value is calculated based on hardness

c - Calculated value

f - Nitrate + nitrite was detected in the field blank at a concentration of 0.034 mg/L

s - Single sample

I - The reported value is between the laboratory method detection limit and the laboratory practical quantitation limit.

Q - Sample held beyond normal holding time

U - Material analyzed for but not detected; value reported is the minimum detection limit

Florida Department of Environmental Protection

Central Laboratory Sample Submittal Form

Request Number: RQ-2006-01-30-13
Orange Park WWTF

Event ID *

Requester: Shannan Gunnoe
Field Report Prepared By:
Collected By: Chet McGhee, Joe Emery / Pat O'Connor / Tom Kallemeyn
Send Final Report To: Melissa Long
Sampling Agency: FDEP

Customer: NE-DIST
Project ID: FYI3
PMAS: 1143

Lab ID *	Location: Orange Park WWTF			☐ Comp ☒ Grab	Collection (begin) Date: 1/30/06 Time: 1800	Eastern Central	Collection (end) Date: Time:	Eastern Central	Bottle Group(s)**	
	Field ID: Effluent Samples			Tot Res Chlorine (mg/L): 1.54 after dechlor 0.0	Diss Oxygen (mg/L):		Storet Station Number:		AC	
	Matrix (Include type e.g. Salt, Fresh, etc):	Temp (C): 22.4	pH: 7.19	Sample Depth: 0 ☐ m ☒ ft			☐ Salinity (PPTh) ☒ Sp Conductance (umhos/cm): 826	NPDES Number:		
	Latitude: ° ' "	Longitude: ° ' "		Comments: other cond. readings 815, 690, 813						
Lab ID *	Location: Orange Park WWTF			☐ Comp ☒ Grab	Collection (begin) Date: Time: 715	Eastern Central	Collection (end) Date: Time:	Eastern Central	Bottle Group(s)**	
	Field ID: Field blanks			Tot Res Chlorine (mg/L):	Diss Oxygen (mg/L):		Storet Station Number:		B	
	Matrix (Include type e.g. Salt, Fresh, etc):	Temp (C):	pH:	Sample Depth: ☐ m ☐ ft			☐ Salinity (PPTh) ☐ Sp Conductance (umhos/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 (umhos/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 (umhos/cm):	NPDES Number:		
	Latitude: ° ' "	Longitude: ° ' "		Comments:						

Relinquished By	Date/Time	Shipping Method	Received By	Date/Time	Relinquished By	Date/Time	Received By	Date/Time

* Shaded Areas for Lab use only

** Please see reverse side for Bottle Group information.

last revised October 1, 2003

4 bxy jars 08 1-31-06 1155

Page 1 of 3

Appendix 1. Chain of Custody form (continued)

DEPARTMENT OF ENVIRONMENTAL PROTECTION-NORTHEAST DISTRICT

SAMPLE SOURCE: _____ DATE: _____ SAMPLERS: _____ EQUIPMENT #: _____ SQ #: _____

STORET/ STATION NUMBER	STATION LOCATION/ DESCRIPTION	TIME	DEPTH	TEMP	pH	CONDUCTIVITY	SECCHI	DO	DO SAT	39	WIND DIR	WIND VEL	WTR V	TIDE
				°C		µmhos/cm			%					
	Bronx Zoo	18:05		22.1	6.90			7.19	92.6					
		13-1		22.2	7.26			7.22	91.7					
		6555		21.9	8.15			7.10	8.16	92.8				
		12:14		22.2	6.13			7.06	8.62	97.4				

Remarks 1: Iced ☐ Nitrogen series preserved to pH < 2 with ___ ml H_2SO_4 ☐ PO_4 filtered ☐ ____, Metals preserved to pH < 2 with ___ ml HNO_3 ☐ ____

Remarks 2: Iced ☐ Nitrogen series preserved to pH < 2 with ___ ml H_2SO_4 ☐ PO_4 filtered ☐ ____, Metals preserved to pH < 2 with ___ ml HNO_3 ☐ ____

Remarks 3: Iced ☐ Nitrogen series preserved to pH < 2 with ___ ml H_2SO_4 ☐ PO_4 filtered ☐ ____, Metals preserved to pH < 2 with ___ ml HNO_3 ☐ ____

Remarks 4: Iced ☐ Nitrogen series preserved to pH < 2 with ___ ml H_2SO_4 ☐ PO_4 filtered ☐ ____, Metals preserved to pH < 2 with ___ ml HNO_3 ☐ ____

Remarks 5: Iced ☐ Nitrogen series preserved to pH < 2 with ___ ml H_2SO_4 ☐ PO_4 filtered ☐ ____, Metals preserved to pH < 2 with ___ ml HNO_3 ☐ ____

Remarks 6: Iced ☐ Nitrogen series preserved to pH < 2 with ___ ml H_2SO_4 ☐ PO_4 filtered ☐ ____, Metals preserved to pH < 2 with ___ ml HNO_3 ☐ ____

CHECK ANALYSIS TO BE PERFORMED:		
B.O.D.	TOT. COLIFORM	
COD	FEC. COLIFORM	✓
COLOR	NUTRIENTS	
COND.	METALS	
T. ALK.	AGP	
TSS	TOXICITY	
VSS	FIST.	
TURB.	SCI / BIORECON	

SAMPLE CUSTODY:				
RELEASED TO:	DATE:	TIME:	PRESERVATION:	RECEIVED BY:
[illegible]	1/24/96	13 30	Ice	Tom E 1/25/96 9:45

Weather Conditions	Moon Phase
1	
2	
3	
4	
5	
6	

FDEP Biology Section - Acute Screen 4/24 Bioassay Bench Sheets

Facility: Orange Park WWTP
Address: 700 Ash Street
City: Orange Park County: Clay
Contact/District: Chet McGhee / NED
NPDES Permit #: FL 0023922
LIMS Job #: TLH-2006 01-31-09 LIMS Sample #: 876733
Lims Data Entry: 2/9/06 SP Data Entry Verification: 2/9/06 MF

Sample Collection: Date: 1-29-06 Time: 1805
Hold Time Start: Date: 1/31/06 Time: 1154 12:18 SP
Sample: 1 of 4

Comments:
(A) transcription error. correct value 84 mg/L SP
(B) transcription error. correct value 86 mg/L SP
(C) field measurement after dechlorination 0.0 mg/L SP

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

Test Type: ~~Definitive~~ Screen SP
Static (Static Renewal / Flow-through
Test Duration: 24 48 96 Hours.
Light Intensity : 50 - 100 ft. candles.
Photoperiod : 16 hours light 8 hours dark.

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: ____

Temperature Range °C
Incubator # 2 Range 24.2 - 24.8
Room B218: 23.6 - 25.2
Waterbath: N/A

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)

Investigators' Signatures
Janet Fairchild
Jaya David
Marshall Fairchild
Bea Nesbitt
Erin Lusk
Mike Wolfe
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	Method	Measured by	Verified by
Field Total Residual Cl2 (mg/L):	N/A	N/A	N/A	N/A	N/A	~~<0.03~~ (C)	unknown	C McGhee	SP
Lab Total Residual Cl2 (mg/L):	<0.03	<0.03				<0.03	HACH	SP/GW	MF
Alkalinity (mg/L as $CaCO_3$):	~~72.5~~ (A)	138.5				155.5	HACH	B	SP
Hardness (mg/L as $CaCO_3$):	~~85.5~~ (B)	131				184	HACH	B	SP
Total Ammonia (mg/L as N):	<0.017	<0.017				8.33	DENVER	MF	SP
Salinity (ppt):	<1	<1				<1	YSI / (Refract)	MF	SP

V 2.0 4/12/05

Appendix 2. Bench Sheets (continued)

FDEP Biology Section - Bioassay Survival Sheet

LIMS Sample #: 8-16335
Test #: 1
Test Started: Date 1-31-06 Time: 1400
Organism: C. dubia
SOP #: TA 07_ C1
Test Ended: Date 2-4-06 Time: 1115
Organism Batch: 1-06
Diluent/ Batch: DMW / 12-06
Organism Age: <24 hrs
Food: YCT P. subcapitata Artemia
Chamber Size: 30 mL
Batch: 5-05 5-05 —
Test Volume: 30 mL
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	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
100%	A	A	5	5	4 1D	4	4	3 1D
	B	B	5	5	5	5	5	4 1D
	C	C	5	5	4 1D	4	3 1D	1 2D
	D	D	5	5	4 1D	4	4	4
Organisms loaded by: MF		Checked by:	[redacted]	[illegible]	MF	MF	MF	MF

Loading Verified by: 6

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

Comments:

Test Results:
% mortality in 100% sample: 40
LC_{50}: >100 If Calculated: 95% CI______ Method: ______

Screening Tests:
Report LC50 as >100%, =100%, or <100%.
Substitue highest test concentration used if other than 100% (example: Ocean outfall tested at 30% concentration).

V 2.1 11/01/2005

Page 000002

Appendix 2. Bench Sheets (continued)



FDEP Biology Section - Bioassay Survival Sheet

LIMS Sample #: 896233 Test #: 2 Test Started: Date 13.06 Time: 1410
Organism: C.leedsi SOP #: TA 07_02 Test Ended: Date 24.06 Time: 1115
Organism Batch: 306 Diluent/ Batch: Well H2O / 1-30-06
Organism Age: 10 days Food: YCT P. subcapitata Artemia
Chamber Size: 1000 mL Batch: – – GSL 530-90
Test Volume: 500 mL 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	A1	5	5	5	5	5	5
	B	A2	5	5	5	5	5	5
	C	A3	5	5	5	5	5	5
	D	A4	5	5	5	5	5	5
100%	A	A5	5	5	5	5	5	5
	B	A6	5	5	5	5	5	5
	C	A7	5	5	5	5	5	5
	D	A8	5	5	5	5	5	5
Organisms loaded by: JD	Checked by: [redacted]		B	JD	EP	EP	MF	
Loading Verified by: EP								

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

Comments:

Test Results:
% mortality in 100% sample: 0
LC_{50}: >100 If Calculated: 95% CI_____ Method:_____

Screening Tests:
Report LC50 as >100%, =100%, or <100%.
Substitue highest test concentration used if other than 100% (example: Ocean outfall tested at 30% concentration).

V 2.1 11/01/2005

Page 000003

FDEP Biology Section - Bioassay Parameter Sheet

LIMS Sample # : 896233 Test #: 1 of 2

TEST SOP #: TA07_ 01 Test Species: (Ceriodaphnia dubia) Cyprinella leedsi Pimephales promelas Americamysis bahia Menidia beryllina Other:_____

Concentration CTRL	0 Hr.	24 Hr.	48 Hr. before renewal (A)	48 Hr. after renewal (A)	72 Hr.	96 Hr.
Replicate	A		B	B		A
pH (S.U.)	8.5		8.2	8.3		8.2
Temperature °C	25.1		24.6	24.6		24.4
Dissolved Oxygen mg/L	7.8		7.9	7.3		7.4
Conductivity (µmhos) mmhos	194		173	222		201
(initials) Measured by:	MF		[illegible]	MF		MF
(initials) Recorded by:	MF		[illegible]	MF		MF

Comments: (A) Before and After readings transposed MF-2-2-06

Concentration 100%	0 Hr.	24 Hr.	48 Hr. before renewal (A)	48 Hr. after renewal (A)	72 Hr.	96 Hr.
Replicate	A		B	B		A
pH (S.U.)	7.5		7.7	8.2 MF		8.1
Temperature °C	24.8		24.8	24.5		24.2
Dissolved Oxygen mg/L	7.6		8.1	7.2		7.4
Conductivity (µmhos) mmhos	843		845	872		876
(initials) Measured by:	MF		[illegible]	MF		MF
(initials) Recorded by:	MF		[illegible]	MF		MF

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

FDEP Biology Section - Bioassay Parameter Sheet

LIMS Sample # : 896233 **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	C	D	A
pH (S.U.)	7.9	8.2	8.3	8.2	8.0	8.2
Temperature °C	25.0	25.0	24.7	24.3	24.7	24.6
Dissolved Oxygen mg/L	8.0	7.3	7.3	7.7	6.4	7.2
Conductivity ~~µmhos~~ mmhos	293	270	276	272	264	271
(initials) Measured by:	[illegible]	B	[illegible]	[illegible]	[illegible]	MF
(initials) Recorded by:	[illegible]	B	[illegible]	[illegible]	[illegible]	MF

Comments:

Concentration 100%	0 Hr.	24 Hr.	48 Hr. before renewal	48 Hr. after renewal	72 Hr.	96 Hr.
Replicate	A	B	C	C	D	A
pH (S.U.)	7.5	7.9	8.1	8.2	7.8	8.0
Temperature °C	25.0	24.7	24.5	24.6	24.5	24.5
Dissolved Oxygen mg/L	7.5	7.2	6.3	7.3	6.2	6.2
Conductivity ~~µmhos~~ mmhos	848	858	851	862	823	848
(initials) Measured by:	[illegible]	B	[illegible]	[illegible]	[illegible]	MF
(initials) Recorded by:	[illegible]	B	[illegible]	[illegible]	[illegible]	MF

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

FDEP Biology Section - Acute Screen 4/24 Bioassay Bench Sheets

Facility: Orange Park WWTP
Address: 700 Ash Street
City: Orange Park County: Clay
Contact/District: Chet McGhee / NED
NPDES Permit #: FL 0023922
LIMS Job #: TLH-20060-01-31-49 LIMS Sample #: 896234
Lims Data Entry: 2-9-06 BB Data Entry Verification: 2-9-06 MF

Sample Collection: Date: 1-30-06 Time: ~~1261~~ 00:01
Hold Time Start: Date: 1-30-06 Time: 12:18
Sample: 2 of 4

Comments:
Before renewal, sample was aerated to lower DO from 9.3 to 7.5 mg/L) for 15 minutes at a rate of about 100 bubbles/minute. ESP 2-2-06

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

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

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

Initial sample handling:
pH adjustment: yes (no) Initial pH: ____ NaOH N HCl N ____ Drops mL Final pH: ____
Aeration (yes) no Initial DO: 9.3 mg/L Final DO: 8.9 mg/L Duration: 15 minutes Rate: ~ 100 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)

Investigators' Signatures
Angela Rickard
Jess Davis
Marshall Fairbanks
Barbara Reading
Erin Lillo
Mindy Wellendorf
REVIEWER

Water Quality Parameters	Original Sample	Method	Measured by	Verified by
Field Total Residual Cl2 (mg/L):	not in	[illegible]	—	MF
Lab Total Residual Cl2 (mg/L):	<0.03	HACH	EW	SP
Alkalinity (mg/L as $CaCO_3$):	148	HACH	B	SP
Hardness (mg/L as $CaCO_3$):	186	HACH	B	SP
Total Ammonia (mg/L as N):	6.69	DENVER	MF	SP
Salinity (ppt):	<1	YSI / (Mettler)	MF	SP

V 2.0 4/12/05

Page 0000006

Appendix 2. Bench Sheets (continued)

FDEP Biology Section - Bioassay Survival Sheet

LIMS Sample #: 891234 Test #: 1 Test Started: Date 1-31-05 Time: 1410
Organism: C. dubia SOP #: TA 07_ 01 Test Ended: Date 2-4-06 Time: 1130
Organism Batch: 6-06 Diluent/ Batch: DMW 12-06
Organism Age: <24 hrs Food: YCT *P. subcapitata* *Artemia*
Chamber Size: 30 mL Batch: 5-05 8-05 —
Test Volume: 20 mL 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	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	~~3~~ 3 3D
100%	A	A	5	5	5	5	5	4 1D
	B	B	5	5	5	5	5	2 3D
	C	C	5	5	4 1D	4	4	1 3D
	D	D	5	5	5	5	5	5
Organisms loaded by: MF		Checked by: [redacted]		SB	MF	MF	MF	MF
Loading Verified by: [redacted]								

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

Comments:

Test Results:
% mortality in 100% sample: 40
LC_{50}: >100 If Calculated: 95% CI_____ Method:_____

Screening Tests:
Report LC50 as >100%, =100%, or <100%.
Substitue highest test concentration used if other than 100% (example: Ocean outfall tested at 30% concentration).

V 2.1 11/01/2005

Page 000007

Appendix 2. Bench Sheets (continued)

FDEP Biology Section - Bioassay Survival Sheet

LIMS Sample #: 896234
Organism: C. leedsi
Organism Batch: 3-06
Organism Age: 10 days
Chamber Size: 1000 mL
Test Volume: 500 mL

Test #: 2
SOP #: TA 07_02
Diluent/ Batch: ~~ASW~~ SP Well H_2O 11-30-06
Food: YCT | P. subcapitata | Artemia
Batch: — | — | GSL 536-90
Feeding: Prior to test / (Prior to renewal) / Daily

Test Started: Date 1.31.06 Time: 1415
Test Ended: Date 2-4.06 Time: 1130

Concentration	Replicate	Chamber #	Test Hour 0 hr	24 hr	48 hr BR	48 hr AR	72 hr	96 hr
CTRL	A	A9	5	5	5	5	5	5
	B	A10	5	5	5	5	5	5
	C	A11	5	5	5	5	5	5
	D	A12	5	5	5	5	5	5
100%	A	A13	5	5	4 1D	~~5~~ SP 4	4	4
	B	A14	5	5	4 1D	~~5~~ SP 4	4	4
	C	A15	5	5	5	5	5	4 1D
	D	A16	5	5	5	5	5	5
Organisms loaded by: JD		Checked by:	[redacted]	B	JD	SP	SP	MF

Loading Verified by: SP

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

Comments:

Test Results:
% mortality in 100% sample: 15
LC_{50}: >100 If Calculated: 95% CI____ Method:____

Screening Tests:
Report LC50 as >100%, =100%, or <100%.
Substitue highest test concentration used if other than 100% (example: Ocean outfall tested at 30% concentration).

V 2.1 11/01/2005

Page 000008

FDEP Biology Section - Bioassay Parameter Sheet

LIMS Sample # : 8961234 Test #: 1 of 2

TEST SOP #: TA07_ 01 Test Species: (Ceriodaphnia dubia) Cyprinella leedsi Pimephales promelas Americamysis bahia Menidia beryllina Other:_____

Concentration CTRL	0 Hr.	24 Hr.	48 Hr. before renewal (A)	48 Hr. after renewal (A)	72 Hr.	96 Hr.
Replicate	A		B	B		A
pH (S.U.)	8.5		8.3	8.3		8.2
Temperature °C	25.0		24.3	24.4		24.1
Dissolved Oxygen mg/L	7.8		7.8	7.5		7.5
Conductivity (µmhos) mmhos	182		179	208		208
(initials) Measured by:	MF		[illegible]	MF		MF
(initials) Recorded by:	MF		[illegible]	MF		MF

Comments: (A) Before and after readings transposed MF 12-06

Concentration 100%	0 Hr.	24 Hr.	48 Hr. before renewal (A)	48 Hr. after renewal (A)	72 Hr.	96 Hr.
Replicate	A		B	B		A
pH (S.U.)	7.8		8.1	8.2		8.2
Temperature °C	24.7		24.8	24.4		24.0
Dissolved Oxygen mg/L	8.4		7.9	7.4		7.5
Conductivity (µmhos) mmhos	807		800	858		863
(initials) Measured by:	MF		[illegible]	MF		MF
(initials) Recorded by:	MF		[illegible]	MF		MF

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:

000009

V 2.0 2/24/04

FDEP Biology Section - Bioassay Parameter Sheet

LIMS Sample # : 896234 **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	C	D	A
pH (S.U.)	7.9	8.2	8.3	8.3	8.1	8.2
Temperature °C	24.9	25.3	24.2	24.4	24.4	24.9
Dissolved Oxygen mg/L	8.0	7.6	7.5	7.6	7.1	7.3
Conductivity ~~µmhos~~ mmhos	272	268	275	278	262	272
(initials) Measured by:	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	MF
(initials) Recorded by:	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	MF

Comments:

Concentration 100%	0 Hr.	24 Hr.	48 Hr. before renewal	48 Hr. after renewal	72 Hr.	96 Hr.
Replicate	A	B	C	C	D	A
pH (S.U.)	7.8	8.0	8.1	C	7.9	8.0
Temperature °C	24.9	24.9	24.2	24.4	24.2	24.5
Dissolved Oxygen mg/L	8.5	7.3	7.0	7.4	5.5	6.4
Conductivity ~~µmhos~~ mmhos	807	796	816	860	785	802
(initials) Measured by:	[illegible]	[illegible]	[illegible]	[illegible]	B	MF
(initials) Recorded by:	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	MF

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

FDEP Biology Section - Acute Screen 4/24 Bioassay Bench Sheets

Facility: Orange Park WWTP
Address: 700 Ash Street
City: Orange Park County: Clay
Contact/District: Chet McGhee / NED
NPDES Permit #: FL 0023922
LIMS Job #: TL4-2006-01-31-49 LIMS Sample #: 876735
Lims Data Entry: 2-9-06 SP Data Entry Verification: 2-9-06 [illegible]

Sample Collection: Date: 1-30-06 Time: 0655
Hold Time Start: Date: 1-30-06 Time: ~~1250~~ 12:18
Sample: 3 of 4

Comments:

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

Test 1 validation:
Control survival ≥90%: (Yes) No
Temperature Range ≤3°C: (Yes) No
Test 1: SOP TA 07_ 01

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

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)

Investigators' Signatures
Sarah Pickard
Jenna Tomlin
Marshall Faircloth
Bill Adams
Erin Wise
Nicole Wetherington
REVIEWER

Water Quality Parameters	Original Sample	Method	Measured by	Verified by
Field Total Residual Cl2 (mg/L):	not impacted		–	SP
Lab Total Residual Cl2 (mg/L):	<0.03	HACH	QAW	SP
Alkalinity (mg/L as $CaCO_3$):	131	HACH	QA	SP
Hardness (mg/L as $CaCO_3$):	188	HACH	QA	SP
Total Ammonia (mg/L as N):	3.245	DENVER	MF	SP
Salinity (ppt):	21	YSI (Mettler)	MF	SP

V 2.0 4/12/05

Page 0000011

3

[redacted]

Appendix 2. Bench Sheets (continued)



FDEP Biology Section - Bioassay Survival Sheet

LIMS Sample #: 8916735 Test #: 1 Test Started: Date 1-31-06 Time: 1420
Organism: C.dubia SOP #: TA 07_ 01 Test Ended: Date 2-4-06 Time: 1145
Organism Batch: 6-06 Diluent/ Batch: DMW / 2-06
Organism Age: <24hrs Food: YCT *P. subcapitata* *Artemia*
Chamber Size: 30 mL Batch: 5-05 8-05 -
Test Volume: 20 mL 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	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
100%	A	A	5	5	5	5	5	5
	B	B	5	5	5	5	5	3 20
	C	C	5	5	5	5	5	5
	D	D	5	5	5	5	5	5
Organisms loaded by: MF	Checked by: [redacted]			SF	MF	MF	MF	MF
Loading Verified by: B								

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

Comments:

Test Results:
% mortality in 100% sample: 10
LC_{50}: >100 If Calculated: 95% CI_____ Method:_____

Screening Tests:
Report LC50 as >100%, =100%, or <100%.
Substitue highest test concentration used if other than 100% (example: Ocean outfall tested at 30% concentration).

V 2.1 11/01/2005

Page 000012

Appendix 2. Bench Sheets (continued)



FDEP Biology Section - Bioassay Survival Sheet

LIMS Sample #: 8961235 Test #: 2 Test Started: Date 1/30/06 Time: 1420
Organism: C. leedsi SOP #: TA 07_02 Test Ended: Date 2-4-06 Time: 1145
Organism Batch: 306 Diluent/ Batch: Well H20 / 1-30-06
Organism Age: 10 days Food: YCT P. subcapitata Artemia
Chamber Size: 1000 mL Batch: - - GSL 536-90
Test Volume: 500 mL 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	A17	5	5	5	5	5	5
	B	A18	5	5	5	5	5	5
	C	A19	5	5	5	5	5	5
	D	A20	5	5	5	5	5	5
100%	A	A21	5	5	5	5	5	5
	B	A22	5	5	5	5	5	5
	C	A23	5	5	5	5	5	5
	D	A24	5	5	5	5	5	5
Organisms loaded by: JD		Checked by: [redacted]		JD	JD	BP	BP	MF
Loading Verified by: BP								

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

Comments:

Test Results:
% mortality in 100% sample: 0
LC_{50}: >100 If Calculated: 95% CI _____ Method: _____

Screening Tests:
Report LC50 as >100%, =100%, or <100%.
Substitue highest test concentration used if other than 100% (example: Ocean outfall tested at 30% concentration).

V 2.1 11/01/2005

Page 000013

FDEP Biology Section - Bioassay Parameter Sheet

LIMS Sample # : 8962735 Test #: 1 of 2

TEST SOP #: TA07_ 01 Test Species: (Ceriodaphnia dubia) Cyprinella leedsi Pimephales promelas Americamysis bahia Menidia beryllina Other:_____

Concentration CTRL	0 Hr.	24 Hr.	48 Hr. before renewal (A)	48 Hr. after renewal (A)	72 Hr.	96 Hr.
Replicate	A	|	B	B	|	A
pH (S.U.)	8.4	|	8.3	8.3	|	8.3
Temperature °C	25.0	|	24.3	24.4	|	24.2
Dissolved Oxygen mg/L	7.8	|	7.8	7.5	|	7.7
Conductivity (µmhos) mmhos	181	|	179	215	|	206
(initials) Measured by:	MF		SF	MF		MF
(initials) Recorded by:	MF		SF	MF		MF

Comments: (A) Before and After readings Transposed MF 2-2-06

Concentration 100%	0 Hr.	24 Hr.	48 Hr. before renewal	48 Hr. after renewal (A)	72 Hr.	96 Hr.
Replicate	A	|	B	B	|	A
pH (S.U.)	7.4	|	7.6	8.2	|	8.3
Temperature °C	24.8	|	24.9	24.4	|	24.0
Dissolved Oxygen mg/L	8.3	|	8.0	7.3	|	7.5
Conductivity (µmhos) mmhos	789	|	790	934	|	906
(initials) Measured by:	MF		SF	MF		MF
(initials) Recorded by:	MF		SF	MF		MF

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:

000014

V 2.0 2/24/04

FDEP Biology Section - Bioassay Parameter Sheet

LIMS Sample # : 896235 Test #: 2 of 2

TEST SOP #: TA07_ 07 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	C	D	A
pH (S.U.)	8.0	8.2	8.3	8.2	8.0	8.2
Temperature °C	24.9	24.1	24.3	24.4	23.8	24.9
Dissolved Oxygen mg/L	8.1	7.3	7.7	7.5	6.1	6.9
Conductivity µmhos mmhos	265	272	279	270	262	271
(initials) Measured by:	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	MF
(initials) Recorded by:	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	MF

Comments:

Concentration 100%	0 Hr.	24 Hr.	48 Hr. before renewal	48 Hr. after renewal	72 Hr.	96 Hr.
Replicate	A	B	C	C	D	A
pH (S.U.)	7.4	7.9	8.0	8.1	7.9	8.0
Temperature °C	25.0	24.9	24.2	24.4	23.7	25.3
Dissolved Oxygen mg/L	8.2	7.2	7.3	7.6	6.5	6.8
Conductivity µmhos mmhos	796	796	819	816	778	795
(initials) Measured by:	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	MF
(initials) Recorded by:	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	MF

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



FDEP Biology Section - Acute Screen 4/24 Bioassay Bench Sheets

Facility: Orange Park WWTP
Address: 700 Ash St
City: Orange Park County: Clay
Contact/District: Chet McGhee / NED
NPDES Permit #: FL 0023922
LIMS Job #: TL11-2006-01-31-49 LIMS Sample #: 8962360
Lims Data Entry: 2-9-06 SP Data Enlry Verification: 2-9-06 MF

Sample Collection: Date: 1-30-06 Time: 1150
Hold Time Start: Date: 1-30-06 Time: ~~1150~~ 12:18
Sample: 4 of 4

Comments:

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

Test 1 validation:
Control survival ≥90%: (Yes) No
Temperature Range ≤3°C: (Yes) No
Test 1: SOP TA 07_ 01

Test 2 validation:
Control survival ≥90%: (Yes) No
Temperature Range ≤3°C: (Yes) No
Test 2: SOP TA 07_ 02

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)

Investigators' Signatures
Sarah Pritchard
Jason Jones
Marshall Faircloth
Cecile Hudson
Sam Meigs
Nicole Wallerstedt
REVIEWER

Water Quality Parameters	Original Sample	Method	Measured by	Verified by
Field Total Residual Cl2 (mg/L):	not-measured		-	SP
Lab Total Residual Cl2 (mg/L):	<0.05	HACH	SW	SP
Alkalinity (mg/L as $CaCO_3$):	118	HACH	JB	SP
Hardness (mg/L as $CaCO_3$):	189	HACH	JB	SP
Total Ammonia (mg/L as N):	3.615	DENVER	MF	SP
Salinity (ppt):	~~2.0~~ 2.1	YSI (Mettler)	MF	SP

MF standardized 1-31-06

V 2.0 4/1/2005

Page 0000016

Appendix 2. Bench Sheets (continued)

[redacted]

FDEP Biology Section - Bioassay Survival Sheet

LIMS Sample #: 8962360 Test #: 1 Test Started: Date 1-31-06 Time: 1430
Organism: C. dubia SOP #: TA 07_01 Test Ended: Date 2-4-06 Time: 1158
Organism Batch: 6-06 Diluent/ Batch: DMW 1-2-06
Organism Age: <24 hrs Food: YCT *P. subcapitata* *Artemia*
Chamber Size: 30 mL Batch: 5-05 8-05 –
Test Volume: 20 mL 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	4 1D	4	4	4
	B	B	5	5	5	5	5	5
	C	C	5	5	5	5	5	5
	D	D	5	5	5	5	4 1D	4
100%	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	5	5	5	5
Organisms loaded by: MF		Checked by:	[redacted]	SS	MF	MF	MF	MF

Loading Verified by: B

Comments:

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

Test Results:
% mortality in 100% sample: 10
LC_{50}: >100 If Calculated: 95% CI________ Method:________
Screening Tests:
Report LC50 as >100%, =100%, or <100%.
Substitue highest test concentration used if other than 100% (example: Ocean outfall tested at 30% concentration).

V 2.1 11/01/2005

Page 000017

Appendix 2. Bench Sheets (continued)

FDEP Biology Section - Bioassay Survival Sheet

LIMS Sample #: 896236 Test #: 2 Test Started: Date 1.31.06 Time: 14:25
Organism: C. leedsi SOP #: TA 07_02 Test Ended: Date 2-4-06 Time: 1200
Organism Batch: 3-06 Diluent/ Batch: Well H_2O 11-30-06
Organism Age: 10 days Food: YCT *P. subcapitata* *Artemia*
Chamber Size: 1000 mL Batch: – – GSL 536-90
Test Volume: 500 mL 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	C1	5	5	5	5	5	5
	B	C2	5	5	5	5	5	5
	C	C3	5	5	5	5	5	5
	D	C4	5	5	5	5	5	5
100%	A	C5	5	5	5	5	5	5
	B	C6	5	5	5	5	5	5
	C	C7	5	5	5	5	5	5
	D	C8	5	5	5	5	5	5
Organisms loaded by: JD		Checked by:	[redacted]	B	JD	SP	SP	MF

Loading Verified by: SP

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

Comments:

Test Results:
% mortality in 100% sample: 0
LC_{50}: >100 If Calculated: 95% CI______ Method:______

Screening Tests:
Report LC50 as >100%, =100%, or <100%.
Substitue highest test concentration used if other than 100% (example: Ocean outfall tested at 30% concentration).

V 2.1 11/01/2005

Page 000018

Appendix 2. Bench Sheets (continued)

FDEP Biology Section - Bioassay Parameter Sheet

LIMS Sample #: 891023(e — Test #: 1 of 2

TEST SOP #: TA07_01 — 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	B		A
pH (S.U.)	8.4		8.3	8.3		8.3 (B)
Temperature °C	24.6		24.5	24.2		24.3
Dissolved Oxygen mg/L	7.7		7.9	7.5		7.7
Conductivity µmhos mmhos	180		179	247		242
(initials) Measured by:	MF		SR	MF		MF
(initials) Recorded by:	MF		SR	MF		MF

Comments: (A) Ceriodaphnia dubia and Cyprinella leedsi transferred MF 3-3-06
(B) ML 3-4-06 Ceriodaphnia dubia DO in pH specs - no measured DO.

Concentration 100%	0 Hr.	24 Hr.	48 Hr. before renewal	48 Hr. after renewal	72 Hr.	96 Hr.
Replicate	A		B	B		A
pH (S.U.)	8.4		7.5	8.1		8.3
Temperature °C	24.5		25.0	24.2		24.3
Dissolved Oxygen mg/L	7.9		7.9	7.7		7.7
Conductivity µmhos mmhos	739		796	883		926
(initials) Measured by:	MF		[illegible]	MF		MF
(initials) Recorded by:	MF		SR	MF		MF

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:

V2.0 2/24/04

Page

FDEP Biology Section - Bioassay Parameter Sheet

LIMS Sample # : 8962736 **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	C	D	A
pH (S.U.)	7.9	8.2	8.3	8.2	8.3	8.3
Temperature °C	25.0	24.6	24.6	24.4	24.2	24.4
Dissolved Oxygen mg/L	8.1	7.4	7.5	7.6	7.5	7.1
Conductivity ~~µmhos~~ mmhos	263	268	284	270	271	267
(initials) Measured by:	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	MA
(initials) Recorded by:	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	MA

Comments:

Concentration 100%	0 Hr.	24 Hr.	48 Hr. before renewal	48 Hr. after renewal	72 Hr.	96 Hr.
Replicate	A	B	C	C	D	A
pH (S.U.)	7.4	7.8	8.1	8.3	7.9	7.9
Temperature °C	24.9	24.3	25.3	25.5	24.4	24.5
Dissolved Oxygen mg/L	7.5	7.2	7.0	7.1	6.7	6.9
Conductivity ~~µmhos~~ mmhos	797	782	824	820	791	796
(initials) Measured by:	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	MA
(initials) Recorded by:	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	MA

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 3. SRT Bench Sheets and Statistical Analysis

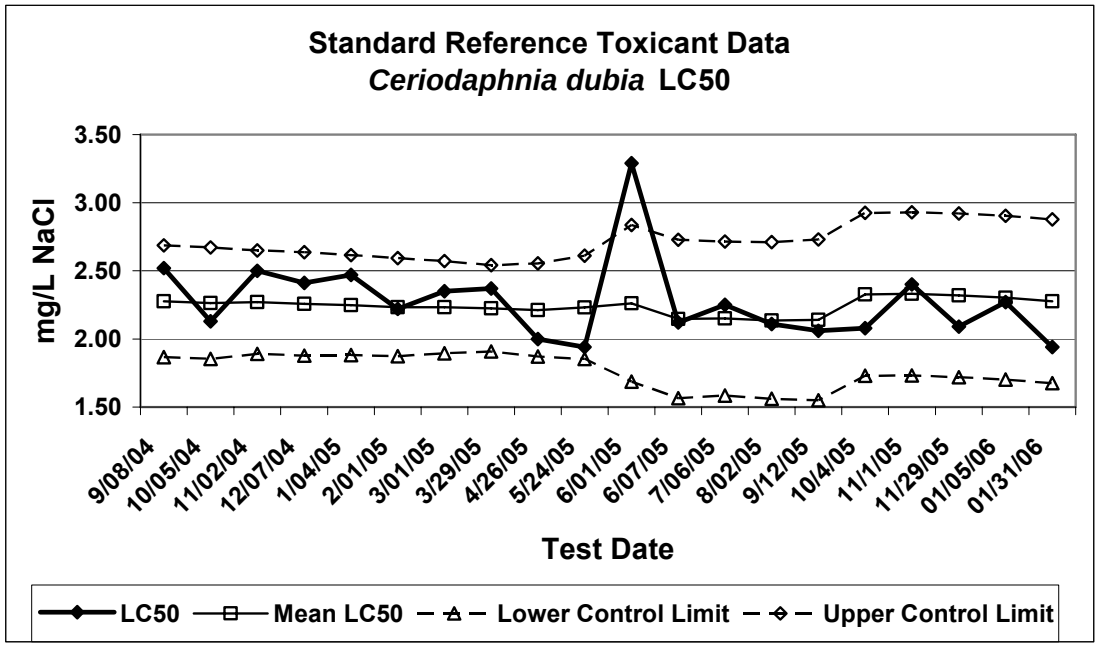
Florida Department of Environmental Protection
Bureau of Laboratories

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

Date	LC50	Mean LC50	Std Dev	Lower Control Limit	Upper Control Limit
7/20/04	2.60	2.30	0.20	1.90	2.69
8/10/04	2.47	2.29	0.20	1.89	2.69
9/08/04	2.52	2.28	0.21	1.87	2.69
10/05/04	2.13	2.26	0.20	1.85	2.67
11/02/04	2.50	2.27	0.19	1.89	2.65
12/07/04	2.41	2.26	0.19	1.88	2.64
1/04/05	2.47	2.25	0.18	1.88	2.61
2/01/05	2.22	2.23	0.18	1.87	2.59
3/01/05	2.35	2.23	0.17	1.89	2.57
3/29/05	2.37	2.22	0.16	1.91	2.54
4/26/05	2.00	2.21	0.17	1.87	2.55
5/24/05	1.94	2.23	0.19	1.85	2.61
6/01/05	3.29	2.26	0.29	1.69	2.84
6/07/05	2.12	2.15	0.29	1.57	2.73
7/06/05	2.25	2.15	0.28	1.58	2.72
8/02/05	2.11	2.14	0.29	1.56	2.71
9/12/05	2.06	2.14	0.29	1.55	2.73
10/4/05	2.08	2.33	0.30	1.73	2.93
11/1/05	2.40	2.33	0.30	1.73	2.93
11/29/05	2.09	2.32	0.30	1.72	2.92
01/05/06	2.27	2.30	0.30	1.70	2.90
01/31/06	1.94	2.28	0.30	1.68	2.88

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

CV = 13.20
EPA reference 75th quartile CV = 29%



Appendix 3. (continued)

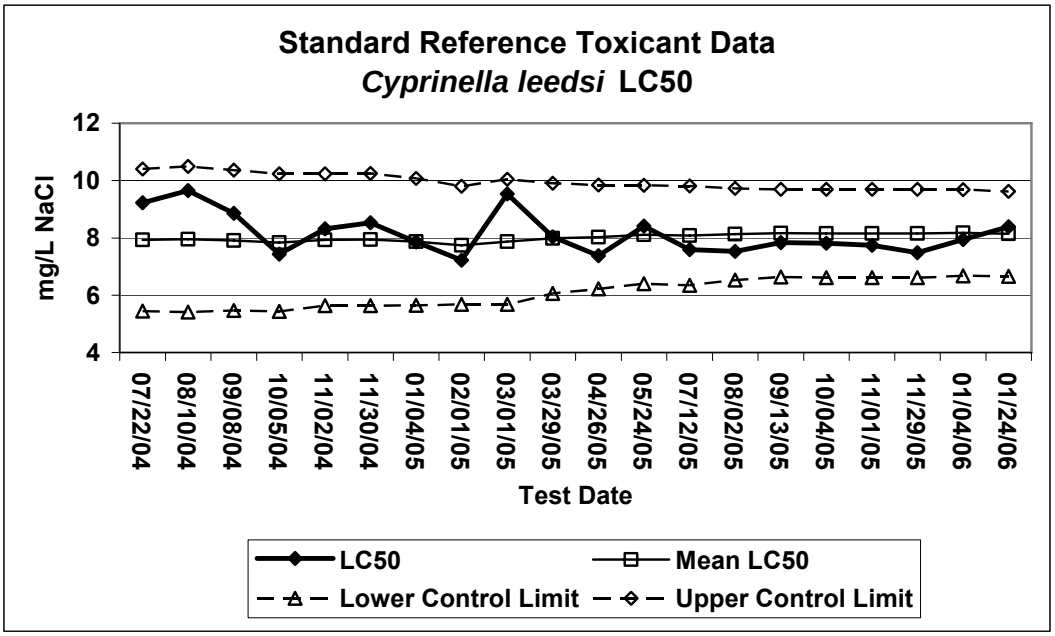
Florida Department of Environmental Protection
Bureau of Laboratories

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

Date	LC50	Mean LC50	Std Dev	Lower Control Limit	Upper Control Limit
05/24/04	7.42	7.88	1.20	5.49	10.27
06/22/04	9.19	7.91	1.23	5.46	10.36
07/22/04	9.23	7.93	1.24	5.45	10.41
08/10/04	9.65	7.95	1.27	5.41	10.49
09/08/04	8.85	7.91	1.22	5.46	10.36
10/05/04	7.43	7.83	1.20	5.43	10.24
11/02/04	8.31	7.93	1.15	5.63	10.24
11/30/04	8.53	7.94	1.15	5.63	10.24
01/04/05	7.86	7.87	1.11	5.65	10.08
02/01/05	7.22	7.74	1.03	5.68	9.80
03/01/05	9.54	7.87	1.09	5.68	10.05
03/29/05	8.04	7.99	0.96	6.07	9.91
04/26/05	7.38	8.03	0.90	6.22	9.84
05/24/05	8.41	8.11	0.86	6.40	9.83
07/12/05	7.59	8.08	0.86	6.35	9.81
08/02/05	7.53	8.13	0.80	6.53	9.72
09/13/05	7.84	8.16	0.77	6.63	9.70
10/04/05	7.81	8.16	0.77	6.62	9.69
11/01/05	7.74	8.15	0.77	6.61	9.69
11/29/05	7.49	8.15	0.77	6.61	9.69
01/04/06	7.94	8.18	0.75	6.68	9.68
01/24/06	8.39	8.14	0.74	6.66	9.62

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

CV = 9.09
EPA reference 75th quartile CV = 19%



Appendix 3. (continued)



FDEP Biology Section - Acute SRT Bench Sheet

Test Organism: *Americamysis bahia* (Method 2007.0) *Menidia beryllina* (Method 2006.0) *Hyalella azteca* (Method 100.1 EPA-600-R-99-064) (*Ceriodaphnia dubia*) (Method 2002.0) *Cyprinella leedsi* (Method 2000.0) *Pimephales promelas* (Method 2000.0) Other:_____

Method from EPA-821-R-02-012
Test Type: Screening / (Definitive)
Static / (Static Renewal)

Diluent/ Batch#: DMW 12-06
Toxicant/ Batch# NaCl 12-06

Test Beginning: Date: 1-31-06 Time: 1220
Test Ending: Date: 2-4-06 Time: 1200

Water Quality Parameters:	Diluent	Method
Total Residual Cl_2 (mg/L):	<0.03	Hach
Alkalinity (mg/L as $CaCO_3$):	84	Hach
Hardness (mg/L as CaCO3):	85.5	Hach
Total Ammonia (mg/L as N):	<0.07	Denver
Salinity (ppth):	<1	Mettler

Organism Batch #/ Age: 6-06 / <24 hrs
Feeding: (Prior to test) (Prior to renewal) / Daily

Food:	YCT	P.subcapitata	Artemia
Food Batch:	5-05	8.05	-

Chamber size: 30 mL
Test volume: 20 mL per replicate

Page 1 of 2

Conc µg/L (g/L)	Rep.	Chamber#	Number Live 0 hr (48 hr)	Number Live 24 hr (72 hr)	Number Live 48 hr (96 hr)	pH (SU) 0 hr (48 hr)	pH (SU) 24 hr (72 hr)	pH (SU) 48 hr (96 hr)	Temperature (° C) 0 hr (48 hr)	Temperature (° C) 24 hr (72 hr)	Temperature (° C) 48 hr (96 hr)	D.O. (mg/l) 0 hr (48 hr)	D.O. (mg/l) 24 hr (72 hr)	D.O. (mg/l) 48 hr (96 hr)	UNCORRECTED Cond. (mmhos/cm) Cond. (µmhom/cm) 0 hr (48 hr)	24 hr (72 hr)	48 hr (96 hr)
Control	A	A	5	5	5	8.1		8.0	24.5		24.8	7.8		7.3	0.115		0.192
	B	B	5	5	5												
	C	C	5	5	5												
	D	D	5	5	5												
1.5	A	A	5	5	5	8.0		8.0	24.8		24.6	7.7		7.4	2.97		3.33
	B	B	5	5	5												
	C	C	5	5	5												
	D	D	5	5	5												
2.0	A	A	5	5	4 ID	8.0		8.0	24.8		24.6	7.6		7.4	3.93		4.03
	B	B	5	5	0 5D												
	C	C	5	5	2 3D												
	D	D	5	5	3 2D												
2.5	A	A	5	5	0 5D	8.0		8.0	24.7		24.5	7.7		7.4	4.74		5.48
	B	B	5	4 ID	0 4D												
	C	C	5	4 ID	0 4D												
	D	D	5	4 ID	0 4D												
3.0	A	A	5	4 ID	0 4D	8.0		8.0	24.5		24.4	7.7		7.2	5.62		6.33
	B	B	5	5	0 5D												
	C	C	5	4 ID	0 4D												
	D	D	5	4 ID	0 5D												
4.0	A	A	5	0 5D	-	8.0			24.4			7.6			7.39		-
	B	B	5	0 5D	-		7.9			24.6			7.9			8.10	
	C	C	5	0 5D	-												
	D	D	5	0 5D	-												
	A																
	B																
	C																
	D																
Loaded / measured by:			MF	SP	SP/MF	MF	SP	MF	MF	SP	MF	MF	SP	MF	MF	SP	MF
Recorded by:			SP	SP	MF	MF	SP	MF	MF	SP	MF	MF	SP	MF	MF	SP	MF

Investigators' Signatures
Sarah Pinchuck
Maitland Faircloth
James Tomé
Bui Phillips

REVIEWER

Concentrations prepared by: MF
Loading verified by: SP
Light Intensity: 50-100 Ft. candles (yes) no)
Photoperiod: 16 hours Light / 8 hours dark (yes) no)
Temperature Range °C:
Incubator # 2 min. 24.2 max. 24.9 mean 24.5
Incubator # ____ min. ____ max. ____ mean ____
Room B246 min. 23.1 max. 25.0 mean 24.4

Remarks: Test run for 96 hours to match duration of ammonia data single test. 48 hour results placed in SRT data file - MF 2/21/06

Statistical Results: Method: Spearman-Karber
(48) 96 hour LC50: 1.44
95% Confidence Interval: lower 1.84 upper 2.06

Revised 01/23/06

Page 000026

Appendix 3. (continued)

FDEP Biology Section - Acute SRT Bench Sheet

Test Organism: *Americamysis bahia* (Method 2007.0) *Menidia beryllina* (Method 2006.0) *Hyalella azteca* (Method 100.1 EPA-600-R-99-064) *Ceriodaphnia dubia* (Method 2002.0) *Cyprinella leedsi* (Method 2000.0) *Pimephales promelas* (Method 2000.0) Other:

Method from EPA-821-R-02-012
Test Type: Screening / Definitive
Static / Static Renewal

Diluent/ Batch#: DMW 12-06
Toxicant/ Batch# NaCl 1-06

Test Beginning: Date: 1-31-06 Time: 1220
Test Ending: Date: 2-4-06 Time: 1200

Water Quality Parameters:	Diluent	Method
Total Residual Cl_2 (mg/L):	<0.03	Hach
Alkalinity (mg/L as $CaCO_3$):	84	Hach
Hardness (mg/L as CaCO3):	85.5	Hach
Total Ammonia (mg/L as N):	<0.097	Denver
Salinity (ppth):	<1	Mettler

Organism Batch #/ Age: 6-06 / <24hrs
Feeding: Prior to test / Prior to renewal / Daily
Food: YCT *P.subcapitata* Artemia
Food Batch: 5-05 8-05 –
Chamber size: 30 mL
Test volume: 20 mL per replicate

Page 2 of 2

Conc. µg/L (g/L)	Rep	Chamber#	Number Live 0 hr / 24 hr / 48 hr			pH (SU) 0 hr / 24 hr / 48 hr			Temperature (°C) 0 hr / 24 hr / 48 hr			D.O. (mg/l) 0 hr / 24 hr / 48 hr			UNCORRECTED Cond. (mmhos/cm) Cond. (µmhom/cm) 0 hr / 24 hr / 48 hr		
			48 hr	72 hr	96 hr	48 hr	72 hr	96 hr	48 hr	72 hr	96 hr	48 hr	72 hr	96 hr	48 hr	72 hr	96 hr
Control	A	A	5	5	5	8.2		8.2	24.0		25.5	7.7		7.5	0.178		0.197
	B	B	5	5	5												
	C	C	5	5	5												
	D	D	5	5	5												
1.5	A	A	5	5	4	8.1		8.1	24.3		25.7	7.7		7.0	2.92		3.27
	B	B	5	5	5												
	C	C	5	5	5												
	D	D	5	0	0												
2.0	A	4	4	0	0	8.0		8.2	24.7		25.6	7.6		7.2	3.85		5.30
	B	B	0	0	0												
	C	C	2	0	0												
	D	D	3	3	3												
2.5	A	A	–	–	–	8.0		–	24.8		–	7.5		–	4.74		–
	B	B	–	–	–												
	C	C	–	–	–												
	D	D	–	–	–												
3.0	A	A	–	–	–	8.0		–	25.0		–	7.6		–	5.63		–
	B	B	–	–	–												
	C	C	–	–	–												
	D	D	–	–	–												
4.0	A	A	–	–	–	–		–	–		–	–		–	–		–
	B	B	–	–	–												
	C	C	–	–	–												
	D	D	–	–	–												
	A																
	B																
	C																
	D																
Loaded / measured by:			MF	MF	MF	SP		MF	SP		MF	SP		MF	SP		MF
Recorded by:			MF	MF	MF	SP		MF	SP		MF	SP		MF	SP		MF

Investigators' Signatures
Jamal Pinchard
Marshall Faircloth
Reid Nichols
Nicole Wetherington
REVIEWER

Concentrations prepared by: SP
Loading verified by: MF for 30 min spot check 2/2/06
Light Intensity: 50-100 Ft. candles (yes) no)
Photoperiod: 16 hours Light / 8 hours dark (yes) no)
Temperature Range °C:
Incubator # 2 min. 24.2 max. 24.8 mean 24.5
Incubator # ___ min. ___ max. ___ mean ___
Room B246 min. 23.7 max. 25.2 mean 24.4

Remarks: TRT run for 96 hours to match duration of environmental sample TRT. 48 hour results placed in SRT Data file MF 2/21/06

Statistical Results: Method: Spearman-Karber
48 / (96) hour LC50: 1.67
95% Confidence Interval: lower 1.55 upper 1.79

Revised 01/23/06

Page 000027

Appendix 3. (continued)

[redacted]

FDEP Biology Section - Acute SRT Bench Sheet

Test Organism: *Americamysis bahia* (Method 2007.0) *Menidia beryllina* (Method 2006.0) *Hyalella azteca* (Method 100.1 EPA-600-R-99-064) *Ceriodaphnia dubia* (Method 2002.0) (*Cyprinella leedsi*) (Method 2000.0) *Pimephales promelas* (Method 2000.0) Other:_____

Method from EPA-821-R-02-012
Test Type: Screening / (Definitive)
Static / (Static Renewal)

Diluent/ Batch#: Well H2O 1-23-06
Toxicant/ Batch# NaCl 1-2-06

Test Beginning: Date: 1-24-06 Time: ~~8~~ 12:15
Test Ending: Date: 1-28-06 Time: 1230

Water Quality Parameters:	Diluent	Method
Total Residual Cl$_2$ (mg/L):	<0.03	Hach
Alkalinity (mg/L as CaCO$_3$):	143	Hach
Hardness (mg/L as CaCO3):	133	Hach
Total Ammonia (mg/L as N):	<0.017	Denver
Salinity (ppth): 21	~~<0.01~~	Mettler

mf 1-24-06

Organism Batch #/ Age: 2-06 / 3days
Feeding: (Prior to test) / Prior to renewal / Daily
Food: YCT P.*subcapitata* Artemia
Food Batch: — — GSL536-90
Chamber size: 1000 mL
Test volume: 500 mL per replicate

Page 1 of 2

Conc. µg/L (Ø)	Rep.	Chamber#	Number Live 0 hr / ~~48 hr~~	Number Live 24 hr / ~~72 hr~~	Number Live 48 hr / ~~96 hr~~	pH (SU) 0 hr / ~~48 hr~~	pH (SU) 24 hr / ~~72 hr~~	pH (SU) 48 hr / ~~96 hr~~	Temperature (° C) 0 hr / ~~48 hr~~	Temperature (° C) 24 hr / ~~72 hr~~	Temperature (° C) 48 hr / ~~96 hr~~	D.O. (mg/l) 0 hr / ~~48 hr~~	D.O. (mg/l) 24 hr / ~~72 hr~~	D.O. (mg/l) 48 hr / ~~96 hr~~	UNCORRECTED Cond. (mmhos/cm) 0 hr / ~~48 hr~~	UNCORRECTED Cond. (mmhos/cm) 24 hr / ~~72 hr~~	UNCORRECTED Cond. (mmhos/cm) 48 hr / ~~96 hr~~
Control	A	F13	10	10	10	8.0		8.3	24.6		24.1	8.4		6.5	0.272		0.309
	B	F14	10	10	10		8.1			24.7			7.3			0.323	
	C																
	D																
6.0	A	F15	10	10	10	7.8		8.1	24.6		24.0	8.2		5.9	10.7		11.01
	B	F16	10	9 D	9		8.0			24.8			7.3			11.0	
	C																
	D																
7.0	A	F17	10	10	10	7.9		8.1	24.7		24.1	8.4		6.0	12.5		12.61
	B	F18	10	9 D	9		8.0			24.7			7.2			12.5	
	C																
	D																
8.0	A	F19	10	9 D	6 3D	7.9		8.1	24.8		24.7	8.4		6.9	14.2		~~14.31~~ 14.6
	B	F20	10	7 3D	7		8.0			24.8			7.1			14.0	
	C																
	D																
9.0	A	F21	10	9 D	~~4 5D~~ A	7.9		8.1	24.8		24.5	8.3		6.1	15.7		15.22
	B	F22	10	8 2D	3 5D		8.0			24.4			7.3			15.7	
	C																
	D																
10.0	A	F23	10	~~8 1D~~ 9 1D	0 1D	7.8		8.1	24.8		24.1	8.4		6.3	17.3		17.09
	B	F24	10	8 2D	1 1D		8.0			24.5			7.3			16.9	
	C																
	D																
	A																
	B																
	C																
	D																
Loaded / measured by:			SP	B	SP	SP	SP	B	SP	B	B	SP	SP	B	SP	SP	B
Recorded by:			B	B	SP	SP	SP	B	SP	B	SP	SP	SP	B	SP	SP	B

Investigators' Signatures
Bonnie Anderson
Joseph Ruthven
Jessica Daniel
Marshall Faircloth

Nigel Willoughby
REVIEWER

Concentrations prepared by: MF
Loading verified by: B
Light Intensity: 50-100 Ft. candles (yes) no)
Photoperiod: 16 hours Light / 8 hours dark (yes) no)
Temperature Range °C:
Incubator 2 min. 24.5 max. 25.1 mean 24.8
Incubator # ___ min. ___ max. ___ mean ___
Room B246 min. 22.9 max. 25.1 mean 24.0

Remarks:
(A) transcription error 3rd SP

48 hr LC50 8.39 95% upper 8.74
95% confidence Lower 8.06

Statistical Results: Method: Spearman-Karber
48 / (96) hour LC50: 7.94
95% Confidence Interval: lower 7.63 upper 8.27

Revised 01/23/06

Page 000024

Appendix 3. (continued)

FDEP Biology Section - Acute SRT Bench Sheet

Test Organism: *Americamysis bahia* (Method 2007.0) *Menidia beryllina* (Method 2006.0) *Hyalella azteca* (Method 100.1 EPA-600-R-99-064) *Ceriodaphnia dubia* (Method 2002.0) **Cyprinella leedsi** (Method 2000.0) *Pimephales promelas* (Method 2000.0) Other:_____

Method from EPA-821-R-02-012
Test Type: Screening / **Definitive**
Static / **Static Renewal**

Diluent/ Batch#: Well H2O 1-23-06
Toxicant/ Batch# NaCl 1-2-06

Test Beginning: Date: 1-24-06 Time: 1215
Test Ending: Date: 1-28-06 Time: 1230

Water Quality Parameters:	Diluent	Method
Total Residual Cl; (mg/L):	<0.03	Hach
Alkalinity (mg/L as $CaCO_3$):	143	Hach
Hardness (mg/L as CaCO3):	133	Hach
Total Ammonia (mg/L as N):	<0.017	Denver
Salinity (ppth):	<1	Mettler

Organism Batch #/ Age: 2-06 13days
Feeding: ~~Prior to test~~ / Prior to renewal / Daily
Food: YCT - P.*subcapitata* - Artemia
Food Batch: GSL536-90
Chamber size: 1000 mL
Test volume: 500 mL per replicate

Page 2 of 2

Conc. µg/L (AI)	Rep.	Chamber#	Number Live ~~0 hr~~ 48 hr	~~24 hr~~ 72 hr	~~48 hr~~ 96 hr	pH (SU) ~~0 hr~~ 48 hr	~~24 hr~~ 72 hr	~~48 hr~~ 96 hr	Temperature (° C) ~~0 hr~~ 48 hr	~~24 hr~~ 72 hr	~~48 hr~~ 96 hr	D.O. (mg/l) ~~0 hr~~ 48 hr	~~24 hr~~ 72 hr	~~48 hr~~ 96 hr	UNCORRECTED Cond. (~~mmhos/cm~~) Cond. (µmhom/cm) ~~0 hr~~ 48 hr	~~24 hr~~ 72 hr	~~48 hr~~ 96 hr
Control	A	F13	10	10	10		8.1			24.2			6.1			0.288	
	B	F211	10	10	10	8.2		8.3	24.1		24.4	8.0		7.5	0.285		0.290
	C																
	D																
6.0	A	F315	10	10	10		8.1			24.3			6.4			16.3	
	B	F16	8[1M]	8	8	8.0		8.1	24.1		24.1	7.8		7.2	10.9		11.3
	C																
	D																
7.0	A	F17	10	10	9[D]		8.1			24.2			6.8			12.7	
	B	F18	9	9	9	8.0		8.1	24.0		24.4	7.7		7.1	12.6		13.2
	C																
	D																
8.0	A	F19	6	6	2[4D]		8.0			24.2			6.8			14.3	
	B	F20	7	7	7	8.0		7.9	24.0		24.4	7.5		6.7	14.1		14.5
	C																
	D																
9.0	A	F21	3[1D]	3[1D]	1[1D]		8.1			24.3			6.6			15.5	
	B	F22	3	3[1D]	2	8.2		8.3	24.1		24.4	7.7		7.4	15.2		15.7
	C																
	D																
10.0	A	F23	-	-	-		8.1			24.7			6.7			17.4	
	B	F24	1	0[1D]	-	8.1		-	24.1		-	7.4		-	17.1		-
	C																
	D																
	A																
	B																
	C																
	D																
Loaded / measured by:			SP	JD	JD	SP	JD	JD	SP	JD	JD	SP	JD	JD	SP	JD	JD
Recorded by:			SP	JD	JD	SP	JD	JD	SP	JD	JD	SP	JD	JD	SP	JD	JD

Investigators' Signatures
Brian Anderson
Sarah Pickland
Marshall Faircloth
James Daniel
Nicole Wetherington
REVIEWER

Renewal
Concentrations prepared by: MF
Loading verified by: MF
Light Intensity: 50-100 Ft. candles (yes) no)
Photoperiod: 16 hours Light / 8 hours dark (yes) no)
Temperature Range °C:
Incubator # 2 min. 24.5 max. 25.1 mean 24.8
Incubator # ___ min. ___ max. ___ mean ___
Room B246 min. 23.8 max. 25.1 mean 24.6

Remarks:
(A) transcription error 3 SP
48 hour results placed in SRT data file SP
48 hour LC50 8.37
95% Confidence level: lower 8.06, upper 8.74

Statistical Results: Method: Spearman-Karber
48 / (96) hour LC50: 7.94
95% Confidence Interval: lower 7.63 upper 8.27

Revised 01/23/06

Page
000025

Appendix 3. (continued)

Ceriodaphnia dubia Acute Standard Reference Toxicant Test

START DATE: 01/31/06 END DATE: 02/04/06 DURATION: 96 H
TOXICANT : NaCl

RAW DATA: Concentration (g/l)	Number Exposed	Mortalities
0.00	20	0
1.50	20	6
2.00	20	17
2.50	20	20
3.00	20	20
4.00	20	20

SPEARMAN-KARBER TRIM: 30.00%

SPEARMAN-KARBER ESTIMATES: LC50: 1.67
95% LOWER CONFIDENCE: 1.55
95% UPPER CONFIDENCE: 1.79

Cyprinella leedsi Acute Standard Reference Toxicant Test

START DATE: 01/24/06 END DATE: 01/28/06 DURATION: 96 H
TOXICANT : NaCl

RAW DATA: Concentration (g/L)	Number Exposed	Mortalities
0 .00	20	0
6.00	20	2
7.00	20	2
8.00	20	11
9.00	20	17
10.00	20	20

SPEARMAN-KARBER TRIM: 10.00%

SPEARMAN-KARBER ESTIMATES: LC50: 7.94
95% LOWER CONFIDENCE: 7.63
95% UPPER CONFIDENCE: 8.27

Appendix 4. Test Organism Receipts

11842 Research Circle
Alachua, FL 32615
T: (386) 462-7889
F: (386) 462-7264
www.hydrosphere.net

HYDROSPHERE
research

Packaging List

Date	Customer Number	Order Shipped Via
1/24/2006	NA	Hydrosphere Research
Customer		**Order Prepared By**
FDEP Marshall Faircloth Biology Mail Stop 6515 2600 Blair Stone Road Tallahassee, Fl 32399-2400		Dustin Onash

Quantity	Description
200	M. bahia from Aquatic Indicators
300	M. beryllina from Aquatic Indicators
120	C. leedsi, #CL 1001, Hatched 1/21/2006

Appendix 4. (continued)

4569 Samuel Street
Sarasota, Florida
34233

NELAC Accredited

MBL
MARINCO
BIOASSAY
LABORATORY

TEL: (941) 925-3594
TEL: (800) 889-0384
FAX: (941) 922-3874
Email: lab@biologylab.com
Internet: biologylab.com

Organism Shipment Record

State of Florida Aquaculture Certificate Number AQ0668007

Date: 1/30/06

Shipped to: FDEP Attn: Marshall Faircloth

P.O. No: _____

Species	Quantity	Age	Brood Number	Temperature	Salinity
Mysidopsis bahia					
Menidia beryllina					
Cyprinella leedsi	1HCT	4 Day	CRLC131	25°C	
Pimephales promelas					

Packed by: Frank Gauvi

Shipped Via: FedEx

Notes: _____

Thank you for your order.

Appendix 5. Chemical Analyses performed on the effluent from the Orange Park Wastewater Treatment Plant Outfall D-001, sampled on January 30, 2006.

Date Sampled	Field ID	Analysis	Component	Results	Units	Remark	MDL	PQL
1/30/2006 7:15	EFFLUENT SAMPLES	Bio-AGP/LimNut	Algal Growth Potential	221	mg DryWt/L		0.3	0.9
1/30/2006 7:15	EFFLUENT SAMPLES	Bio-BOD	Biochemical Oxygen Demand-5 Day,N-Inhib	2.7	mg/L	Q	0.2	2.0
1/30/2006 7:15	EFFLUENT SAMPLES	Bio-Chl-a	Chlorophyll-A, Monochromatic, Water	0.96	ug/L	U	0.96	2.9
1/30/2006 7:15	EFFLUENT SAMPLES	Bio-Chl-a	Phaeophytin-A, Monochromatic, Water	1.7	ug/L	U	1.7	5.1
1/30/2006 12:18	EFFLUENT SAMPLES	Bio-Toxicology	Bioassay-Acute-Screen-FW-C.dubia, LC50	100	LC50	L		
1/30/2006 12:18	EFFLUENT SAMPLES	Bio-Toxicology	Bioassay-Acute-Screen-FW-Fish, LC50	100	LC50	L		
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	1,2,4-Trichlorobenzene	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	1,2-Dichlorobenzene	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	1,3-Dichlorobenzene	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	1,4-Dichlorobenzene	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	2,4,6-Trichlorophenol	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	2,4-Dichlorophenol	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	2,4-Dimethylphenol	49	ug/L	U	49	190
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	2,4-Dinitrophenol	15	ug/L	U	15	58
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	2,4-Dinitrotoluene	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	2,6-Dinitrotoluene	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	2-Chloronaphthalene	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	2-Chlorophenol	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	2-Methyl-4,6-dinitrophenol	2.9	ug/L	U	2.9	12
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	2-Nitrophenol	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	3,3'-Dichlorobenzidine	39	ug/L	U	39	160
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	4,4'-DDD	1.5	ug/L	U	1.5	5.8
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	4,4'-DDE	1.5	ug/L	U	1.5	5.8
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	4,4'-DDT	1.5	ug/L	U	1.5	5.8
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	4-Bromophenyl phenyl ether	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	4-Chloro-3-methylphenol	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	4-Chlorophenyl phenyl ether	1.9	ug/L	U	1.9	7.8
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	4-Nitrophenol	15	ug/L	U	15	58
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Acenaphthene	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Acenaphthylene	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Aldrin	1.5	ug/L	U	1.5	5.8
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	alpha-BHC	1.5	ug/L	U	1.5	5.8

Appendix 5. Chemical Analyses (continued)

Date Sampled	Field ID	Analysis	Component	Results	Units	Remark	MDL	PQL
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Anthracene	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Benzidine	97	ug/L	U	97	390
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Benzo(a)anthracene	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Benzo(a)pyrene	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Benzo(b)fluoranthene	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Benzo(g,h,i)perylene	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Benzo(k)fluoranthene	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	beta-BHC	1.5	ug/L	U	1.5	5.8
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Bis(2-chloroethoxy)methane	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Bis(2-chloroethyl)ether	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Bis(2-chloroisopropyl)ether	2.9	ug/L	U	2.9	12
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Bis(2-ethylhexyl)phthalate	15	ug/L	U	15	58
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Butyl benzyl phthalate	4.9	ug/L	U	4.9	19
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Chrysene	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	delta-BHC	1.5	ug/L	U	1.5	5.8
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Dibenzo(a,h)anthracene	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Dieldrin	1.5	ug/L	U	1.5	5.8
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Diethyl phthalate	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Dimethyl phthalate	49	ug/L	U	49	190
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Di-n-butyl phthalate	4.9	ug/L	U	4.9	19
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Di-n-octyl phthalate	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Endosulfan I	3.9	ug/L	U	3.9	16
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Endosulfan II	3.9	ug/L	U	3.9	16
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Endosulfan sulfate	1.5	ug/L	U	1.5	5.8
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Endrin	1.5	ug/L	U	1.5	5.8
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Endrin aldehyde	3.9	ug/L	U	3.9	16
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Fluoranthene	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Fluorene	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	gamma-BHC	1.5	ug/L	U	1.5	5.8
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Heptachlor	1.5	ug/L	U	1.5	5.8
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Heptachlor epoxide	1.5	ug/L	U	1.5	5.8
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Hexachlorobenzene	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Hexachlorobutadiene	2.9	ug/L	U	2.9	12
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Hexachlorocyclopentadiene	2.9	ug/L	UJ	2.9	12
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Hexachloroethane	2.9	ug/L	U	2.9	12
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Indeno(1,2,3-cd)pyrene	0.97	ug/L	U	0.97	3.9

Appendix 5. Chemical Analyses (continued)

Date Sampled	Field ID	Analysis	Component	Results	Units	Remark	MDL	PQL
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Isophorone	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Naphthalene	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Nitrobenzene	1.9	ug/L	U	1.9	7.8
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	N-Nitrosodimethylamine	1.9	ug/L	U	1.9	7.8
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	N-Nitrosodi-n-propylamine	1.9	ug/L	U	1.9	7.8
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	N-Nitrosodiphenylamine	2.9	ug/L	U	2.9	12
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Pentachlorophenol	2.9	ug/L	U	2.9	12
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Phenanthrene	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Phenol	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	EFFLUENT SAMPLES	BNA-Water	Pyrene	0.97	ug/L	U	0.97	3.9
1/30/2006 7:15	FIELD BLANKS	BNA-Water	1,2,4-Trichlorobenzene	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	1,2-Dichlorobenzene	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	1,3-Dichlorobenzene	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	1,4-Dichlorobenzene	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	2,4,6-Trichlorophenol	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	2,4-Dichlorophenol	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	2,4-Dimethylphenol	48	ug/L	U	48	190
1/30/2006 7:15	FIELD BLANKS	BNA-Water	2,4-Dinitrophenol	14	ug/L	U	14	58
1/30/2006 7:15	FIELD BLANKS	BNA-Water	2,4-Dinitrotoluene	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	2,6-Dinitrotoluene	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	2-Chloronaphthalene	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	2-Chlorophenol	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	2-Methyl-4,6-dinitrophenol	2.9	ug/L	U	2.9	12
1/30/2006 7:15	FIELD BLANKS	BNA-Water	2-Nitrophenol	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	3,3'-Dichlorobenzidine	38	ug/L	U	38	150
1/30/2006 7:15	FIELD BLANKS	BNA-Water	4,4'-DDD	1.4	ug/L	U	1.4	5.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	4,4'-DDE	1.4	ug/L	U	1.4	5.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	4,4'-DDT	1.4	ug/L	U	1.4	5.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	4-Bromophenyl phenyl ether	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	4-Chloro-3-methylphenol	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	4-Chlorophenyl phenyl ether	1.9	ug/L	U	1.9	7.7
1/30/2006 7:15	FIELD BLANKS	BNA-Water	4-Nitrophenol	14	ug/L	U	14	58
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Acenaphthene	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Acenaphthylene	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Aldrin	1.4	ug/L	U	1.4	5.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	alpha-BHC	1.4	ug/L	U	1.4	5.8

Appendix 5. Chemical Analyses (continued)

Date Sampled	Field ID	Analysis	Component	Results	Units	Remark	MDL	PQL
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Anthracene	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Benzidine	96	ug/L	U	96	380
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Benzo(a)anthracene	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Benzo(a)pyrene	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Benzo(b)fluoranthene	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Benzo(g,h,i)perylene	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Benzo(k)fluoranthene	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	beta-BHC	1.4	ug/L	U	1.4	5.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Bis(2-chloroethoxy)methane	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Bis(2-chloroethyl)ether	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Bis(2-chloroisopropyl)ether	2.9	ug/L	U	2.9	12
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Bis(2-ethylhexyl)phthalate	14	ug/L	U	14	58
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Butyl benzyl phthalate	4.8	ug/L	U	4.8	19
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Chrysene	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	delta-BHC	1.4	ug/L	U	1.4	5.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Dibenzo(a,h)anthracene	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Dieldrin	1.4	ug/L	U	1.4	5.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Diethyl phthalate	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Dimethyl phthalate	48	ug/L	U	48	190
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Di-n-butyl phthalate	4.8	ug/L	U	4.8	19
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Di-n-octyl phthalate	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Endosulfan I	3.8	ug/L	U	3.8	15
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Endosulfan II	3.8	ug/L	U	3.8	15
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Endosulfan sulfate	1.4	ug/L	U	1.4	5.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Endrin	1.4	ug/L	U	1.4	5.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Endrin aldehyde	3.8	ug/L	U	3.8	15
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Fluoranthene	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Fluorene	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	gamma-BHC	1.4	ug/L	U	1.4	5.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Heptachlor	1.4	ug/L	U	1.4	5.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Heptachlor epoxide	1.4	ug/L	U	1.4	5.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Hexachlorobenzene	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Hexachlorobutadiene	2.9	ug/L	U	2.9	12
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Hexachlorocyclopentadiene	2.9	ug/L	UJ	2.9	12
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Hexachloroethane	2.9	ug/L	U	2.9	12
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Indeno(1,2,3-cd)pyrene	0.96	ug/L	U	0.96	3.8

Appendix 5. Chemical Analyses (continued)

Date Sampled	Field ID	Analysis	Component	Results	Units	Remark	MDL	PQL
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Isophorone	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Naphthalene	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Nitrobenzene	1.9	ug/L	U	1.9	7.7
1/30/2006 7:15	FIELD BLANKS	BNA-Water	N-Nitrosodimethylamine	1.9	ug/L	U	1.9	7.7
1/30/2006 7:15	FIELD BLANKS	BNA-Water	N-Nitrosodi-n-propylamine	1.9	ug/L	U	1.9	7.7
1/30/2006 7:15	FIELD BLANKS	BNA-Water	N-Nitrosodiphenylamine	2.9	ug/L	U	2.9	12
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Pentachlorophenol	2.9	ug/L	U	2.9	12
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Phenanthrene	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Phenol	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	FIELD BLANKS	BNA-Water	Pyrene	0.96	ug/L	U	0.96	3.8
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Alachlor	0.58	ug/L	U	0.58	2.3
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Ametryn	0.049	ug/L	U	0.049	0.2
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Atrazine	0.056	ug/L	U	0.056	0.23
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Azinphos Methyl	0.19	ug/L	U	0.19	0.76
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Bromacil	0.19	ug/L	U	0.19	0.76
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Butylate	0.19	ug/L	U	0.19	0.76
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Chlorpyrifos Ethyl	0.049	ug/L	U	0.049	0.2
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Chlorpyrifos Methyl	0.097	ug/L	U	0.097	0.39
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Diazinon	0.049	ug/L	UJ	0.049	0.2
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Ethion	0.049	ug/L	U	0.049	0.2
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Ethoprop	0.097	ug/L	U	0.097	0.39
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Fenamiphos	0.19	ug/L	U	0.19	0.76
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Fonofos	0.097	ug/L	U	0.097	0.39
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Hexazinone	0.097	ug/L	U	0.097	0.39
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Malathion	0.15	ug/L	UJ	0.15	0.6
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Metalaxyl	0.24	ug/L	U	0.24	0.96
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Metolachlor	0.49	ug/L	U	0.49	2
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Metribuzin	0.097	ug/L	U	0.097	0.39
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Mevinphos	0.19	ug/L	UJ	0.19	0.76
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Naled	0.78	ug/L	UJ	0.78	3.1
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Norflurazon	0.15	ug/L	U	0.15	0.6
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Parathion Ethyl	0.15	ug/L	U	0.15	0.6
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Parathion Methyl	0.097	ug/L	U	0.097	0.39
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Phorate	0.049	ug/L	U	0.049	0.2
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Prometryn	0.15	ug/L	U	0.15	0.6
1/30/2006 7:15	EFFLUENT SAMPLES	GC-Water	Simazine	0.049	ug/L	U	0.049	0.2

Appendix 5. Chemical Analyses (continued)

Date Sampled	Field ID	Analysis	Component	Results	Units	Remark	MDL	PQL
1/30/2006 7:15	FIELD BLANKS	GC-Water	Alachlor	0.59	ug/L	U	0.59	2.4
1/30/2006 7:15	FIELD BLANKS	GC-Water	Ametryn	0.049	ug/L	U	0.049	0.2
1/30/2006 7:15	FIELD BLANKS	GC-Water	Atrazine	0.049	ug/L	U	0.049	0.2
1/30/2006 7:15	FIELD BLANKS	GC-Water	Azinphos Methyl	0.2	ug/L	U	0.2	0.8
1/30/2006 7:15	FIELD BLANKS	GC-Water	Bromacil	0.2	ug/L	U	0.2	0.8
1/30/2006 7:15	FIELD BLANKS	GC-Water	Butylate	0.2	ug/L	U	0.2	0.8
1/30/2006 7:15	FIELD BLANKS	GC-Water	Chlorpyrifos Ethyl	0.049	ug/L	U	0.049	0.2
1/30/2006 7:15	FIELD BLANKS	GC-Water	Chlorpyrifos Methyl	0.098	ug/L	U	0.098	0.39
1/30/2006 7:15	FIELD BLANKS	GC-Water	Diazinon	0.049	ug/L	UJ	0.049	0.2
1/30/2006 7:15	FIELD BLANKS	GC-Water	Ethion	0.049	ug/L	U	0.049	0.2
1/30/2006 7:15	FIELD BLANKS	GC-Water	Ethoprop	0.098	ug/L	U	0.098	0.39
1/30/2006 7:15	FIELD BLANKS	GC-Water	Fenamiphos	0.2	ug/L	U	0.2	0.8
1/30/2006 7:15	FIELD BLANKS	GC-Water	Fonofos	0.098	ug/L	U	0.098	0.39
1/30/2006 7:15	FIELD BLANKS	GC-Water	Hexazinone	0.098	ug/L	U	0.098	0.39
1/30/2006 7:15	FIELD BLANKS	GC-Water	Malathion	0.15	ug/L	UJ	0.15	0.6
1/30/2006 7:15	FIELD BLANKS	GC-Water	Metalaxyl	0.25	ug/L	U	0.25	1
1/30/2006 7:15	FIELD BLANKS	GC-Water	Metolachlor	0.49	ug/L	U	0.49	2
1/30/2006 7:15	FIELD BLANKS	GC-Water	Metribuzin	0.098	ug/L	U	0.098	0.39
1/30/2006 7:15	FIELD BLANKS	GC-Water	Mevinphos	0.2	ug/L	UJ	0.2	0.8
1/30/2006 7:15	FIELD BLANKS	GC-Water	Naled	0.78	ug/L	UJ	0.78	3.1
1/30/2006 7:15	FIELD BLANKS	GC-Water	Norflurazon	0.15	ug/L	U	0.15	0.6
1/30/2006 7:15	FIELD BLANKS	GC-Water	Parathion Ethyl	0.15	ug/L	U	0.15	0.6
1/30/2006 7:15	FIELD BLANKS	GC-Water	Parathion Methyl	0.098	ug/L	U	0.098	0.39
1/30/2006 7:15	FIELD BLANKS	GC-Water	Phorate	0.049	ug/L	U	0.049	0.2
1/30/2006 7:15	FIELD BLANKS	GC-Water	Prometryn	0.15	ug/L	U	0.15	0.6
1/30/2006 7:15	FIELD BLANKS	GC-Water	Simazine	0.049	ug/L	U	0.049	0.2
1/30/2006 7:15	EFFLUENT SAMPLES	Metals-Water	Aluminum	30	ug/L		5	20
1/30/2006 7:15	EFFLUENT SAMPLES	Metals-Water	Arsenic	4	ug/L	U	4	16
1/30/2006 7:15	EFFLUENT SAMPLES	Metals-Water	Cadmium	0.05	ug/L	U	0.05	0.2
1/30/2006 7:15	EFFLUENT SAMPLES	Metals-Water	Calcium	43.7	mg/L		0.05	0.2
1/30/2006 7:15	EFFLUENT SAMPLES	Metals-Water	Chromium	2	ug/L	U	2	8
1/30/2006 7:15	EFFLUENT SAMPLES	Metals-Water	Copper	3.05	ug/L		0.25	1
1/30/2006 7:15	EFFLUENT SAMPLES	Metals-Water	Iron	93	ug/L		10	40
1/30/2006 7:15	EFFLUENT SAMPLES	Metals-Water	Lead	0.35	ug/L		0.075	0.3
1/30/2006 7:15	EFFLUENT SAMPLES	Metals-Water	Magnesium	19.7	mg/L		0.01	0.04
1/30/2006 7:15	EFFLUENT SAMPLES	Metals-Water	Nickel	1.5	ug/L	I	1	4

Appendix 5. Chemical Analyses (continued)

Date Sampled	Field ID	Analysis	Component	Results	Units	Remark	MDL	PQL
1/30/2006 7:15	EFFLUENT SAMPLES	Metals-Water	Selenium	0.5	ug/L	U	0.5	2
1/30/2006 7:15	EFFLUENT SAMPLES	Metals-Water	Silver	0.34	ug/L		0.025	0.1
1/30/2006 7:15	EFFLUENT SAMPLES	Metals-Water	Zinc	42	ug/L		3	12
1/30/2006 7:15	FIELD BLANKS	Metals-Water	Aluminum	5	ug/L	U	5	20
1/30/2006 7:15	FIELD BLANKS	Metals-Water	Arsenic	4	ug/L	U	4	16
1/30/2006 7:15	FIELD BLANKS	Metals-Water	Cadmium	0.05	ug/L	U	0.05	0.2
1/30/2006 7:15	FIELD BLANKS	Metals-Water	Calcium	0.05	mg/L	U	0.05	0.2
1/30/2006 7:15	FIELD BLANKS	Metals-Water	Chromium	2	ug/L	U	2	8
1/30/2006 7:15	FIELD BLANKS	Metals-Water	Copper	0.25	ug/L	U	0.25	1
1/30/2006 7:15	FIELD BLANKS	Metals-Water	Iron	10	ug/L	U	10	40
1/30/2006 7:15	FIELD BLANKS	Metals-Water	Lead	0.075	ug/L	U	0.075	0.3
1/30/2006 7:15	FIELD BLANKS	Metals-Water	Magnesium	0.01	mg/L	U	0.01	0.04
1/30/2006 7:15	FIELD BLANKS	Metals-Water	Nickel	1	ug/L	U	1	4
1/30/2006 7:15	FIELD BLANKS	Metals-Water	Selenium	0.5	ug/L	U	0.5	2
1/30/2006 7:15	FIELD BLANKS	Metals-Water	Silver	0.025	ug/L	U	0.025	0.1
1/30/2006 7:15	FIELD BLANKS	Metals-Water	Zinc	3	ug/L	U	3	12
1/30/2006 7:15	EFFLUENT SAMPLES	Nutrients-Liquid	Alkalinity	113	mg CaCO3/L		0.65	2.5
1/30/2006 7:15	EFFLUENT SAMPLES	Nutrients-Liquid	Ammonia-N	4.9	mg N/L		0.5	1
1/30/2006 7:15	EFFLUENT SAMPLES	Nutrients-Liquid	Kjeldahl Nitrogen	6.6	mg N/L		0.24	0.6
1/30/2006 7:15	EFFLUENT SAMPLES	Nutrients-Liquid	NO2NO3-N	11	mg N/L		0.08	0.2
1/30/2006 7:15	EFFLUENT SAMPLES	Nutrients-Liquid	O-Phosphate-P	3.3	mg P/L		0.16	0.4
1/30/2006 7:15	EFFLUENT SAMPLES	Nutrients-Liquid	Specific_Conductance	751	umhos/cm		2	8
1/30/2006 7:15	EFFLUENT SAMPLES	Nutrients-Liquid	Total-P	3.7	mg P/L		0.1	0.3
1/30/2006 7:15	EFFLUENT SAMPLES	Nutrients-Liquid	TSS	4	mg/L	U	4	16
1/30/2006 7:15	FIELD BLANKS	Nutrients-Liquid	Ammonia-N	0.012	mg N/L	I	0.01	0.02
1/30/2006 7:15	FIELD BLANKS	Nutrients-Liquid	Kjeldahl Nitrogen	0.08	mg N/L	U	0.08	0.2
1/30/2006 7:15	FIELD BLANKS	Nutrients-Liquid	NO2NO3-N	0.034	mg N/L		0.004	0.01
1/30/2006 7:15	FIELD BLANKS	Nutrients-Liquid	O-Phosphate-P	0.004	mg P/L	U	0.004	0.01
1/30/2006 7:15	FIELD BLANKS	Nutrients-Liquid	Total-P	0.02	mg P/L	U	0.02	0.06

The Bioassay of Orange Park Wastewater Treatment Plant effluent sampled on January 29 and 30, 2006. NPDES #FL0023922.

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 2 3 9 2 2 11	12 0 6 0 1 3 0	18 **B**	19 **S**	20 1

Remarks

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The Priority Pollutants Analysis of Orange Park Wastewater Treatment Plant effluent sampled on January 30, 2006. NPDES #FL0023922.

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 2 3 9 2 2 11	12 0 6 0 1 3 0	18 **X**	19 **S**	20 1

Remarks

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Biological Analyses of Orange Park Wastewater Treatment Plant effluent sampled on January 30, 2006. NPDES #FL0023922.

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 2 3 9 2 2 11	12 0 6 0 1 3 0	18 **S**	19 **S**	20 1

Remarks

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