

Environment Testing Bio-Aquatics



Natural Energy Laboratory of Hawaii Authority Hawaii Ocean Science and Technology Park OUTFALL SSW-28 Ocean Intake

Chronic Biomonitoring Report

98036

Americamysis bahia Menidia beryllina

August 22, 2025

Approved by: Lab director

Eurofins Environment Testing Bio-Aquatics • 2501 Mayes Rd. Ste. 100 • Carrollton, Texas • 75006

TABLE OF CONTENTS

TOXICITY TEST REPORT

SURVIVAL TEST SUMMARY

6

STATISTICAL & CHEMICAL ANALYSIS

Appendix A

REFERENCE TOXICANTS

Appendix B

LITERATURE REFERENCES

Appendix C

CHAIN-OF-CUSTODY SHEETS

Appendix D

REGULATORY AGENCY TABLES

Appendix E

Unless otherwise noted in the body of the report, all data reported in this document are in compliance with TNI standards and apply only to the samples referenced within. This report document may not be edited or reproduced in part or in full by any other entity, unless Eurofins Environment Testing Bio-Aquatics issues written approval.

*HAND-WRITTEN RAW DATA TABLES ARE AVAILABLE UPON REQUEST

2501 Mayes Road, Suite 100 Carrollton, Texas 75006 Tel: (972) 242-7750 Fax: (972) 242-7749

TOXICITY TEST REPORT - Chronic

Client:Natural Energy Laboratory of Hawaii Authority
Facility:Hawaii Ocean Science and Technology Park
Permit No.

Sample: SSW-28 Ocean Intake
Laboratory Number: 98036
Date: August 22, 2025

Americamysis bahia passed survival and growth testing requirements. Menidia beryllina passed survival and growth testing requirements.

SAMPLE COLLECTION:

A grab sample from the Natural Energy Laboratory of Hawaii Authority, Hawaii Ocean Science and Technology Park, was transported to Eurofins Environment Testing Bio-Aquatics on August 22, 2025. The sample was collected from the SSW-28 Ocean Intake by facility personnel.

The sample was analyzed for total residual chlorine using the Hanna Ion Specific Meter #711 and contained <0.10 mg/L. The sample and laboratory dilution water pH, temperature, salinity, and dissolved oxygen data were collected daily.

TEST PROCEDURES:

Americamysis bahia

EPA METHOD: 1007

The seven-day Chronic *Americamysis bahia* survival and growth test was initiated at 14:52 hours on August 22, 2025. One concentration of 100% was prepared utilizing synthetic water as dilution water. The test was set up with 266mL plastic cups containing 200mL of test solution or control dilution water. Each concentration consisted of five replicate cups with five organisms each, giving a total of 25 (twenty-five) per treatment. The control was run concurrently with the test. Test organisms were 7-day old laboratory cultured juveniles. Juveniles were randomly introduced into test solutions and controls. The number of surviving organisms, and water quality parameters in the old test solutions, were recorded after each 24-hour period. Water quality parameters were again measured after the test was renewed with fresh solutions. Surviving organisms in each test chamber were fed freshly hatched brine shrimp two times per day. The test proceeded for seven days.

At the end of the test, all organisms were sacrificed, dried, and weighed. The test ended at 16:13 hours on August 29, 2025. Survival and growth (weight) data were statistically (p=0.05) analyzed according to EPA procedures to determine the Lowest Observable Effect Concentration (LOEC) and the No Observable Effect Concentration (NOEC).

SURVIVAL:

Americamysis bahia

The Equal and Unequal variance t-test performed on survival test data demonstrated no statistically significant differences between the control and any of the sample concentrations tested.

LOEC: Not Calculable (Q)

NOEC: 100%

GROWTH:

Americamysis bahia

The *Americamysis bahia* growth data were normally distributed at the alpha level of 0.01 (13.277) using the Chi-square test for normality. Using the Equal and Unequal variance t-test on *Americamysis bahia* growth data demonstrated no statistically significant differences between the control and any of the sample concentrations tested.

LOEC: Not Calculable (Q)

NOEC: 100%

TEST PROCEDURES:

Menidia beryllina

EPA METHOD: 1006

The seven-day Chronic *Menidia beryllina* survival and growth test was initiated at 14:50 hours on August 22, 2025. One concentration of 100% was prepared utilizing synthetic water as dilution water. The test was set up with 650mL plastic cups containing 600mL of test solution as test chambers. The test organisms were initiated in synthetic lab water 24 hours before the test began. Each concentration consisted of three replicate chambers containing eight laboratory-cultured larvae each, giving a total of 24 (twenty-four) per treatment. The control was run concurrently with the test. Test organisms were laboratory cultured *Menidia beryllina* between seven and eleven days old. Juveniles were randomly introduced into test solutions and controls. The number of surviving juveniles, and water quality parameters in the old test solutions were recorded after each 24-hour period. Water quality parameters were again measured after the test was renewed with fresh solutions. Surviving organisms in each test chamber were fed freshly hatched brine shrimp two times per day. The test proceeded for seven days.

At the end of the test, all organisms were sacrificed, dried, and weighed. The test ended at 16:54 hours on August 29, 2025. Survival and growth (weight) were statistically (p=0.05) analyzed according to EPA procedures to determine the Lowest Observable Effect Concentration (LOEC) and the No Observable Effect Concentration (NOEC).

SURVIVAL:

Menidia beryllina

The Equal and Unequal variance t-test performed on *Menidia beryllina* survival data demonstrated no statistically significant differences between the control and any of the sample concentrations tested.

LOEC: Not Calculable (Q)

NOEC: 100%

GROWTH:

Menidia beryllina

The *Menidia beryllina* growth data were normally distributed at the alpha level of 0.01 (0.900) using Shapiro Wilk's test for normality. Using the Equal and Unequal variance t-test on *Menidia beryllina* growth data demonstrated no statistically significant differences between the control and any of the sample concentrations tested.

LOEC: Not Calculable (Q)

NOEC: 100%

Eurofins Environment Testing Bio-Aquatics TOXICITY TEST

Chronic Americamysis bahia

Client: Natural Energy Laboratory of Hawaii Hawaii Ocean Science and Technology Park Lab ID: 98036

Permit Number: N/A Test Temperature (oC): 25 ± 1

Sample Type: Grab Outfall Name: SSW-28 Ocean Intake Photo Period: 16 Hours Light

8 Hours Dark

Receiving Water Name:

Begin Date: 8/22/2025

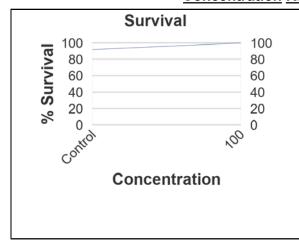
Test Start Time: 14:52 Test End Time: 16:13 **End Date:** 8/29/2025

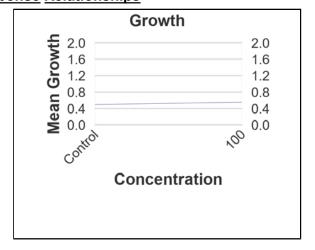
SURVIVAL

Effluent Con. % 8/22 8/23 8/24 8/25 8/26 8/27 8/28 8/29 Surv. A S 4 4 4 4 4 4 4 4 B S S S S S S S S S	E-604					~ ~	14 1 11				
% 8/22 8/23 8/24 8/25 8/26 8/27 8/28 8/29 Surv. Control A 5 4			Number of Alive								
Control B	%		8/22	8/23	8/24	8/25	8/26	8/27	8/28	8/29	
Control C		A	5	4	4	4	4	4	4	4	
C S S S S S S S S S		В	5	5	5	4	4	4	4	4	
E 5 5 5 5 5 5 5 5 5 100.0% A 5 5 5 5 5 5 5 5 5 100.0% C 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	Control	С	5	5	5	5	5	5	5	5	92.0%
A 5 5 5 5 5 5 5 5 5 5 5 100.0% 100 C 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5		D	5	5	5	5	5	5	5	5	
B 5 5 5 5 5 5 5 5 100.0% C 5 5 5 5 5 5 5 5 5 5 5 100.0% D 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5		Е	5	5	5	5	5	5	5	5	
100 C 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5		A	5	5	5	5	5	5	5	5	
D 5		В	5	5	5	5	5	5	5	5	
E 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	100	С	5	5	5	5	5	5	5	5	100.0%
A B C C C C C C C C C C C C C C C C C C		D	5	5	5	5	5	5	5	5	
B		Е	5	5	5	5	5	5	5	5	
C		A									
D		В									
B		С									
A		D									
B		Е									
B		A									
		H									
		Ħ									
		D									
		Е									

Effluent	[0.0.11				
Con. %	Į	8/22	8/23	8/24	Number 8/25	Of Alive 8/26	8/27	8/28	8/29	Avg% Surv.
7.0		0.22	0.25	<u> </u>	1		<u> </u>			Sui v.
	A									
	В									
	С									
	D									
	Е									
					<u> </u>					
	Α									
	В									
	С									
	D									
	Е									
	E									
	A									
	В									
	С									
	H									
	D									
	Е									
	A									
	H									
	В									
	С									
	D									
	Е									
					<u> </u>					

Concentration Response Relationships





BIU-AQUATIC TESTING, INC.

	Chronic Americamysis bahia SURVIVAL Lab ID: 98036																									
C1	ier	t:Na	tural	Ener	gy La	borat	tory o	of Hav	waii	Faci	ility	Haw	vaii	Ocea	n Sci	ence a	nd T	echno	logy	_	C	utfal de Ty		SSW. Grab	-28	
TES	ΤI	NSTF	RUCT	IONS	: Му	sid te	est is	Abbr	eviate	d Rep	os (01	nly n	eec	1 5 N	OT 8))								-114		
Cult	ure	No.	:_^	My-	25-	22	1			_ <u>P</u>	hoto	Perio	<u>od</u> : 1	l 6hr 1	light, 8	Bhr da	rk	RAN	DOM	IZA	TIO	<u>V:</u> [SC-5		1	
		D	iluti	on:	1/	Cont	rol		, 	11	100	-Tr				1		ı 	1	- 						_
		DATE/I TECHN	IME/ ICIAN	A	В	С	D	Е	A	В	С	D		Е	A	В	С	D	Е		Α	В	С	D	Е	
0Hr	- 1	8-22 16 14		5	E-BOLOGOZIANON	n eld Marches on algebra.		********	5	· Agrand Street Visites	The second district of	ita e senden ered		aude												
24Hr		77 /1		4.	2	Approximately		lanenso.	S	p. 3900000	es especial construction of the second															
48Hr	12	5-24	-25	e de la company	6	C and Delicembre	ary account on the same of the	an control and	5	**************************************		or Market scott		000												
72Hr		8-25. ((1	25	4	40	5-	Andreicher Bei Spenson	-	5-			oo I To a series														
96Hr	8	20-2 B0	25	H	4	5		ganin-	5		. everatas, o			queoti ^{ne}												
5 days	95	3-2			4	5	en mantenner.	entrectus	5	·				,												
6 days	3		25 354	1-1	4	5		Anna annual	5																	_
7 days	Ι.	29		4	4	5	A village and a second	**Citatrong	5	- Andrewson		Pile trade control of the		*******											1	_
Dil	uti							! <u></u>	J		<u> </u>		1						<u> </u>	J L						_
		A	В	С	D	Е		A	В	С	D	Е		A	В	С	D	Е		A	В	C	: 1)	Е	
0Н	[r																									
24H	[r						Ī																			
48H	r																		Ī							
72H	r																			***************************************						
96Н	r																		_ [
5 da	ys																									
6 day	ys																									
7 day	ys																									

Lined through spaces preceded by a number represent the same number. Lined spaces without a preceding number indicate unused or not applicable sx

DIU-AQUATIC LESTING, INC.

	Chronic	American	nysis bahia SURVIVAL	La	b ID: 980 :	36
Client: Natural	Energy Laborat	ory of Hawaii	Facility Hawaii Ocean Science and Tech	nology	Outfall: Sample Type	SSW-28 Grab
TEST INSTRUCTI	ONS: Mysid te	est is Abbreviat	ed Reps (only need 5 NOT 8)			

Test Temperatures

	0Hr	24Hr	48Hr	72Hr	<u>96Hr</u>	5 days	6 days	7 days
Control 100	new 25.9	old / new	old / new 25 / 25 /	old / new 26 / 25 /	old / new 25/5 25/5	old / new 27,3 25,3	old / new	old 25-8
TIME/DATE TECH IR GUN ID #	8-22-28 56 1452	8-13-25 DT 1/26	8-24-28 50 1010	9-29-29 CCC 1489	9-20-25 45 0820 013	8-27-25 8-1130 013	11 1	0-29-25 56 1613

Lined through spaces preceded by a number represent the same number. Lined spaces without a preceding number indicate unused or not applicable spaces.

TOXICITY TEST

Chronic Americamysis bahia

Client: Natural Energy Laboratory of Hawaii Ocean Science and Technology Park

Lab ID: 98036

Permit Number: N/A

Sample Type: Grab Outfall Name: SSW-28 Ocean Intake

Receiving Water Name:

Synthetic 100 SN Avg. Avg. ON Avg. 5 2.52 0.504 0.630 5 3.06 0.612 5 5 2.49 0.498 В 2.70 0.540 В В 0.623 0.442 0.442 0.556 2.21 2.78 5 5 5 2.59 0.518 0.518 D 2.30 0.460 D D 0.526 0.526 3.04 0.608 C.V. % C.V. % Mean Mean C.V. % Mean Mean C.V. % 0.498 6.63 0.555 11.14

SN Mean	SN C.V. %
0.548	14.4

A ON Wt.	Avg. ON	Wt. Avg.	ON W	/t. Avg.	ON	Wt. Avg.
В	В		В		В	
С	С		С		С	
D	D		D		D	
E	Е		Е		Е	
Mean C.V	V. % Mean	C.V. %	Mean	C.V. %	Mean	C.V. %

Note: ON stands for original number per replicate, while SN refers to the number surviving after test completion.

^{* =} spilled cup

Chronic

Americamysis bahia TOXICITY TEST

Client:	Natural Energy Labor	Authority	Lab ID:	8036	
Facility:	Hawaii Ocean Sci	ence and Technol	ogy Park	Balance: Radwag BAL-	·007
Begin Date: 8/2: Analyst: Weigh Date: _	2/2025 End Date: 9/2/28	8/29/2025	Organism: Americamy Date/Time placed ir Date/Time removed	1	1740
Qty.	Control Wt. 2-52	Qty.	100 % Wt. 3.06	Qty.	Wt.
в - 7 с 5 р 5	2.21 2.59 2.63	B	2.78 2.30 3.04	B	
Qty.	Wt.	Qty.	Wt.	Qty.	Wt.
B C D		В С С D		B C D	
Е		Е		Е	
Qty.	Wt.	Qty.	Wt.	Qty.	Wt.
В		ВС		B C	
D E	ANNA CALL ACTION AND ACTION ACTION AND ACTION ACTI	D E		D E	

Lined through spaces preceded by a number represent the same number. Lined spaces without a preceding number indicate unused or not applicable spaces.

Eurofins Environment Testing Bio-Aquatics TOXICITY TEST

Chronic Menidia beryllina

Client:	Natural Energy Laboratory of	Hawaii Ocean Science and Technology	Lab ID:	98036
---------	------------------------------	-------------------------------------	---------	-------

Permit Number: N/A **Test Temperature (oC):** 25 ± 1

Sample Type: Grab Outfall Name: SSW-28 Ocean Intake Photo Period: 16 Hours Light

8 Hours Dark

Receiving Water Name:

Begin Date: 8/22/2025

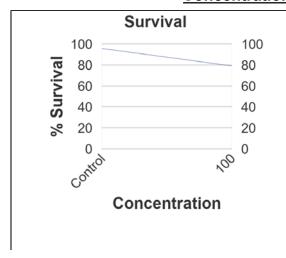
Test Start Time: 14:50 Test End Time: 16:54 **End Date:** 8/29/2025

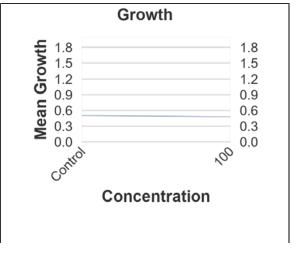
SURVIVAL

Effluent						Of Alive				Avg%
Con.		8/22	8/23	8/24	8/25	8/26	8/27	8/28	8/29	Surv.
	A	8	8	8	8	8	8	7	7	
Control	В	8	8	8	8	8	8	8	8	95.8%
	С	8	8	8	8	8	8	8	8	
	D									
	A	8	8	8	8	8	8	8	8	
	В	8	8	8	8	8	7	6	5	79.2%
100	С	8	8	8	8	8	7	6	6	
	D									
	A									
	В									
	С									
	D									
	_									
	A									
	В	_			_					
	С									
	D									

Effluent				Number	Of Alive				Avg%
Con.	8/22	8/23	8/24	8/25	8/26	8/27	8/28	8/29	Surv.
	A								
 									
	В								
	С								
	D								
L		<u> </u>	II						
	A								
1 - 1	_	<u> </u>							
	В								
	С								
	D								
		JI	II						
	A								
	В								
	D								
	С								
	D								
	_'!		!!			!			
	A								
	В								
	- 								
	С								
	D								
		,							

Concentration Response Relationships





Eurofins Environment Testing Bio-Aquatics Menidia beryllina SURVIVAL Chronic Lab ID: 98036 Client: Natural Energy Laboratory of Hawaii Facility Outfall:SSW-28 Ocean Hawaii Ocean Science and Sample TypGrab TEST INSTRUCTIONS: Mysid test is Abbreviated Reps (only need 5 NOT 8) Culture No.: Mn-27-223 Photo Period: 16hr light, 8hr dark **RANDOMIZATION:** Control Dilution: 100 DATE/TIME/ TECHNICIAN C \mathbf{C} В \mathbf{C} D E 8 0Hr 1450 24Hr X DT-1059 -2+-Cr 48Hr 1023 -25-25 72Hr a 1512 3.24.25 96Hr 5 days 6 days 7 days 56 1454 Dilution: В C C D E В C D C В E 0Hr 24Hr 48Hr 72Hr 96Hr 5 days

Lined through spaces preceded by a number represent the same number. Lined spaces without a preceding number indicate unused or not applicable spa

6 days

7 days

BIO-AQUATIC TESTING, INC.

	Chronic	Menidia b	eryllina	SURVIVAL	Lab ID: 98036
·	l Energy Labora		_	Hawaii Ocean Science and	Outfall:SSW-28 Ocean Sample Typ@rab
FEST INSTRUCT	Mysid te	st is Abbreviate	ed Reps (or	nly need 5 NOT 8)	

Test Temperatures

	<u>OHr</u>	24Hr	48Hr	<u>72Hr</u>	<u>96Hr</u>	5 days	6 days	7 days
	new	old / new	old / new	old / new	old / new	old / new	old / new	_old
Control	25.4		25 / 25 /	257 25.3	25/5 25/5	253 2514	25 8 25/5	25/3
100		13.6						

TIME/DATE TECH	8-22-25	8.29. 25	8-24-25	8-25-29	8:24-25	8-27-25		079-25
	56 1450	1059	SG 1023	CCC 1512	BB 0810	3 12	B 1407	86 1654
IR GUN ID#	013	0/3	013	ol3	013	013	013	013

Lined through spaces preceded by a number represent the same number. Lined spaces without a preceding number indicate unused or not applicable spaces.

2nd Organism Eurofins Environment Testing Bio-Aquatics TOXICITY TEST

Chronic Menidia beryllina

Client: Natural Energy Laboratory of Hawaii Ocean Science and Technology Lab ID: 98036

Permit Number: N/A

Sample Type: Grab Outfall Name: SSW-28 Ocean Intake

Receiving Water Name:

	S	ynth	netic		SN		10	0												
	ON SI	<u> </u>	Wt.	Avg.	Avg.		ON	Wt.	_	Avg.		ON	Wt.	Avg.			ON	Wt.		Avg.
A	8 7		3.88	0.485	0.554	Α	8	4.38		0.548	A					A			∐Լ	
В	8 8	IJĹ	4.00	0.500	0.500	В	8	3.71		0.464	В			<u> </u>		В				
С	8 8		4.20	0.525	0.525	С	8	3.45		0.431	С					С			╛	
D						D					D					D			$\rfloor \lfloor$	
	Me	an	(C.V. %			Mean		C	c.V. %	N	1ean	(C.V. %		I	Mean		C.V	V. %
	0.5	503		4.01			0.481		1	2.47										
	SN N	1ean	SN	V C.V. %	- -					· ·					-					
	1 05	26	- II	5 16																

_	ON	Wt.	Avg.		ON	Wt.	Avg.	,		ON	Wt.	Avg.	ı		ON	Wt.	Avg.
Α				A					Α					Α			<u> </u>
В				В					В					В			
С				С					С					С			
D				D					D					D			
	Mean	C.	.V. %		Mean	(C.V. %		I	Mean	C	.V. %		1	Mean	(C.V. %

Note: ON stands for original number per replicate, while SN refers to the number surviving after test comple

TOXICITY TEST

Chronic

Menidia beryllina

Lab ID:

98036

Client: Natural Energy Laboratory of Hawaii - Hawaii Ocean Science and

Balance: Radwag BAL-007

Begin Date: 8/22/2025

End Date: 8/29/2025

Organism: Menidia beryllina

Analyst: -Weigh Date:

Date/Time placed in Oven:

Date/Time removed from Oven

Wt.

Control

	Qty.	Wt.
Α	7	3-88
В	8	4.00
С	\mathcal{S}_{-}	4.20
-D-		

100 %

	Qt <u>y</u> .	Wt.
A	S	4.38
В	5	3.71
С	φ	3.45
Đ-	A COMPANY OF THE PROPERTY OF T	

Qty.

A	
В	
С	
D	

Qty. Wt.

	Qty.	Wt.
A		
В		
С		
D		

	Qty.	Wt.
A		
В		
С		
D		

	Qty.	Wt.				
A						
В						
С						
D						

	Qty.	Wt.
A		
В		
С		
D		

	Qty.	Wt.
 Α		
 В		
С		
D		

Lined through spaces preceded by a number represent the same number. Lined spaces without a preceding number indicate unused or not applicable spaces.

APPENDIX A

STATISTICS SUMMARY

Both the lethal and sub-lethal endpoints were statistically calculated according to their respective EPA guidelines. The Chronic Freshwater organisms were calculated according to EPA-821-R-02-013, October 2002 Fourth Edition. The Chronic Marine and Estuarine organisms were calculated according to EPA-821-R-02-014, October 2002 Third Edition. The Acute Freshwater and Marine organisms were calculated according to EPA-821-R-02-012, October 2002 Fifth Edition. The fertilization organisms were calculated according to EPA-600-R-95-136 or EPA-600-R-12-022, dependent upon the species. Listed below are the basic principles of these guidelines. If you would like a copy of the raw statistical calculations for your test then please contact us.

The chronic and acute *Pimephales promelas* and *Menidia beryllina* survival data is analyzed using Shapiro Wilks Test and Bartlett's Test. If the data passes both tests then the data is run through ANOVA and Dunnetts (parametric). If the data fails Shapiro Wilks Test or Bartlett's Test then Steels Many One Test (non-parametric) is used. The chronic *Pimephales promelas* and *Menidia beryllina* growth data is analyzed using Shapiro Wilks Test and Bartlett's Test. If the data passes one of these tests then the data is run through ANOVA and Dunnetts. If the data fails Shipiro Wilks Test and Bartlett's Test then Steels Many One Test is used. Point estimation may also be used.

The chronic *Mysidopsis bahia* survival data is analyzed using Chi-square test and Bartlett's Test. If the data passes both tests then the data is run through ANOVA and Dunnetts. If the data fails Chi-square test or Bartlett's Test then Steels Many One Test is used. *Mysidopsis bahia* growth data is analyzed using Chi-square test and Bartlett's Test. If the data passes one of these tests then the data is run through ANOVA and Dunnetts. If the data fails Chi-square test and Bartlett's Test then Steels Many One Test is used. Point estimation may also be used.

The acute *Mysidopsis bahia* survival data is analyzed using Shapiro Wilks Test and Bartlett's Test. If the data passes both tests then the data is run through ANOVA and Dunnetts. If the data fails Shipiro Wilks Test or Bartlett's Test then Steels Many One Test is used. Point estimation may also be used.

The chronic *Ceriodaphnia dubia* survival data are analyzed using the Fisher's Exact Test. The chronic *Ceriodaphnia dubia* reproduction and are analyzed using the Chi-square test and Bartlett Test. If the data passes one of these tests then the data is run through ANOVA and Dunnetts. If the data fails Chi-square test and Bartlett's Test then Steels Many One Test is used. Point estimation may also be used.

The acute *Daphnia pulex* and *Ceriodaphnia dubia* survival data is analyzed using Shapiro Wilks Test and Bartlett's Test. If the data passes both tests then the data is run through ANOVA and Dunnetts. If the data fails Shapiro Wilks Test or Bartlett's Test then Steels Many One Test is used. Point estimation may also be used.

The fertilization data is analyzed using Shapiro Wilks Test and Bartlett's Test. If the data passes both tests then the data is run through ANOVA and Dunnetts. If the data fails Shapiro Wilks Test or Bartlett's Test then Steels Many One Test is used. Point estimation or TST methodology may also be used.

mysid growth

File: 98036.myg Transform: NO TRANSFORMATION

Chi-square test for normality: actual and expected frequencies

Calculated Chi-Square goodness of fit test statistic = 6.7702 Table Chi-Square value (alpha = 0.01) = 13.277

Data PASS normality test. Continue analysis.

mysid growth

File: 98036.myg Transform: NO TRANSFORMATION

F-Test for equality of two variances

GROUP IDENTIFICATION VARIANCE F

1 con 0.001
2 100 0.004 3.515

Critical F = 23.20 (P=0.01, 4, 4)

Since F <= Critical F, FAIL TO REJECT Ho: Equal Variances.

mysid growth

File: 98036.myg Transform: NO TRANSFORMATION

ANOVA TABLE

 SOURCE
 DF
 SS
 MS
 F

 Between
 1
 0.008
 0.008
 3.374

 Within (Error)
 8
 0.020
 0.002

 Total
 9
 0.028

Critical F value = 5.32 (0.05,1,8)

Since F < Critical F FAIL TO REJECT Ho: All equal

mysid growth

File: 98036.myg Transform: NO TRANSFORMATION

EQUAL	VARIANCE t-TEST -	TABLE 1 OF 2	Ho:Contro	1 <treatm< th=""><th>ent</th></treatm<>	ent					
GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG					
1 2	con 100	0.498 0.555	0.498 0.555	-1.837						
2 Sample	2 Sample t table value = 1.86 (1 Tailed Value, P=0.05, df=8,1)									
UNEQUA	L VARIANCE t-TEST		Ho:Contro	l <treatm< td=""><td>ent</td></treatm<>	ent					
GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG					
1 2	con 100	0.498 0.555	0.498 0.555	-1.837						

2 Sample t table value = 1.94 (1 Tailed Value, P=0.05, df=6,1)

mysid growth

File: 98036.myg Transform: NO TRANSFORMATION

EQUAL	VARIANCE t-TEST -	TABLE	2 OF 2	Ho:Contr	ol <treatment< th=""></treatment<>
GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)		DIFFERENCE FROM CONTROL
1 2	con 100	5 5	0.058	11.7	-0.058
UNEQUAL	VARIANCE t-TEST			Ho:Cont	rol <treatment< td=""></treatment<>
GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)		DIFFERENCE FROM CONTROL
1 2	con 100	5 5	0.061	12.2	-0.058

menidia growth

File: 98036.meg Transform: NO TRANSFORMATION

Shapiro - Wilk's test for normality

D = 0.008

W = 0.949

Critical W (P = 0.05) (n = 6) = 0.788Critical W (P = 0.01) (n = 6) = 0.713

Data PASS normality test at P=0.01 level. Continue analysis.

menidia growth

File: 98036.meg Transform: NO TRANSFORMATION

F-Test for equality of two variances

GROUP IDENTIFICATION VARIANCE F
---- 1 con 0.000

con 0.000 2 100 0.004 8.912

Critical F = 199.00 (P=0.01, 2, 2)

Since F <= Critical F, FAIL TO REJECT Ho: Equal Variances.

menidia growth

File: 98036.meg Transform: NO TRANSFORMATION

ANOVA TABLE

 SOURCE
 DF
 SS
 MS
 F

 Between
 1
 0.001
 0.001
 0.370

 Within (Error)
 4
 0.008
 0.002

 Total
 5
 0.009

Critical F value = 7.71 (0.05,1,4)

Since F < Critical F FAIL TO REJECT Ho: All equal

menidia growth

File: 98036.meg Transform: NO TRANSFORMATION

EQUAL	VARIANCE t-TEST -	TABLE 1 OF 2	Ho:Contro	l <treatm< th=""><th>ent</th></treatm<>	ent
GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1 2	con 100	0.503 0.481	0.503 0.481	0.608	
2 Sampl	e t table value = 2.1	 13 (1 Tailed	d Value, P=0.05, df=	4,1)	
UNEQU	AL VARIANCE t-TEST		Ho:Contro	l <treatm< td=""><td>ent</td></treatm<>	ent
GROUP	IDENTIFICATION	TRANSFORMED MEAN		T STAT	SIG
1 2		0.503 0.481	0.503 0.481	0.608	

2 Sample t table value = 2.92 (1 Tailed Value, P=0.05, df=2,1)

menidia growth

File: 98036.meg Transform: NO TRANSFORMATION

EQUAL	VARIANCE t-TEST -	TABLE	2 OF 2	Ho:Contr	ol <treatment< th=""></treatment<>
GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)		
1 2	con 100	3 3	0.078	15.6	0.022
UNEQUAL	VARIANCE t-TEST			Ho:Cont	rol <treatment< td=""></treatment<>
GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)		
1	con	3			

2 100 3 0.107 21.3 0.022

SALT WATER TEST SETUP FORM Client: Natural Energy Laboratory of Hawaii Permit N/A Lab Number 98036 Hawaii Ocean Science and Technology Facility: Number of samples Outfall Name: SSW-28 Ocean Intake **Sampling Times** Sampling Dates Rcvd Rcvd SxDilution Water: Synthetic Lab Date Time Begin Date End Date Start End 08/22/25 13:26 08/20/25 08/20/25 10:53 10:53 Receiving Water Name: Dechlorinate Sample: Start Sx # 1 Date: _ 8/22/2025 Type of Test(s)Renew Sx # 1 Date: _ 8/23/2025 Americamysis bahia Chronic Renew Sx # ____1 Date: ___ 8/24/2025 Menidia beryllina Chronic Renew Sx # ____1 Date: ___ 8/25/2025 Renew Sx # ____1 Date: _ 8/26/2025 Renew Sx # ____1 ___ Date: ___ 8/27/2025 Controls: Synthetic Renew Sx # 1 Date: ___ 8/28/2025 pH Match: Test End Date: Test Start Date: Hardness Match: 8/29/2025 8/22/2025 Americamysis Test Set Up: 5 Reps & 5 Organisms per Rep Menidia beryllina Test Set Up: 3 Reps & 8 Organism per Rep Concentrations: 100 Test Chemistry on these dilutions: 100

Samples received by:				ODHL
	Federal Express	O the Client	O Bio-Aquatic personnel	
-				
Other:				

Hardness, Alkalinity, Residual Chlorine, Specific Conductivity, and Salinity Analysis Data

Client: Natural Energy Laboratory of Lab ID: 98036

Facility: Hawaii Ocean Science and Dilution Water(s): Synthetic Lab

Outfall: SSW-28 Ocean Intake Test Date: August 22, 2025

EFFLUENT PARAMETERS

Effluent	Recei		Residual	DeChlor	Ammonia	Analyst	Temp.
Sample #	Date	Time	Cl ₂ (mg/L)	$(ml/L)^1$	(mg/L)	Initials	Received
1	8/22/25	13:26	< 0.10	N/A	< 0.25	DT	3.1

¹**Dechlorination Reagent:** 0.025 N Sodium Thiosulfate

Effluent Sample #	рН	DO (mg/L)	Init. Salinity (ppt)	Adjusted Salinity	Analyst Initials
1	8.4	7.2	32.6	N/A	DT

pH, Dissolved Oxygen, Salinity

Chronic Americamysis bahia

Client: Natural Energy Laboratory of Hawaii Lab Number: 98036

Facility: Hawaii Ocean Science and
Outfall: SSW-28 Ocean Intake

Dilution Water(s): Synthetic Lab
Test Begin Date: August 22, 2025

NR indi	cates that tl	ne test wa	s not renev	wed		Concer	tration		
ANALYST	DATE	TIME	SX#	UNIT %	Control 100				
SG	8/22	Start 25 ± 1		pH DO (mg/L) Salinity (ppt)	8.1 8.3 7.1 6.9 21.5 34.7				
GJ	8/23	24 Hr 25 ± 1 Renew		pH DO (mg/L) Salinity (ppt) pH DO (mg/L) Salinity (ppt)	7.9 7.8 6.5 35.8 7.8 8.0 6.7 21.3 35.1				
CAP	8/24	48 Hr 25 ± 1 Renew		pH DO (mg/L) Salinity (ppt) pH DO (mg/L) Salinity (ppt)	7.7 7.8 7.0 6.4 21.9 35.9 7.9 8.0 7.1 6.6 30.4 34.0				
CCC	8/25	72 Hr 25 ± 1 Renew		pH DO (mg/L) Salinity (ppt) pH DO (mg/L) Salinity (ppt)	7.7				
JR	8/26	96 Hr 25 ± 1		pH DO (mg/L) Salinity (ppt) pH	8.0 8.1 6.4 7.2 24.6 32.3 8.0 8.0				
		Renew		DO (mg/L) Salinity (ppt)	6.5 7.0 24.3 35.8				
CCC	8/27	120 Hr 25 ± 1		pH DO (mg/L) Salinity (ppt) pH	7.9 7.9 6.9 6.2 30.7 37.7 8.0 8.1				
		Renew		DO (mg/L) Salinity (ppt)	6.8 6.8 36.6 32.5				
MM	8/28	144 Hr 25 ± 1 Renew		pH DO (mg/L) Salinity (ppt) pH DO (mg/L) Salinity (ppt)	8.0 8.0 6.9 6.9 34.1 32.5 8.1 8.1 7.0 7.0 27.1 32.5				
SG	8/29	168 Hr 25 ± 1	1	pH DO (mg/L) Salinity (ppt)	7.9 7.9 6.4 6.2 26.9 37.9				

pH, Dissolved Oxygen, Salinity

Chronic Menidia beryllina

Client: Natural Energy Laboratory of Lab Number: 98036

Facility: Hawaii Ocean Science and
Outfall: SSW-28 Ocean Intake

Dilution Water(s): Synthetic Lab
Test Begin Date: August 22, 2025

NR indicates that the test was not renewed

							Conce	ntration		
ANALYST	DATE	TIME	SX#	UNIT %	Control	100				
		Start		рН	8.1	8.3				
SG	8/22		1	DO (mg/L)	7.1	6.9				
		25 ± 1		Salinity (ppt)	21.5	34.7				
		24 Hr		рН	7.4	7.9				
		25 ± 1	1	DO (mg/L)	7.5	6.6				
GJ	8/23	23 ± 1		Salinity (ppt)	21.5	35.8				
		D	,	pH DO (mg/L)	7.8	8.0	l 			
		Renew	1	Salinity (ppt)	7.2	6.7 35.1				
		40.44		рН	7.8	7.8)			
		48 Hr	1	DO (mg/L)	7.0	6.4				
CAP	0.15.4	25 ± 1		Salinity (ppt)	21.2	35.1				
CAP	8/24	一		pН	7.9	8.0				
		Renew	1	DO (mg/L)	7.1	6.6				
				Salinity (ppt)	30.4	34.0				
		72 Hr		рН	7.7	7.7				
		25 ± 1	1	DO (mg/L)	7.0	6.3				
CCC	8/25	23 ± 1		Salinity (ppt)	21.6	35.9				
		Renew	,	pH DO (mg/L)	7.8 6.9	6.5				
		Kellew	1	Salinity (ppt)	23.6	34.2				
		96 Hr		рН	8.0	8.0				
		90 ПГ	1	DO (mg/L)	6.5	6.9				
JR	8/26	25 ± 1		Salinity (ppt)	24.3	34.2				
JK	8/20			pН	8.0	8.0				
		Renew	1	DO (mg/L)	6.5	7.0				
				Salinity (ppt)	24.3	35.8				
		120 Hr		pH PO (// T)	7.9	7.9				
		25 ± 1	1	DO (mg/L) Salinity (ppt)	24.2	36.5				
CCC	8/27			pH	8.0	8.1				
		Renew	1	DO (mg/L)	6.8	6.8				
				Salinity (ppt)	36.6	32.5				
		144 Hr		рН	8.0	8.1				
		\vdash	1	DO (mg/L)	6.9	6.8				
MM	8/28	25 ± 1		Salinity (ppt)	34.1	32.7				
141141				pH	8.1	8.1	 			
		Renew	1	DO (mg/L) Salinity (ppt)	7.0 27.1	7.0 32.5				\vdash
		\vdash	\square							
SG	8/29	168 Hr		pH DO (mg/L)	7.8 6.6	7.7 6.0		<u> </u>		\vdash
	0/27	25 ± 1	1	Salinity (ppt)	24.9	37.5				\vdash
		ш		-7 (FF-)		27.5			 	

Appendix B

Americamysis bahia

EUROFINS ENVIRONMENT TESTING BIO-AQUATICS

Carrollton, TX

REFERENCE TOXICANTS

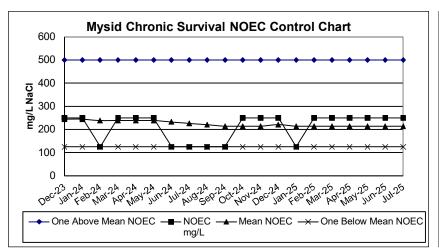
Eurofins Environment Testing Bio-Aquatics conducts reference toxicant testing monthly for organisms cultured in-house. For studies requiring purchased organisms, reference toxicant testing is performed simultaneously. Reference toxicant testing validates data and measures organism consistency. Only reagent grade chemicals are used of the following choices: sodium laurel sulfate (SLS), copper sulfate, copper chloride, potassium chloride, and sodium chloride. Organism responses are tracked with control charts for each reference toxicant/organism combination. The data are examined for sensitivity trends and to determine if results are within EPA described limits.

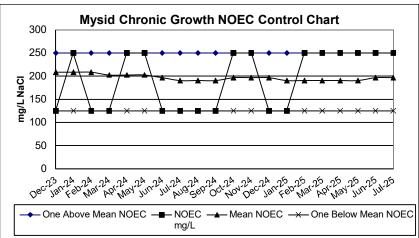
Standard Synthetic Saltwater

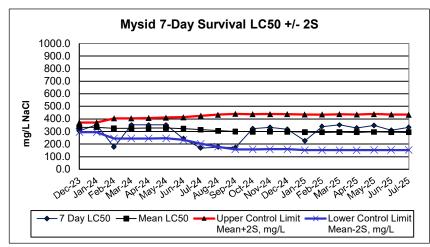
CHRONIC REFERENCE TOXICANT TEST RESULTS

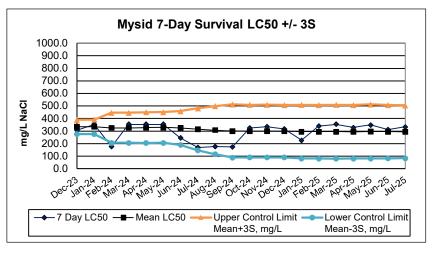
CHEMICAL:	Potassiu	ım Ch	loride				
DURATION:	7 Days						
TEST NUMBER:	169						
PROJECT NUMBER:	98122						
START DATE:	7/30/20	25					
START TIME:	14:08						
TOTAL NUMBER EXPOSED:	40 or	ganisr	ns per co	oncentra	ation		
CONCENTRATIONS (mg/L):	CON	25	50	125	250	500	1000
NUMBER DEAD PER CONCENTRATION:	0	1	0	1	3	40	40
TEST METHODS:	Method	1007	.0 As lis	ted in E	EPA-82	1-R-02	-014
STATISTICAL METHODS:	SURVI GROW FECUN	TH: A	NOVA	w/Dunr	nett's Te		
NOEC FOR SURVIVAL:	25	0	mg/L				
LOEC FOR SURVIVAL:	50	0	mg/L				
NOEC FOR GROWTH:	25	-	mg/L				
LOEC FOR GROWTH:	50	0	mg/L				
PMSD: 20.6							

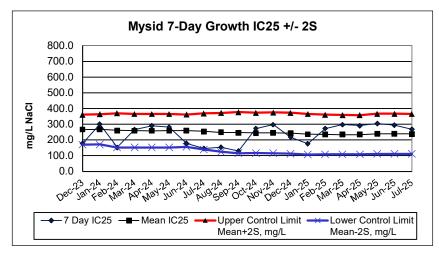
DILUTION WATER:

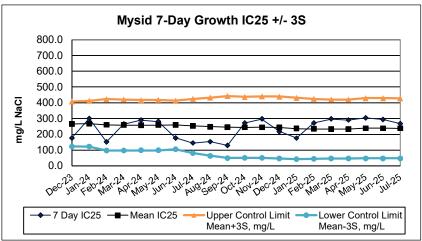












Appendix B

Menidia beryllina

EUROFINS ENVIRONMENT TESTING BIO-AQUATICS

Carrollton, TX

REFERENCE TOXICANTS

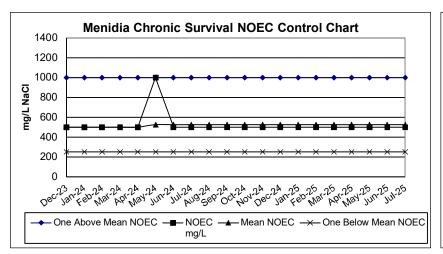
Eurofins Environment Testing Bio-Aquatics conducts reference toxicant testing monthly for organisms cultured in-house. For studies requiring purchased organisms, reference toxicant testing is performed simultaneously. Reference toxicant testing validates data and measures organism consistency. Only reagent grade chemicals are used of the following choices: sodium laurel sulfate (SLS), copper sulfate, copper chloride, potassium chloride, and sodium chloride. Organism responses are tracked with control charts for each reference toxicant/organism combination. The data are examined for sensitivity trends and to determine if results are within EPA described limits.

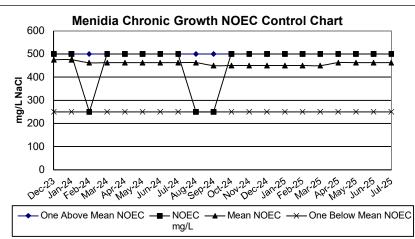
CHRONIC REFERENCE TOXICANT TEST RESULTS

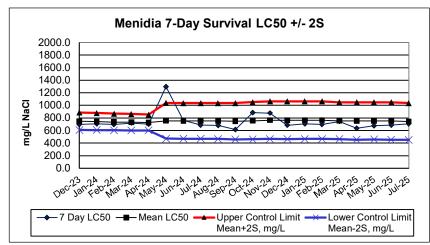
DILUTION WATER:	Standar	rd Synt	hetic Sa	ıltwate	r		
CHEMICAL:	Potassi	um Chl	oride				
DURATION:	7 Days						
TEST NUMBER:	169						
PROJECT NUMBER:	98123						
START DATE:	7/30/20)25					
START TIME:	13:40						
TOTAL NUMBER EXPOSED:	40 or	rganism	s per co	oncentr	ation		
CONCENTRATIONS (mg/L):	CON	125	250	500	1000	2000	4000
NUMBER DEAD PER CONCENTRATION:	0	0	0	0	40	40	40
TEST METHODS:	Method	1 1006.	0 As lis	ted in 1	EPA-82	1-R-02	-014
STATISTICAL METHODS:					ne Ran nett's Te		
NOEC FOR SURVIVAL:	50	00	mg/L				
LOEC FOR SURVIVAL:	10	00	mg/L				
NOEC FOR GROWTH:	50	00	mg/L				
LOEC FOR GROWTH:	10	00	mg/L				

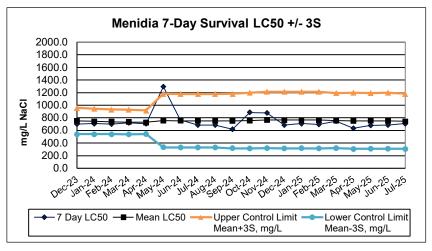
21.4

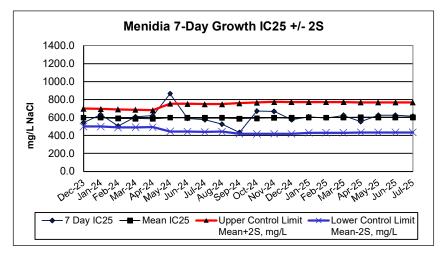
PMSD:

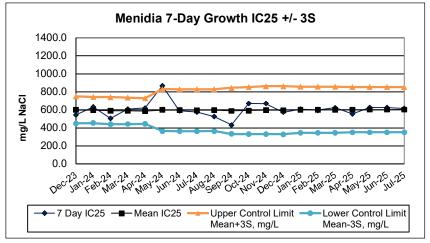












APPENDIX C

LITERATURE REFERENCES

- U.S.E.P.A., 2002. Short-Term Methods For Estimating The Chronic Toxicity Of Effluents And Receiving Water To Freshwater Organisms (Fifth Edition) U.S. Environmental Protection Agency, Office of Water, Washington D.C., EPA-821-R-02-012.
- U.S.E.P.A., 2002. Short-Term Methods For Estimating The Chronic Toxicity Of Effluents and Receiving Water To Marine And Estuarine Organisms (Third Edition) U.S. Environmental Protection Agency, Office of Water, Washington D.C., EPA-821-R-02-014.
- U.S.E.P.A., 2002. Short-Term Methods For Estimating The Chronic Toxicity Of Effluents And Receiving Water To Freshwater Organisms (Fourth Edition) U.S. Environmental Protection Agency, Office of Water, Washington D.C., EPA-821-R-02-013.
- U.S.E.P.A., 2012. Tropical Collector Urchin, *Tripneustes gratilla* (First Edition) U.S. Environmental Protection Agency, Office of Research and Development and Region 9, EPA-600-R-12-022.
- U.S.E.P.A., 1995. Short-Term Methods For Estimating The Chronic Toxicity Of Effluents And Receiving Water To West Coast Marine and Estuarine Organisms (First Edition) U.S. Environmental Protection Agency, EPA-600-R-95-136.
- U.S.E.P.A., 2010. National Pollutant Discharge Elimination System Test of Significant Toxicity Technical Document, U.S. Environmental Protection Agency, Office of Wastewater, Washington D.C., EPA-833-R-10-004.
- U.S.E.P.A., 1991. Technical Support Document For Water Quality-Based Toxics Control, U.S. Environmental Protection Agency, EPA-505-2-90-001.
- Zarr, Jerrold, H., 1984. Biostatistical Analysis, (Second Edition). Prentice-Hall, Inc., Englewood Cliffs, N.J.

CHAIN-OF-CUSTODY SHEETS

Appendix D

1	BIG	D-AQU	BIO-AQUATIC TESTING, INC.	TING, IN	<u>ن</u>	CHAII	NOF	CHAIN OF CUSTODY		Bio Only: No Sample Left	Lab id:	98036
	CAF CAF	1 MAYES ROLLTO	2501 MAYES RD., S1E. 100 CARROLLTON, TX 75006		Please	Review &	Complete	Please Review & Complete Sections A, B, C, &	A, B, C, & [D. S	Sample No:	98036 - Effective Dule 97302017
Rend		9/2-242-/	PH: 9/2-242-//50 FAX: 9/2-242-//49	2-242-1149	Che	Check Sample No. :		First,Secor	Second, orTh	Third. P.O	P.O. No:	
Client:	Natural Energy Laboratory of Hawaii	ergy Labo	oratory of H	awaii		9	1		0 0) 1 - E	.; (V)	
Facility:	Hawaii Ocean	an Science	ice and Technology	hnology P	Park Use	e area ber	ow to make	area below to make changes, in	If the Schedu	s)ısəl pəir	scheduled lest(s) in A are incorrect:	correct:
Permit No: N/A	M/A						LIESTIM	riesiiwatei opecies			Sallwale	Saltwater Species
Onttall :	SSW-28 Ocean Intake	cean Inta	ke					ejas Liea)		(əɐɓ) ພານງ:	enilly (wc	
Client Contact:	ontact:				t date grapit	nd :	еш	ater	ouuju		luuju Vuoju	lopis mind
Client Phone:	ione:				?M) Э	<u> </u>		<u></u>				
NEW PRESE	REVIEW SCHEDULED		TEST(s):		□Chronic □96 Hour	Chronic Chour		□Chronic □C	□Chronic □	☐96 Hour	□Chronic □96 Hour	☐Chronic ☐96 Hour
Chronic	1	Americamysis bahia	is bahia		□48 Hour		_			724 Hour	□48 Hour	148 Hour
Chronic		Menidia beryllina	ıryllina	To Ship the		\exists	_	□24 Hour □24	□24 Hour		□24 Hour	□24 Hour
				1st Sample on:		1-Routine/Spe	cialty Testing	Notes: Non-Routine/Specialty Testing for Information purpose	nrpose			
Concentration:	ation: 100	The second secon		7/21/2025	- 5/4							
5 3	1X) Setup separate 24hr Acute Test?	Acute Test?	No		EA.							
ට of 35	S. S	Sample Type: E = Effluent		Sample Date	Sam (m	Sample Time (military)	Grab		Sam	Sampled By:		Number Of
Outfall I		S = Rec. Stream S = Sediment	From	To	From	<u>و</u>	Composite		(Sign and	(Sign and Print Name)	ne)	Shipped
1C,SW.	Statu.		8/20/25		1083	١	6	Bur Madler	de B	MM	Noce	
2		1000)		7			/
က				- Vare								
Ö.	Relinquished By:	i By:		Date	Time			Received Bv:			Date	Time
The Charle	Maer			8/20/2	1230	Tea	Federa					
> ∧l	We consider the second						~	Ì		8	872.25	1326
ෆ atic La												
	Bio-Aquatic Sample Login	Login	BAT sample personnel: O Yes O No		Date: 8-27.27	. Time:/326	326	By: D7		Temperature: 3	/	(C) IR#: 026
8036			Dechlorinate Sample:		Chlorine: 🟒 , 🚶	mg/I An	Ammonia: 🂪	oll mg	mg/l Int. Sal\Cond: $\Im \mathcal{Z}_{\mathcal{L}}$		ppt/uS Adj. Salinity	ity ppt
			Dilution	ater:	, ,	H	Hardness:	l/gm	Other			
			Synthetic Lab	ab	DO: 7,7	mg/I All	Alkalinity:	l/gm	Condition: 9 00 d	dood		



Bio-Aquatic Testing, Inc.



Report Revision Form

Report Revision Number 0 for Lab ID 98036 was revised on 09/11/2025 .
The revision was issued for the following reason(s):
☐ Typo in the report document or tables
☐ Missing sheets or tables
☐ Hard data was not scanned in as required by the client
☐ Missing specially requested forms or data for the client
Other (Please Specify):
Updated sampling information

FORM 10.6 Revision 1 Effective: 08/07/2018