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CITY OF SANTA ANA

PUBLIC WORKS AGENCY
20 Civic Center Plaza • P.O. Box 1988
Santa Ana, California 92702
www.santa-ana.org

January 18, 2022

Mr. Warren Teitz
Metropolitan Water District of Southern California
FSAfundingprogram@mwdh20.com

RE: Future Supply Actions Funding Program Final Report for *Restoration of Local Recharge Sources from Invasive Dreissenid Mussels*

Dear Mr. Teitz:

This final report on the *Restoration of Local Recharge Sources from Invasive Dreissenid Mussels* provides a comprehensive summary of the project efforts, findings, and lessons learned. During the period of May 2019 through September 2021, the project team completed coordination, planning, and implementation of five rounds of testing with EarthTec QZ for both 1) quagga veliger dose-response and 2) non-target acute toxicity from three project test locations.

A total of $122,773.84 was disbursed by Metropolitan Water District of Southern California as part of the cost-sharing arrangements for the Future Supply Actions Funding Program for a total of $245,547.65 incurred by the project team for work completed between May 2019 through September 2021.

I am informed and believe that the information contained in this report is true and that the supporting data is accurate and complete.

Sincerely,

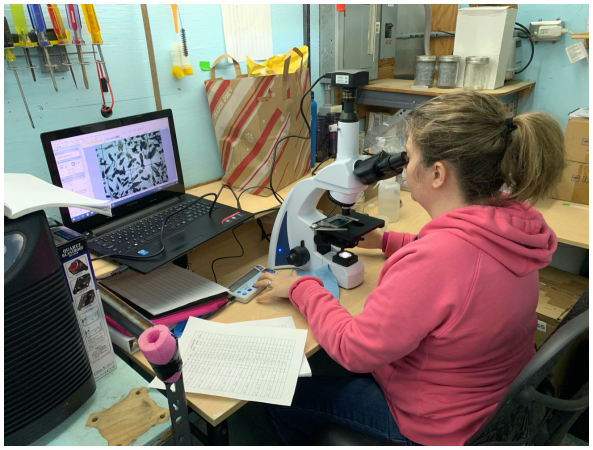
Nabil Saba, P.E.
Executive Director
City of Santa Ana Public Works Agency

SANTA ANA CITY COUNCIL

Vicente Sarmiento, Mayor, vsarmiento@santa-ana.org
Phil Bacerra, Mayor Pro Tem, Ward 4, pbacerraa@santa-ana.org
Thai Viet Phan, Ward 1, tphan@santa-ana.org
David Penaloza, Ward 2, dpenaloza@santa-ana.org
Jessie Lopez, Ward 3, jessielopez@santa-ana.org
Johnathan Ryan Hernandez, Ward 5, jryanhernandez@santa-ana.org
Nelida Mendoza, Ward 6, nmendoza@santa-ana.org

Final Report

Restoration of Local Recharge Sources from Invasive Dreissenid Mussels



Prepared for:

Metropolitan Water District of Southern California

City of Santa Ana

Orange County Water District

January 2022

KASF Consulting

Trussell TECHNOLOGIES INC

Prepared for: Metropolitan Water District of Southern California
City of Santa Ana
Orange County Water District

Copies to: Water Replenishment District of Southern California
United Water Conservation District
Main San Gabriel Basin Watermaster
Central Basin Municipal Water District
California Department of Fish and Wildlife
California Department of Water Resources

Authors: Kelly Stockton-Fiti, KASF Consulting
Emily Owens-Bennett, P.E., Trussell
Liana Olivas, Trussell
Chao-Chun Yang, Trussell

Reviewers: David Hokanson, Ph.D., P.E., BCEE, Trussell
Rhodes Trussell, Ph.D., P.E., BCEE, Trussell
Adam Hutchinson, PG, CHG, OCWD
Christine Pham, OCWD
Megan H. Plumlee, Ph.D., P.E., OCWD

Table of Contents

List of Figures

List of Tables

1 Executive Summary

Invasive quagga mussels (*Dreissena bugensis*) have impacted critical imported water systems in Southern California, which restricts the beneficial uses of affected reservoirs, rivers, streams, and recharge basins. A study entitled "Restoration of Local Recharge Sources from Invasive Dreissenid Mussels" (Study) was conducted to evaluate the efficacy of the molluscicide EarthTec QZ™ (active ingredient is copper sulfate pentahydrate) in controlling the veliger stage of quagga mussels, which is the larval stage when the quagga mussels are mobile. For the remainder of this report, quagga mussel veligers will be referred to as veligers. Veliger mortality was evaluated via bench-scale testing using veligers collected from three Southern California locations identified in Table 1-1 – Lake Piru, Lake Mathews, and Weymouth Water Treatment Plant (WTP). Lake Piru is a mix of State Water Project water and local run-off and Lake Mathews is Colorado River Aqueduct water. The Weymouth WTP receives untreated water from two sources: Colorado River water from Lake Mathews via the Upper Feeder and State Water Project (SWP) water from the East Branch of the SWP via the Rialto Pipeline or from the Upper Feeder via the Etiwanda Pipeline from the Rialto Pipeline. During this study, sampling at the Weymouth WTP was during periods of exclusive delivery of Colorado River water from Lake Mathews that had been chlorinated prior to entering the Upper Feeder. The travel time from Lake Matthews to the Weymouth WTP is approximately 8 hours. The same experimental conditions were used for each of the locations: a range of EarthTec QZ concentrations and exposure times at two different testing temperatures.

Table 1-1. Sampling site location and owner

Sampling Site	Location	Owner
Lake Piru	Ventura County, CA	United Water Conservation District
Lake Mathews	Riverside County, CA	Metropolitan Water District of Southern California
Weymouth WTP	La Verne, CA	Metropolitan Water District of Southern California

EarthTec QZ was able to produce 100% veliger mortality; however, several variables contributed to the efficacy of EarthTec QZ on veliger mortality, including EarthTec QZ dose, exposure duration, sampling location, and site-specific water quality. Veliger mortality was found to increase with both EarthTec QZ dose and exposure time. The veligers collected from Weymouth WTP exhibited the highest response to EarthTec QZ at lower concentrations and shorter exposure times, compared to the same conditions tested at Lake Mathews and Lake Piru. This is likely due to the cumulative exposure to chlorine and EarthTec QZ for the veligers collected from Weymouth WTP.

The toxicity of EarthTec QZ to veligers from each test location was compared to that of non-target indicator organisms (water flea, rainbow trout, or fathead minnow) from all locations using a time-dose variable representing total exposure (Figure 1-1), presented in terms of hours x mg/L of copper. Non-target organisms were exposed to EarthTec QZ for 96 hours, following standard test methods for measuring the acute toxicity of

effluents and receiving water to freshwater and marine organisms (EPA-821-R-02-012, 2002; EPA/600/4-90/027F, 1993). Water fleas and rainbow trout were very sensitive to EarthTec QZ and fathead minnows were the least susceptible. Veligers had similar sensitivity to EarthTec QZ as the water fleas and rainbow trout.

The Study results can be applied to other Southern California waterbodies with high hardness values and similar alkalinity and DOC levels. Since variations of mortality results were observed, which were attributed to water quality parameters, it is recommended that site-specific field assessments be conducted to assess the appropriate dose and exposure time to achieve the desired mortality. During field treatment, veliger mortality should be monitored along with relevant water quality parameters – hardness, alkalinity, dissolved organic carbon, pH, dissolved oxygen, temperature, and ions. To limit exposure of EarthTec QZ and resulting toxicity to non-target aquatic organisms, it is ideal to identify treatment configurations that allow for long exposure times to veligers with a low dose of EarthTec QZ (e.g., within an aqueduct or pipeline) prior to discharge to a lake, river, or stream.

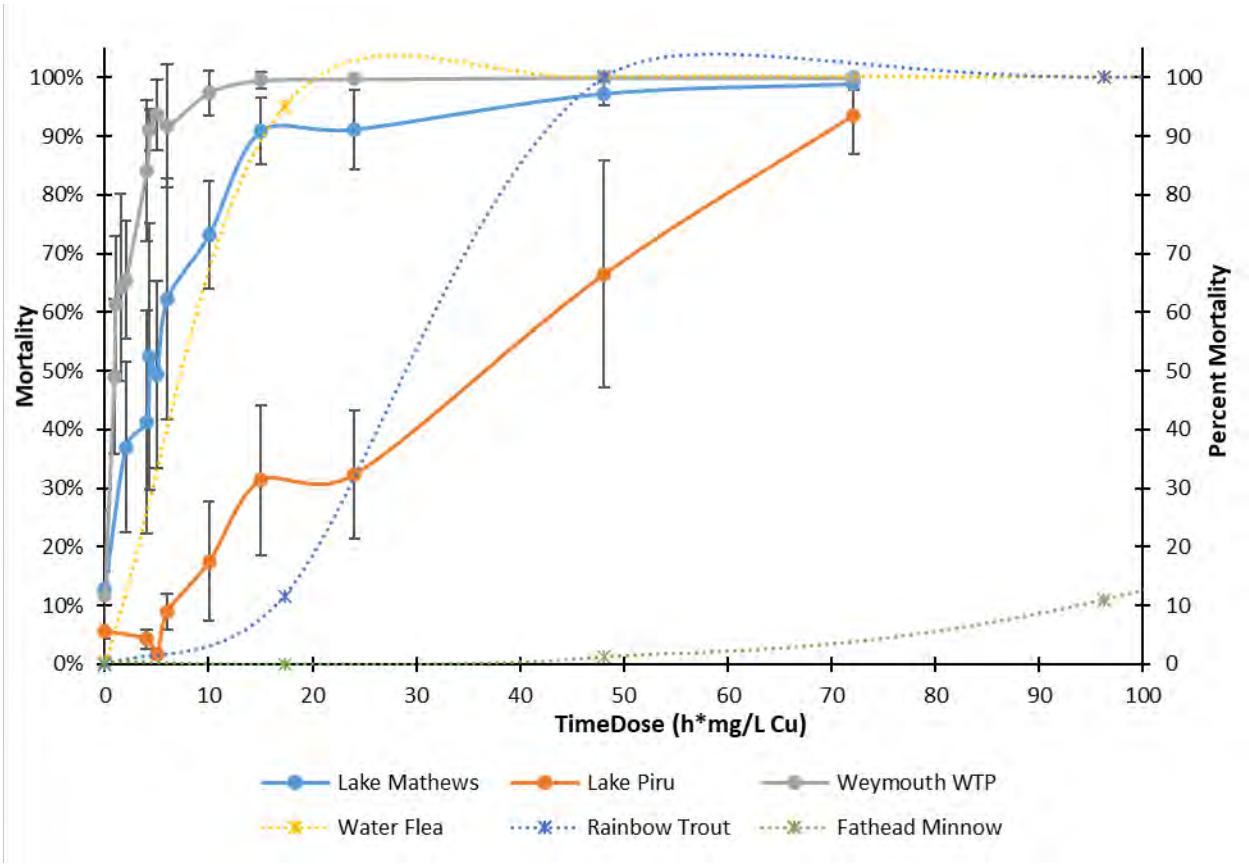


Figure 1-1. Mortality with cumulative exposure to EarthTec QZ (hours x mg/L copper)
For veligers by location, plus water flea, rainbow trout, and fathead minnow (non-target indicator results for all locations combined).

Key lessons learned from the Study include the following:

- Although a prior study with veligers exposed to EarthTec QZ in water from Lake Mead found 100% mortality at 3 µL/L EarthTec QZ with 30 minutes of exposure time, the 3 µL/L EarthTec QZ dose did not result in 100% veliger mortality with any of the southern California waters tested as part of this Study, even after 24 hours of exposure time.
- No tested concentration of EarthTec QZ achieved 100% veliger mortality with an exposure duration of 30 minutes.
- The use of a 24-hour recovery period, during which veligers were exposed to clean water following each tested exposure duration with EarthTec QZ, allowed for improved determination of veliger mortality.
- Lower doses of EarthTec QZ are safer for non-target aquatic organisms but require longer contact time to achieve high veliger mortality.

2 Introduction

The communities of Southern California rely heavily on water supplied from the Colorado River Aqueduct (CRA) and California State Water Project (SWP). These vital water supplies are conveyed hundreds of miles from their sources and stored within a network of reservoirs. Since 2007, invasive quagga mussels (*Dreissena bugensis*) have spread from Lake Mead in Nevada through the CRA system to many California lakes, reservoirs, and watersheds that receive raw water from the CRA. MWD implements quagga mussel controls on the CRA system, including chlorination, desiccation, and physical removal. Quagga mussels in the SWP system have been more limited, to-date, with detections in Pyramid Lake, Castaic Lake, and Lake Piru, as well as downstream rivers and streams.

The presence of quagga mussels in imported water systems threatens the beneficial uses of the reservoirs, rivers, streams, and recharge basins. Key uses include providing habitat for aquatic species, recharging groundwater with local and imported water, storing local runoff, and facilitating flood control. The California Department of Fish and Wildlife (CDFW) requires impacted facilities to implement control strategies and complete monitoring to prevent the spread of invasive quagga mussels. Desiccation (allowing a basin or stream bed to dry) is the primary control method for quagga mussels, but this strategy can only be implemented on a small scale. Without a viable quagga mussel control strategy for rivers and reservoirs, the use of the imported water systems for valuable groundwater replenishment has been restricted.

This Study, entitled “Restoration of Local Recharge Sources from Invasive Dreissenid Mussels” was conducted to evaluate the efficacy of the molluscicide EarthTec QZ (active ingredient is copper sulfate pentahydrate) for controlling quagga veligers (veligers for the remainder of this report), which are the larval stage of the mussels. Five rounds of bench-scale dose-response testing were completed between December 2019 and July 2021 using veligers collected from three Southern California locations impacted by quagga mussels representing distinct water qualities (SWP with local

runoff, CRA, and combined impact of CRA water with pipeline chlorine treatment). For each round of testing, veligers were exposed to five concentrations of EarthTec QZ at two temperatures and mortality was evaluated over a range of hold times. This robust testing matrix allowed for comparison of veliger mortality based on EarthTec QZ concentration, exposure time, cumulative time-dose exposure, veliger size, and site-specific water quality.

In addition to assessing veliger mortality with exposure to EarthTec QZ, the Study also measured residual free and total copper for all test conditions along with other water quality parameters. Acute toxicity of EarthTec QZ to three non-target aquatic indicator species was also evaluated. Limiting the EarthTec QZ dose and residual total copper concentration is important if this treatment strategy is going to be viable for use on a larger scale. Residual copper levels must be compatible with the United States Environmental Protection Agency (USEPA) regulations for copper in drinking water, which include an action level of 1.3 mg/L as Cu and a secondary maximum contaminant level (sMCL) of 1.0 mg/L as Cu. The toxicity of EarthTec QZ in aquatic organisms is also an important consideration, as the imported water systems, associated reservoirs, and potential conveyance systems for recharge provide habitat for a variety of aquatic organisms.

This Final Report provides an overview of the Study efforts, summarizes results and key findings, and puts these findings in context by identifying lessons learned and potential next steps. In addition, this document compares the costs incurred and funds distributed to support the Study with the planned budget. Finally, the project schedule is discussed, along with adjustments made over the course of the Study, and the tasks accomplished.

Overall, EarthTec QZ was found to be effective in controlling veligers with exposure times of 24 hours or less. The efficacy of this treatment method varied as a function of the EarthTec QZ dose concentration, exposure duration, and site-specific water quality. Transitioning from the proof-of-concept bench-scale testing approach adopted for the Study, it is recommended to complete additional site-specific testing to optimize EarthTec QZ dosing and exposure times based on available full-scale treatment configurations (e.g., pipeline, basin, tank, lake).

The Study was conducted by Trussell Technologies (Trussell), KASF Consulting, and Orange County Water District (OCWD). Funding was provided by the Metropolitan Water District of Southern California (MWD) Future Supply Actions Funding Program, along with project partners including OCWD, the Water Replenishment District of Southern California (WRD), the United Water Conservation District (UWCD), the Main San Gabriel Basin Watermaster, and the Central Basin Municipal Water District (CBMWD). Additional technical assistance and support were provided by CDFW and the California Department of Water Resources (DWR). As a member agency of MWD, the City of Santa Ana was involved as a partner with OCWD and thus the FSA funding agreement with MWD was between MWD and the City of Santa Ana.

3 Cost Summary

Cumulative costs and funds disbursed (May 2019 through September 2021) associated with the original Study scope of work (testing at 4 locations) are summarized by invoice and task in Table 3-1. All project costs that are reported in Table 3-1 were incurred by Trussell and project partner KASF Consulting. The FSA funds were dispersed from MWD to the City of Santa Ana, then to the project team, and the totals are summarized in Table 3-1.

The originally planned budget that was funded through the FSA Funding Program with additional financial support from the aforementioned project partners included a single round of testing at four different locations. Challenges with scheduling in the first half of 2020 related to Covid-19 restrictions precluded sampling from one of the original test locations, OCWD's OC-28 turnout. The project team met with MWD to identify an alternative sampling location with representative water quality (chlorinated water from Lake Mathews) and Weymouth WTP was determined to be the most appropriate location. Both Weymouth WTP and OC-28 receive water from Lake Matthews with nearly the same travel time. As a result, both locations are essentially equivalent having equal contact times with the chlorine dosage. Since Weymouth WTP was already one of the project test locations, it was decided to complete another round of testing at both Weymouth WTP and the upstream Lake Mathews location in 2021. This resulted in five rounds of testing overall, which would allow for investigation of seasonality variability and variability of the quagga veliger size distribution. The fifth round of testing was funded separately by OCWD, WRD, Main San Gabriel Basin Watermaster and CBMWD, without additional funding from the FSA Funding Program; the associated costs from the fifth round of testing are thus not reflected in Table 3-1.

Table 3-1. Summary of project costs incurred and FSA funds dispersed

Cost Incurred and Funds Disbursed Thoughout the Duration of the Study																						
Task No.	Cost Estimate from FSA Agreement between City of Santa Ana and Metropolitan		Invoice No. 18928001Rev3 (2019)		Invoice No. 18928002Rev2 (2020 Q1)		Invoice No. 18928003 (2020 Q2)		Invoice No. 18928004 (2020 Q3)		Invoice No. 18928005 (2020 Q4)		Invoice No. 18928006 (2021 Q1)		Invoice No. 18928007 (2021 Q2)		Invoice No. 18928008 (2021 Q3)		Invoice Totals		Invoice No. 18928009 Retention	Total Funds Disbursed (Invoice Totals plus Retention)
	Total Study Cost	Requested Metropolitan Funding Match	Costs Incurred	Funds Disbursed	Costs Incurred	Funds Disbursed	Costs Incurred	Funds Disbursed	Costs Incurred	Funds Disbursed	Costs Incurred	Funds Disbursed	Costs Incurred	Funds Disbursed	Costs Incurred	Funds Disbursed	Costs Incurred	Funds Disbursed	Costs Incurred	Funds Disbursed	Funds Disbursed	
Task 1: Evaluation of EarthTec QZ Veliger Treatment	$154,180	$77,090	$71,039.09	$35,519.55	$6,504.82	$3,252.41	$500.00	$250.00	$36,935.93	$18,467.97	$3,747.09	$1,873.55	$9,352.50	$4,676.25	$200.00	$100.00	$25,865.72	$12,932.86	$154,145.15	$77,072.59		$77,072.59
Task 2: Toxicity Assessment	$62,228	$31,114	$19,364.56	$9,682.28	$1,910.00	$955.00	$0.00	$0.00	$10,509.94	$5,254.97	$12,890.00	$6,445.00	$12,520.00	$6,260.00	$540.00	$270.00	$4,492.50	$2,246.25	$62,227.00	$31,113.50		$31,113.50
Task 3: Project Management	$29,184	$14,592	$16,062.50	$8,031.25	$4,778.00	$2,389.00	$5,155.00	$2,577.50	$2,910.00	$1,455.00	$0.00	$0.00	$270.00	$135.00	$0.00	$0.00	$0.00	$0.00	$29,175.50	$14,587.75		$14,587.75
Subtotals	$245,592	$122,796	$106,466.15	$53,233.08	$13,192.82	$6,596.41	$5,655.00	$2,827.50	$50,355.87	$25,177.94	$16,637.09	$8,318.55	$22,142.50	$11,071.25	$740.00	$370.00	$30,358.22	$15,179.11	$245,547.65	$122,773.84		$122,773.84
Retention @ 25%				-$13,308.27		-$1,649.10		-$706.88		-$6,294.48		-$2,079.64		-$2,767.81		-$92.50		-$3,794.78		-$30,693.46	$30,693.46	$0.00
Totals	$245,592	$122,796	$106,466.15	$39,924.81	$13,192.82	$4,947.31	$5,655.00	$2,120.62	$50,355.87	$18,883.46	$16,637.09	$6,238.91	$22,142.50	$8,303.44	$740.00	$277.50	$30,358.22	$11,384.33	$245,547.65	$92,080.38	$30,693.46	$122,773.84

Note: Reported costs and anticipated disbursement for Invoice Nos. 18928005 through 18928009 have been submitted and are still under review as of the date of this Final Report, but are not anticipated to change.
Invoice No. 18928002Rev2 Costs Incurred for Task 1 includes a $0.18 deduction due to a prior overcharge in Invoice No. 18928001Rev3.
Invoice No. 18928005 Costs Incurred for Task 1 includes a $32.91 deduction due to a prior overcharge in Invoice No. 18928004.

4 Schedule Summary

This collaborative Study involved multiple meetings with various stakeholders, planning efforts for the veliger and non-target toxicity testing, permitting for collection of invasive quagga mussels, field sampling and testing, as well as reporting. A summary of the main project efforts and deliverables is provided in Table 4-1.

Table 4-1. Summary of project efforts by task

Project Effort	Timing
Task 1 Evaluation of EarthTec QZ Veliger Treatment	
Veliger Dose-Response Test Plan	Q2 2019
CDFW Scientific Collecting Permit Approval	Q4 2019
Lake Piru Testing	Q4 2019
Lake Mathews Testing, Part 1	Q3 2020
Weymouth WTP Testing, Part 1	Q3 2020
Draft Technical Memorandum 1	Q4 2020
Lake Mathews Testing, Part 2	Q3 2021
Weymouth WTP Testing, Part 2	Q3 2021
Final Technical Memorandum 1	Q3 2021
Task 2 Toxicity Assessment	
Non-Target Toxicity Test Plan	Q2 2019
Lake Piru Testing	Q4 2019
Lake Mathews Testing, Part 1	Q3 2020
Weymouth WTP Testing, Part 1	Q3 2020
Draft Technical Memorandum 2	Q4 2020
Lake Mathews Testing, Part 2	Q3 2021
Weymouth WTP Testing, Part 2	Q3 2021
Final Technical Memorandum 2	Q3 2021
Combined Task 1 and 2 Efforts	
Project Kickoff Meeting	Q2 2019
Stakeholder Review Meeting (Internal)	Q2 2019
FSA Funding Agreement Meeting	Q1 2020
Semi Annual Progress Report 2019 No. 1	Q1 2020
Semi Annual Progress Report 2020 No. 1	Q3 2020
Semi Annual Progress Report 2020 No. 2	Q1 2021
Draft Final Report	Q3 2021
Final Report	Q4 2021
Task 3 Project Management	
Webinar Presentation of Final Report	Q4 2021

The Study schedule is provided in Table 4-2. Yellow shading represents the original schedule established in the Final Agreement between MWD and the City of Santa Ana,

and blue shading (or cross hatching where the original and actual schedules coincided) represents the actual timing of project activities.

A Specific Use Scientific Collecting Permit is required by CDFW prior to collecting, transporting, or using quagga veligers in any tests. This effort was not anticipated as part of the original project planning, and testing was delayed until the permit was secured. The project team worked with CDFW to apply for this permit and provide the supporting documentation. The permit was finalized in November 2019, and the first round of testing at Lake Piru was completed in December 2019.

As discussed in Section 3, testing planned for Lake Mathews, Weymouth WTP, and OCWD's OC-28 turnout in quarter 2 of 2020 was delayed due to Covid-19 restrictions. The OC-28 turnout location was unavailable in fiscal year 2021, and the project team worked with MWD to identify Weymouth WTP as the most appropriate surrogate location for OC-28. Repeating the testing at Weymouth WTP was expected to also introduce some seasonal variability in the veliger size and water quality. Considering the benefit of the repeat testing, as well as the desired correlation of the veliger dose-response for Weymouth WTP with the upstream, unchlorinated water from Lake Mathews, OCWD, WRD, Main San Gabriel Basin Watermaster and CBMWD funded one additional round of testing at Lake Mathews in July 2021. These additional 2021 testing efforts delayed the development of the Final Report.

Table 4-2. Study schedule

The original plan is indicated in yellow, with actual timing indicated in blue; where these period coincide, the shading is striped.

Task	Task Name	Jan-Mar	Apr-Jun	Jul-Sep	Oct-Dec	Jan-Mar	Apr-Jun	Jul-Sep	Oct-Dec	Jan-Mar	Apr-Jun	Jul-Sep	Oct-Dec
		2019				2020				2021			
		Q1	Q2	Q3	Q4	Q1	Q2	Q3	Q4	Q1	Q2	Q3	Q4
1	Collect and compile available data												
1	Develop Draft Veliger Dose Response Test Plan												
1	Stakeholder Review Meeting 1												
1	Finalize Veliger Dose Response Test Plan												
1	Obtain Scientific Collecting Permit from CDFW												
1	Conduct Field Sampling/Dose Response Testing												
1	Compile Results												
1,2	Semi-Annual Progress Report 2019 No. 1 (May 2019 – December 2019)												
1	Conduct Additional Sampling (If needed)												
1,2	Semi-Annual Progress Report 2020 No. 1 (January 2020 – June 2020)												
1	Compile Season 2 Results (If needed)												
1	Stakeholder Review Meeting 2 (If needed)												
1	Prepare Draft Technical Memorandum 1												
1	Review Draft Technical Memorandum 1												
1	Submit Final Technical Memorandum 1												
1,2	Semi-Annual Progress Report 2020 No. 2 (July 2020 – December 2020)												
2	Develop Toxicity Test Plan												
2	Stakeholder Review Meeting 1												

Task	Task Name	Jan-Mar	Apr-Jun	Jul-Sep	Oct-Dec	Jan-Mar	Apr-Jun	Jul-Sep	Oct-Dec	Jan-Mar	Apr-Jun	Jul-Sep	Oct-Dec
		2019				2020				2021			
		Q1	Q2	Q3	Q4	Q1	Q2	Q3	Q4	Q1	Q2	Q3	Q4
2	Finalize Toxicity Test Plan												
2	Conduct Toxicity Testing												
2	Compile Results												
2	Conduct Additional Testing (If needed)												
2	Compile Season 2 Results (If needed)												
2	Stakeholder Review Meeting 2 (If needed)												
2	Prepare Draft Technical Memorandum 2												
2	Review Draft Technical Memorandum 2												
2	Submit Final Technical Memorandum 2												
1,2	Draft Final Report												
1,2	Final Report												
3	Symposium Presentation of Study Findings												
3	Project Management												

5 Study Results and Analysis

Three locations, indicated in Figure 5-1, were sampled over a two-year period of December 2019 to July 2021: Lake Piru, Lake Mathews, and Weymouth WTP. Lake Piru is managed by UWCD. This reservoir, located in Ventura County, CA, impounds storm flows from the Piru Creek watershed and supplemental imported SWP water supplied by DWR via Pyramid Lake. Lake Mathews, a reservoir located in Riverside County, CA owned and operated by MWD, is primarily used to store CRA water. Water from Lake Mathews is delivered into raw water pipelines that are dosed with chlorine for quagga mussel control. One location that receives chlorinated Lake Mathews water is MWD's Weymouth WTP which is connected to the Upper Feeder pipeline (Figure 5-1). Testing was completed on samples from a) Lake Piru in December 2019, b) Lake Mathews in September 2020 and July 2021, and c) Weymouth WTP in September 2020 and June-July 2021.

Figure 5-1. Map of Study sample locations in Southern California

5.1 Susceptibility of Quagga Veligers to Earth Tec QZ

Live quagga veligers and raw water were collected from each aforementioned location. Subsets of the collected veligers were placed in beakers with raw water for testing a range of EarthTec QZ doses, various exposure times, and two different temperatures (15°C and 20°C). The same test conditions were repeated using water from each location. The range of EarthTec QZ (ET) doses tested include 0, 3.0, 16.7, 33.4, 50.1 µL/L (equivalent to 0, 0.18, 1.0, 2.0, and 3.0 mg/L as Cu). Copper ions are the active ingredient in EarthTec QZ that cause veliger mortality. Veliger mortality was evaluated with the Fast Green method (Stockton-Fiti and Claudi 2017) after 0.5, 2, 5, and 24 hours of exposure time (i.e., end mortality) for each EarthTec QZ dose and temperature condition. Directly following this count at the end of the exposure time, the veligers were placed in a beaker and exposed to raw sample water (without the addition of EarthTec QZ) for a 24-hour recovery period. A final mortality assessment was made at the end of the recovery time to allow for analysis of delayed mortality. The complete set of quagga veliger dose-response results from each location is summarized in a separate Technical Memorandum 1 (TM1) in Appendix A. The discussion in this Final Report is focused on significant trends and findings from TM1.

In general, veliger mortality increased with EarthTec QZ dose at all sampling locations. Veliger mortality increased with exposure time to EarthTec QZ (see TM1 in Appendix A for individual site results). Recovery mortality results for all three locations were greater

than the corresponding end mortality assessment for each condition. Considering the objective of the study to characterize the impact of EarthTec QZ treatment on the quagga veligers, the recovery results are displayed in this Final Report; all data can be found in TM1 (Appendix A). Statistical analysis showed that EarthTec QZ concentration, exposure duration, and location were significant variables. Recovery mortality results for veliger dose-response test conditions are presented in Figure 5-2.

Weymouth WTP veligers were the most susceptible to EarthTec QZ, exhibiting higher mortality in shorter times at lower concentrations (Figure 5-2) when compared with the corresponding results from Lake Piru and Lake Mathews. The results for the 16.7 µL/L EarthTec QZ dose (1.0 mg/L as Cu) test condition (see Figure 5-2 part B) are the main point of reference for assessing the efficacy of EarthTec QZ as a viable treatment strategy. This corresponds to the established drinking water limits for copper of 1.0 mg/L as Cu. In the Weymouth WTP tests, greater than 85% mortality was achieved at the 5 hours of exposure at 16.7 µL/L EarthTec QZ. Mortality increased with exposure time and concentration beyond this condition. Veligers from Lake Mathews were not as susceptible to EarthTec QZ: greater than 70% mortality after 24-hour exposure time at 16.7 µL/L EarthTec QZ (1.0 mg/L as Cu). Lake Piru only achieved approximately 30% mortality at the 24-hour exposure time at 16.7 µL/L EarthTec QZ (1.0 mg/L as Cu). The two higher concentrations tested, 33.4 and 50.1 µL/L EarthTec QZ, exhibited higher mortality and quicker response (Figure 5-2 parts C and D).

Control condition veliger mortality was usually less than 20% with two exceptions for 24-hour recovery (Figure 5-2, part A): Lake Piru at 20°C and Lake Mathews 2020 at 20°C. The control showed that there is a baseline mortality, which can increase with time for batch tests. Holding the veligers in beakers for 48 hours contributes to mortality regardless of EarthTec QZ dosing, so longer duration studies are not advisable without flowing water. The lowest dose tested was 3 µL/L EarthTec QZ. Although this condition achieved high mortality (>85%) in the Weymouth WTP samples at 24 hours of exposure, the Lake Mathews and Lake Piru samples did not exhibit significant increased mortality when compared with the control. For this reason, the 3 µL/L EarthTec QZ results are not presented in Figure 5-2. Additional discussion of the 3 µL/L EarthTec QZ experimental results is provided in Section 6 (Figure 6-1).

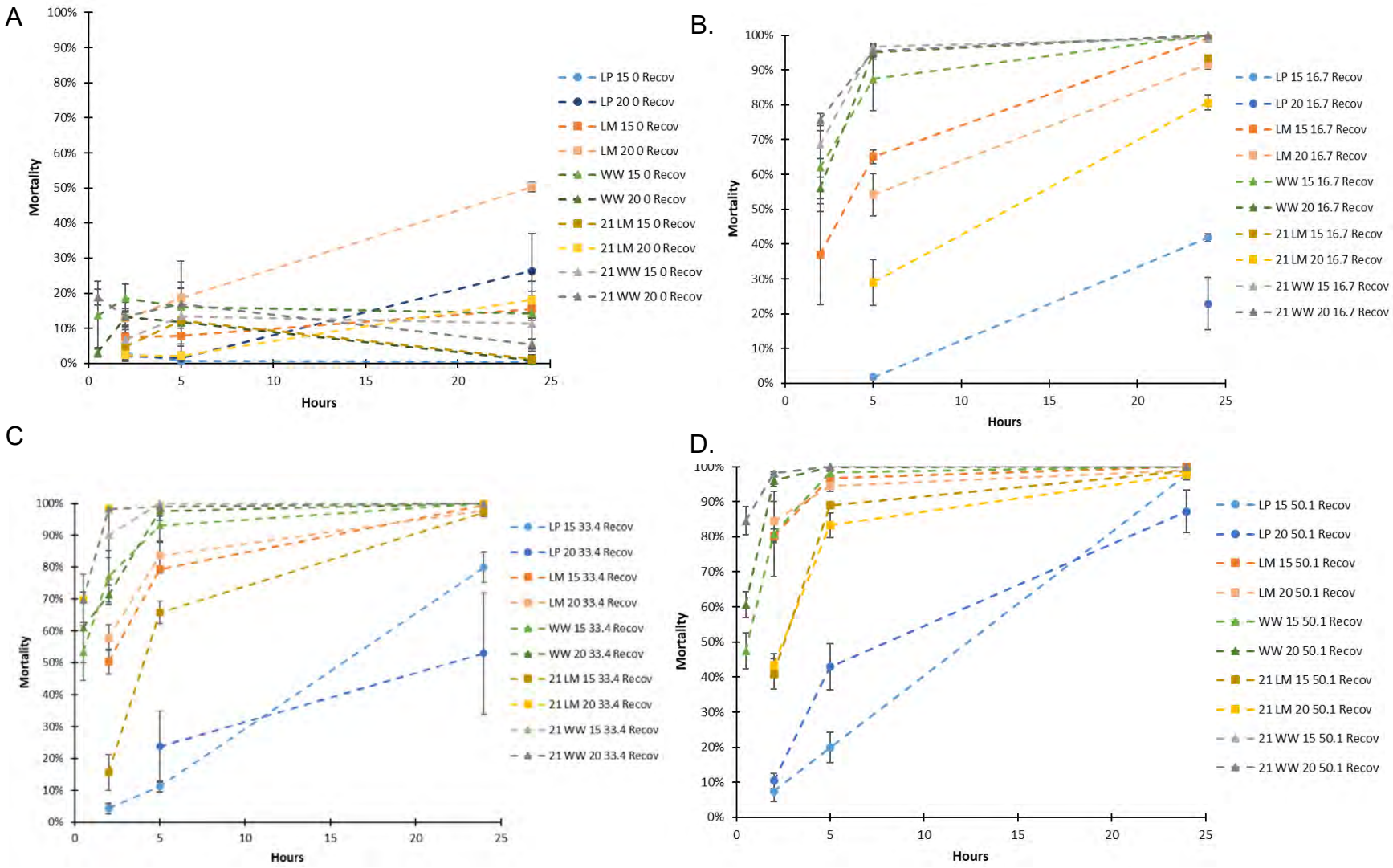


Figure 5-2. Veliger dose-response mortality

Standard deviation bars for the 24-hour recovery (Recov) measurement for each test condition from Lake Piru (LP), Lake Mathews (LM) in 2020 and 2021, and Weymouth WTP (WW) in 2020 and 2021 at the two test temperatures (15 & 20℃). (A) 0 µL/L control mortality; (B) 16.7 µL/L EarthTec QZ; (C) 33.4 µL/L EarthTec QZ; (D) 50.1 µL/L EarthTec QZ.

The water quality while testing was suitable for veliger survival. As expected, the pH decreased with increasing EarthTec QZ concentration. The free and total copper concentrations increased with increased EarthTec QZ concentration. Over the 24-hour exposure time for each experimental condition (EarthTec QZ dose and temperature), copper concentrations decreased but were still within 20% of the initial dosed concentration for all sampling locations. The water at Weymouth WTP represents CRA water from Lake Mathews that had been exposed to free chlorine during the approximate 8 hours of travel time within the Upper Feeder pipeline (chlorine is dosed at the pipeline inlet, as the water leaves Lake Mathews). The measured residual chlorine levels in the Weymouth WTP influent samples were very low (0.07 mg/L), consistent with levels measured from Lake Mathews samples (0.06 mg/L); these levels, which approach the method detection limit (0.02 mg/L), are expected to be related to interferences from other ions in the water and not residual oxidant.

Further analysis was completed on the veliger mortality data to look for significant trends. The recovery mortality data was used to perform linear regression modeling. In order to get the data to fit normality assumptions, the recovery mortality was transformed with a square root of the arcsine value of the recovery mortality. Linear regression modeling showed that concentration, duration, and location were significant variables. For simplicity, the copper concentration of each dose was multiplied by the testing duration to get a time-dose variable (Table 5-1). Testing temperature (15 or 20°C) and year conducted were not significant variables in mortality results, therefore results were combined for each location to get a simplified model with a good fit (linear regression fit of r^2=0.91) (Figure 5-3).

Veligers collected from Weymouth WTP had the highest mortality and quickest response. Veligers from Lake Mathews were not as susceptible to the copper: longer times and higher concentration required to achieve close to 100% mortality. Similarly, Lake Piru water veligers were less susceptible to copper.

Table 5-1. Time-Dose variable calculation

*Units are hours of exposure duration multiplied by concentration of EarthTec QZ as Cu (h*mg/L Cu)*

Concentration as EarthTec QZ (as Cu)	Duration (hours)			
	0.5	2	5	24
3 µL/L (0.18 mg/L)	0.09	0.36	0.9	4.32
16.7 µL/L (1 mg/L)	0.5	2	5	24
33.4 µL/L (2 mg/L)	1	4	10	48
50.1 µL/L (3 mg/L)	1.5	6	15	72

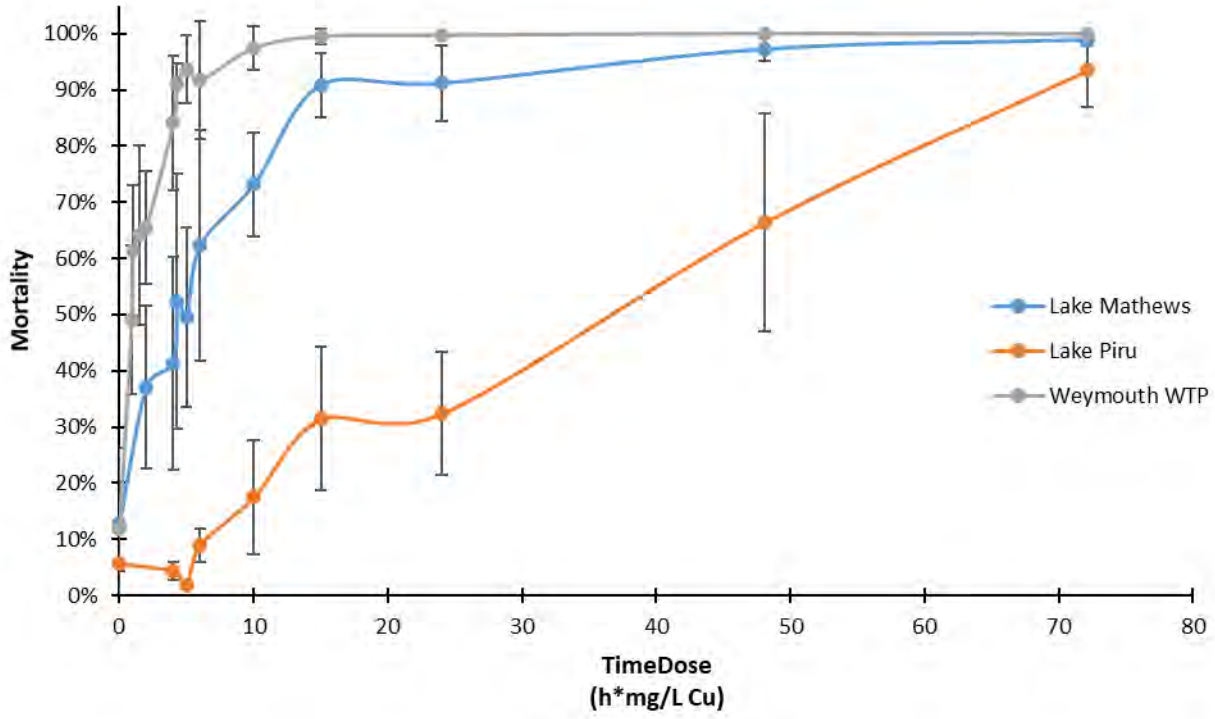


Figure 5-3. Veliger mortality for cumulative exposure to EarthTec QZ
Standard deviation bars for each sample location combine all temperatures and years tested.

5.2 Susceptibility of Non-targets to EarthTec QZ

Large volumes of water were collected from each test location and delivered to Aquatic Bioassay and Consulting Laboratories (ABC Labs) in Ventura, CA to conduct acute toxicity testing on three non-targeted indicator species: water flea (*Ceriodaphnia dubia*), rainbow trout (*Oncorhynchus mykiss*), and fathead minnow (*Pimephales promelas*).

ABC Labs completed acute toxicity tests using guidelines prescribed in *Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms* (EPA-821-R-02-012, 2002; EPA/600/4-90/027F, 1993), where the indicator species are exposed to the test water (sample water collected a given test location and spiked with the selected dose of EarthTec QZ) and evaluated over a 96-hour exposure period. This testing was completed in parallel with the dose-response veliger testing. Each round of toxicity testing utilized the same site-specific water and EarthTec QZ stock that was used during the veliger dose-response testing and results are presented in Technical Memorandum 2 (TM2) in Appendix B. The selected EarthTec QZ doses used for the acute toxicity testing were determined based on the most effective dose identified from the preliminary veliger dose-response results for each location.

For all sampling locations, the water flea was the most susceptible to EarthTec QZ (Figure 5-4). One round of testing with the Weymouth WTP influent water reported 0%

mortality for the 8.35 and 16.7 µL/L EarthTec QZ (0.5 and 1.0 mg/L as Cu), but test condition repeats demonstrated 100% mortality which caused the large standard deviation bars. The reason for the discrepancy between the two tests is unknown: possible hypotheses include different size classes of test subjects used, laboratory error (though ABC Labs provided quality assurance/quality control (QA/QC) review and did not identify any deviation from their protocol), or water quality differences. The chlorine for the 2021 testing at Weymouth WTP was higher than in 2020 and the water flea test subjects are very sensitive to chlorine exposures (the lethal concentration for 50% of the sample population (LC50) for free chlorine species hypochlorous acid and hypochlorite ion are 0.14 mg/L and 0.08 mg/L, respectively (Taylor 1993)). The repeated test results with 100% mortality were used to provide conservative estimates for evaluating EarthTec QZ toxicity for the non-target indicator species.

Rainbow trout were very sensitive to Earth Tec QZ concentrations (Figure 5-4). All sample locations had the same mortality curves. Fathead minnows were not as sensitive to copper as EarthTec QZ (Figure 5-4). In Lake Mathews and Weymouth WTP water, there was little to no mortality observed even at 16.7 µL/L EarthTec QZ or 1.0 mg/L as Cu. In Lake Piru water 42.5% mortality was observed at 16.7 µL/L EarthTec QZ (1.0 mg/L as Cu). However, it should be noted that Lake Piru water had an initial copper concentration of 0.15 mg/L (Table 5-2) whereas Lake Mathews and Weymouth had an initial copper concentration of 0.01 mg/L. This could have increased the dose of copper to 1.15 mg/L, which could be a tipping point in the toxicity to fathead minnows given the Lake Piru water quality.

The difference in acute toxicity response observed between the two different waters can potentially be further explained by the differences in water quality that affect the bioavailability of metals in water as well as competitive ions. Copper toxicity in fish can be predicted using the biotic ligand model which uses a wide array of water quality to predict bioavailability and toxicity: pH, dissolved organic carbon (DOC), ions, alkalinity, and temperature (USEPA 2016). As discussed in Section 5.3, these water quality parameters affect the chemical composition of copper in the water and determine the bioavailability. For example, alkalinity, pH, hardness, and anions affect the saturation indices for copper and the percentage of total copper existing in dissolved form. It has been demonstrated that copper toxicity decreases when pH and hardness increase (Welsh et al. 1995). Meanwhile, other cations, especially sodium, can compete with free copper ions for binding sites in the gills of fish (Nelson et al., 1986; Welsh et al. 1995, Parametrix and HydroQual 2006; USEPA 2016). In other studies conducted by KASF Consulting, sodium levels in Lake Piru in 2019 were lower (65 mg/L) than those measured in Colorado River water (100 mg/L) in 2015. While sodium measurements were not included in this study, the higher sodium concentration for Lake Mathews and Weymouth WTP should have provided more protection from the copper toxicity. The suite of water quality for predicting toxicity with the biotic ligand model was not measured for the tests; however, the water quality clearly affects copper toxicity and could be the reason for the difference in Lake Piru fathead minnow mortality.

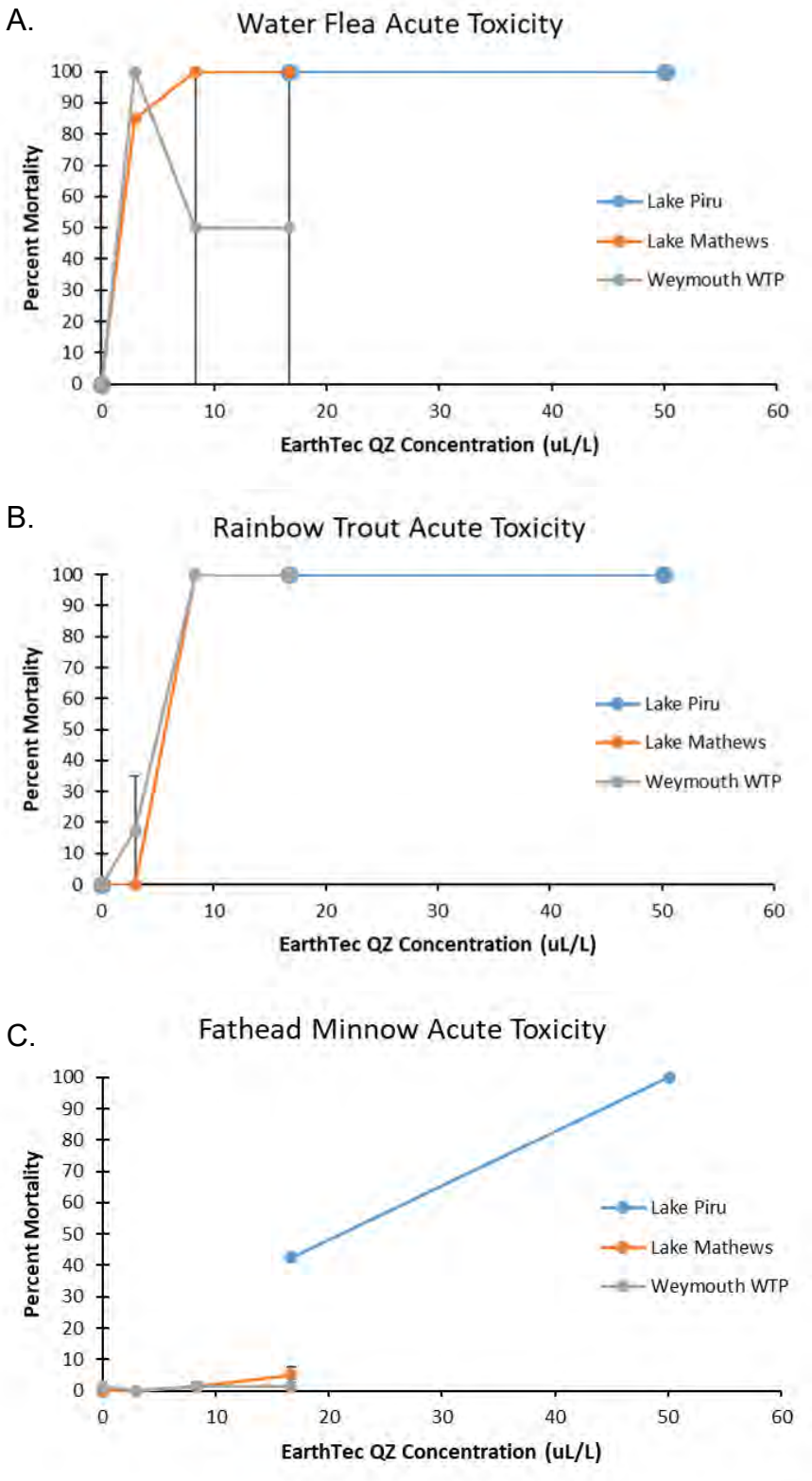


Figure 5-4. Average acute toxicity for (A)water flea; (B)rainbow trout; and (C)fathead minnow with EarthTec QZ following 96=hour exposure durations
Standard deviation bars for each sample location combine all acute toxicity results.

The acute toxicity tests were 96-hour exposure durations for the water flea, rainbow trout, and the fathead minnow. To obtain a similar metric to compare the veliger mortality, the exposure time was multiplied by the EarthTec QZ concentration as mg/L of copper tested to obtain a time-dose variable. Mortality is correlated with cumulative EarthTec QZ exposure in Figure 5-5 for veligers from each test location (data from each temperature condition is combined) and for all non-target indicator species (combined data from all locations) for exposures of up to 100 h*mg/L.

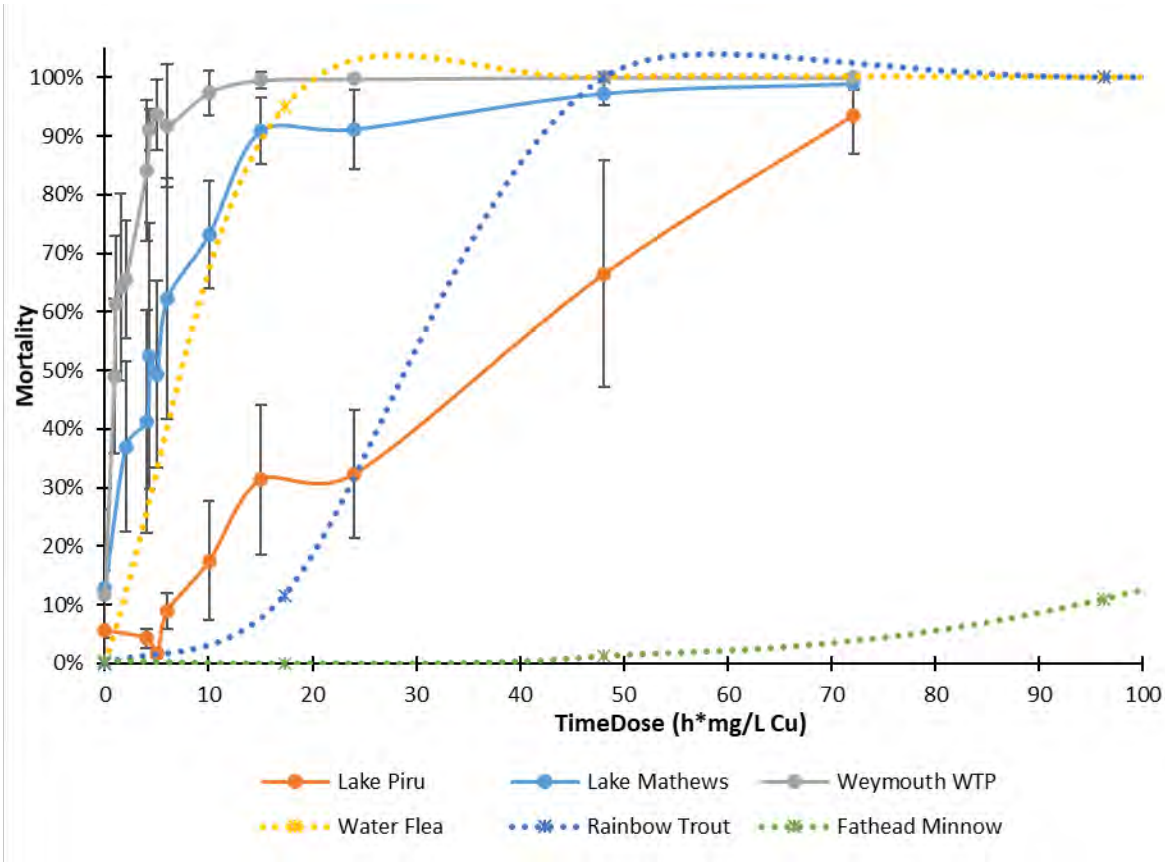


Figure 5-5. Mortality with cumulative exposure to EarthTec QZ
For veligers by location, plus water flea, rainbow trout, and fathead minnow (non-target indicator results for all locations combined).

Higher veliger mortality was observed in Weymouth WTP water with lower cumulative exposure (e.g., shorter time exposure, lower EarthTecQZ dose, or both) to EarthTec QZ relative to the exposure required for similar mortality levels in the water flea, rainbow trout, or fathead minnow (Figure 5-5). In Lake Mathews water, the veligers had approximately the same mortality rate as the water flea and died with lower EarthTec QZ exposure relative to the rainbow trout and the fathead minnow. Veligers in Lake Piru survived at a higher rate relative to the water flea and rainbow trout (above 24 h*mg/L exposure), but had higher mortality with lower exposure compared to the fathead minnow. This time-dose assessment allowed for a broad comparison of the impact of EarthTec QZ exposure on both the veligers and non-target indicators, given the

differences in experimental setup; the veliger exposure times were limited to 24 hours, while the non-target indicators were all tested using a 96-hour exposure time. Further testing of co-mortalities of veligers and the non-target indicators is recommended using a wider range of EarthTec QZ exposures with field-based test methods, such as a mesocosm (e.g., pilot-scale water enclosure system adjacent to treatment site), to better replicate expected full-scale conditions.

5.3 Raw Water Quality

Raw water quality was measured for each veliger collection and acute toxicity sampling event. Many of the water quality parameters were the same for Lake Mathews and Weymouth WTP, which was expected since Weymouth WTP influent water is the chlorinated water downstream of Lake Mathews (Table 5-2). Background copper levels were higher at Lake Piru compared to Lake Mathews and Weymouth WTP. Chlorine levels were also higher in Lake Piru water, but this could have been due to interferences in testing or residual levels from washing the test vials with tap water. Copper and chlorine levels did not impact veliger results but in some instances were high enough to impact the acute toxicity results. Dissolved oxygen, conductivity, and pH were at levels suitable for organism growth and survival. Oxidation reduction potential (ORP) was consistent for all samples.

The alkalinity of Lake Piru (mean=171 mg/L as $CaCO_3$) was higher than Lake Mathews and Weymouth WTP (mean=126 mg/L as $CaCO_3$), which indicated that Lake Piru water was able to buffer more of the effects of the addition of EarthTec QZ (a very acidic compound). Hardness measurements were also highest for Lake Piru (mean=315 mg/L as $CaCO_3$). Tests conducted in June 2021 for Lake Mathews and Weymouth WTP had higher hardness measurements (mean=278 mg/L as $CaCO_3$) than sampling 2 weeks later in 2021 (mean=182 mg/L as $CaCO_3$). Water from the 2020 sampling events had the lowest hardness (mean=143 mg/L as $CaCO_3$). Calcium competes with copper for binding sites in low hardness waters but is not a factor in waters that are considered hard like both Lake Piru and Lake Mathews waters (Parametrix and HydroQual 2006). DOC levels were different by location, with the highest levels measured at Lake Piru (Table 5-2). Chemical oxygen demand (COD) varied widely, and no trend was apparent.

Researchers have shown that along with calcium, other competing cations such as magnesium, sodium, and hydrogen, as well as complexing ligands (DOC, OH^-, Cl^- and CO_3^{2-}) decrease the toxicity of copper as their concentrations increase in natural waters (Parametrix and HydroQual 2006). Alkalinity can affect copper by changing which complex is bioavailable (Parametrix and HydroQual 2006). When the water quality parameters were put into the veliger logistic regression model, alkalinity, DOC, and COD were important variables, but hardness was not a significant factor in the model. Water quality was an important factor in determining the toxicity of EarthTec QZ to veligers. Lake Piru water quality was different from Lake Mathews water, especially the alkalinity and DOC measurements.

Table 5-2. Raw water quality
Readings from each sample location at the time of veliger testing.

Parameter			Total Copper	Free Copper	Total Chlorine	Oxidative Reduction Potential	Dissolved Oxygen	Conductivity	pH	Alkalinity	Hardness	Dissolved Organic Carbon	Chemical Oxygen Demand
Location	**Year**	**Temperature Condition**	**mg/L**	**mg/L**	**mg/L**	**mV**	**mg/L**	**µS/cm**	**pH unit**	**mg/L as CaCO3**		**mg/L**	**mg/L**
Lake Piru	**Raw Water Quality**												
	2019	15ºC Tests	0.15	0.14	0.08	210.4	10.45	927	8.43	187	314	5.33	10.00
		20ºC Tests*	0.16	0.16	0.12	191.9	8.88	913	8.25	154	316	5.05	5.65
Lake Mathews	**Raw Water Quality**												
	2020	15ºC Tests	0.02	0.02	0.02	212.5	7.57	946	8.18	115	138	3.01	4.67
		19ºC Tests	0.01	0.01	0.02	245	8.08	937	8.20	115	142	2.95	5.69
	2021	15ºC Tests	0.00	0.00	0.01	185.6	8.46	950	8.24	138	281	2.98	4.65
		20ºC Tests	0.00	0.00	0.00	208.2	8.34	957	8.29	135	178	2.91	3.30
Weymouth WTP	**Raw Water Quality**												
	2020	15ºC Tests	0.01	0.01	0.04	214	8.27	944	8.02	119	150	4.09	8.50
		20ºC Tests	0.00	0.00	0.07	223.5	8.14	941	8.13	121	139	4.19	8.24
		Additional Sampling **	0.02	0.02	0.05	211	8.19	955	8.12	119	145	4.05	7.88
	2021	15ºC Tests	0.02	0.03	0.10	195.6	8.69	969	8.18	137	275	3.01	4.46
		20ºC Tests	0.00	0.00	0.02	218.2	8.34	961	8.28	135	186	3.12	4.86

*Some parameters may be different from normal as it was raining during the sampling and testing period.
** Re-sample of Weymouth WTP influent for acute toxicity testing with 3 µL/L EarthTec QZ. This water was not used for veliger testing.

While working with the samples, we observed that the larger sized veligers were taking longer to succumb to the toxic effects of copper and many of these larger sized veligers were still alive during the final readings. Each sample collection was evaluated to determine the proportion of the four veliger size classes (i.e., D-shaped, small umbonal, large umbonal, and pediveliger) present, as indicated in Table 5-3. Samples taken during the same week from a given location were combined. The size distributions from all of the sample collections were then ranked by size class to compare the overall veliger sizes among the different sample collections. The ranking system assigned lower numerical values to a sample collection with higher proportions of larger sized veligers. The two larger size classes (i.e., pediveliger and large umbonal) were ranked from the location where they were most abundant to least (1 to 8). The opposite approach was used for the smaller two size classes (i.e., small umbonal and D-shaped), where the ranking values indicated least abundance to most (1 to 8). The scores for each sample collection were added and then ranked from A (lowest value) to H (highest value). This showed which samples had a relatively higher proportion of large veligers compared to those containing small veligers (Table 5-3).

Table 5-3. Size class distribution of veligers used in testing
Each sample site was ranked by density of large veligers to small veligers.

Sample Site	Size Class				Rank (score wt)
	D-shaped (50-150 µm)	**Small umbonal** (150-250 µm)	**Large umbonal** (200-350 µm)	**Pediveliger** (350-500 µm)	
2021 Lake Mathews (20°C)	32%	18%	38%	11%	A (8)
Lake Piru (15 & 20°C)	45%	15%	20%	20%	B (9)
2021 Lake Mathews (15°C)	52%	22%	23%	3%	C (16)
2021 Weymouth WTP (15°C)	74%	8%	17%	2%	D (17)
2020 Lake Mathews (20°C)	50%	30%	15%	5%	E (20)
2021 Weymouth WTP (20ºC)	53%	25%	22%	1%	F (22)
2020 Weymouth WTP (15 & 20°C)	80%	15%	3%	2%	G (23)
2020 Lake Mathews (15°C)	75%	20%	4%	1%	H (26)

Veligers in Lake Mathews during the 2021 sampling event for the 20ºC test conditions had the most abundant amount of the largest size classes and had a very similar veliger size class distribution to Lake Piru. The veliger mortality for this sampling event was lower than the other Lake Mathews events, and more consistent with the low mortality achieved in the Lake Piru testing (Figure 5-6). This trend was also observed with the

test sites containing mostly small veligers, where the smallest veligers utilized during testing were from the 2020 Lake Mathews sampling event for the 15ºC test conditions and the mortality was similar to the Weymouth WTP results (Figure 5-6).

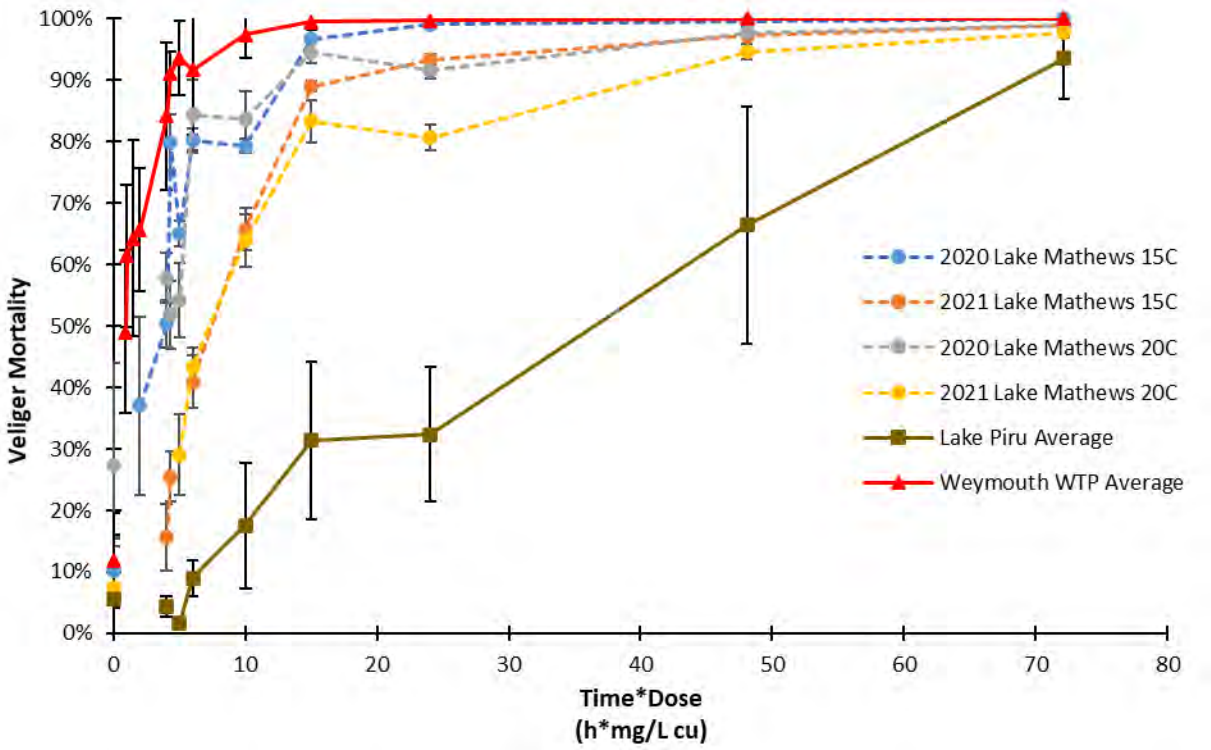


Figure 5-6. Veliger mortality with exposure to EarthTec QZ
Standard deviation bars combine all temperatures and years tested for Lake Piru and Weymouth WTP. Lake Mathews veliger mortality with standard deviation bars was not combined by temperature or sampling year.

The simplest and best fitting model showed that mortality was a function of location, concentration, and exposure duration. The location variable could be replaced with the alkalinity and DOC. Additionally, adding in veliger size class helped explain some of the variation in the model, but increased the complexity of the model.

5.4 Study goals and objectives

The study goals and objectives were to evaluate the effectiveness of EarthTec QZ, an EPA–registered molluscicide for prevention and control of quagga and zebra mussels, in Southern California water.

EarthTec QZ was effective in reducing the number of live veligers in all systems treated. Veliger mortality increased with increased concentration of EarthTec QZ and with increased duration of exposure. The water that had the highest mortality was the chlorinated Lake Mathews water sampled at Weymouth WTP. The lowest mortality per concentration and duration was observed in veligers from Lake Piru. Alkalinity and

dissolved organic carbon were significant water quality factors in influencing the toxicity of EarthTec QZ in the hard waters of Southern California.

5.5 Major problems in achieving study goals and objectives

There were two major problems that were encountered while conducting this study. The first was the impact of Covid-19 on the sampling schedule and the second was obtaining enough veligers at Lake Mathews in 2020.

Covid-19 stay at home orders and associated restrictions delayed sampling events at Lake Mathews, Weymouth WTP, and OCWD's OC-28 turnout (original location) from May or June 2020 to September 2020. Testing was completed at the original study testing locations, with the exception of OC-28 turnout. The study was designed with the intention of correlating the dose-response efficacy of EarthTec QZ at Lake Mathews with that of two downstream locations. Water from Lake Mathews is chlorinated prior to entering pipelines supplying both OC-28 and Weymouth WTP. Considering the connection of these water supplies, the Study aimed to correlate testing from Lake Mathews with the two downstream locations – OC-28 turnout and Weymouth WTP influent. Testing these three locations in a series would have allowed for comparison of the effects of EarthTec QZ with and without chlorine exposure. However, due to prior budgeting, OCWD only had access to Lake Mathews water at the OC-28 turnout through June 2020, and not during the 2021 fiscal year (July 2020 – June 2021). Given the similarity in travel time from Lake Matthews to Weymouth WTP and Lake Mathews to OC-28, the Weymouth WTP results would be expected to be the same at OC-28.

Samples of quagga veligers were initially collected at Lake Mathews on September 3, 2020 for the 20°C dose-response testing with EarthTec QZ. However, veliger densities were too low to provide greater than 50 veligers per testing replicate and the control mortality was greater than 50% for all exposure durations. An additional sampling and subsequent testing event was conducted on September 18, 2020 at deeper depths which yielded a higher veliger density acceptable for dose-response testing. These results were originally labeled as 19°C to avoid confusion with the initial 20°C results that had high mortality in the control group. In the summary of the data, the initial 20°C results were removed from the analysis and discussion; the complete results are available in TM1 in Appendix A.

5.6 Application of study to other areas of the region

EarthTec QZ was effective in reducing the number of live veligers in all systems treated. This copper treatment was most effective after the veligers had been exposed to chlorine between Lake Mathews and Weymouth WTP. EarthTec QZ treatments were less effective on veligers collected in Lake Piru, which had higher alkalinity and DOC measurements, and a large variety of veliger size classes present.

These results can be applied to other waterbodies within Southern California that have high hardness values and similar alkalinity and DOC levels, but site-specific testing would be required to assess the appropriate dose and exposure time to achieve the desired mortality. Some deviations from the results are expected if applied in other waterbodies because copper toxicity is influenced by high alkalinity levels and dissolved organic carbon levels. These parameters also change with seasons, as we observed different values among different sampling events at the same location with seasonally variable mortality curves. Though we did see seasonal variation in mortality results, the variability was only about 15%.

This Study evaluated veliger toxicity at a bench-scale level that has its own limitations when comparing it to a field application. Veligers collected from each study location were filtered to remove large non-target organisms and represent a size range of 50-500 micrometers. Likewise, the sample waters used for the beaker tests were filtered to remove particles above 10 micrometers. If the organisms that were between 10 and 40 micrometers or larger than 500 micrometers were present, then the efficacy of the EarthTec QZ on the veligers may be lower because these organisms could uptake copper, making it less bioavailable. This would be monitored by measuring and comparing the residual copper at the end of the exposure duration, which if true would be less than found in these study results. As discussed in conjunction with the results from the control conditions, natural veliger mortality increased with residence time in the test beakers, which will obscure the results for test conditions after approximately 48 hours. Utilizing a mesocosm set up or a flow-through apparatus to test site-specific conditions (dose and exposure time) is recommended to optimize the potential use of EarthTec QZ for controlling veligers at each candidate location.

Copper is regulated by the State Water Resources Control Board Division of Drinking Water in California and has a secondary maximum containment level of 1.0 mg/L (State of California 2018). This will regulate the maximum dose of copper to be used in treatments of imported water supplies. If locations around Southern California were to use EarthTec QZ, they could expect similar results to this study (Table 5-4). For example, if EarthTec QZ was applied to the water at OC-28 at a dosage of 16.7 µL/L EarthTec QZ dosage (1.0 mg/L as Cu), then we would expect mortality results similar to what we observed in the Weymouth WTP testing. This would be applicable when OC-28 is receiving MWD's CRA water.

Table 5-4. Average percent veliger mortality observed at each location
Data presented for 16.7 µL/L EarthTec QZ dosage (1.0 mg/L as Cu) with standard deviation.

Location	Exposure Duration (hours)		
	2	5	24
Lake Piru	Not Assessed	1.7 (±0.6)%	32.4(±10.9)%
Lake Mathews	37.0(±14.5)%	49.4(±16.0)%	91.2(±6.8)%
Weymouth WTP	65.6(±10.0)%	93.6(±6.0)%	99.8(±0.5)%

6 Conclusion

EarthTec QZ is an effective tool to use in Southern California waters to achieve high mortality of quagga mussel veligers. EarthTec QZ effectiveness is driven by bioavailable copper ions in the water which is affected by hardness, alkalinity, and DOC. EarthTec QZ was more toxic to veligers in water that had previously been exposed to a chlorine treatment. EarthTec QZ is also toxic to non-target species, including water fleas and rainbow trout. This study showed that fathead minnows are impacted less by the toxicity of EarthTec QZ.

The design of this Study allowed for comparison of multiple variables expected to potentially influence the toxicity of EarthTec QZ. Temperature was not an explanatory factor in determining mortality, however, using a broader range of temperatures could be considered if a treatment were to be applied at temperatures outside of the range included in the study, such as at 25 to 30°C. COD did not correlate to mortality of veligers and was not found to be a parameter of interest. The size distribution of the veligers correlated with mortality differences for an individual sampling location (Lake Mathews). Small veligers succumbed quickly to the toxic effects of EarthTec QZ, but the larger veligers (pediveligers) were still alive at the end of both the treatment duration and the recovery period.

6.1 Lessons Learned

We found that it was important to add in a recovery period after the exposure duration to determine final mortality. During the Study, we observed veligers that were moving slowly and bloated, but were not stained green with the Fast Green dye used to determine veliger mortality. With the additional 24 hours of exposure to clean water, the veligers succumbed to the toxicity and were confirmed dead.

An example of how a recovery period would work in a field application would be as follows. OC-28 could be treated with EarthTec QZ at the inlet area for a 5-hour exposure duration, and then the treated water could be mixed with either reclaimed water or Santa Ana River water (which is veliger and copper free water), and the

expected mortality of veliger would be around 94% (Table 5-4) instead of 58% mortality without a recovery period. This post-treatment dilution is also expected to be beneficial for reducing the impact of EarthTec QZ on non-target aquatic species present in natural systems (e.g., Santa Ana River) and reducing the residual copper concentration.

One of the initial EarthTec QZ studies found that 100% veliger mortality occurred in 30 min at 3 µL/L EarthTec QZ (Watters et al. 2013). This Study did not achieve these results in any of the waters tested. The water used in the Watters study was Colorado River water at Lake Mead fish hatchery. Lake Mathews receives the same water downstream of this location. Our results did not obtain 100% mortality with a dosage of 3 µL/L EarthTec QZ even after a 24-hour exposure duration (Figure 6-1). Additionally, no tested concentration of EarthTec QZ achieved 100% morality with an exposure duration of 30 min (Figure 5-2).

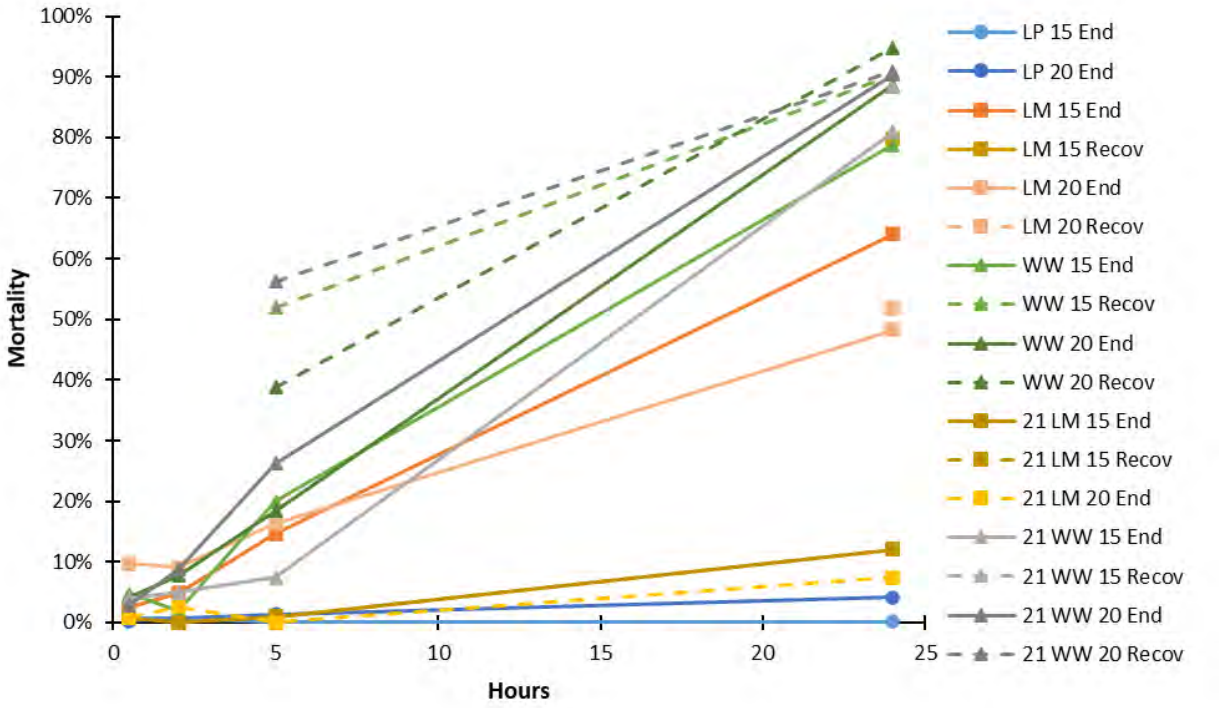


Figure 6-1. Mortality of quagga mussel veligers for 3 µL/L EarthTec QZ.
Data presented for mortality measured at the end (End) of each exposure time and after a 24-hour recovery (Recov) period with no additional exposure to EarthTec QZ.

6.2 Next Steps of the Study

The data from this Study can be used as a baseline to conduct full scale field treatments, but site-specific assessments would be required to assess the appropriate dose and exposure time to achieve the desired mortality for each particular site. Utilizing a mesocosm set-up or a flow-through apparatus to test site-specific conditions to determine dose and exposure time is recommended to optimize the potential use of

EarthTec QZ for controlling veligers at each candidate location. Site-specific testing should also evaluate potential effects on non-target organisms related to full-scale treatment, in coordination with CDFW and other regulators. During field treatment, veliger mortality should be monitored along with relevant water quality parameters – hardness, alkalinity, dissolved organic carbon, pH, dissolved oxygen, temperature, and ions. Data from a field study will be useful for more broadly assessing full-scale implementation of a chemical control method for quagga, such as EarthTec QZ.

Since this Study did not include all the organisms present in the water from each test location, site-specific testing prior to large scale testing is recommended. In this study the organisms between 10 and 50 micrometers and those larger than 500 micrometers were excluded in the veliger testing. Additionally, the acute toxicity data did not show the full impact of an EarthTec QZ treatment because testing water was filtered and represented only organisms less than 10 micrometers, in addition to the indicator species. Moreover, the treatment duration was 96 hours at full copper dosage. As such the results may not be representative of full-scale field treatment for these locations. The organisms that were not present in our study could have the potential of reducing the bioavailability of copper and decreasing the effectiveness of EarthTec QZ; and to make it as effective as our Study results, longer exposure durations or higher concentrations at the point of dosing may be required.

Additional studies that evaluate the extent of copper decay with a wide range of organisms present and/or similar to prospective treatment locations is recommended to fully understand EarthTec QZ's effect in the environment. This data would then be used to determine what type of remediation could be needed after EarthTec QZ treatment in locations with short exposure durations. If there are organisms that reduce the bioavailability of copper in the environment, then a high dose, short duration use of EarthTec QZ could be considered and not be toxic to other more sensitive organisms. For example, dosing EarthTec QZ in an aqueduct or pipeline may be acceptable, as long as the residual copper and toxicity to sensitive aquatic species no longer remains once the water is discharged into a downstream lake or river.

In a site-specific study, the toxicity to non-target aquatic organisms of concern would need to be evaluated. Part of the evaluation should include consideration of the size class of non-target organisms, as the small size classes are used in acute toxicity testing and are the most susceptible to a toxicant. In Hammond and Ferris (2019) no non-target mortality was observed, and zooplankton densities returned to high densities after treatment.

Finally, the data showed that low doses of EarthTec QZ work as well as higher doses, but longer exposure times are needed to obtain the same mortality. The field results and current practices of using EarthTec QZ in the Midwest show that low doses of 0.5 mg/L as Cu are effective at reducing and removing quagga mussels in lotic systems, but the treatment duration is weeks to a month (Hammond and Ferris 2019). Further site-

specific evaluation of the use of lower EarthTec QZ is recommended, with correlation of water quality.

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References

Hammond D, and G Ferris. 2019. Low doses of EarthTec QZ ionic copper used in effort to eradicate quagga mussels from an entire Pennsylvania lake. Management of Biological Invasions 10: 500-516.

Nelson H, D Benoit, R Erickson, V Mattson, and J Lindberg. 1986. The Effects of Variable Hardness, pH, and Alkalinity, and Humics on the Chemical Speciation and Aquatic Toxicity of Copper. USEPA (Duluth, MN 55804).

Parametrix and HydroQual. 2006. Evaluation of the Reliability of Biotic Ligand Model Predictions for Copper Toxicity in Waters Characteristic of the Ardi West. Final Report for Arid West Water Quality Research Project prepared for Pima County Wastewater Management. Available: https://webcms.pima.gov/UserFiles/Servers/Server_6/File/Government/Wastewater%20Reclamation/AAWQRP/BioticLigandModel.pdf

State of California. 2018. Maximum Contaminant Levels and Regulatory Dates for Drinking Water U.S. EPA VS California. Available: https://www.waterboards.ca.gov/drinking_water/certlic/drinkingwater/documents/ccr/mcls_epa_vs_dwp.pdf.

Stockton-Fiti KA and R Claudi. 2017. Use of a differential simple stain to confirm mortality of dreissenid mussel veligers in field and laboratory experiments. Management of Biological Invasions 8:325-333.

Taylor PA. 1993. An Evaluation of the toxicity of various forms of chlorine to *Ceriodaphnia dubia.* Environmental Toxicology and Chemistry 12: 925-930.

USEPA. 1993. *Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms*, Fourth Edition. EPA/600/4-90/027F, August 1993.

USEPA. 2002. *Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms*, Fifth Edition. EPA-821-R-02-012, October 2002.

USEPA. 2016. Water Quality Standards Academy: Biotic Ligand Model and Copper Criteria.

Watters A, SL Gerstenberger, and WH Wong. 2013. Effectiveness of EarthTec® for killing invasive quagga mussels (*Dreissena rostriformis bugensis*) and preventing their colonization in the Western United States. Biofouling 29: 21-28.

Welsh P, J Parrott, D G Dixon, P Hodson, D Spry, and G Mierle. 2011. Estimating acute copper toxicity to larval fathead minnow (*Pimephales promelas*) in soft water from measurements of dissolved organic carbon, calcium, and pH. Canadian Journal of Fisheries and Aquatic Sciences 53: 1263-1271.

Appendix A. Technical Memorandum 1 Evaluation of EarthTec QZ Veliger Treatment

TECHNICAL MEMORANDUM #1

Evaluation of EarthTec QZ Veliger Treatment

Fina Date: December 20, 2021
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Prepared for: Metropolitan Water District of Southern California
City of Santa Ana
Orange County Water District

Copies to: Water Replenishment District of Southern California
United Water Conservation District
Main San Gabriel Basin Watermaster
Central Basin Municipal Water District
California Department of Fish and Wildlife
California Department of Water Resources

Authors: Kelly Stockton-Fiti, KASF Consulting
Emily L. Owens-Bennett, P.E., Trussell
Liana Olivas, Trussell

Reviewers: David Hokanson, Ph.D., P.E., BCEE, Trussell
Adam Hutchinson, PG, CHG, OCWD
Christine Pham, OCWD
Tessa Lenz, UWCD

Subject: Technical Memorandum 1

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1 Introduction

1.1 Background

Since their discovery in the Colorado River system in January 2007, Quagga mussels (*Dreissena bugensis*) have rapidly spread through the Metropolitan Water District of Southern California's (MWD's) Colorado River Aqueduct (CRA) and have been detected at many sites in California. The presence of this invasive species presents a barrier to the use of imported water and storm water for groundwater replenishment. Considering these challenges, the City of Santa Ana proposed a study entitled "Restoration of Local Recharge Sources from Invasive Dreissenid Mussels" to MWD's Future Supply Actions Funding Program. The Study was conducted by Trussell Technologies (Trussell), KASF Consulting, and Orange County Water District (OCWD), with support from project partners including the Water Replenishment District of Southern California (WRD), the United Water Conservation District (UWCD), the Main San Gabriel Basin Watermaster,

and the Central Basin Municipal Water District (CBMWD). Although not directly contributing financially to the study, the California Department of Fish and Wildlife (CDFW) and California Department of Water Resources are supporting the Study by providing technical review and assistance.

1.2 Study Description

The purpose of the Study was to evaluate a potential treatment method, other than desiccation, for use in eradicating quagga mussel infestations as well as preventing the spread of this invasive species.

Trussell led the Study in partnership with KASF Consulting, a firm specializing in quagga mussel sampling, identification, and testing. The Study evaluated the effectiveness of EarthTec QZ™[1] for control of quagga mussel veligers (the mussel's final larval stage). A fourth location, OCWD's OC-28 turnout at Anaheim Lake, was originally planned but later eliminated based on scheduling issues and identification of a surrogate location. The Study included testing with samples collected from multiple locations, to allow the project team to evaluate the impact of site-specific water quality on the performance of EarthTec QZ.

The following objectives were evaluated at each testing location:

1. Quagga veliger dose-response testing with EarthTec QZ
2. Toxicity assessment of EarthTec QZ with non-target species

The purpose of this technical memorandum ™ is to describe the results of **Objective #1**.

2 Schedule Summary

The status of veliger dose-response tests completed at each testing location is summarized in Table 2.1.

[1] EarthTec QZ® is registered with the United States Environmental Protection Agency (USEPA) as a molluscicide for the control of quagga and zebra mussels. It is also registered with the California Department of Pesticide Regulation as a molluscicide for use in lakes, streams, rivers, waterways, canals, ponds, reservoirs, irrigation systems, and catch basins.

Table 2.1. Study status at each sampling location

Location	Study Status	Testing Date
Lake Piru	Completed	December 2 – 6, 2019
Lake Mathews	Completed	September 9 – 11, 2020 September 18 – 20, 2020
Weymouth WTP Influent	Completed	September 14 – 18, 2020
OC-28 Turnout	N/A[2]	
Weymouth WTP Influent 2021	Completed	June 28 – 30, 2021 July 12 – 14, 2021
Lake Mathews 2021	Completed	June 30 – July 2, 2021 July 14 – 16, 2021

Testing was completed for each of the study testing locations (Table 2.1), with the exception of the OC-28 turnout. The study was designed with the intention of correlating the dose-response efficacy of EarthTec QZ at Lake Mathews with that of two downstream locations. Water delivered from Lake Mathews is chlorinated prior to entering transmission pipelines supplying both OC-28 and Weymouth WTP. Considering the connection of these water supplies, the Study aimed to correlate testing from Lake Mathews with the two downstream locations – OC-28 turnout and Weymouth WTP influent. Testing these three locations in a series would allow for comparison of the effects of EarthTec QZ with and without chlorine exposure. However, due to prior budgeting, OCWD only had access to Lake Mathews water at the OC-28 turnout through June 2020, and not during the 2021 fiscal year (July 2020 – June 2021). Covid-19 restrictions precluded the project team from sampling at Lake Mathews until September 2020. For these reasons, it was not possible to complete sampling from the OC-28 turnout in conjunction with the sampling and testing at Lake Mathews and Weymouth WTP influent.

The project team met with MWD staff in August 2020 to discuss alternative sampling locations for the OC-28 turnout. Although already included as a sampling location for the Study, Weymouth WTP was identified as the most appropriate surrogate monitoring location for the OC-28 turnout, as both sites can receive water from Lake Mathews that is chlorinated and transmitted within a pipeline for a similar travel time and therefore, similar exposure time to chlorine. For this reason, sampling at OC-28 is no longer being considered for this Project. The project team proceeded with testing at Lake Mathews and Weymouth WTP during September 2020 and completed follow up testing at both of these locations in 2021, as well.

3 Study Results

Objective #1 of the Study, to conduct quagga veliger dose-response tests with EarthTec QZ, was completed at three locations: (1) Lake Piru, (2) Lake Mathews and (3) MWD's

[2] Sampling at OC-28 is no longer applicable given the similarity of this location with that of Weymouth WTP, as is later discussed.

Weymouth WTP influent. The Project Team developed a Test Plan for the study, providing details for the expected methods, materials, and test conditions related to collecting quagga mussel veligers and establishing a dose-response relationship to EarthTec QZ (Trussell 2019b).

The results from the completed three testing locations are presented in the following subsections. As discussed in **Section 2**, it was not possible to complete testing with samples collected from the OC-28 turnout due to restrictions from COVID-19, as well as water purchasing agreements between OCWD and MWD. The project team met with MWD staff in August 2020 to discuss alternative sampling locations for the OC-28 turnout. Although already included as a sampling location for the Study, Weymouth WTP was identified as the most appropriate surrogate monitoring location for the OC-28 turnout, as both sites can receive water from Lake Mathews that is chlorinated and transmitted within a pipeline for a similar travel time and therefore, similar contact time with chlorine. Thus, sampling at OC-28 was eliminated as a sampling location for the Study. The project team proceeded with testing at Lake Mathews and Weymouth WTP during September 2020 and completed follow up testing at both of these locations in 2021, as well.

Trussell worked with KASF Consulting to complete and submit an application for a Specific Use Scientific Collecting Permit to CDFW (Trussell 2019a). The permit was approved on November 6, 2019.

Live veligers were collected from each location, and subsets of the collected veligers were tested at two different temperatures (15°C and 20°C). Each temperature condition included veliger exposure to five concentrations of EarthTec QZ (ET) doses (0, 3.0, 16.7, 33.4, 50.1 µL/L). Copper is the active ingredient in EarthTec QZ molluscicide, thus the equivalent copper doses are also provided (0, 0.18, 1.0, 2.0, and 3.0 mg/L as Cu). Veliger mortality was evaluated at the end of each exposure time. Directly following this count at the end of the exposure time, the veligers were placed in a beaker and exposed to raw sample water (without the addition of EarthTec QZ) for a 24-hour recovery period. A final mortality assessment was made at the end of the recovery time to allow for analysis of delayed mortality.

The raw water quality of the sample water for each round of testing was measured during each round of testing, and can be found in **Appendix A**.

3.1 Lake Piru

The first round of dose-response quagga veliger testing was completed at UWCD's Lake Piru. This reservoir, located in Ventura County, CA, impounds storm flows from the Piru Creek watershed and supplemental imported State Project Water supplied by the Department of Water Resources (DWR) via Pyramid Lake. It is used both for recreation and storage of water used to recharge downstream groundwater basins between Lake Piru and the coastal Oxnard Plain. Quagga mussel infestation of Lake Piru was first discovered in late 2014.

Veligers and water samples were collected on December 2nd and 4th, 2019 for dose-response testing from December 2nd through the 6th, 2019. KASF Consulting led the quagga veliger collection and testing efforts, while Trussell supported veliger collection, dose-response testing, and provided water quality assessment. UWCD staff supported veliger collection efforts with plankton tow nets from a research-use boat. Veligers were collected on the first and third day of testing; both days resulted in a sufficient count of quagga mussel veligers for testing.

The veliger mortality results for Lake Piru are summarized in **Table 3.1** and plotted for each temperature condition in **Appendix B**.

Table 3.1. Average mortality with standard deviation (n=3) of veligers at Lake Piru as a function of EarthTec QZ dose, temperature, and exposure time

		0.5 hr.	2 hr.	5 hr.	24 hr.
Control					
15º C	End*	0.8±0.5%	0.2±0.3%	0.3±0.3%	0.2±0.4%
	Recovery**	NA	3.0±2.7%	0.7±0.6%	0.6±0.7%
20º C	End	0.0±0.0%	0.5±0.6%	1.2±0.2%	2.9±2.8%
	Recovery	NA	2.2±0.4%	1.5±1.3%	33.4±13.2%
3 µL/L as EarthTec (0.18 mg/L as Cu)					
15º C	End	0.7±0.2%	0.7±0.7%	0.0±0.0%	0.1±0.2%
20º C	End	0.2±0.3%	0.7±0.4%	1.4±1.2%	4.1±3.2%
16.7 µL/L as EarthTec (1.0 mg/L as Cu)					
15º C	End	1.0±0.9%	1.3±0.7%	2.1±2.1%	42.4±3.0%
	Recovery	NA	NA	1.7±0.7%	41. 9±1.3%
20º C	End	6.0±5.9%	2.1±1.3%	7.9±6.3%	23.5±12.3%
	Recovery	NA	NA	NA	22.9±9.2%
33.4 µL/L as EarthTec (2.0 mg/L as Cu)					
15º C	End	2.1±1.8%	4.7±0.3%	19.3±2.8%	74.2±4.4%
	Recovery	NA	4.3±2.0%	11.2±2.1%	79.9±5.8%
20º C	End	2.6±0.8%	8.6±2.1%	28.0±4.3%	51.0±17.0%
	Recovery	NA	NA	23.8±13.7%	53.0±23.3%
50.1 µL/L as EarthTec (3.0 mg/L as Cu)					
15º C	End	1.8±1.1%	5.1±0.2%	37.4±10.0%	99.0±0.8%
	Recovery	NA	7.5±3.6%	19.9±5.4%	NA
20º C	End	NA	NA	NA	97.7±1.8%
	Recovery	3.6±0.9%	15.9±4.7%	41.8±4.1%	85.0±4.3%

* End is the end of the exposure period
** Recovery is at the end of the 24-hour recovery period following the exposure period.
NA=Not Assessed

As shown in **Table 3.1**, veliger mortality occurred at 16.7(1.0), 33.4(2.0), and 50.1(3.0) µL/L as EarthTec QZ (as Cu in mg/L) at both temperatures. Mortality of the veligers at 16.7 µL/L EarthTec QZ was similar to the control. At 5 hours of exposure, the veligers exposed to the two highest concentrations (33.4 and 50.1 µL/L EarthTec QZ) began to demonstrate mortality. It took 24 hours for the veligers exposed to 50.1 µL/L EarthTec QZ to reach mortality of 85% or greater. Higher veliger mortality was observed at the lower temperature of 15ºC, compared with the 20ºC test conditions; however, using linear regression modeling temperature was not a significant variable in predicting mortality.

The recovery procedure did not show a significant difference statistically or observationally in mortality results compared with the mortality measured at the end of the exposure period. The main exception to this trend was observed in the 20ºC control after the recovery period, with mortality greater than 33% compared to approximately 3% mortality measured at the end of the 24-hour exposure. Natural mortality was

observed when veligers were held in static conditions; mortality was increased with time and was amplified by higher temperatures.

Water quality parameters that were measured throughout testing included temperature, dissolved oxygen, pH, specific conductance, free copper, and total copper. The average temperatures of the 15°C and 20°C test conditions were 15.1°C and 19.7°C, respectively. Varying the dose of EarthTec QZ did not produce notable changes in either the dissolved oxygen or the specific conductance in any of the test conditions. A decrease in pH was observed with increasing EarthTec QZ concentrations, however the pH for each individual test condition remained mostly consistent over time. The full range of tested conditions resulted in pH values that were consistent with those under which veligers can thrive. Free and total copper readings were very similar for each tested condition. Over time the copper concentrations decreased slightly but was not significantly different for a given dose of EarthTec QZ. After 24 hours of exposure, the measured copper concentration was similar to the original treatment concentration. Copper concentrations in the recovery were near zero. A table of detailed water quality measurements from Round 1 Lake Piru testing are included in **Appendix C**.

3.2 Lake Mathews 2020

The second round of dose-response veliger testing was completed at Lake Mathews, a reservoir located in Riverside County, CA that is owned and operated by MWD. Lake Mathews is primarily used to store CRA water. Water from Lake Mathews is delivered into raw water pipelines that are dosed with chlorine for quagga control.

Samples of quagga veligers were initially collected at Lake Mathews on September 3rd, 2020 for the 20°C dose-response testing with EarthTec QZ. However, veliger densities were too low to provide greater than 50 veligers per testing replicate and the control mortality was greater than 50% for all exposure durations. The low veliger counts can be attributed to an insufficient depth at which the veligers were collected. During additional sampling at Lake Mathews, the towing depth was increased from 50 feet to 100 feet from the top of the intake tower. This increase in collection depth yielded a higher veliger density acceptable for dose-response testing.

Additional veligers were collected on September 18th, 2020 to repeat the testing of the 20°C conditions (September 18-20); these results are recorded in Table 3 as 19°C to avoid confusion with the initial 20°C results that had high mortality in the control group. Testing at 15°C was completed September 9th through 11th, 2020 using veligers and water samples collected from Lake Mathews on September 9th. KASF Consulting led the quagga veliger collection and testing efforts while Trussell and OCWD supported veliger collection, dose-response testing, and water facility and towing/collection locations.

The veliger mortality results for the 2020 Lake Mathews are summarized in **Table 3.2** and graphed by temperature condition in **Appendix D**.

Table 3.2. Average mortality and standard deviation (n=3) of veligers from Lake Mathews in 2020 as a function of EarthTec QZ dose, temperature, and exposure time (results labeled as 20ºC are not considered representative due to low veliger counts in the sample collection and high mortality of the control; these test conditions were repeated and labeled as 19ºC)

		0.5 hr.	2 hr.	5 hr.	24 hr.
Control					
15°C	End*	3.4±0.8%	5.7±1.7%	4.1±1.6%	3.8±2.3%
	Recovery**	NA	7.5±2.9%	7.8±2.2%	15.5±5.0%
19°C	End	5.5±0.7%	5.2±3.1%	8.2±1.3%	19.6±9.0%
	Recovery	NA	12.9±2.2%	18.6±3.0%	50.3±1.3%
20°C	*End*	*50.7±9.6%*	*63.1±4.0%*	*60.9±3.7%*	*60.5±8.6%*
	Recovery	*NA*	*66.4±2.6%*	*87.1±3.8%*	*72.9±4.2%*
3 µL/L as EarthTec (0.18 mg/L as Cu)					
15°C	End	2.4±1.1%	4.9±0.7%	14.7±2.5%	64.1±7.7%
	Recovery	NA	NA	NA	79.8±4.6%
19°C	End	9.7±1.2%	9.1±2.9%	16.3±0.6%	48.3±6.7%
	Recovery	NA	NA	NA	51.8±5.6%
20°C	*End*	*65.0±9.6%*	*64.6±10.3%*	*67.4±5.4%*	*68.0±8.4%*
	Recovery	*NA*	*NA*	*NA*	*NA*
16.7 µL/L as EarthTec (1.0 mg/L as Cu)					
15°C	End	6.8±3.2%	32.0±9.9%	37.7±9.4%	95.6±1.7%
	Recovery	NA	37.0±14.5%	65.1±2.0%	99.1±0.7%
19°C	End	8.5±3.3%	16.9±4.2%	23.8±6.8%	90.2±3.4%
	Recovery	NA	NA	54.2±6.0%	91.6±1.3%
20°C	*End*	*53.7±15.0%*	*64.2±8.8%*	*52.3±7.0%*	*97.8±1.8%*
	Recovery	*NA*	*NA*	*NA*	*98.2±1.6%*
33.4 µL/L as EarthTec (2.0 mg/L as Cu)					
15°C	End	7.2±1.4%	55.7±15.8%	60.1±7.8%	99.0±0.8%
	Recovery	NA	50.4±3.9%	79.3±1.2%	99.5±0.4%
19°C	End	11.5±4.9%	22.7±3.4%	40.5±0.6%	97.9±1.3%
	Recovery	NA	57.8±4.1%	83.6±4.6%	97.8±1.0%
20°C	*End*	*69.7±6.0%*	*73.2±4.2%*	*94.2±2.3%*	*100±0.0%*
	Recovery	*NA*	*NA*	*100±0.0%*	*NA*
50.1 µL/L as EarthTec (3.0 mg/L as Cu)					
15°C	End	7.9±2.7%	60.5±6.1%	66.6±8.2%	100.0±0.0%
	Recovery	NA	80.2±2.0%	96.7±2.1%	NA
19°C	End	18.2±1.5%	25.3±3.6%	43.9±2.0%	98.4±0.4%
	Recovery	NA	84.4±5.7%	94.5±1.7%	98.9±0.8%
20°C	*End*	*66.4±3.7%*	*89.6±7.5%*	*77.5±29.6%*	*100±0.0%*
	Recovery	*NA*	*98.5±1.2%*	*98.2±2.5%*	*NA*

* End is the end of the exposure period.
** Recovery is at the end of the 24-hour recovery period following the exposure period.
NA=Not Assessed

In the Lake Mathews samples, significant quagga veliger mortality was observed initially with the 16.7(1.0), 33.4(2.0), and 50.1(3.0) µL/L as EarthTec QZ (mg/L as Cu) test conditions at both temperatures after 24 hours of exposure. Mortality of the 3 µL/L EarthTec QZ was greater than the control at 15 and 19°C, and mortality at 3 µL/L EarthTec QZ after 24 h was around 50%. At 5 hours of exposure, the veligers exposed to the two highest concentrations (33.4 and 50.1 µL/L EarthTec QZ) began to demonstrate mortality greater than 50% mortality. It took 24 hours for the veligers exposed to 16.7, 33.4 and 50.1 mg/L µL/L EarthTec QZ to reach mortality of 85% or greater. Higher veliger mortality was observed at the lower temperature of 15ºC, compared with the 19ºC test conditions. Temperature was not a significant variable in the linear regression model.

Only the end mortality results greater than 20% were checked for additional recovery mortality after 24 hours. The recovery procedure did show a significant increase in mortality results both observationally and was statistically significant. Increased mortality in the recovery was due to veligers showing signs of stress at the initial reading but were not quite dead enough to be stained by the fast green and counted as dead. With the additional recovery time, the stressed veligers were dead and stained. The recovery mortality was high with greater than 90% mortality being achieved in the three highest concentrations. Interestingly, the 2 h 50.1 µL/L EarthTec QZ reached high mortality (greater than 80%). At the 24-hour exposure in the 19ºC study, the control mortality was over 50%. Natural mortality was observed when veligers were held in static conditions; mortality increased with time and was amplified by higher temperatures. Laboratory conditions in the OCWD laboratory were hotter than at the Lake Piru laboratory, which could have contributed to additional stress on the veligers. Steps were minimized to limit the time veligers were out of the water bath.

Water quality parameters that were measured throughout testing included temperature, dissolved oxygen, pH, specific conductance, free and total copper, and chlorine. The average temperatures recorded for the 15°C and 19°C test conditions were 14.8°C and 21.3°C, respectively. Varying the dose of EarthTec QZ did not produce notable changes in either the dissolved oxygen or the specific conductance in any of the test conditions. The pH decreased with increased concentration and each concentrations pH was significantly different from each other during testing. The pH for each individual test condition remained mostly consistent over time. The full range of tested conditions resulted in pH values that are consistent with those under which veligers can thrive. Free and total copper readings were very similar for each tested condition. Over time the copper concentrations decreased slightly for a given dose of EarthTec QZ, but after 24 hours of exposure, the measured copper concentration was not significantly different from the original treatment concentration. Copper concentrations in the recovery were near zero. The measured chlorine concentrations were negligible. A table of detailed water quality measurements from the 2020 Lake Mathews testing is included in **Appendix E**.

3.3 Weymouth Water Treatment Plant Influent 2020

The Weymouth WTP receives untreated water from two sources: Colorado River water from Lake Mathews via the Upper Feeder and State Project Water (SWP) from the East Branch of the SWP via the Rialto Pipeline or from the Upper Feeder via the Etiwanda Pipeline from the Rialto Pipeline. The WRD historically had access to this same source water through the Upper Feeder pipeline at CenB-28 for use in groundwater recharge via spreading. Detections of quagga in the nearby San Gabriel River in October 2017 were linked to deliveries of imported water. Since these detections, imported water has not been approved for use in groundwater recharge, due to the potential for quagga mussel infestations.

For the purpose of this study, the influent water sampling location at Weymouth WTP provides an access point for understanding the condition of the quagga veligers present in the pipeline downstream of Lake Mathews, following exposure to chlorine, and the cumulative impacts of exposure to both chlorine and EarthTec QZ. This location is of interest to the study when MWD is supplying only imported water from CRA via Lake Mathews. Water from Lake Mathews is dosed with chlorine as it enters the downstream water supply pipelines, as a control measure for quagga. The pipeline travel time from Lake Mathews to the Weymouth WTP is approximately 8 hours via the Upper Feeder. Another branch of pipeline downstream of the chlorine dosing station supplies raw water from Lake Mathews to the OC-28 turnout and MWD's Diemer WTP. The travel time from Lake Mathews to the Diemer WTP is approximately 11 hours, with the OC-28 turnout located upstream, but nearby the Diemer WTP. The approximate pipeline travel times were provided per personal communication with project partner Paul Rochelle of MWD. As such, the study aimed to complete parallel dose-response EarthTec QZ testing with quagga veligers collected from both Lake Mathews (no chlorine exposure) and Weymouth WTP (exposed to chlorine through pipeline) as a surrogate location for expected veliger populations at WRD's CenB-28 (Upper Feeder) or OCWD's OC-28 turnout. The Upper Feeder pipeline supply in late 2019 was from SWP, but changed to CRA via Lake Mathews in 2020.

The third round of dose-response veliger testing was completed from September 14th through 18th, 2020 using veligers and raw water collected from the influent water to MWD's Weymouth WTP. Collection of veligers and water samples were taken September 14th and 16th, 2020; both days resulted in a sufficient count of quagga mussel veligers for testing. KASF Consulting led the quagga veliger collection and testing efforts while Trussell and OCWD supported veliger collection, dose-response testing, and water quality assessment. MWD staff supported veliger collection efforts with access to the facility and towing/collection locations.

The veliger mortality results for the 2020 Weymouth WTP influent are summarized in **Table 3.3** and presented graphically by temperature condition in **Appendix F.**

Table 3.3. Average mortality with standard deviation (n=3) of veligers from Weymouth Water Treatment Plant in 2020 as a function of EarthTec QZ dose, temperature, and exposure time

		0.5 hr.	2 hr.	5 hr.	24 hr.
Control					
15°C	End*	2.2±1.6%	1.2±1.7%	6.1±5.1%	2.7±2.0%
	Recovery**	13.7±9.7%	18.6±4.0%	16.2±13.0%	14.3±2.0%
20°C	End	3.4±2.4%	1.9±1.4%	4.3±1.7%	0.8±1.2%
	Recovery	3.1±1.3%	13.2±2.4%	11.8±6.9%	0.9±1.2%
3 µL/L as EarthTec (0.18 mg/L as Cu)					
15°C	End	4.7±3.5%	2.0±0.5%	20.0±3.6%	78.8±9.6%
	Recovery	NA	NA	52±14.7%	90.4±4.7%
20°C	End	4.3±2.4%	7.8±2.4%	18.4±1.9%	88.7±1.3%
	Recovery	NA	NA	38.8±8.0%	94.8±1.4%
16.7 µL/L as EarthTec (1.0 mg/L as Cu)					
15°C	End	3.1±2.2%	27.4±10.9%	39.7±3.2%	100±0.0%
	Recovery	NA	62.0±12.6%	87.5±9.1%	NA
20°C	End	4.1±2.9%	22.5±2.8%	36.1±4.4%	100±0.0%
	Recovery	NA	56.1±3.0%	95±1.8%	NA
33.4 µL/L as EarthTec (2.0 mg/L as Cu)					
15°C	End	27.9±9.4%	27.9±0.7%	68.6±10.2%	100±0.0%
	Recovery	53.5±9.2%	77.1±8.1%	93.1±5.2%	NA
20°C	End	20.1±7.7%	44.7±7.4%	57.6±10.1%	100±0.0%
	Recovery	61.1±11.1%	71.3±3.1%	97.6±1.7%	NA
50.1 µL/L as EarthTec (3.0 mg/L as Cu)					
15°C	End	15.4±8.4%	38.1±9.5%	59.7±4.5%	100.0±0.0%
	Recovery	47.7±5.2%	80.8±12.1%	98.3±2.4%	NA
20°C	End	18.3±6.8%	34.7±8.7%	75.9±0.6%	99.7±0.5%
	Recovery	60.7±3.7%	96.1±1.7%	99.8±0.3%	99.9±0.1%

* End is the end of the exposure period.
** Recovery is at the end of the 24-hour recovery period following the exposure period.
NA=Not Assessed

In the Weymouth WTP influent samples, significant veliger mortality was observed at 3 (0.18), 16.7(1.0), 33.4(2.0), and 50.1(3.0) µL/L as EarthTec QZ (mg/L as Cu) at both temperatures, following 24 hours of exposure time. Mortality of the 3 µL/L EarthTec QZ was much higher than control at 5 and 24 hours of exposure. At 5 hours of exposure, the veligers exposed to all concentrations tested (3, 16.7, 33.4 and 50.1 µL/L EarthTec QZ) began to demonstrate mortality greater than the control mortality. It took 24 hours for the veligers exposed to 16.7, 33.4 and 50.1 µL/L EarthTec QZ to reach mortality of 100%. Higher veliger mortality was observed at the lower temperature of 15ºC, compared with the 20ºC test conditions; however, temperature was not a significant variable in the probit regression model to predict mortality.

End mortality results greater than 15% were checked for additional recovery mortality. The recovery procedure did show a significant increase in mortality results, both observationally and statistically.

Increased mortality in the recovery was due to veligers that showed signs of stress at the initial reading but were not quite dead enough to be stained by the fast green and counted as dead. With the additional recovery time, the stressed veligers were dead and stained. With this set of test subjects, the control mortality was less than 20% for all exposure durations and temperatures. The recovery mortality was generally higher than the initial end mortality. After the recovery period in the 5-hour treatments greater than 90% mortality was achieved in the three highest doses (16.7, 33.4, and 50.1 µL/L EarthTec QZ). The 2-hour treatments reached greater than 70% mortality in the two highest concentrations (33.4 and 50.1 µL/L EarthTec QZ).

Water quality parameters that were measured throughout testing included temperature, dissolved oxygen, pH, specific conductance, free and total copper, and chlorine. The average temperatures measured for the 15°C and 20°C test conditions were 15.3°C and 20.3°C, respectively. Varying the dose of EarthTec QZ did not produce notable changes in either the dissolved oxygen or the specific conductance in any of the test conditions. A statistically significant decrease in pH was observed with increasing EarthTec QZ concentrations, however the pH for each individual test condition remained mostly consistent over time. The full range of tested conditions resulted in pH values that were consistent with those under which veligers can thrive. Consistent with the findings from Lake Piru and Lake Mathews, free and total copper readings were very similar for each tested condition. As with the other test locations, the copper concentrations decreased slightly over time for a given dose of EarthTec QZ, but after 24 hours of exposure, the measured copper concentration was not significantly different from the original treatment concentration. Copper concentrations in the recovery were near zero. The measured chlorine concentrations were negligible. A table of detailed water quality measurements from the 2020 Weymouth WTP testing is included in **Appendix G**.

3.4 Lake Mathews 2021

For the fourth round of dose-response veliger testing was completed at Lake Mathews, a reservoir located in Riverside County, CA that is owned and operated by MWD. Samples of quagga veligers were collected at Lake Mathews on June 30, 2021 for the 15°C dose-response testing with EarthTec QZ with testing occurring June 30 to July 2. The 20°C dose-response testing with EarthTec QZ occurred July 14 to 16 with veligers and raw water collection occurring on the morning of July 14. Both sampling events resulted in a sufficient count of quagga mussel veligers for testing. KASF Consulting led the quagga veliger collection and testing efforts while Trussell and OCWD supported veliger collection, dose-response testing, and water quality assessment. MWD staff supported veliger collection efforts with access to the facility and towing/collection locations.

The veliger mortality results for the 2021 Lake Mathews testing are summarized in **Table 3.4** and graphed by temperature condition in **Appendix H**.

Table 3.4. Average mortality and standard deviation (n=3) of veligers from Lake Mathews in 2021 as a function of EarthTec QZ dose, temperature, and exposure time

		0.5 hr.	2 hr.	5 hr.	24 hr.
Control					
15°C	End*	0.0±0.0%	0.5±0.7%	0.0±0.0%	0.7±1.0%
	Recovery**	NA	4.7±4.1%	12.3±10.9%	1.2±1.2%
20°C	End	1.8±0.9%	0.0±0.0%	0.0±0.0%	1.0±0.8%
	Recovery	NA	2.3±1.4%	2.2±0.3%	18.1±5.3%
3 µL/L as EarthTec (0.18 mg/L as Cu)					
15°C	End	0.8±0.6%	0.0±0.0%	0.9±1.0%	12.1±3.3%
	Recovery	NA	NA	NA	25.5±4.1%
20°C	End	0.8±0.6%	2.6±0.9%	0.0±0.0%	7.4±0.9%
	Recovery	NA	NA	NA	NA
16.7 µL/L as EarthTec (1.0 mg/L as Cu)					
15°C	End	0.8±0.2%	2.4±1.7%	8.7±3.0%	85.6±1.0%
	Recovery	NA	NA	NA	93.3±0.8%
20°C	End	1.2±0.6%	0.3±0.4%	19.5±8.3%	68.5±2.1%
	Recovery	NA	NA	29.1±6.6%	80.7±2.1%
33.4 µL/L as EarthTec (2.0 mg/L as Cu)					
15°C	End	1.3±1.0%	3.3±2.4%	31.5±8.9%	96.7±1.0%
	Recovery	NA	15.6±5.5%	65.8±3.5%	97.2±1.4%
20°C	End	5.3±2.0%	11.2±6.2%	48.5±3.6%	87.3±8.6%
	Recovery	NA	NA	64.0±4.3%	94.7±1.2%
50.1 µL/L as EarthTec (3.0 mg/L as Cu)					
15°C	End	1.9±0.7%	1.8±0.6%	40.0±9.2%	99.5±0.7%
	Recovery	NA	40.9±4.3%	89.0±0.8%	98.9±0.8%
20°C	End	7.3±1.0%	20.7±5.6%	48.0±2.0%	94.4±0.8%
	Recovery	NA	43.4±3.1%	83.3±3.5%	97.8±0.8%

* End is the end of the exposure period.
** Recovery is at the end of the 24-hour recovery period following the exposure period.
NA=Not Assessed

In the Lake Mathews samples, significant quagga veliger mortality was observed with the 16.7(1.0), 33.4(2.0), and 50.1(3.0) µL/L as EarthTec QZ (mg/L as Cu) test conditions at both temperatures after 24 hours of exposure. Mortality of the 3 µL/L EarthTec QZ was greater than the control at 15 and 20°C, but was not greater than 20%. At 5 hours of exposure, the veligers exposed to the two highest concentrations (33.4 and 50.1 µL/L EarthTec QZ) began to demonstrate mortality greater than 30%, but 50% mortality was not achieved. It took 24 hours for the veligers exposed to 16.7, 33.4 and 50.1 mg/L µL/L EarthTec QZ to reach mortality of 85% or greater, except in the 24 h 16.7 µL/L EarthTec QZ tests. Higher veliger mortality was observed at the lower temperature of 15ºC, compared with the 19ºC test conditions. Temperature was not a significant variable in the probit regression model.

Only the end mortality results greater than 12% were checked for additional recovery mortality after 24 hours. Additionally, there were two testing concentrations at 2 hours (33.4 and 50.1 µL/L EarthTec QZ) that had low initial mortality that were checked for additional mortality to ensure that the results were less than 50%. These two concentrations at 15°C resulted in increased mortality but was less than 50%. The recovery procedure did show a significant increase in mortality results both observationally and was statistically significant. Increased mortality in the recovery was due to veligers showing signs of stress at the initial reading but were not quite dead enough to be stained by the fast green and counted as dead. With the additional recovery time, the stressed veligers were dead and stained. The recovery mortality was high with greater than 80% mortality being achieved in the three highest concentrations. The 5 h 50.1 µL/L EarthTec QZ reached high mortality (greater than 80%).

Water quality parameters that were measured throughout testing included temperature, dissolved oxygen, pH, specific conductance, free and total copper, and chlorine. The average temperatures recorded for the 15°C and 20°C test conditions were 15.0°C and 20.9°C, respectively. Varying the dose of EarthTec QZ did not produce notable changes in either the dissolved oxygen or the specific conductance in any of the test conditions. The pH decreased with increased concentration and each concentrations pH was significantly different from each other during testing. The pH for each individual test condition remained mostly consistent over time. The full range of tested conditions resulted in pH values that were consistent with those under which veligers can thrive. Free and total copper readings were very similar for each tested condition. Over time the copper concentrations decreased slightly for a given dose of EarthTec QZ, but after 24 hours of exposure, the measured copper concentration was not significantly different from the original treatment concentration. Copper concentrations in the recovery were near zero. The measured chlorine concentrations were negligible. A table of detailed water quality measurements from the 2021 Lake Mathews testing is included in **Appendix I**.

3.5 Weymouth Water Treatment Plant Influent 2021

The fifth round of dose-response veliger testing was completed on June 28th through 30th, 2021 and July 12th through 14th, 2021 using veligers and raw water collected from the influent water to MWD's Weymouth WTP. Collection of veligers and water samples were taken June 30th and July 12th, 2021; both days resulted in a sufficient count of quagga mussel veligers for testing. KASF Consulting led the quagga veliger collection and testing efforts while Trussell and OCWD supported veliger collection, dose-response testing, and water quality assessment. MWD staff supported veliger collection efforts with access to the facility and towing/collection locations.

The veliger mortality results for the Weymouth WTP influent are summarized in **Table 3.5** and presented graphically by temperature condition in **Appendix J.**

Table 3.5. Average mortality with standard deviation (n=3) of veligers from Weymouth Water Treatment Plant in 2021 as a function of EarthTec QZ dose, temperature, and exposure time

		0.5 hr.	2 hr.	5 hr.	24 hr.
Control					
15°C	End*	1.8±0.8%	3.4±1.2%	4.3±0.4%	5.2±5.8%
	Recovery**	NA	6.7±1.8%	13.4±2.3%	11.3±7.8%
20°C	End	2.5±0.3%	5.0±1.7%	4.2±2.6%	5.6±2.3%
	Recovery	18.9±2.2%	13.5±4.0%	17.0±4.3%	5.5±1.5%
3 µL/L as EarthTec (0.18 mg/L as Cu)					
15°C	End	4.0±1.8%	5.0±1.3%	7.4±2.5%	81.0±7.0%
	Recovery	NA	NA	NA	88.4±1.4%
20°C	End	2.9±0.9%	8.8±0.9%	26.2±4.8%	90.4±3.8%
	Recovery	NA	NA	56.3±8.9%	91.0±1.5%
16.7 µL/L as EarthTec (1.0 mg/L as Cu)					
15°C	End	4.7±0.3%	32.7±10.1%	77.4±10.5%	98.8±0.9%
	Recovery	NA	68.6±4.0%	96.7±1.0%	99.2±0.6%
20°C	End	11.1±1.9%	43.2±3.5%	79.4±2.5%	99.8±0.3%
	Recovery	NA	75.7±1.7%	95.4±1.8%	100±0.0%
33.4 µL/L as EarthTec (2.0 mg/L as Cu)					
15°C	End	11.5±4.5%	66.1±9.4%	96.9±2.6%	100±0.0%
	Recovery	NA	90.0±7.2%	100±0.0%	NA
20°C	End	17.7±1.4%	53.2±12.1%	94.3±2.1%	99.7±0.4%
	Recovery	69.7±8.0%	98.2±0.7%	99.0±0.8%	100±0.0%
50.1 µL/L as EarthTec (3.0 mg/L as Cu)					
15°C	End	12.3±5.6%	94.2±1.8%	98.4±2.2%	100.0±0.0%
	Recovery	NA	NA	100±0.0%	NA
20°C	End	36.0±14.1%	57.2±9.0%	97.7±1.0%	100±0.0%
	Recovery	84.6±4.0%	98.2±0.3%	100±0.0%	NA

* End is the end of the exposure period.
** Recovery is at the end of the 24-hour recovery period following the exposure period.
NA=Not Assessed

In the 2021 Weymouth WTP influent samples, significant veliger mortality was observed at 3 (0.18), 16.7(1.0), 33.4(2.0), and 50.1(3.0) µL/L as EarthTec QZ (mg/L as Cu) at both temperatures, following 24 hours of exposure time. Mortality of the 3 µL/L EarthTec QZ was much higher than control at 5 (20 ºC) and 24 hours of exposure. At 5 hours of exposure, the veligers exposed to all concentrations tested (3, 16.7, 33.4 and 50.1 µL/L EarthTec QZ) began to demonstrate mortality greater than the control mortality. It took 24 hours for the veligers exposed to 50.1 µL/L EarthTec QZ to reach mortality of 100%. Higher veliger mortality was observed at the lower temperature of 15ºC, compared with the 20ºC test conditions; however, temperature was not a significant variable in the probit regression model to predict mortality.

End mortality results greater than 15% were checked for additional recovery mortality. The recovery procedure did show a significant increase in mortality results, both observationally and statistically. The recovery mortality was higher than the initial end mortality. After the recovery period in the 5-hour treatments greater than 95% mortality was achieved in the three highest doses (16.7, 33.4, and 50.1 µL/L EarthTec QZ). The 2-hour treatments reached greater than 65% mortality in the three highest concentrations (16.7, 33.4 and 50.1 µL/L EarthTec QZ). An error in testing occurred where the 15°C 50.1 µL/L EarthTec QZ 2 hour was not read for recovery, but the initial end mortality showed that high veliger mortality was achieved. Increased mortality in the recovery was due to veligers that showed signs of stress at the initial reading but were not quite dead enough to be stained by the fast green and counted as dead. With the additional recovery time, the stressed veligers were dead and stained. With this set of test subjects, the control mortality was less than 20% for all exposure durations and temperatures.

Water quality parameters that were measured throughout testing included temperature, dissolved oxygen, pH, specific conductance, free and total copper, and chlorine. The average temperatures measured for the 15°C and 20°C test conditions were 14.2°C and 20.9°C, respectively. Varying the dose of EarthTec QZ did not produce notable changes in either the dissolved oxygen or the specific conductance in any of the test conditions. A statistically significant decrease in pH was observed with increasing EarthTec QZ concentrations, however the pH for each individual test condition remained mostly consistent over time. The full range of tested conditions resulted in pH values that were consistent with those under which veligers can thrive. Consistent with the findings from Lake Piru and Lake Mathews, free and total copper readings were very similar for each tested condition. As with the other test locations, the copper concentrations decreased slightly over time for a given dose of EarthTec QZ, but after 24 hours of exposure, the measured copper concentration was not significantly different from the original treatment concentration. Copper concentrations in the recovery were near zero. The measured chlorine concentrations were negligible. A table of detailed water quality measurements from Weymouth WTP testing is included in **Appendix K**.

4 Analysis

The following is a high-level analysis of the results shown in **Section 3**. Further discussion, correlations, statistical analysis, integrated analyses and recommendations will be provided in the Final Report.

To begin comparing dose-response results at the three sites, veliger mortality of all tested locations were grouped by EarthTec QZ concentration in **Appendix L**. As previously mentioned, a more comprehensive analysis of these results will be included in the Final Report.

Mortality of veligers was achieved using EarthTec QZ at the three sample locations over the three years. Weymouth WTP veligers in both 2020 and 2021 had the highest mortality (>75% mortality) using the lowest dose of EarthTec QZ (3 µL/L EarthTec QZ)

after 24 hours exposure time. The higher concentrations tested on Weymouth WTP veligers resulted in 100% mortality at both temperatures. Mortality of veligers from the 2020 Lake Mathews testing was greater than 90% at 16.7, 33.4, and 50.1 µL/L EarthTec QZ for both temperatures tested at 24 hours of exposure time, but mortality in 2021 was lower for 16.7 µL/L EarthTec QZ achieving 86 and 69% mortality for the 15 and 20°C tests. Veligers from Lake Piru had the lowest mortality after 24 hours of exposure, but >85% mortality was achieved at the highest concentration tested (50.1 µL/L EarthTec QZ). In all test locations, the 15°C tests had higher mortality than the 20°C tests.

This study design used the fast green stain method to quickly analyze the health status of the veligers. In the Lake Piru samples, the end mortality was similar to the recovery mortality. The stain easily identified the veligers that were dead and additional mortality was not observed. In both the Lake Piru and Weymouth WTP samples, the recovery mortality was much higher than the end mortality, indicating that there was additional mortality after being evaluated with the fast green stain. Mortality in the 2- and 5-hour exposure times in the Weymouth WTP and Lake Mathews water increased at least 30% with the additional recovery time. Control mortality for the same exposure time increased about 10%. The fast green stain was working well, but stressed individuals did not stain. Their tissue did look abnormal and swollen, but there was still movement in some individuals with this condition. With the additional time of recovery, the fast green stain adhered to the dead tissue showing that mortality increased. If fast green stain is used and veligers are observed with abnormal and swollen tissue, the use of a recovery period is recommended.

The water quality parameters measured during testing did not show any variances around the potential differences in mortalities seen. The temperature, dissolved oxygen, pH, and conductivity were within ranges that veligers survive and flourish. Chlorine concentrations were low, showing that the test results were not confounded by the presence of chlorine. The free and total copper concentrations were consistently similar, indicating that the free copper ion (i.e., Cu^{2+}) was dominant, rather than copper in the particulate or complexed form. In the recovery step, the copper concentrations were minimal in all testing water, suggesting that any copper absorption by the veligers with exposure to EarthTec QZ was not released once the veligers were returned to source water without EarthTec QZ.

Differences in source water quality were observed (**Appendix A**), which likely factored into the efficacy of EarthTec QZ on the veligers. Lake Piru is a reservoir that is filled mainly with local runoff with some SWP, whereas Lake Mathews is supplied by the CRA system. The Weymouth WTP influent at the time of the sampling was supplied from Lake Mathews, but is chlorinated as it enters the pipeline. The differences in water quality include the following:

- Alkalinity and hardness were significantly higher at Lake Piru compared with the other locations; average values of 171 and 315 mg/L as $CaCO_3$, respectively, were measured at Lake Piru. The alkalinity and hardness of the 2020 Lake Mathews testing waters were the same as Weymouth WTP

influent samples (average values of 118 and 142 mg/L as $CaCO_3$, respectively. The 2021 alkalinity for Lake Mathews and Weymouth WTP (average 136 mg/L as $CaCO_3$) were similar to each other and higher than the 2020 levels. Hardness for 2021 did vary significantly between the two sampling events, but Lake Mathews and Weymouth WTP values were similar. During the first week of testing at 15ºC the hardness mean value was 275 mg/L as $CaCO_3$ and the second week of testing at 20 ºC the hardness mean value was 182 mg/L as $CaCO_3$.

- Water temperatures at Lake Piru (average 13.4ºC) were significantly lower compared to those measured from samples collected from Lake Mathews (average 23ºC) or Weymouth WTP influent (average 26.5ºC). These temperature differences account for seasonal changes in the surface water. Lake Piru testing was completed in December 2019, whereas the testing associated with the Lake Mathews and Weymouth WTP influent samples were completed in September 2020 and July 2021. The higher water temperatures in September 2020 and July 2021 at Lake Mathews and post-pipeline at the downstream Weymouth WTP influent increases veliger metabolism, which could contribute to the sensitivity to EarthTec QZ.
- Lake Piru had a slightly higher average pH (8.34) compared to that of the 2020 Lake Mathews and Weymouth WTP influent (8.19 and 8.08, respectively). The 2021 pH of Lake Mathews and Weymouth WTP influent (8.27 and 8.23, respectively) were higher than 2020, but still lower than Lake Piru.
- The turbidity values at Lake Piru (average 7.94 NTU) were also elevated, compared to the 2020 and 2021 Lake Mathews and Weymouth WTP influent (0.74 and 0.67 NTU, respectively).

The size distribution of the veligers collected from Lake Piru had a large number of each size class present (**Table 4.1**). The veligers from the 2020 Lake Mathews and Weymouth WTP were mostly comprised of the smaller size class of D-shaped veligers. In the 2021 Lake Mathews samples the veligers were larger with less D-shaped veligers present. The 2021 Weymouth WTP had many small veligers present and only a few larger veligers, but the distribution of larger veligers increased in the 20°C tests. The veligers that were observed to survive EarthTec QZ addition tended to be pediveliger to large umbonal in size in all treatment waters. Therefore, the toxicity of EarthTec QZ for larger sized veligers may be lower in these two locations. Collection of veligers was completed within 2 days for the Lake Piru and 2020 Weymouth WTP testing and resulted in the same distribution of size classes. The 2020 and 2021 Lake Mathews and 2021 Weymouth WTP collections were taken 2 weeks apart and the distribution of size class was different with the later sampling event (20°C) having larger veligers, as reflected in **Table 4.1**.

Table 4.1. Size class distribution of veligers used in testing at each sample site

	Size Class			
Sample Site	**D-shaped (50-150 µm)**	**Small umbonal (150-250 µm)**	**Large umbonal (200-350 µm)**	**Pediveliger (350-500 µm)**
Lake Piru (15 & 20°C)	45%	15%	20%	20%
2020 Lake Mathews (15ºC)	75%	20%	4%	1%
2020 Lake Mathews (19ºC)	50%	30%	15%	5%
2020 Weymouth WTP (15 & 20°C)	80%	15%	3%	2%
2021 Lake Mathews (15ºC)	52%	22%	23%	3%
2021 Lake Mathews (20ºC)	32%	18%	38%	11%
2021 Weymouth WTP (15ºC)	74%	8%	17%	2%
2021 Weymouth WTP (20ºC)	53%	25%	22%	1%

5 References

Trussell Technologies (2019a). "Scientific Collection Permit, Specific Use – Application." Application for *California Department of Fish and Wildlife*. November 6.

Trussell Technologies (2019b). "Test Plan Evaluation of EarthTec QZ® for Use in the Restoration of Local Recharge Sources from Invasive Dreissenid Mussels." Report for *Orange County Water District*. November 15.

Appendix List

Appendix A: Raw Water Quality of Lake Piru, Lake Mathews and Weymouth WTP Influent

Appendix B: Lake Piru Veliger Dose-Response Figures

Appendix C: Lake Piru Dose-Response Veliger Testing – Water Quality Data

Appendix D: 2020 Lake Mathews Veliger Dose-Response Figures

Appendix E: 2020 Lake Mathews Dose-Response Veliger Testing – Water Quality Data

Appendix F: 2020 Weymouth Water Treatment Plant Influent Veliger Dose-Response Figures

Appendix G: 2020 Weymouth WTP Dose-Response Veliger Testing – Water Quality Data

Appendix H: 2021 Lake Mathews Veliger Dose-Response Figures

Appendix I: 2021 Lake Mathews Dose-Response Veliger Testing – Water Quality Data

Appendix J: 2021 Weymouth Water Treatment Plant Influent Veliger Dose-Response Figures

Appendix K: 2021 Weymouth WTP Dose-Response Veliger Testing – Water Quality Data

Appendix L: Veliger Mortality at All Sites Grouped by EarthTec QZ Concentration

Appendix A

Raw Water Quality of Lake Piru, Lake Mathews and Weymouth WTP Influent

Table A1. Individual raw water quality measurements for Lake Piru, Lake Mathews, and Weymouth WTP

Location		Parameter	Total Copper	Free Copper	Total Chlorine	Oxidation Reduction Potential	Dissolved Oxygen	Conductivity	pH	Temperature	Alkalinity	Hardness	Turbidity	Dissolved Organic Carbon	Chemical Oxygen Demand
			mg/L	mg/L	mg/L	mV	mg/L	µS/cm	pH unit	°C	mg/L as CaCO3		NTU	mg/L	mg/L
Lake Piru	**Raw Water Quality**														
	2019	15ºC Tests	0.15	0.14	0.08	210.4	10.45	927	8.43	12.0	187	314	9.29	5.33	10.00
		20ºC Tests	0.16	0.16	0.12	191.9	8.88	913	8.25	14.8	154	316	6.59	5.05	5.65
Lake Mathews	**Raw Water Quality**														
	2020	15ºC Tests	0.02	0.02	0.02	212.5	7.57	946	8.18	25.1	115	138	0.61	3.01	4.67
		19ºC Tests	0.01	0.01	0.02	245	8.08	937	8.20	18.9	115	142	0.89	2.95	5.69
		20ºC Tests *	0.02	0.02	0.10	244.3	7.86	948	8.21	25.8	116	144	0.92	2.91	7.83
	2021	15ºC Tests	0.00	0.00	0.01	185.6	8.46	950	8.24	24.6	138	281	0.56	2.98	4.65
		20ºC Tests	0.00	0.00	0.00	208.2	8.34	957	8.29	23.5	135	178	0.92	2.91	3.30
Weymouth WTP	**Raw Water Quality**														
	2020	15ºC Tests	0.01	0.01	0.04	214	8.27	944	8.02	25.6	119	150	0.76	4.09	8.50
		20ºC Tests	0.00	0.00	0.07	223.5	8.14	941	8.13	27.1	121	139	0.69	4.19	8.24
		Additional Sampling **	0.02	0.02	0.05	211	8.19	955	8.12	26.8	119	145	0.64	4.05	7.88
	2021	15ºC Tests	0.02	0.03	0.10	195.6	8.69	969	8.18	21.9	137	275	0.61	3.01	4.46
		20ºC Tests	0.00	0.00	0.02	218.2	8.34	961	8.28	23.9	135	186	0.61	3.12	4.86

* During Round #2 testing, the Lake Mathews 20ºC sample water was recollected due to insufficient veliger counts in the original collection. The Lake Mathews 19ºC sample water represents the recollected sample water used for successful veliger testing.
** Re-sample of Weymouth Water Treatment Plant influent during Round #2 of testing for additional acute toxicity testing. This water was NOT used for veliger testing.

Appendix B
Lake Piru Veliger Dose Response Figures

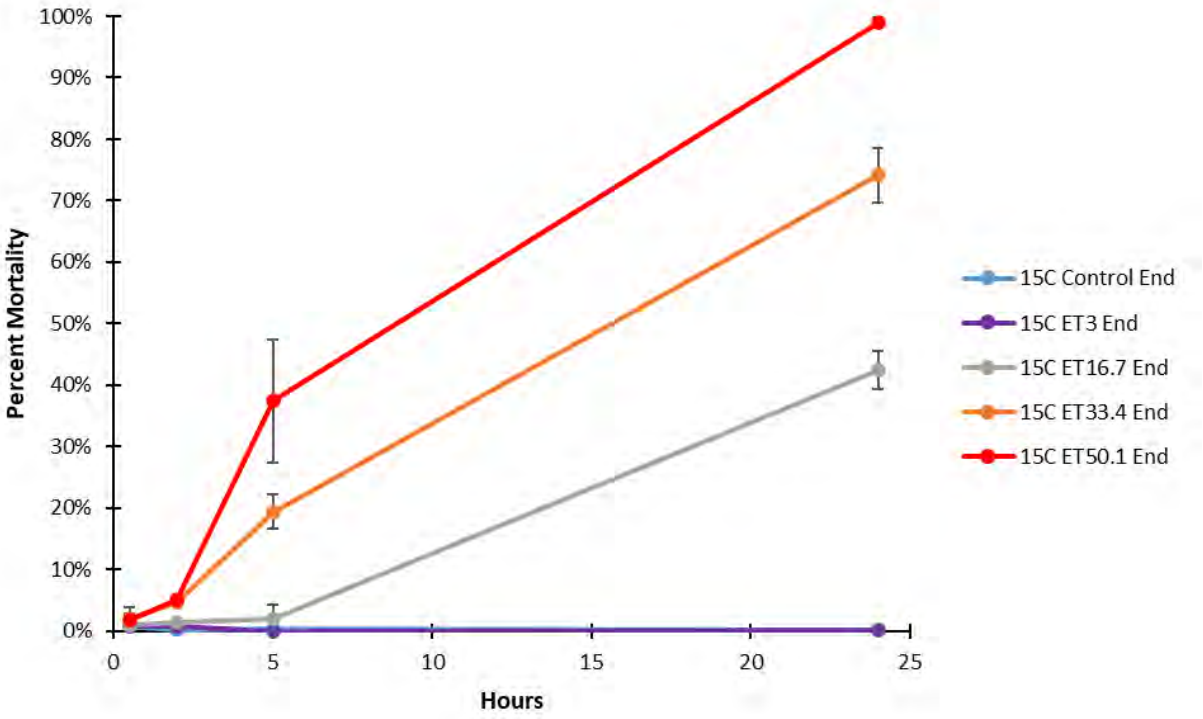

Figure B1. Veliger mortality with standard deviation bars (n=3) at the end of each exposure period at 15°C for Lake Piru trials

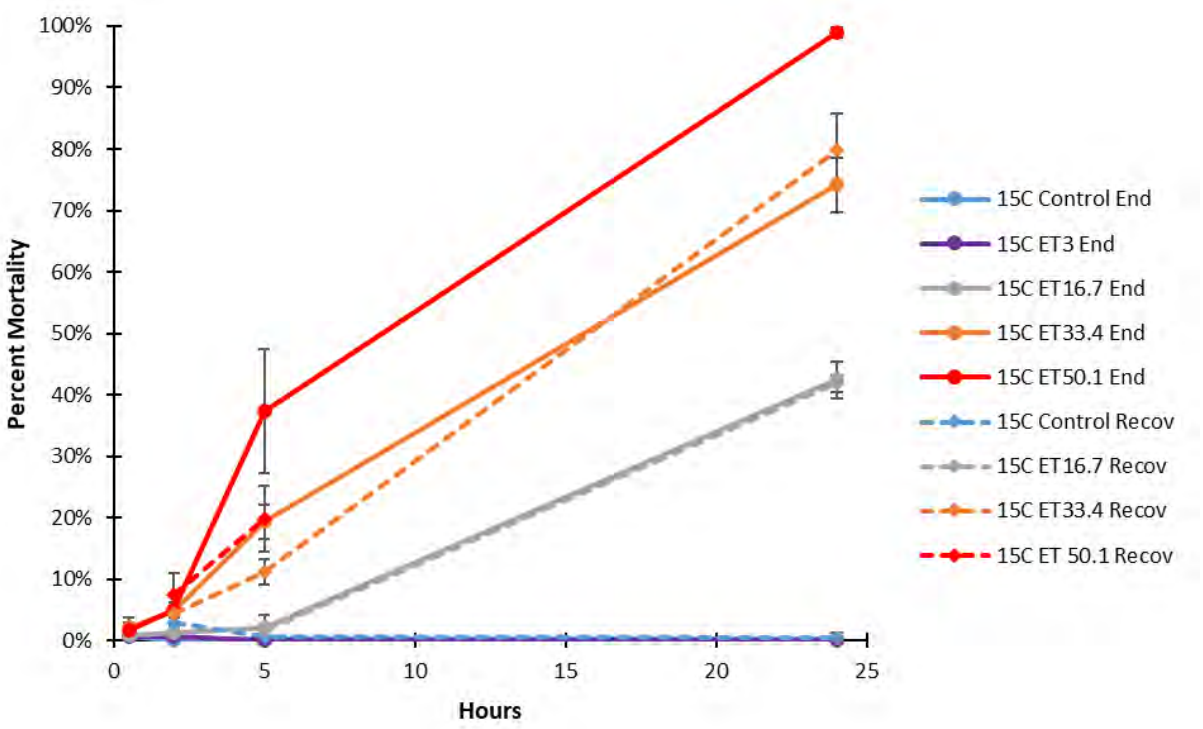

Figure B2. Veliger mortality with standard deviation bars (n=3) at the end of each exposure and recovery period at 15°C for Lake Piru trials

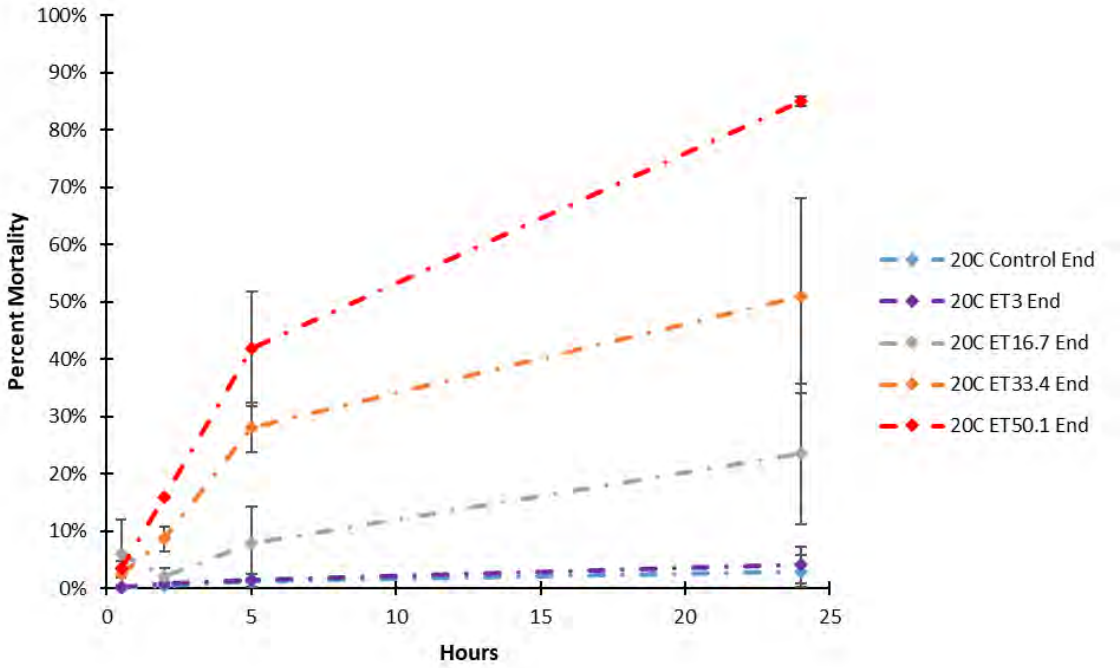

Figure B3. Veliger mortality with standard deviation bars (n=3) at the end of each exposure period at 20°C for Lake Piru trials

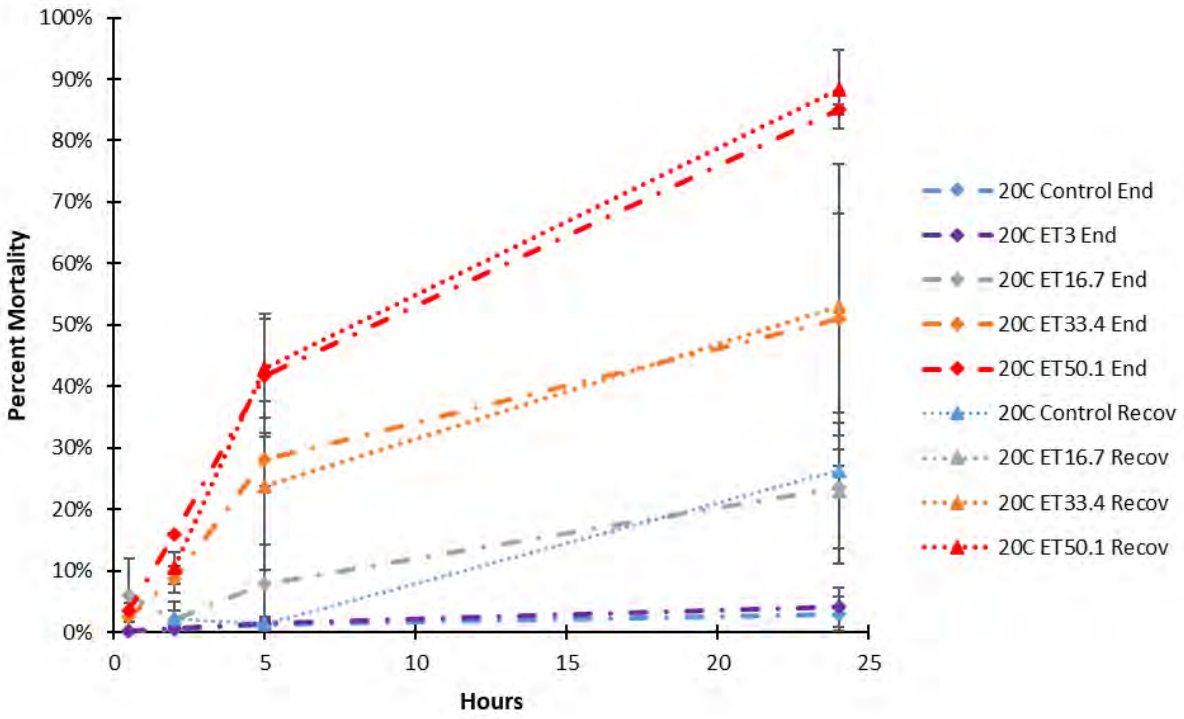

Figure B4. Veliger mortality with standard deviation bars (n=3) at the end of each exposure and recovery period at 20°C for Lake Piru trials

Appendix C

Lake Piru Dose-Response Veliger Testing – Water Quality Data

Table C1. Average with standard deviation of water quality data for each Lake Piru dose-response test condition

		15ºC Test Conditions						20ºC Test Conditions					
	Earth Tec QZ (µL/L)	0 h	0.5 h	2 h	5 h	24 h	Recovery	0 h	0.5 h	2 h	5 h	24 h	Recovery
Temp. (°C)	0.0	14.3 ±0.5	14.9 ±0.0	15.2 ±0.0	15.3 ±0.0	15.1 ±0.0	14.8 ±0.3	17.2 ±1.4	20.7 ±0.1	20.1 ±0.0	20.4 ±0.0	20.3 ±0.0	20.0 ±0.3
	3.0	14.5 ±0.4	14.9 ±0.0	15.4 ±0.2	15.4 ±0.0	15.1 ±0.0	14.8 ±0.2	16.7 ±1.4	21.1 ±0.0	20.0 ±0.0	20.3 ±0.0	20.3 ±0.0	NM
	16.7	14.5 ±0.6	16.1 ±0.0	15.7 ±0.0	15.5 ±0.0	15.0 ±0.0	14.8 ±0.4	17.4 ±1.4	20.4 ±0.0	20.6 ±0.0	19.8 ±0.0	20.2 ±0.0	20.1 ±0.0
	33.4	14.9 ±0.6	15.9 ±0.0	15.5 ±0.0	15.5 ±0.0	15.0 ±0.0	14.8 ±0.2	17.5 ±1.4	20.6 ±0.0	20.6 ±0.0	19.9 ±0.0	20.2 ±0.0	19.9 ±0.3
	50.1	14.6 ±0.6	15.9 ±0.0	15.5 ±0.0	15.6 ±0.0	15.2 ±0.0	15.1 ±0.1	17.1 ±1.5	20.7 ±0.0	20.3 ±0.0	20.1 ±0.0	20.0 ±0.0	20.2 ±0.3
DO (mg/L)	0.0	9.07 ±0.03	8.96 ±0.03	8.80 ±0.04	8.56 ±0.04	9.21 ±0.11	9.07 ±0.25	9.40 ±0.13	9.13 ±0.05	9.16 ±0.52	8.20 ±0.19	5.65 ±0.45	7.11 ±0.44
	3.0	9.05 ±0.05	8.97 ±0.01	8.84 ±0.03	8.55 ±0.06	9.19 ±0.05	9.14 ±0.13	9.60 ±0.12	9.16 ±0.09	9.09 ±0.04	8.28 ±0.18	5.15 ±0.31	NM
	16.7	8.96 ±0.08	8.75 ±0.03	8.82 ±0.02	8.58 ±0.01	9.26 ±0.04	9.24 ±0.08	9.47 ±0.16	8.25 ±0.06	9.07 ±0.06	8.37 ±0.26	6.78 ±0.34	7.06 ±0.36
	33.4	8.98 ±0.12	8.77 ±0.01	8.78 ±0.02	8.60 ±0.02	9.26 ±0.05	9.33 ±0.07	9.54 ±0.12	9.12 ±0.02	8.88 ±0.09	8.21 ±0.13	7.61 ±0.46	7.23 ±0.66
	50.1	8.69 ±0.09	8.75 ±0.02	8.85 ±0.10	8.62 ±0.06	9.71 ±0.00	9.40 ±0.10	9.53 ±0.15	8.17 ±0.07	8.71 ±0.19	8.26 ±0.17	8.09 ±0.02	7.47 ±0.35
pH	0.0	8.50 ±0.10	8.47 ±0.06	8.45 ±0.03	8.49 ±0.02	8.47 ±0.02	8.46 ±0.05	8.31 ±0.03	8.22 ±0.02	8.21 ±0.03	8.15 ±0.03	8.16 ±0.04	8.35 ±0.07
	3.0	8.53 ±0.05	8.47 ±0.04	8.45 ±0.05	8.46 ±0.03	8.5 ±0.01	8.52 ±0.02	8.33 ±0.01	8.22 ±0.01	8.24 ±0.00	8.02 ±0.15	8.12 ±0.02	NM
	16.7	8.46 ±0.09	8.40 ±0.05	8.36 ±0.04	8.32 ±0.07	8.46 ±0.00	8.51 ±0.03	8.19 ±0.03	8.09 ±0.05	8.12 ±0.02	8.06 ±0.00	8.22 ±0.02	8.44 ±0.04
	33.4	8.26 ±0.03	8.35 ±0.02	8.21 ±0.03	8.31 ±0.00	8.40 ±0.01	8.53 ±0.02	8.09 ±0.04	8.02 ±0.01	8.03 ±0.01	7.98 ±0.01	8.24 ±0.04	8.44 ±0.07
	50.1	8.20 ±0.40	8.25 ±0.01	8.16 ±0.01	8.24 ±0.01	8.36 ±0.01	8.53 ±0.02	7.99 ±0.03	7.95 ±0.01	7.94 ±0.01	7.92 ±0.01	8.23 ±0.01	8.41 ±0.09
Specific Conductance (µS/cm)	0.0	950 ±23	938 ±1	936 ±9	940 ±1	949 ±1	946 ±2	930 ±5	938 ±2	944 ±1	947 ±0	966 ±2	965 ±6
	3.0	936 ±1	938 ±1	940 ±1	940 ±1	948 ±1	945 ±2	940 ±2	944 ±1	944 ±1	947 ±1	966 ±1	NM
	16.7	939 ±3	942 ±1	944 ±3	942 ±1	949 ±1	944 ±3	943 ±2	946 ±0	947 ±1	948 ±1	965 ±1	964 ±2
	33.4	941 ±1	943 ±0	944 ±2	943 ±1	950 ±1	944 ±3	943 ±2	948 ±2	949 ±1	951 ±1	964 ±2	962 ±3
	50.1	951 ±6	944 ±1	946 ±2	945 ±0	951 ±0	947 ±1	943 ±4	950 ±0	950 ±1	953 ±0	958 ±1	958 ±3
Free Copper (mg/L)	0.0	0.19 ±0.04	0.21 ±0.03	0.17 ±0.01	0.13 ±0.03	0.13 ±0.01	0.15 ±0.04	0.18 ±0.05	0.23 ±0.02	0.12 ±0.01	0.13 ±0.02	0.18 ±0.04	NM
	3.0	0.32 ±0.04	0.29 ±0.00	0.27 ±0.03	0.32 ±0.06	0.25 ±0.01	0.19 ±0.04	0.31 ±0.04	0.29 ±0.03	0.28 ±0.01	0.28 ±0.00	0.33 ±0.02	NM
	16.7	1.05 ±0.03	1.02 ±0.01	0.89 ±0.04	0.95 ±0.02	1.00 ±0.09	0.21 ±0.04	1.10 ±0.04	1.05 ±0.01	1.06 ±0.05	1.04 ±0.01	1.08 ±0.01	NM
	33.4	1.96 ±0.02	1.93 ±0.03	1.9 ±0.02	1.86 ±0.02	2.13 ±0.20	0.22 ±0.03	2.05 ±0.05	1.98 ±0.01	1.95 ±0.02	1.93 ±0.01	1.91 ±0.02	NM
	50.1	2.93 ±0.09	2.84 ±0.02	2.77 ±0.03	2.75 ±0.04	2.93 ±0.10	0.24 ±0.04	2.97 ±0.09	2.84 ±0.04	2.84 ±0.04	2.77 ±0.01	2.80 ±0.08	NM

	Earth Tec QZ (µL/L)	15ºC Test Conditions						20ºC Test Conditions					
		0 h	0.5 h	2 h	5 h	24 h	Recovery	0 h	0.5 h	2 h	5 h	24 h	Recovery
Total Copper (mg/L)	0.0	0.19 ±0.00	0.14 ±0.02	0.18 ±0.02	0.13 ±0.02	0.10 ±0.02	0.14 ±0.05	0.13 ±0.02	0.13 ±0.00	0.12 ±0.01	0.12 ±0.01	0.12 ±0.02	NM
	3.0	0.29 ±0.02	0.28 ±0.00	0.24 ±0.01	0.29 ±0.03	0.23 ±0.00	0.16 ±0.07	0.32 ±0.02	0.29 ±0.02	0.27 ±0.01	0.30 ±0.01	0.37 ±0.02	NM
	16.7	1.04 ±0.02	1.00 ±0.02	0.98 ±0.02	0.98 ±0.01	1.03 ±0.07	0.17 ±0.08	1.10 ±0.06	1.04 ±0.03	1.07 ±0.06	1.02 ±0.02	1.09 ±0.02	NM
	33.4	1.96 ±0.01	1.93 ±0.05	1.91 ±0.02	1.88 ±0.03	2.14 ±0.18	0.20 ±0.04	2.05 ±0.04	1.96 ±0.01	1.97 ±0.04	1.93 ±0.01	1.93 ±0.02	NM
	50.1	2.91 ±0.07	2.85 ±0.03	2.81 ±0.03	2.78 ±0.05	2.97 ±0.11	0.23 ±0.05	2.96 ±0.09	2.85 ±0.02	2.85 ±0.05	2.80 ±0.00	2.79 ±0.08	NM

NM: Not Measured
Recovery: Follow up measurement at the end of the 24 h recovery period following the exposure period.

Appendix D
2020 Lake Mathews Veliger Dose-Response Figures

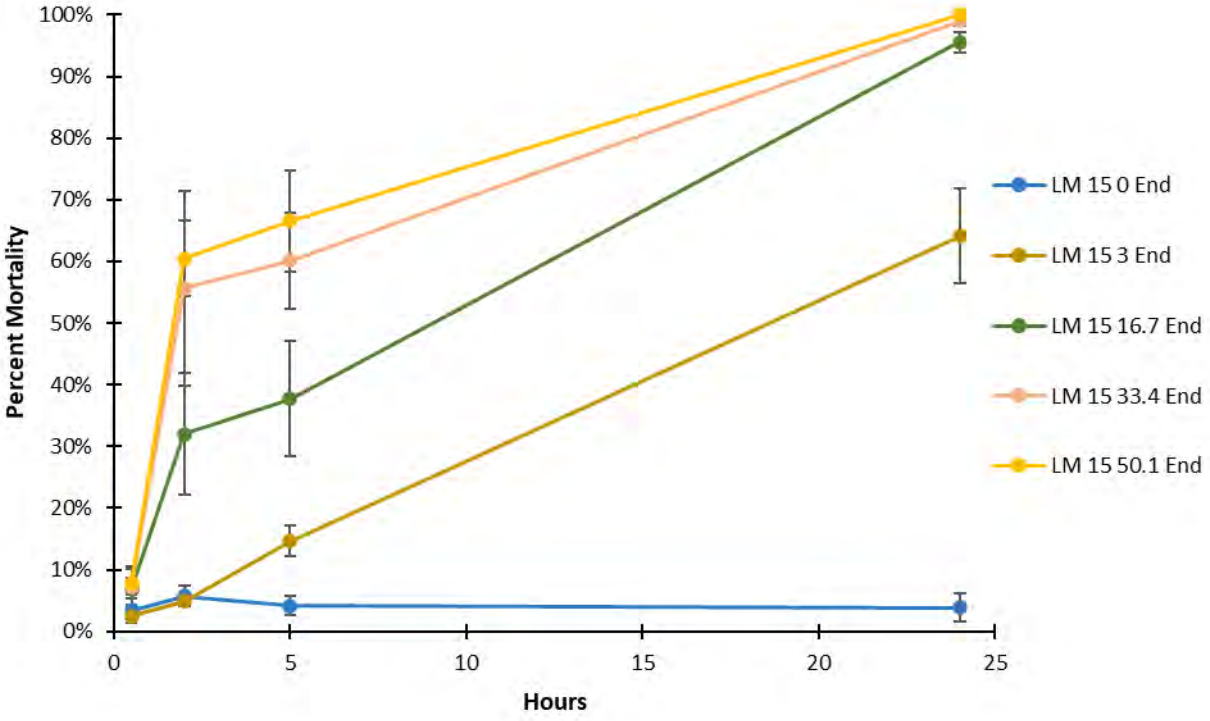

Figure D1. Veliger mortality with standard deviation bars (n=3) at the end of each exposure period at 15°C for 2020 Lake Mathews (LM) trials

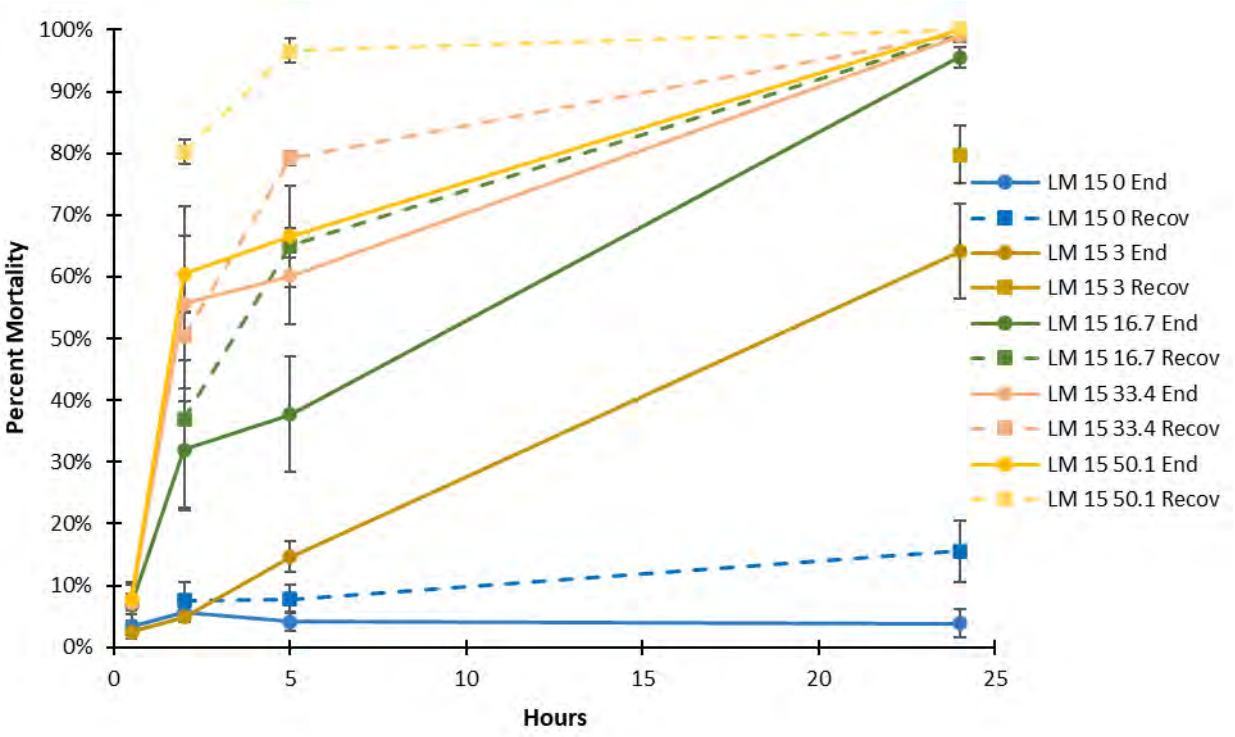

Figure D2. Veliger mortality with standard deviation bars (n=3) at the end of each exposure and recovery period at 15°C for 2020 Lake Mathews (LM) trials

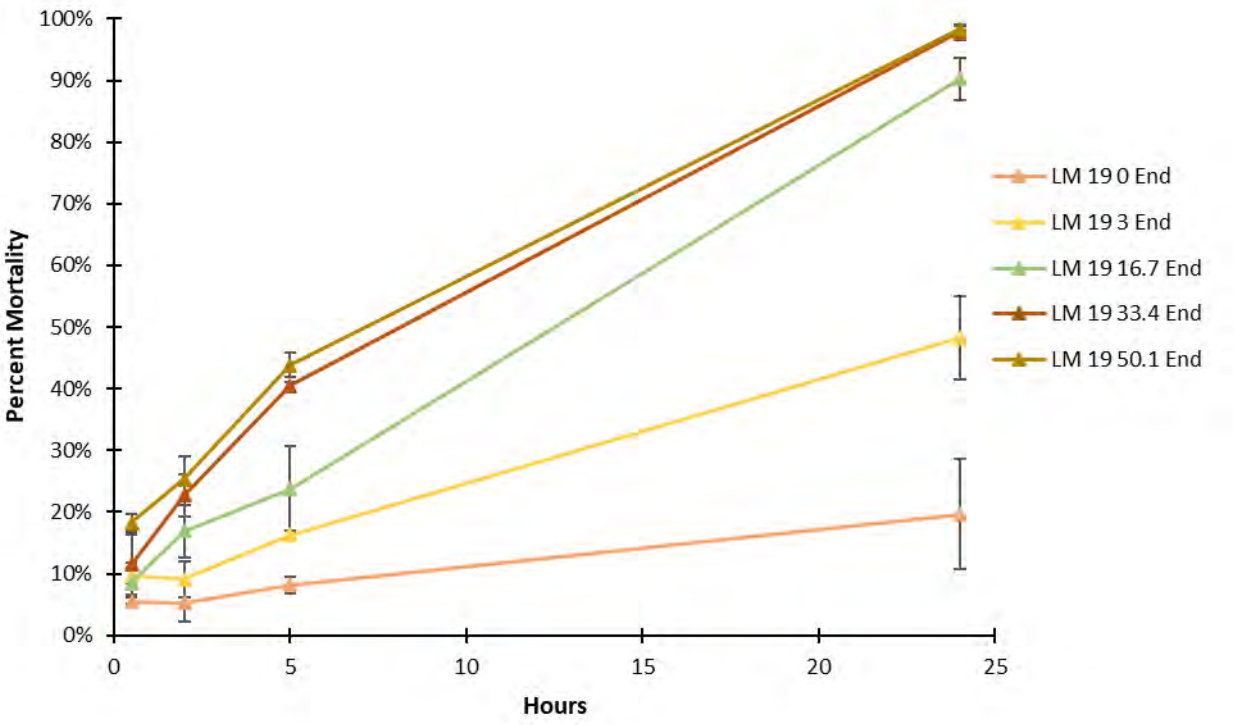

Figure D3. Veliger mortality with standard deviation bars (n=3) at the end of each exposure period at 19°C for 2020 Lake Mathews (LM) trials

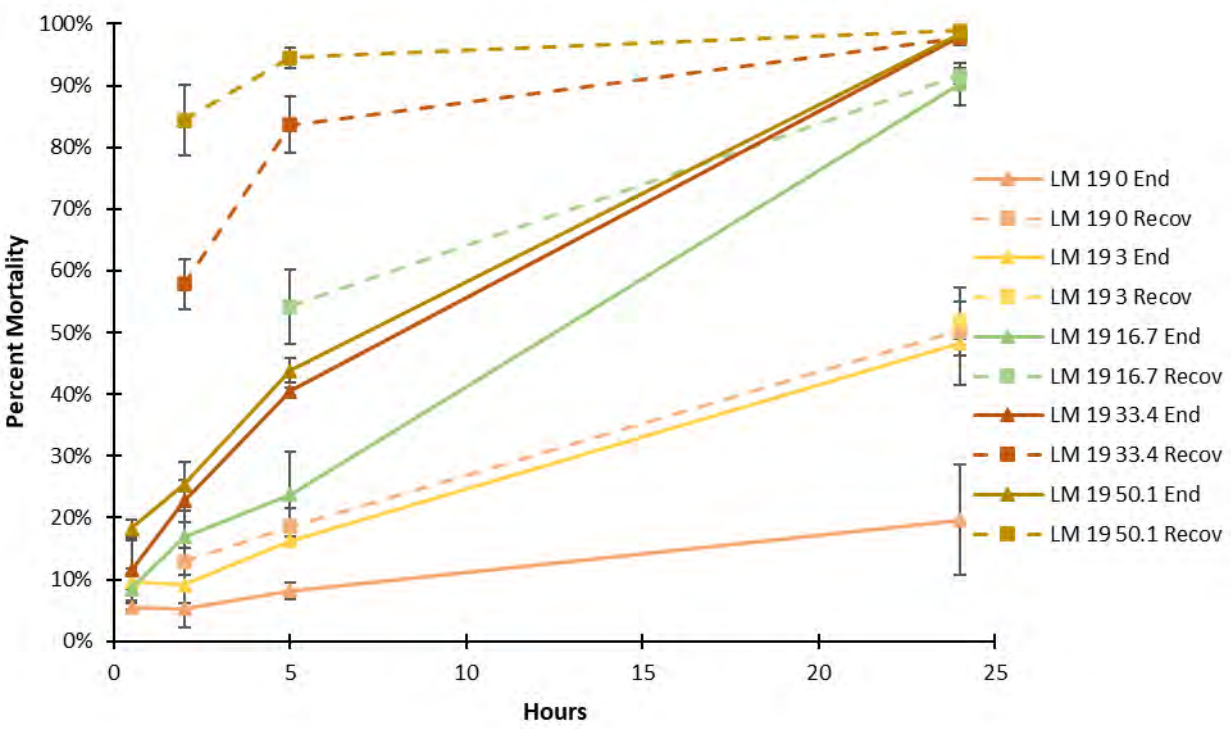

Figure D4. Veliger mortality with standard deviation bars (n=3) at the end of each exposure and recovery period at 19°C for 2020 Lake Mathews (LM) trials

Appendix E

2020 Lake Mathews Dose-Response Veliger Testing – Water Quality Data

Table E1. Average water quality data for each 2020 Lake Mathews dose-response test condition

		15°C Test Conditions						19°C Test Conditions					
	Earth Tec QZ (µL/L)	0 h	0.5 h	2 h	5 h	24 h	Recovery	0 h	0.5 h	2 h	5 h	24 h	Recovery
Temp. (°C)	0.0	17.3 ±0.1	14.9 ±0.1	14.2 ±0.0	14.2 ±0.1	15.1 ±0.0	14.5 ±0.2	21.2 ±0.6	22.5 ±0.1	21.0 ±0.0	21.2 ±0.1	21.2 ±0.8	21.0 ±0.1
	3.0	15.0 ±0.7	14.8 ±0.0	14.2 ±0.0	14.2 ±0.1	15.1 ±0.0	14.5 ±0.1	21.5 ±0.6	20.9 ±0.1	20.9 ±0.0	21.2 ±0.1	21.3 ±0.0	21.3 ±0.0
	16.7	16.9 ±1.2	14.5 ±0.1	13.9 ±0.1	14.4 ±0.0	15.1 ±0.0	14.4 ±0.3	21.7 ±0.5	20.6 ±0.1	20.7 ±0.0	21.2 ±0.0	21.3 ±0.1	21.3 ±0.1
	33.4	15.7 ±2.0	13.6 ±0.3	13.5 ±0.1	15.1 ±0.0	15.1 ±0.0	14.4 ±0.3	22.3 ±0.5	20.8 ±0.1	21.0 ±0.0	21.5 ±0.0	21.3 ±0.0	21.3 ±0.1
	50.1	15.6 ±2.9	13.2 ±0.1	13.6 ±0.0	15.1 ±0.0	15.0 ±0.0	14.5 ±0.4	22.5 ±0.5	20.6 ±0.0	21.0 ±0.0	21.6 ±0.0	21.3 ±0.1	21.2 ±0.1
DO (mg/L)	0.0	7.63 ±0.16	8.10 ±0.21	7.96 ±0.04	7.94 ±0.06	8.68 ±0.10	8.84 ±0.28	8.01 ±0.07	8.11 ±0.05	8.09 ±0.05	7.93 ±0.01	7.58 ±0.07	8.01 ±0.15
	3.0	8.31 ±0.11	8.71 ±0.03	8.59 ±0.06	8.48 ±0.05	8.99 ±0.04	9.11 ±0.07	8.07 ±0.04	8.12 ±0.02	8.02 ±0.02	8.05 ±0.03	7.61 ±0.09	8.09 ±0.04
	16.7	8.33 ±0.27	9.02 ±0.04	8.43 ±0.12	8.40 ±0.03	8.98 ±0.03	8.90 ±0.23	8.14 ±0.05	8.13 ±0.02	8.01 ±0.01	8.06 ±0.02	8.11 ±0.06	8.13 ±0.09
	33.4	8.45 ±0.38	9.24 ±0.03	8.41 ±0.12	8.35 ±0.06	8.92 ±0.07	8.81 ±0.12	8.10 ±0.07	8.17 ±0.06	7.96 ±0.02	7.98 ±0.01	8.33 ±0.09	8.22 ±0.10
	50.1	8.61 ±0.53	9.72 ±0.02	8.44 ±0.09	8.45 ±0.07	9.12 ±0.06	8.73 ±0.13	8.09 ±0.07	8.12 ±0.04	7.99 ±0.01	8.00 ±0.04	8.26 ±0.04	8.49 ±0.14
pH	0.0	8.31 ±0.02	8.35 ±0.00	8.34 ±0.01	8.36 ±0.00	8.26 ±0.01	8.26 ±0.03	8.08 ±0.03	8.13 ±0.00	8.13 ±0.00	8.14 ±0.01	8.15 ±0.03	8.24 ±0.03
	3.0	8.29 ±0.01	8.32 ±0.01	8.32 ±0.01	8.32 ±0.01	8.22 ±0.00	8.27 ±0.00	8.09 ±0.01	8.14 ±0.00	8.11 ±0.01	8.12 ±0.01	8.08 ±0.01	8.25 ±0.01
	16.7	8.20 ±0.03	8.27 ±0.02	8.27 ±0.02	8.23 ±0.02	8.18 ±0.02	8.26 ±0.04	8.06 ±0.02	8.11 ±0.01	8.06 ±0.01	8.09 ±0.02	8.12 ±0.01	8.25 ±0.01
	33.4	8.15 ±0.08	8.19 ±0.04	8.21 ±0.06	8.19 ±0.04	8.12 ±0.03	8.25 ±0.05	7.93 ±0.06	8.05 ±0.03	7.92 ±0.03	7.93 ±0.03	8.09 ±0.03	8.27 ±0.01
	50.1	8.00 ±0.08	8.08 ±0.03	8.08 ±0.02	8.09 ±0.05	8.08 ±0.03	8.25 ±0.05	8.03 ±0.06	7.90 ±0.04	7.75 ±0.03	7.84 ±0.04	8.00 ±0.03	8.30 ±0.02
Specific Conductance (µS/cm)	0.0	939 ±1	938 ±2	937 ±1	936 ±1	940 ±0	939 ±4	947 ±3	947 ±1	948 ±1	948 ±1	954 ±1	954 ±1
	3.0	941 ±0	938 ±0	938 ±2	940 ±0	943 ±1	941 ±0	945 ±2	946 ±0	946 ±0	947 ±0	955 ±1	946 ±0
	16.7	942 ±3	940 ±1	939 ±0	941 ±0	943 ±0	941 ±1	951 ±1	948 ±0	949 ±0	949 ±0	954 ±1	954 ±2
	33.4	945 ±6	942 ±0	944 ±0	946 ±0	947 ±0	941 ±1	955 ±3	954 ±2	952 ±0	954 ±0	958 ±0	952 ±1
	50.1	946 ±4	945 ±0	943 ±0	946 ±0	948 ±1	942 ±1	957 ±1	955 ±1	955 ±0	957 ±0	957 ±0	953 ±2
Free Copper (mg/L)	0.0	0.09 ±0.03	0.10 ±0.02	0.08 ±0.04	0.04 ±0.00	0.05 ±0.01	0.05 ±0.02	0.09 ±0.00	0.11 ±0.00	0.10 ±0.02	0.05 ±0.02	0.07 ±0.01	0.04 ±0.03
	3.0	0.43 ±0.00	0.41 ±0.00	0.41 ±0.01	0.37 ±0.04	0.36 ±0.2	0.07 ±0.02	0.36 ±0.02	0.35 ±0.01	0.34 ±0.02	0.32 ±0.01	0.32 ±0.02	0.05 ±0.00
	16.7	1.25 ±0.02	1.21 ±0.01	1.20 ±0.02	1.18 ±0.05	1.16 ±0.00	0.09 ±0.03	1.03 ±0.01	1.04 ±0.01	1.00 ±0.03	0.95 ±0.01	0.95 ±0.01	0.10 ±0.01
	33.4	2.00 ±0.04	2.02 ±0.02	1.96 ±0.02	1.94 ±0.02	1.86 ±0.01	0.11 ±0.05	1.96 ±0.03	1.94 ±0.01	1.90 ±0.01	1.87 ±0.02	1.64 ±0.05	0.18 ±0.04
	50.1	2.93 ±0.08	2.98 ±0.04	2.90 ±0.05	2.78 ±0.02	2.56 ±0.02	0.13 ±0.04	2.95 ±0.01	2.91 ±0.05	2.83 ±0.01	2.84 ±0.03	2.27 ±0.04	0.36 ±0.13

	Earth Tec QZ (µL/L)	15°C Test Conditions						19°C Test Conditions					
		0 h	0.5 h	2 h	5 h	24 h	Recovery	0 h	0.5 h	2 h	5 h	24 h	Recovery
Total Copper (mg/L)	0.0	0.09 ±0.02	0.06 ±0.01	0.07 ±0.02	0.04 ±0.02	0.06 ±0.01	0.05 ±0.01	0.09 ±0.01	0.10 ±0.00	0.09 ±0.02	0.07 ±0.05	0.08 ±0.01	0.04 ±0.02
	3.0	0.42 ±0.02	0.39 ±0.00	0.40 ±0.01	0.37 ±0.03	0.37 ±0.03	0.07 ±0.02	0.36 ±0.01	0.36 ±0.01	0.33 ±0.04	0.34 ±0.03	0.36 ±0.01	0.05 ±0.00
	16.7	1.25 ±0.03	1.21 ±0.02	1.18 ±0.02	1.19 ±0.06	1.20 ±0.01	0.09 ±0.03	1.04 ±0.02	1.01 ±0.03	0.99 ±0.04	0.94 ±0.01	0.98 ±0.02	0.10 ±0.02
	33.4	2.00 ±0.05	2.01 ±0.01	1.95 ±0.01	1.91 ±0.02	1.86 ±0.02	0.12 ±0.06	1.94 ±0.03	1.93 ±0.02	1.90 ±0.00	1.88 ±0.00	1.64 ±0.04	0.18 ±0.04
	50.1	2.94 ±0.06	2.95 ±0.04	2.87 ±0.05	2.77 ±0.02	2.56 ±0.03	0.14 ±0.05	2.95 ±0.01	2.90 ±0.02	2.84 ±0.02	2.85 ±0.02	2.29 ±0.05	0.36 ±0.15
Total Chlorine (mg/L)	0.0	0.04 ±0.01	0.04 ±0.01	0.03 ±0.01	0.06 ±0.00	0.04 ±0.01	NM	0.07 ±0.00	0.06 ±0.01	0.06 ±0.01	0.05 ±0.02	0.05 ±0.01	NM
	3.0	0.05 ±0.01	0.07 ±0.01	0.03 ±0.00	0.04 ±0.01	0.04 ±0.01	NM	0.06 ±0.01	0.06 ±0.00	0.06 ±0.02	0.07 ±0.01	0.04 ±0.00	NM
	16.7	0.06 ±0.00	0.05 ±0.02	0.05 ±0.00	0.06 ±0.01	0.04 ±0.01	NM	0.08 ±0.01	0.08 ±0.01	0.06 ±0.02	0.06 ±0.00	0.05 ±0.00	NM
	33.4	0.07 ±0.02	0.09 ±0.01	0.07 ±0.00	0.08 ±0.02	0.05 ±0.01	NM	0.07 ±0.02	0.10 ±0.01	0.07 ±0.01	0.10 ±0.00	0.06 ±0.00	NM
	50.1	0.08 ±0.02	0.11 ±0.01	0.08 ±0.01	0.10 ±0.01	0.09 ±0.00	NM	0.08 ±0.01	0.11 ±0.01	0.09 ±0.02	0.11 ±0.02	0.08 ±0.01	NM

NM: Not Measured
Recovery: Follow up measurement at the end of the 24 h recovery period following the exposure period.

Appendix F

2020 Weymouth Water Treatment Plant Influent Veliger Dose-Response Figures

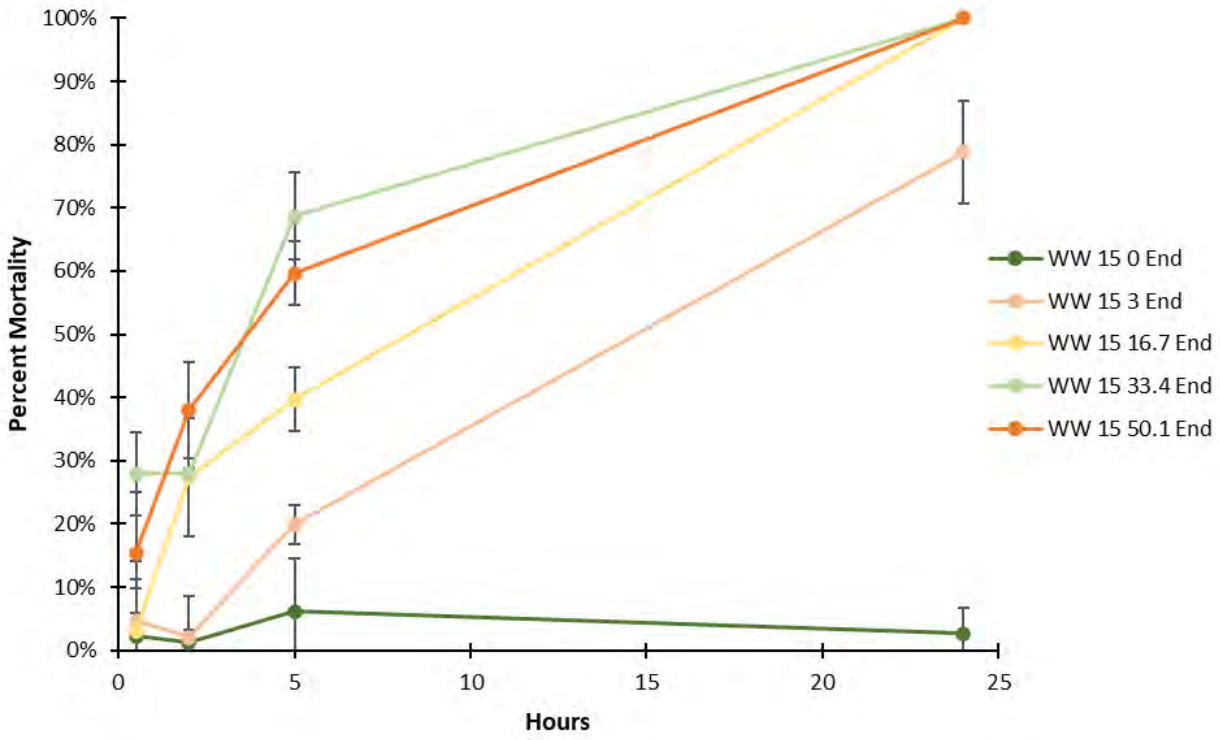

Figure F1. Veliger mortality with standard deviation bars (n=3) at the end of each exposure period at 15°C for the 2020 Weymouth Water Treatment Plant (WW) trials

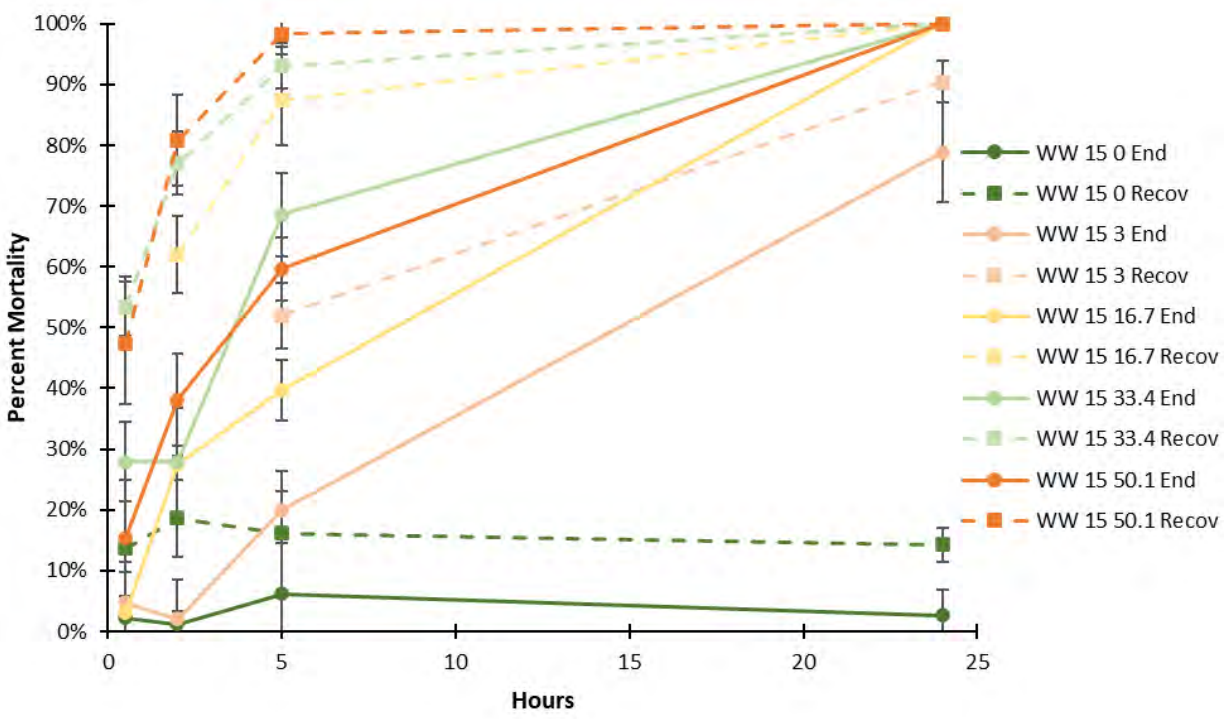

Figure F2. Veliger mortality with standard deviation bars (n=3) at the end of each exposure and recovery period at 15°C for the 2020 Weymouth Water Treatment Plant (WW) trials

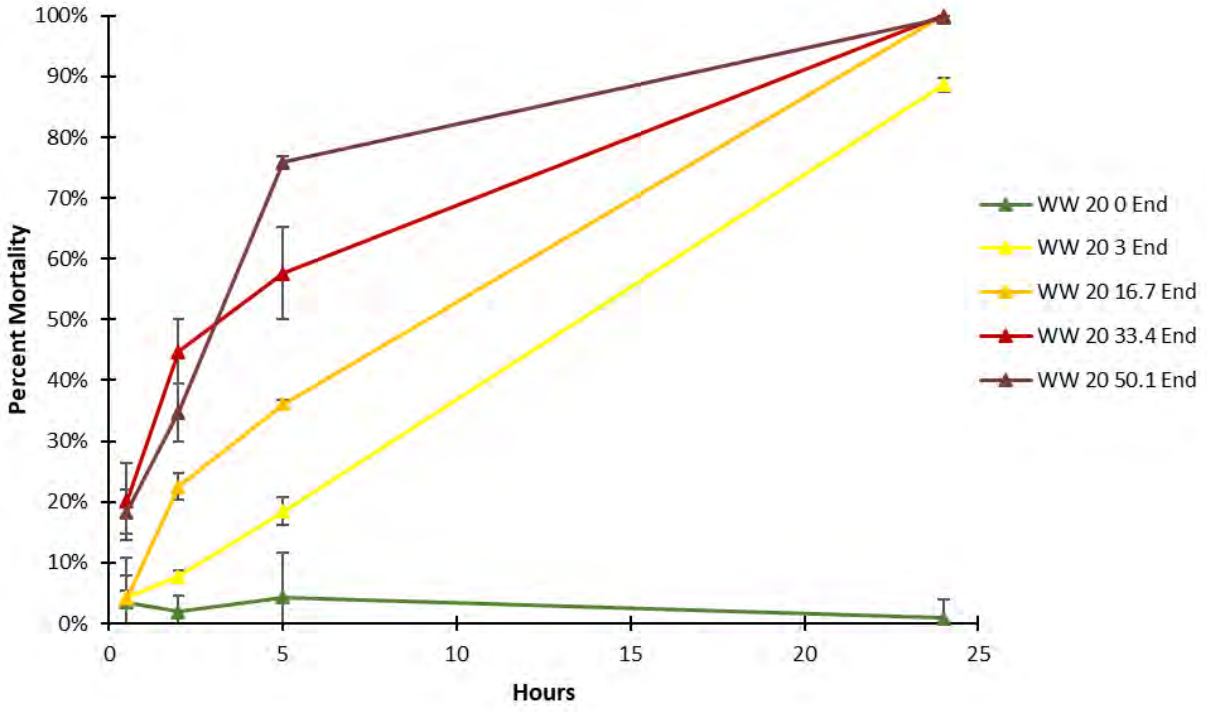

Figure F3. Veliger mortality with standard deviation bars (n=3) at the end of each exposure period at 20°C for the 2020 Weymouth Water Treatment Plant (WW) trials

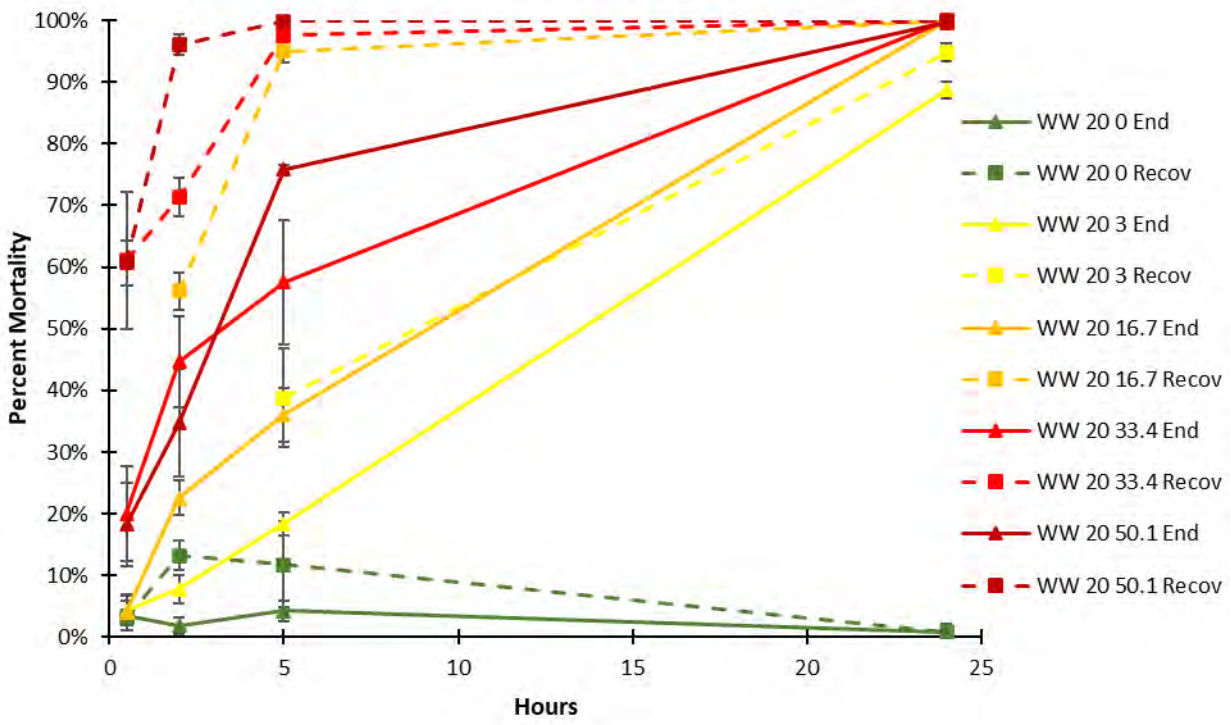

Figure F4. Veliger mortality with standard deviation bars (n=3) at the end of each exposure and recovery period at 20°C for the 2020 Weymouth Water Treatment Plant (WW) trials

Appendix G

2020 Weymouth WTP Dose-Response Veliger Testing – Water Quality Data

Table G1. Average water quality data for each dose-response test condition using Weymouth WTP influent in 2020

		15°C Test Conditions						20°C Test Conditions					
	Earth Tec QZ (µL/L)	0 h	0.5 h	2 h	5 h	24 h	Recovery	0 h	0.5 h	2 h	5 h	24 h	Recovery
Temp. (°C)	0.0	16.6 ±0.5	15.2 ±0.5	14.8 ±0.5	14.5 ±0.0	15.4 ±0.1	15.4 ±0.3	22.0 ±0.5	20.1 ±0.1	19.7 ±0.1	20.9 ±0.0	19.7 ±0.1	20.0 ±0.3
	3.0	16.6 ±0.8	14.8 ±0.1	14.4 ±0.1	14.4 ±0.0	15.2 ±0.0	15.5 ±0.3	22.0 ±0.4	19.9 ±0.0	19.6 ±0.1	21.1 ±0.3	19.7 ±0.0	20.0 ±0.3
	16.7	16.9 ±0.8	14.6 ±0.0	14.5 ±0.0	14.2 ±0.1	14.9 ±0.1	15.4 ±0.3	22.2 ±0.4	19.7 ±0.1	19.6 ±0.0	20.8 ±0.0	19.7 ±0.1	19.9 ±0.1
	33.4	16.9 ±1.7	15.8 ±0.1	15.0 ±0.1	15.1 ±0.0	14.5 ±0.0	15.2 ±0.3	22.3 ±0.6	19.9 ±0.1	19.9 ±0.1	19.9 ±0.0	NM	20.0 ±0.2
	50.1	16.8 ±1.7	15.7 ±0.0	15.2 ±0.0	15.2 ±0.1	14.4 ±0.1	15.2 ±0.2	22.4 ±0.4	19.8 ±0.0	20.0 ±0.0	19.6 ±0.0	20.3 ±0.1	20.0 ±0.2
DO (mg/L)	0.0	8.73 ±0.20	9.22 ±0.03	8.73 ±0.06	8.88 ±0.02	9.35 ±0.10	9.24 ±0.06	8.19 ±0.04	8.37 ±0.07	8.37 ±0.05	8.33 ±0.02	8.63 ±0.04	8.61 ±0.12
	3.0	8.73 ±0.17	9.18 ±0.10	8.76 ±0.04	8.76 ±0.17	9.42 ±0.03	9.13 ±0.06	8.21 ±0.08	8.33 ±0.03	8.32 ±0.02	8.40 ±0.05	8.73 ±0.01	8.68 ±0.09
	16.7	8.72 ±0.17	9.13 ±0.10	8.78 ±0.14	8.97 ±0.09	9.55 ±0.03	9.23 ±0.09	8.22 ±0.04	8.28 ±0.06	8.36 ±0.03	8.39 ±0.06	8.74 ±0.03	8.53 ±0.08
	33.4	8.86 ±0.06	8.88 ±0.05	8.85 ±0.01	9.02 ±0.02	9.55 ±0.08	9.25 ±0.08	8.25 ±0.09	8.22 ±0.07	8.37 ±0.02	8.34 ±0.02	NM	8.56 ±0.11
	50.1	8.77 ±0.05	8.80 ±0.01	8.89 ±0.12	8.87 ±0.05	9.64 ±0.05	9.12 ±0.10	8.27 ±0.12	8.28 ±0.07	8.34 ±0.08	8.37 ±0.02	8.77 ±0.02	8.64 ±0.14
pH	0.0	8.29 ±0.05	8.31 ±0.00	8.33 ±0.00	8.31 ±0.00	8.31 ±0.00	8.32 ±0.01	8.21 ±0.06	8.28 ±0.00	8.30 ±0.00	8.30 ±0.01	8.27 ±0.00	8.23 ±0.10
	3.0	8.28 ±0.02	8.30 ±0.00	8.31 ±0.00	8.30 ±0.01	8.30 ±0.00	8.32 ±0.01	8.22 ±0.02	8.26 ±0.00	8.28 ±0.01	8.28 ±0.01	8.26 ±0.00	8.31 ±0.01
	16.7	8.18 ±0.01	8.23 ±0.02	8.24 ±0.02	8.23 ±0.03	8.23 ±0.00	8.33 ±0.01	8.18 ±0.05	8.26 ±0.08	8.19 ±0.02	8.21 ±0.02	8.22 ±0.02	8.27 ±0.04
	33.4	8.08 ±0.05	8.10 ±0.02	8.13 ±0.03	8.09 ±0.03	8.13 ±0.03	8.33 ±0.01	8.10 ±0.04	8.18 ±0.06	8.16 ±0.05	8.04 ±0.03	NM	8.23 ±0.08
	50.1	7.92 ±0.06	8.01 ±0.05	7.97 ±0.09	7.95 ±0.05	8.05 ±0.03	8.34 ±0.01	8.01 ±0.07	8.05 ±0.04	8.02 ±0.04	7.96 ±0.04	8.11 ±0.04	8.26 ±0.06
Specific Conductance (µS/cm)	0.0	937 ±3	931 ±1	936 ±1	936 ±1	940 ±1	941 ±2	937 ±2	937 ±2	934 ±1	937 ±0	941 ±1	942 ±1
	3.0	938 ±1	937 ±0	936 ±0	936 ±0	939 ±0	941 ±0	935 ±1	936 ±0	935 ±1	937 ±0	941 ±0	944 ±1
	16.7	940 ±0	938 ±1	938 ±0	937 ±1	939 ±0	940 ±0	937 ±1	937 ±0	937 ±0	938 ±0	942 ±0	942 ±1
	33.4	943 ±2	944 ±1	940 ±1	940 ±0	942 ±0	940 ±1	945 ±4	939 ±0	939 ±0	941 ±0	NM	942 ±0
	50.1	944 ±1	945 ±0	943 ±0	943 ±1	942 ±0	941 ±1	944 ±3	944 ±0	943 ±0	944 ±0	945 ±0	942 ±2
Free Copper (mg/L)	0.0	0.02 ±0.01	0.05 ±0.01	0.03 ±0.00	0.03 ±0.01	0.03 ±0.00	0.02 ±0.01	0.02 ±0.02	0.04 ±0.01	0.03 ±0.00	0.03 ±0.02	0.03 ±0.01	0.01 ±0.01
	3.0	0.28 ±0.00	0.29 ±0.01	0.28 ±0.01	0.28 ±0.00	0.26 ±0.00	0.02 ±0.01	0.29 ±0.01	0.30 ±0.02	0.27 ±0.00	0.20 ±0.00	0.32 ±0.12	0.03 ±0.09
	16.7	0.95 ±0.02	0.95 ±0.01	0.94 ±0.02	0.92 ±0.02	0.91 ±0.00	0.03 ±0.01	1.08 ±0.01	1.08 ±0.02	1.06 ±0.02	1.03 ±0.01	0.98 ±0.01	0.02 ±0.01
	33.4	1.99 ±0.04	1.92 ±0.01	1.95 ±0.02	1.92 ±0.00	1.87 ±0.01	0.04 ±0.01	1.93 ±0.01	1.91 ±0.02	1.95 ±0.02	1.90 ±0.01	NM	0.04 ±0.01
	50.1	2.86 ±0.03	2.81 ±0.03	2.85 ±0.04	2.78 ±0.05	2.57 ±0.03	0.05 ±0.02	2.89 ±0.02	2.84 ±0.00	2.86 ±0.04	2.79 ±0.02	2.41 ±0.04	0.10 ±0.07

	Earth Tec QZ (µL/L)	15°C Test Conditions						20°C Test Conditions					
		0 h	0.5 h	2 h	5 h	24 h	Recovery	0 h	0.5 h	2 h	5 h	24 h	Recovery
Total Copper (mg/L)	0.0	0.02 ±0.01	0.05 ±0.01	0.03 ±0.01	0.03 ±0.01	0.03 ±0.01	0.02 ±0.01	0.03 ±0.02	0.04 ±0.02	0.02 ±0.00	0.04 ±0.01	0.02 ±0.00	0.01 ±0.01
	3.0	0.27 ±0.01	0.29 ±0.01	0.28 ±0.00	0.28 ±0.00	0.26 ±0.01	0.02 ±0.01	0.29 ±0.00	0.28 ±0.01	0.27 ±0.01	0.28 ±0.00	0.28 ±0.05	0.03 ±0.01
	16.7	0.95 ±0.01	0.95 ±0.01	0.94 ±0.01	0.93 ±0.01	0.92 ±0.01	0.03 ±0.01	1.08 ±0.01	1.07 ±0.01	1.06 ±0.01	1.03 ±0.02	0.98 ±0.01	0.02 ±0.00
	33.4	1.98 ±0.04	1.91 ±0.02	1.93 ±0.02	1.91 ±0.01	1.88 ±0.03	0.04 ±0.02	1.93 ±0.00	1.90 ±0.02	1.95 ±0.02	1.90 ±0.02	NM	0.04 ±0.01
	50.1	2.83 ±0.02	2.80 ±0.03	2.83 ±0.04	2.78 ±0.03	2.57 ±0.03	0.06 ±0.02	2.88 ±0.02	2.82 ±0.01	2.83 ±0.02	2.76 ±0.03	2.42 ±0.03	0.10 ±0.07
Total Chlorine (mg/L)	0.0	0.06 ±0.02	0.06 ±0.00	0.06 ±0.00	0.06 ±0.00	0.03 ±0.00	NM	0.08 ±0.01	0.08 ±0.01	0.06 ±0.01	0.05 ±0.00	0.05 ±0.00	NM
	3.0	0.05 ±0.01	0.06 ±0.00	0.05 ±0.00	0.06 ±0.00	0.03 ±0.00	NM	0.05 ±0.00	0.06 ±0.00	0.05 ±0.00	0.05 ±0.00	0.04 ±0.01	NM
	16.7	0.05 ±0.00	0.06 ±0.01	0.05 ±0.01	0.06 ±0.01	0.04 ±0.01	NM	0.05 ±0.01	0.07 ±0.01	0.05 ±0.01	0.05 ±0.00	0.08 ±0.02	NM
	33.4	0.07 ±0.01	0.07 ±0.00	0.08 ±0.01	0.06 ±0.01	0.04 ±0.00	NM	0.09 ±0.02	0.06 ±0.01	0.05 ±0.00	0.05 ±0.00	NM	NM
	50.1	0.06 ±0.01	0.07 ±0.01	0.09 ±0.01	0.08 ±0.00	0.05 ±0.01	NM	0.10 ±0.02	0.07 ±0.01	0.08 ±0.02	0.08 ±0.02	0.07 ±0.03	NM

NM: Not Measured
Recovery: Follow up measurement at the end of the 24 h recovery period following the exposure period.

Appendix H
2021 Lake Mathews Veliger Dose-Response Figures

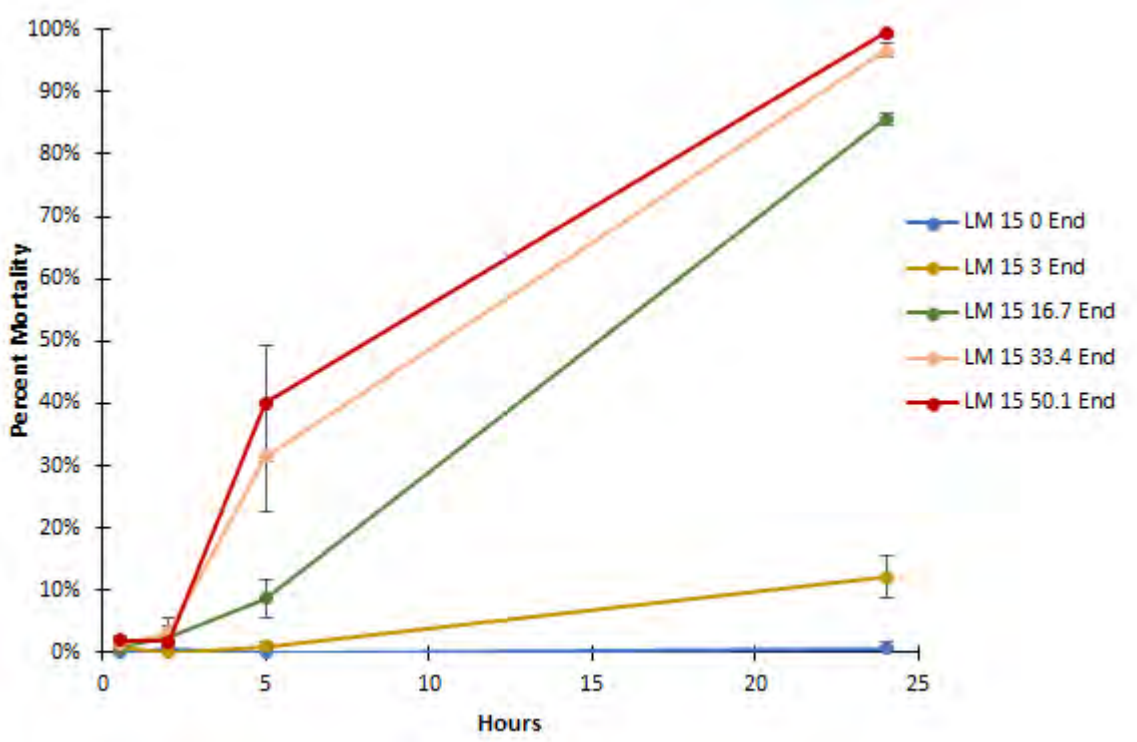

Figure H1. Veliger mortality with standard deviation bars (n=3) at the end of each exposure period at 15°C for 2021 Lake Mathews (LM) trials

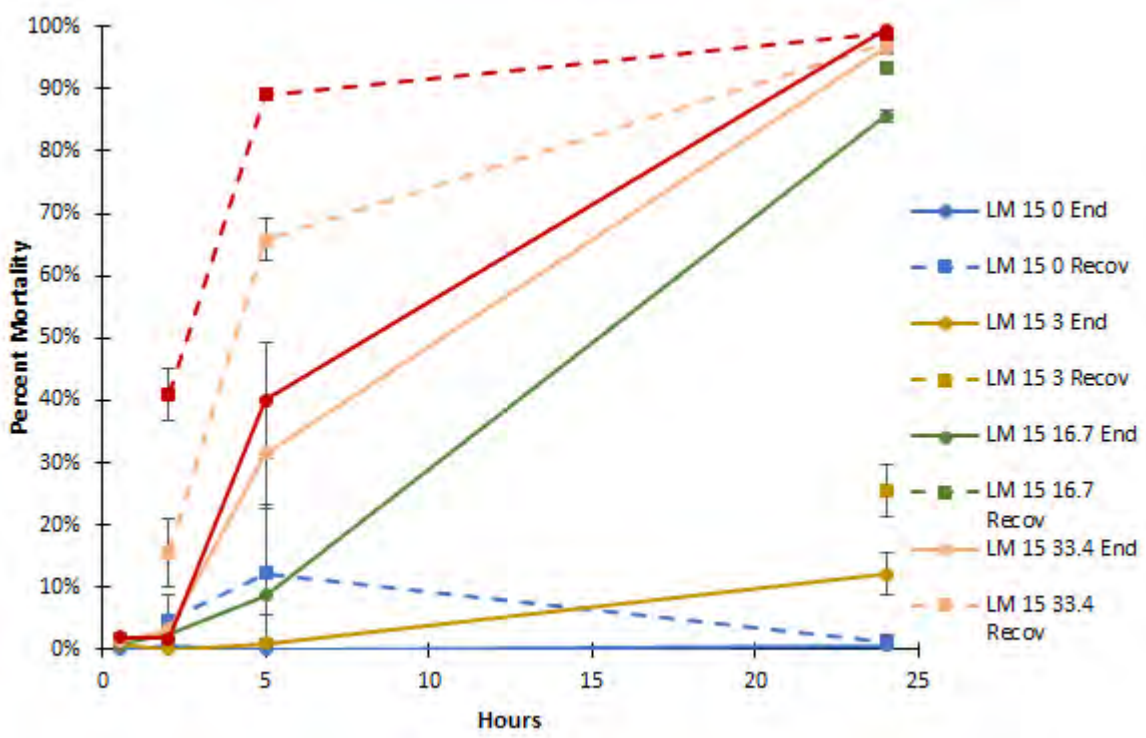

Figure H2. Veliger mortality with standard deviation bars (n=3) at the end of each exposure and recovery period at 15°C for 2021 Lake Mathews (LM) trials

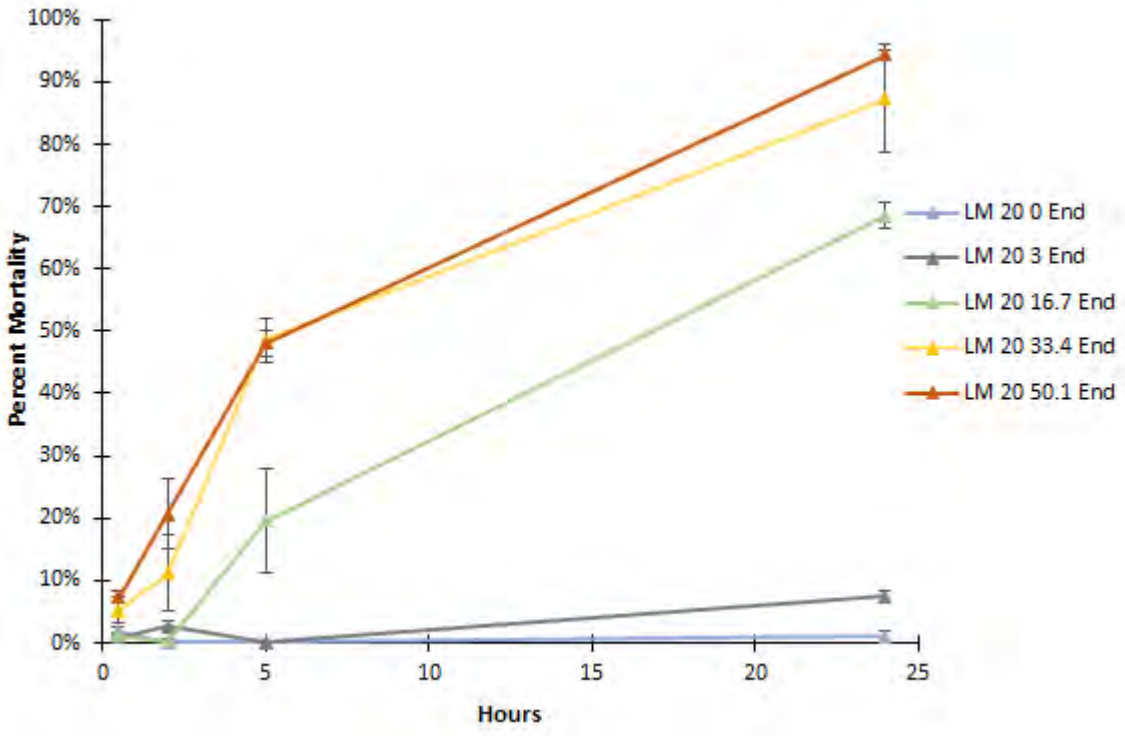

Figure H3. Veliger mortality with standard deviation bars (n=3) at the end of each exposure period at 20°C for 2021 Lake Mathews (LM) trials

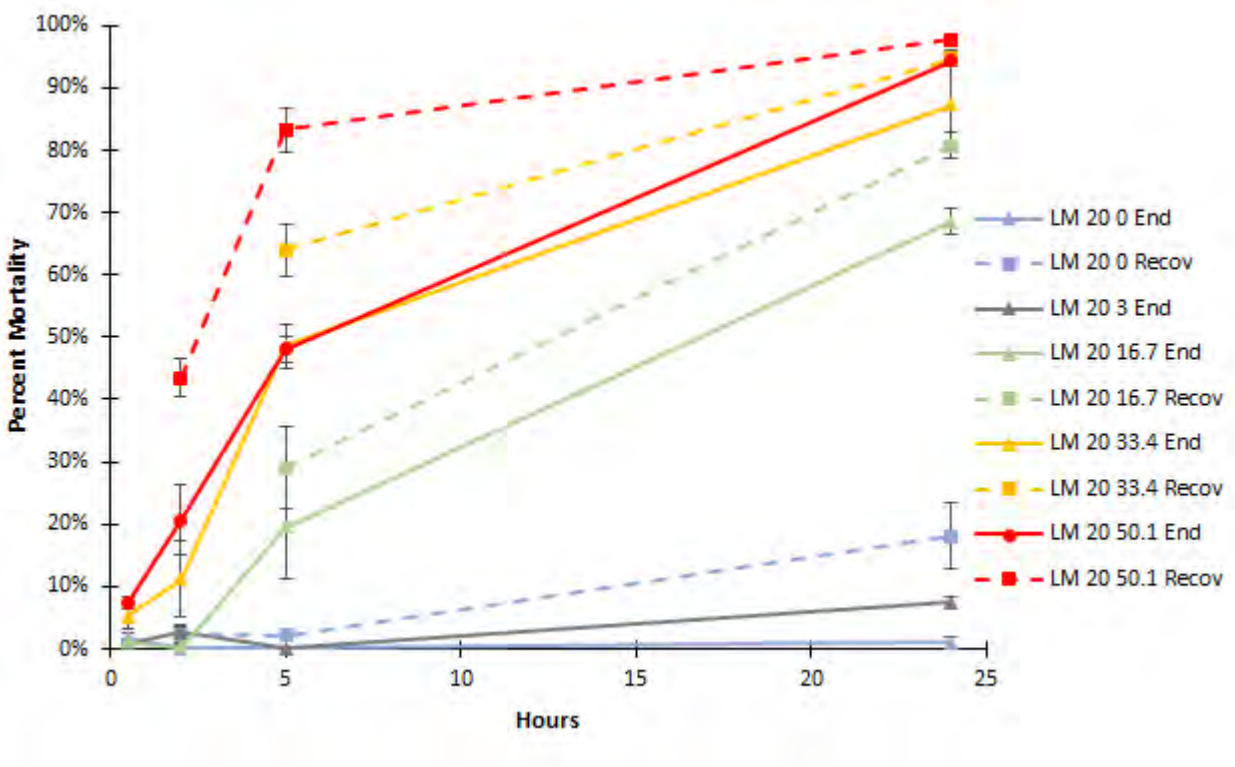

Figure H4. Veliger mortality with standard deviation bars (n=3) at the end of each exposure and recovery period at 20°C for 2021 Lake Mathews (LM) trials

Appendix I

2021 Lake Mathews Dose-Response Veliger Testing – Water Quality Data

Table I1. Average water quality data for each 2021 Lake Mathews dose-response test condition

		15°C Test Conditions						20°C Test Conditions					
	Earth Tec QZ (µL/L)	0 h	0.5 h	2 h	5 h	24 h	Recovery	0 h	0.5 h	2 h	5 h	24 h	Recovery
Temp. (°C)	0.0	14.5 ±0.0	14.9 ±0.5	15.5 ±0.1	14.8 ±0.1	14.9 ±0.0	15.3 ±0.2	22.1 ±0.6	20.0 ±0.1	20.1 ±0.1	21.1 ±0.0	21.1 ±0.0	21.1 ±0.3
	3.0	14.2 ±0.1	14.8 ±0.0	15.5 ±0.0	14.8 ±0.0	14.9 ±0.0	15.3 ±0.1	23.2 ±0.9	20.0 ±0.1	20.0 ±0.1	21.1 ±0.1	21.1 ±0.0	NM
	16.7	14.5 ±0.1	14.4 ±0.0	14.6 ±0.0	14.4 ±0.1	14.5 ±0.0	15.2 ±0.1	23.3 ±0.4	19.9 ±0.1	20.1 ±0.1	21.1 ±0.0	20.1 ±0.1	21.0 ±0.1
	33.4	14.3 ±0.2	14.1 ±0.1	14.5 ±0.2	14.4 ±0.1	14.5 ±0.0	15.0 ±0.0	21.8 ±0.2	20.9 ±0.0	20.0 ±0.1	20.3 ±0.0	20.1 ±0.1	20.1 ±0.1
	50.1	14.0 ±0.5	14.4 ±0.1	15.2 ±0.1	15.6 ±0.1	15.6 ±0.1	15.3 ±0.1	21.7 ±0.2	20.9 ±0.0	20.0 ±0.1	20.2 ±0.0	20.0 ±0.0	20.8 ±0.3
DO (mg/L)	0.0	9.59 ±0.20	9.86 ±0.11	9.43 ±0.07	9.36 ±0.08	9.22 ±0.05	8.83 ±0.10	8.04 ±0.21	8.17 ±0.04	8.05 ±0.02	8.01 ±0.02	7.47 ±0.09	7.81 ±0.12
	3.0	9.82 ±0.07	10.02 ±0.01	9.83 ±0.01	9.71 ±0.03	9359 ±0.02	8.92 ±0.05	8.00 ±0.16	8.19 ±0.04	7.97 ±0.03	8.00 ±0.05	7.85 ±0.04	NM
	16.7	9.64 ±0.12	9.79 ±0.06	9.53 ±0.06	9.49 ±0.03	9.40 ±0.03	9.06 ±0.03	7.99 ±0.13	8.18 ±0.11	8.04 ±0.07	8.06 ±0.16	7.95 ±0.01	8.07 ±0.17
	33.4	9.87 ±0.19	10.05 ±0.06	9.68 ±0.06	9.54 ±0.03	9.52 ±0.03	9.01 ±0.03	8.16 ±0.14	8.28 ±0.08	8.04 ±0.05	8.02 ±0.06	8.10 ±0.08	8.30 ±0.11
	50.1	9.97 ±0.17	10.19 ±0.01	9.70 ±0.05	9.78 ±0.02	9.93 ±0.02	9.23 ±0.07	8.16 ±0.07	8.33 ±0.04	7.99 ±0.05	8.01 ±0.03	7.95 ±0.01	8.24 ±0.24
pH	0.0	8.49 ±0.07	8.23 ±0.18	8.36 ±0.00	8.29 ±0.01	8.35 ±0.01	8.38 ±0.03	8.30 ±0.02	8.35 ±0.03	8.33 ±0.03	8.32 ±0.01	8.31 ±0.05	8.31 ±0.03
	3.0	8.38 ±0.05	8.34 ±0.00	8.34 ±0.00	8.29 ±0.00	8.24 ±0.01	8.39 ±0.00	8.27 ±0.01	8.32 ±0.00	8.29 ±0.01	8.31 ±0.02	8.26 ±0.00	NM
	16.7	8.30 ±0.04	8.27 ±0.02	8.27 ±0.03	8.23 ±0.02	8.26 ±0.01	8.41 ±0.00	8.20 ±0.04	8.17 ±0.04	8.20 ±0.02	8.27 ±0.05	8.23 ±0.01	8.32 ±0.04
	33.4	8.18 ±0.04	8.13 ±0.01	8.18 ±0.05	8.16 ±0.05	8.19 ±0.02	8.40 ±0.02	8.15 ±0.10	8.23 ±0.04	8.13 ±0.04	8.17 ±0.05	8.22 ±0.02	8.34 ±0.01
	50.1	8.04 ±0.05	8.03 ±0.05	8.04 ±0.05	8.05 ±0.03	8.13 ±0.02	8.42 ±0.02	8.01 ±0.05	8.12 ±0.02	8.00 ±0.05	8.02 ±0.04	8.14 ±0.03	8.34 ±0.03
Specific Conductance (µS/cm)	0.0	948 ±1	952 ±0	950 ±0	953 ±1	950 ±0	952 ±1	949 ±4	949 ±1	949 ±1	949 ±0	951 ±0	955 ±1
	3.0	950 ±2	952 ±0	951 ±0	952 ±1	949 ±0	952 ±0	945 ±3	948 ±0	948 ±0	949 ±1	951 ±0	NM
	16.7	954 ±2	954 ±0	953 ±1	956 ±1	949 ±0	952 ±0	947 ±3	951 ±0	951 ±0	952 ±0	953 ±0	951 ±1
	33.4	954 ±3	954 ±2	958 ±2	954 ±3	957 ±0	956 ±1	953 ±2	954 ±0	954 ±0	954 ±0	955 ±1	951 ±1
	50.1	958 ±1	959 ±0	959 ±2	956 ±1	958 ±0	952 ±1	957 ±1	956 ±0	956 ±1	956 ±0	957 ±0	950 ±1
Free Copper (mg/L)	0.0	0.01 ±0.01	0.01 ±0.01	0.01 ±0.01	0.03 ±0.00	0.01 ±0.01	0.01 ±0.01	0.05 ±0.01	0.05 ±0.01	0.03 ±0.02	0.03 ±0.03	0.06 ±0.03	0.02 ±0.02
	3.0	0.20 ±0.00	0.20 ±0.01	0.19 ±0.00	0.23 ±0.01	0.24 ±0.03	0.00 ±0.00	0.23 ±0.05	0.24 ±0.02	0.17 ±0.01	0.19 ±0.00	0.21 ±0.01	NM
	16.7	0.00 ±0.02	0.94 ±0.01	0.94 ±0.00	0.95 ±0.01	0.94 ±0.03	0.00 ±0.00	1.12 ±0.15	1.04 ±0.02	1.12 ±0.16	0.94 ±0.01	0.98 ±0.01	0.05 ±0.01
	33.4	1.93 ±0.05	1.88 ±0.02	1.89 ±0.02	1.83 ±0.01	1.73 ±0.02	0.05 ±0.03	2.02 ±0.01	1.93 ±0.01	1.93 ±0.02	1.88 ±0.01	1.81 ±0.02	0.14 ±0.03
	50.1	2.94 ±0.01	2.85 ±0.03	2.87 ±0.01	2.68 ±0.02	2.49 ±0.03	0.12 ±0.07	2.95 ±0.02	2.82 ±0.02	2.77 ±0.02	2.66 ±0.02	2.34 ±0.02	0.18 ±0.07

	Earth Tec QZ (µL/L)	15°C Test Conditions						20°C Test Conditions					
		0 h	0.5 h	2 h	5 h	24 h	Recovery	0 h	0.5 h	2 h	5 h	24 h	Recovery
Total Copper (mg/L)	**0.0**	0.02 ±0.01	0.00 ±0.00	0.01 ±0.01	0.02 ±0.01	0.00 ±0.00	0.01 ±0.01	0.03 ±0.01	0.05 ±0.01	0.04 ±0.02	0.03 ±0.02	0.04 ±0.01	0.03 ±0.03
	3.0	0.20 ±0.00	0.20 ±0.01	0.18 ±0.00	0.23 ±0.03	0.22 ±0.01	0.00 ±0.00	0.23 ±0.05	0.24 ±0.02	0.17 ±0.00	0.19 ±0.01	0.19 ±0.01	NM
	16.7	0.96 ±0.01	0.94 ±0.01	0.95 ±0.01	0.93 ±0.00	0.92 ±0.01	0.01 ±0.01	1.11 ±0.15	1.03 ±0.02	1.12 ±0.16	0.94 ±0.01	0.98 ±0.03	0.05 ±0.01
	33.4	1.93 ±0.02	1.88 ±0.03	1.89 ±0.03	1.83 ±0.01	1.75 ±0.03	0.06 ±0.03	2.02 ±0.01	1.94 ±0.00	1.93 ±0.01	1.88 ±0.00	1.84 ±0.02	0.14 ±0.03
	50.1	2.92 ±0.01	2.84 ±0.02	2.85 ±0.02	2.68 ±0.03	2.79 ±0.04	0.12 ±0.07	2.96 ±0.01	2.83 ±0.02	2.75 ±0.02	2.65 ±0.02	2.35 ±0.03	0.19 ±0.07
Total Chlorine (mg/L)	**0.0**	0.03 ±0.01	0.03 ±0.00	0.00 ±0.00	0.00 ±0.00	0.00 ±0.00	NM	0.04 ±0.01	0.04 ±0.00	0.03 ±0.00	0.01 ±0.00	0.03 ±0.01	NM
	3.0	0.03 ±0.00	0.02 ±0.01	0.01 ±0.00	0.03 ±0.00	0.02 ±0.00	NM	0.05 ±0.03	0.03 ±0.01	0.04 ±0.02	0.01 ±0.01	0.04 ±0.01	NM
	16.7	0.04 ±0.01	0.02 ±0.00	0.03 ±0.01	0.03 ±0.01	0.02 ±0.00	NM	0.06 ±0.01	0.05 ±0.01	0.04 ±0.01	0.01 ±0.01	0.05 ±0.01	NM
	33.4	0.04 ±0.01	0.01 ±0.01	0.04 ±0.01	0.04 ±0.01	0.07 ±0.01	NM	0.06 ±0.02	0.07 ±0.02	0.06 ±0.03	0.05 ±0.01	0.07 ±0.01	NM
	50.1	0.05 ±0.01	0.06 ±0.02	0.05 ±0.00	0.05 ±0.01	0.06 ±0.01	NM	0.08 ±0.00	0.09 ±0.01	0.05 ±0.01	0.08 ±0.01	0.07 ±0.03	NM

NM: Not Measured

Recovery: Follow up measurement at the end of the 24 h recovery period following the exposure period.

Appendix J

2021 Weymouth Water Treatment Plant Influent Veliger Dose-Response Figures

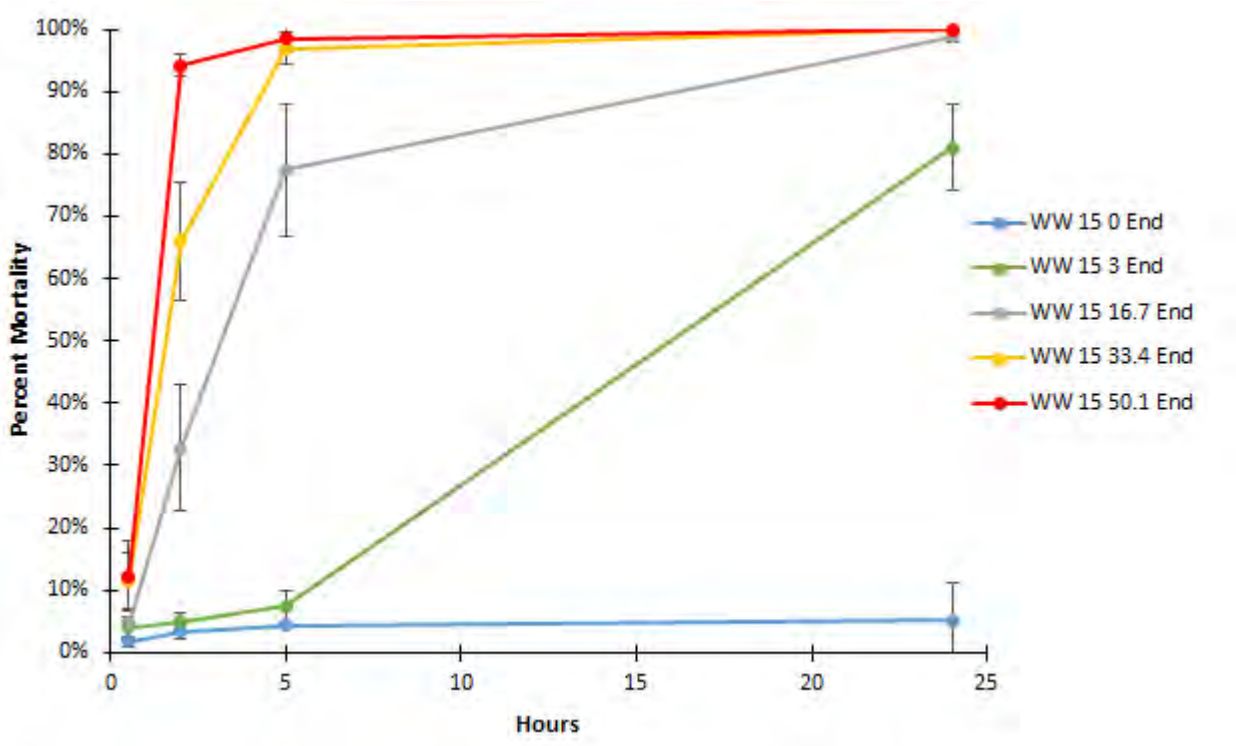

Figure J1. Veliger mortality with standard deviation bars (n=3) at the end of each exposure period at 15°C for the 2021 Weymouth Water Treatment Plant (WW) trials

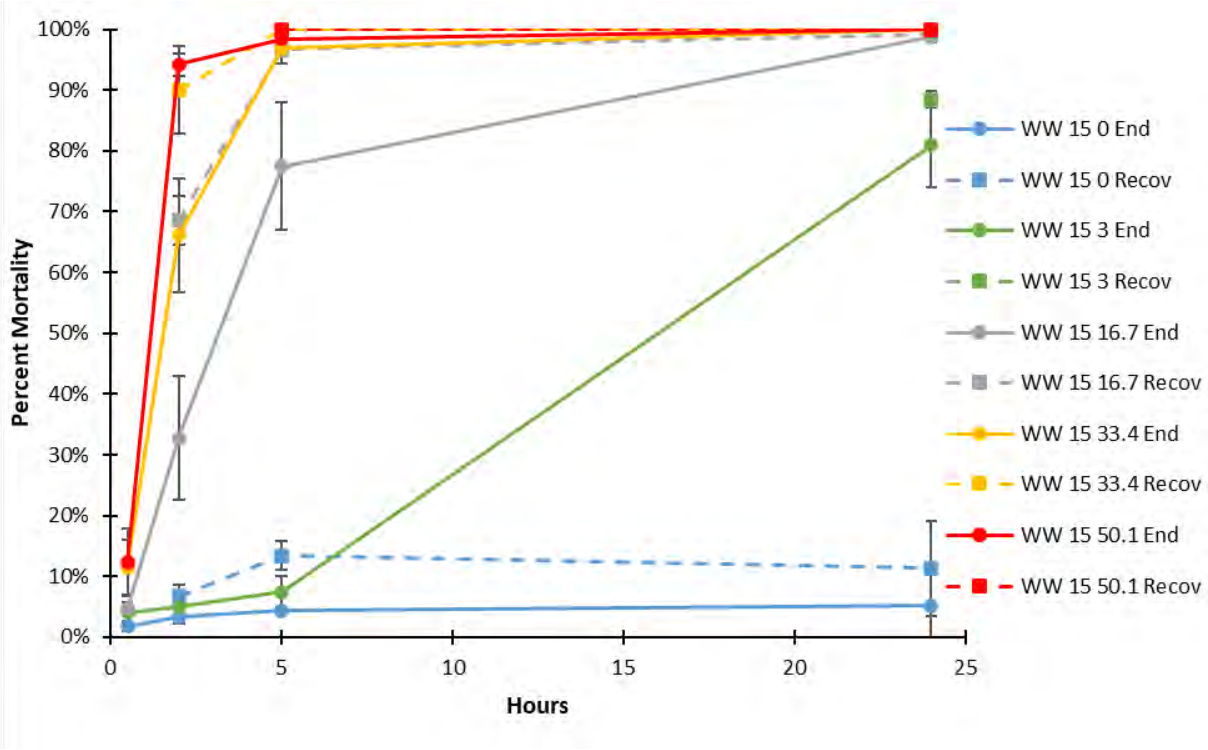

Figure J2. Veliger mortality with standard deviation bars (n=3) at the end of each exposure and recovery period at 15°C for the 2021 Weymouth Water Treatment Plant (WW) trials

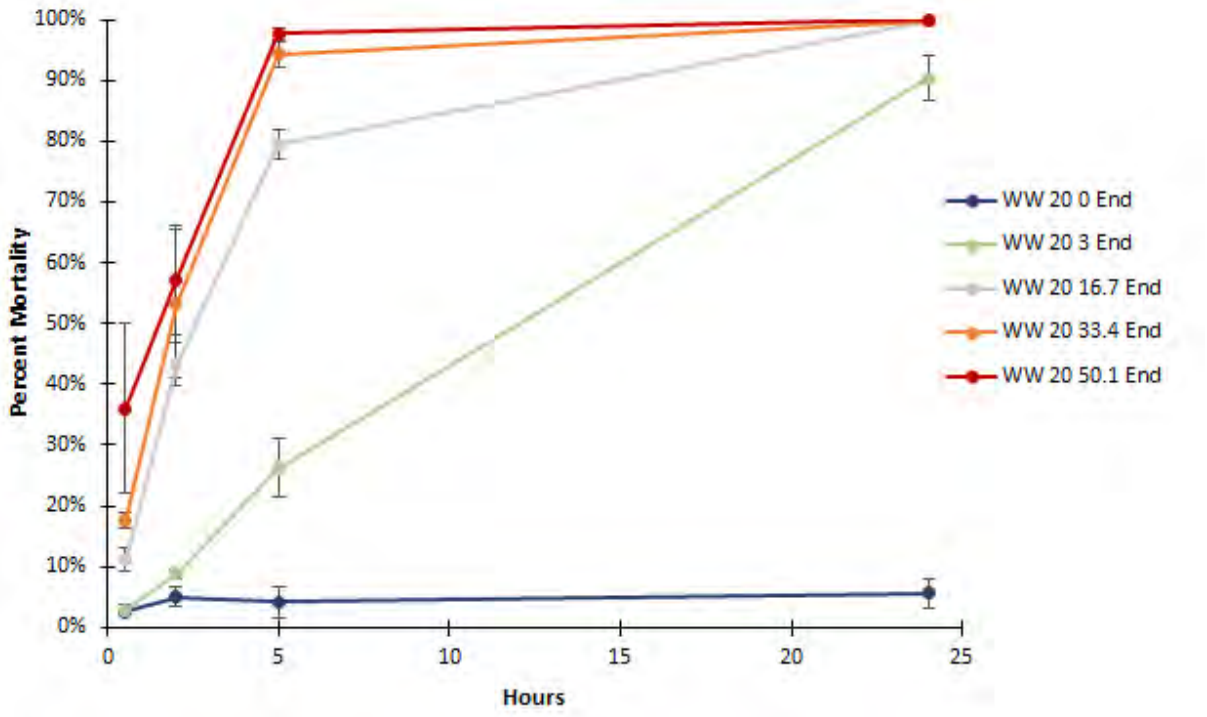

Figure J3. Veliger mortality with standard deviation bars (n=3) at the end of each exposure period at 20°C for the 2021 Weymouth Water Treatment Plant (WW) trials

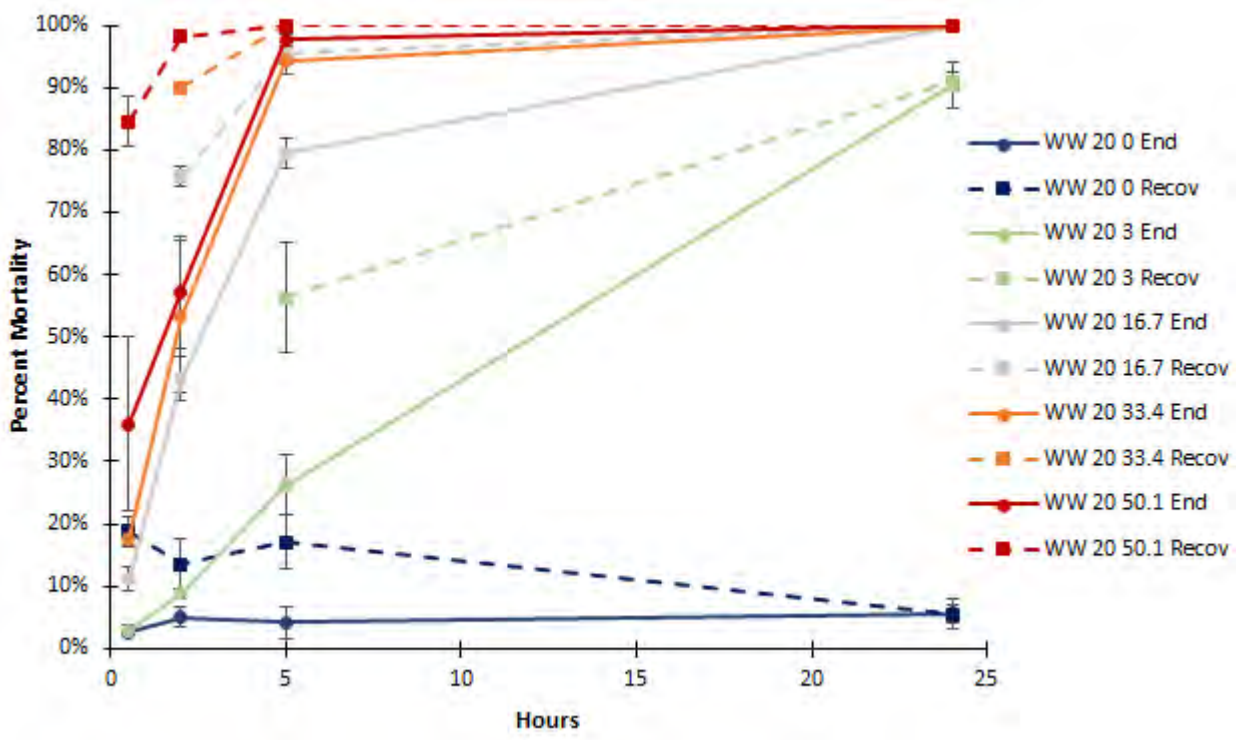

Figure J4. Veliger mortality with standard deviation bars (n=3) at the end of each exposure and recovery period at 20°C for the 2021 Weymouth Water Treatment Plant (WW) trials

Appendix K

2021 Weymouth WTP Dose-Response Veliger Testing – Water Quality Data

Table G1. Average water quality data for each dose-response test condition using Weymouth WTP influent in 2021

		15°C Test Conditions						20°C Test Conditions					
	Earth Tec QZ (µL/L)	0 h	0.5 h	2 h	5 h	24 h	Recovery	0 h	0.5 h	2 h	5 h	24 h	Recovery
Temperature (°C)	0.0	11.8 ±0.7	14.0 ±0.1	14.4 ±0.1	14.9 ±0.1	15.7 ±0.1	14.7 ±0.2	21.7 ±1.3	20.1 ±0.2	20.5 ±0.0	20.6 ±0.1	21.0 ±0.1	20.7 ±0.5
	3.0	10.5 ±0.5	14.1 ±0.0	14.8 ±0.1	14.9 ±0.0	15.5 ±0.3	14.5 ±0.0	21.8 ±1.1	20.2 ±0.1	20.6 ±0.1	20.5 ±0.0	21.0 ±0.0	20.7 ±0.6
	16.7	10.6 ±0.4	14.1 ±0.1	14.5 ±0.0	15.0 ±0.1	15.8 ±0.1	14.5 ±0.1	22.0 ±1.1	20.2 ±0.0	20.5 ±0.0	20.6 ±0.0	21.0 ±0.0	20.7 ±0.4
	33.4	13.2 ±1.0	13.6 ±0.1	14.6 ±0.0	15.3 ±0.1	15.2 ±0.1	15.0 ±0.3	22.1 ±0.5	19.9 ±0.1	20.7 ±0.1	21.0 ±0.1	21.3 ±0.0	20.3 ±0.4
	50.1	11.6 ±0.5	13.6 ±0.1	14.6 ±0.1	15.0 ±0.0	15.0 ±0.0	15.2 ±0.3	22.6 ±0.7	19.8 ±0.0	20.9 ±0.1	21.0 ±0.0	21.4 ±0.0	20.7 ±0.2
DO (mg/L)	0.0	9.85 ±0.25	10.47 ±0.06	10.17 ±0.02	10.13 ±0.01	10.15 ±0.01	9.10 ±0.12	8.24 ±0.09	8.37 ±0.10	8.36 ±0.01	8.36 ±0.06	8.68 ±0.03	8.66 ±0.08
	3.0	10.62 ±0.17	11.17 ±0.01	10.91 ±0.07	10.83 ±0.16	10.46 ±0.20	9.22 ±0.05	8.20 ±0.02	8.34 ±0.04	8.38 ±0.04	8.39 ±0.07	8.64 ±0.01	8.65 ±0.10
	16.7	11.47 ±0.24	11.56 ±0.04	11.45 ±0.07	11.20 ±0.14	10.58 ±0.11	9.33 ±0.08	8.27 ±0.03	8.46 ±0.10	8.38 ±0.01	8.45 ±0.10	8.66 ±0.07	8.61 ±0.11
	33.4	11.82 ±0.20	11.61 ±0.23	11.80 ±0.08	11.67 ±0.08	10.49 ±0.15	8.85 ±0.08	8.27 ±0.05	8.38 ±0.04	8.33 ±0.10	8.35 ±0.00	8.69 ±0.04	8.61 ±0.04
	50.1	11.11 ±0.15	11.49 ±0.20	11.37 ±0.14	11.27 ±0.06	10.52 ±0.03	9.10 ±0.29	8.29 ±0.03	8.36 ±0.04	8.35 ±0.03	8.42 ±0.02	8.66 ±0.03	8.59 ±0.03
pH	0.0	8.33 ±0.08	8.30 ±0.00	8.30 ±0.00	8.28 ±0.01	8.37 ±0.02	8.39 ±0.02	8.32 ±0.05	8.36 ±0.02	8.38 ±0.01	8.37 ±0.01	8.42 ±0.02	8.40 ±0.02
	3.0	8.28 ±0.06	8.28 ±0.00	8.30 ±0.01	8.28 ±0.01	8.37 ±0.01	8.39 ±0.00	8.32 ±0.02	8.33 ±0.01	8.36 ±0.01	8.36 ±0.01	8.38 ±0.00	8.39 ±0.02
	16.7	8.21 ±0.04	8.21 ±0.01	8.24 ±0.02	8.23 ±0.03	8.30 ±0.03	8.38 ±0.03	8.25 ±0.02	8.28 ±0.02	8.31 ±0.04	8.28 ±0.03	8.34 ±0.02	8.40 ±0.02
	33.4	8.19 ±0.06	8.12 ±0.05	8.16 ±0.02	8.12 ±0.03	8.28 ±0.03	8.34 ±0.02	8.18 ±0.04	8.24 ±0.04	8.18 ±0.04	8.18 ±0.02	8.34 ±0.04	8.39 ±0.01
	50.1	8.08 ±0.08	8.01 ±0.05	8.05 ±0.05	8.02 ±0.05	8.18 ±0.03	8.36 ±0.01	8.08 ±0.07	8.03 ±0.06	8.05 ±0.02	8.06 ±0.02	8.21 ±0.05	8.39 ±0.01
Specific Conductance (µS/cm)	0.0	958 ±1	961 ±2	963 ±2	961 ±0	963 ±0	956 ±2	953 ±4	951 ±1	950 ±1	950 ±0	953 ±0	952 ±1
	3.0	949 ±3	954 ±2	954 ±0	954 ±1	955 ±0	956 ±0	949 ±2	951 ±1	951 ±1	951 ±0	953 ±0	954 ±1
	16.7	955 ±4	956 ±0	955 ±0	956 ±1	956 ±0	955 ±1	951 ±2	952 ±0	953 ±0	954 ±0	956 ±0	953 ±1
	33.4	955 ±6	957 ±3	958 ±1	960 ±1	959 ±0	955 ±1	956 ±4	957 ±1	957 ±0	957 ±0	958 ±0	953 ±1
	50.1	953 ±5	963 ±2	962 ±0	962 ±1	962 ±1	955 ±1	958 ±3	961 ±3	960 ±0	959 ±0	960 ±0	952 ±1
Free Copper (mg/L)	0.0	0.03 ±0.02	0.00 ±0.00	0.02 ±0.01	NM	0.01 ±0.01	0.00 ±0.01	0.01 ±0.01	0.01 ±0.02	0.00 ±0.00	0.01 ±0.01	0.00 ±0.00	0.00 ±0.00
	3.0	0.23 ±0.01	0.16 ±0.01	0.15 ±0.00	NM	0.17 ±0.01	0.00 ±0.00	0.22 ±0.01	0.20 ±0.01	0.18 ±0.00	0.19 ±0.01	0.21 ±0.01	0.01 ±0.01
	16.7	0.96 ±0.04	0.94 ±0.02	0.93 ±0.01	NM	0.92 ±0.04	0.00 ±0.01	1.03 ±0.01	1.04 ±0.01	1.03 ±0.02	1.03 ±0.02	1.02 ±0.02	0.00 ±0.01
	33.4	1.91 ±0.02	1.87 ±0.01	1.86 ±0.01	1.84 ±0.00	1.79 ±0.01	0.02 ±0.01	1.95 ±0.01	1.92 ±0.02	1.89 ±0.01	1.87 ±0.01	1.76 ±0.01	0.04 ±0.03
	50.1	2.93 ±0.03	2.87 ±0.03	2.85 ±0.02	2.74 ±0.01	2.56 ±0.02	0.04 ±0.01	2.89 ±0.03	2.84 ±0.02	2.84 ±0.02	2.76 ±0.01	2.59 ±0.03	0.05 ±0.02
	0.0	0.04 ±0.05	0.01 ±0.01	0.02 ±0.01	NM	0.01 ±0.01	0.00 ±0.01	0.01 ±0.01	0.01 ±0.01	0.01 ±0.01	0.00 ±0.00	0.00 ±0.00	0.00 ±0.00

	Earth Tec QZ (µL/L)	15°C Test Conditions						20°C Test Conditions					
		0 h	0.5 h	2 h	5 h	24 h	Recovery	0 h	0.5 h	2 h	5 h	24 h	Recovery
Total Copper (mg/L)	**3.0**	0.17 ±0.01	0.16 ±0.01	0.16 ±0.01	NM	0.15 ±0.00	0.00 ±0.00	0.21 ±0.00	0.19 ±0.00	0.20 ±0.00	0.20 ±0.01	0.20 ±0.00	0.22 ±0.05
	16.7	0.93 ±0.01	0.93 ±0.01	0.92 ±0.01	NM	0.91 ±0.01	0.00 ±0.00	1.03 ±0.00	1.02 ±0.01	1.02 ±0.02	1.02 ±0.02	1.02 ±0.01	0.01 ±0.01
	33.4	1.89 ±0.01	1.86 ±0.01	1.85 ±0.00	1.85 ±0.00	1.79 ±0.00	0.02 ±0.02	1.94 ±0.01	1.90 ±0.01	1.89 ±0.01	1.87 ±0.01	1.76 ±0.00	0.04 ±0.03
	50.1	2.90 ±0.02	2.84 ±0.03	2.83 ±0.02	2.75 ±0.02	2.55 ±0.01	0.05 ±0.02	2.88 ±0.03	2.84 ±0.03	2.83 ±0.02	2.75 ±0.01	2.59 ±0.02	0.04 ±0.02
Total Chlorine (mg/L)	**0.0**	0.11 ±0.01	0.11 ±0.00	0.10 ±0.02	NM	0.07 ±0.02	NM	0.08 ±0.00	0.09 ±0.01	0.06 ±0.03	0.06 ±0.01	0.06 ±0.00	NM
	3.0	0.12 ±0.01	0.06 ±0.04	0.09 ±0.01	NM	0.06 ±0.02	NM	0.07 ±0.00	0.08 ±0.01	0.05 ±0.03	0.05 ±0.03	0.05 ±0.01	NM
	16.7	0.11 ±0.02	0.10 ±0.01	0.09 ±0.01	NM	0.06 ±0.00	NM	0.07 ±0.00	0.09 ±0.02	0.07 ±0.01	0.05 ±0.00	0.04 ±0.01	NM
	33.4	0.09 ±0.05	0.07 ±0.02	0.09 ±0.01	0.09 ±0.00	0.08 ±0.00	NM	0.06 ±0.03	0.07 ±0.00	0.08 ±0.01	0.07 ±0.01	0.05 ±0.00	NM
	50.1	0.10 ±0.01	0.06 ±0.02	0.13 ±0.00	0.11 ±0.00	0.12 ±0.01	NM	0.08 ±0.03	0.08 ±0.01	0.12 ±0.00	0.07 ±0.01	0.09 ±0.02	NM

NM: Not Measured
Recovery: Follow up measurement at the end of the 24 h recovery period following the exposure period.

Appendix L

Veliger Dose Response at All Sites Grouped by EarthTec QZ Concentration

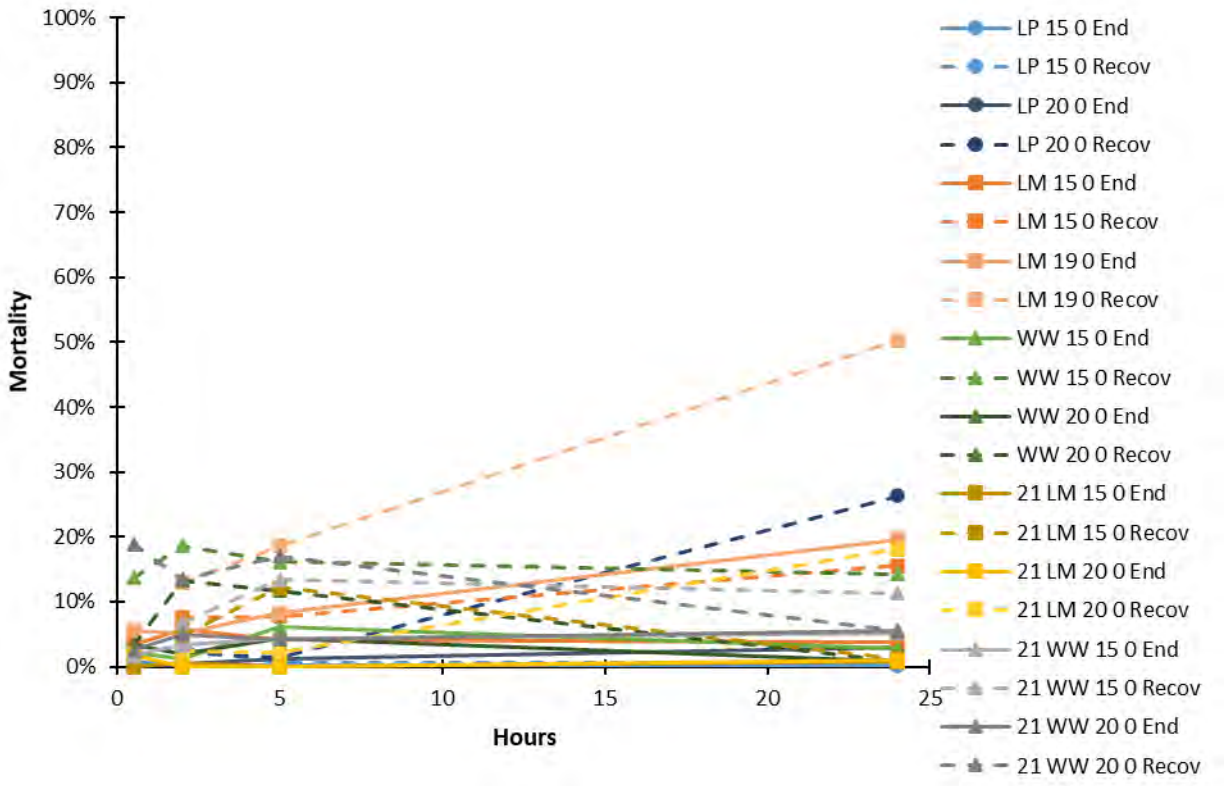

Figure L1. Veliger mortality of the control treatment at the end of each exposure and recovery period at 15 & 20°C for Lake Piru (LP), 2020 and 2021 Lake Mathews (LM), and 2020 and 2021 Weymouth WTP (WW) trials

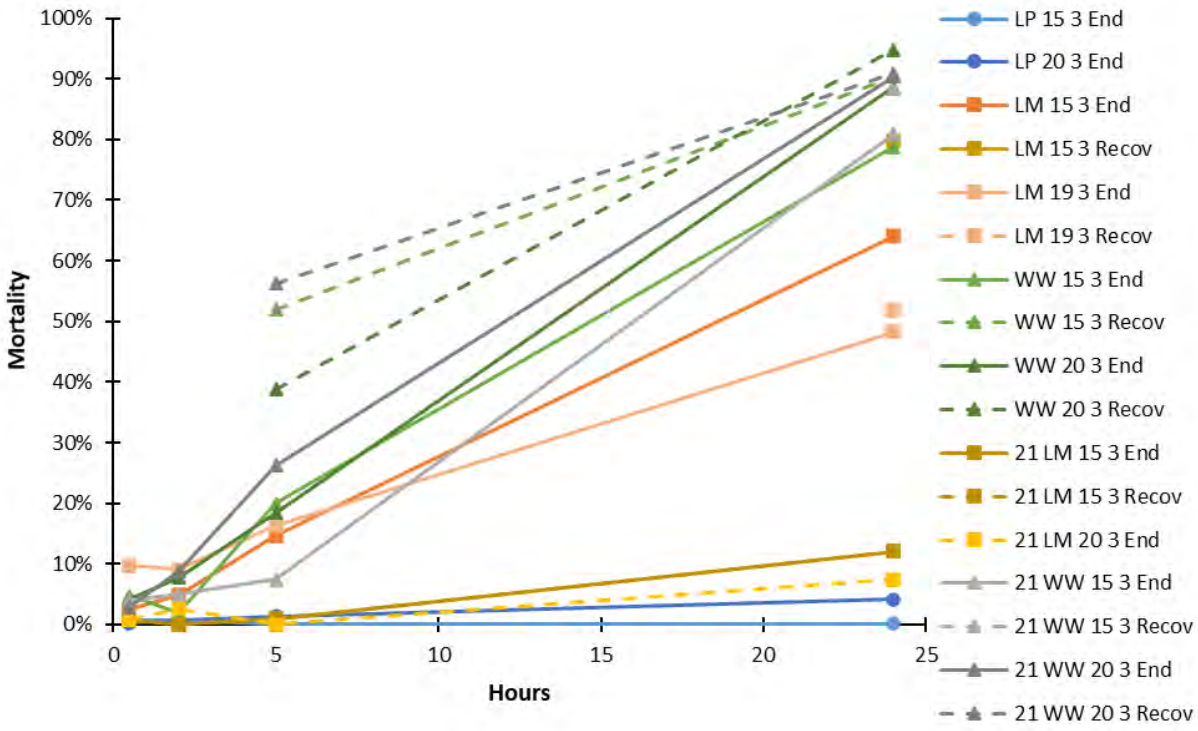

Figure L2. Veliger mortality of the EarthTec QZ 3 µl/L treatment at the end of each exposure and recovery period at 15 & 20°C for Lake Piru (LP), 2020 and 2021 Lake Mathews (LM), and 2020 and 2021 Weymouth WTP (WW) trials

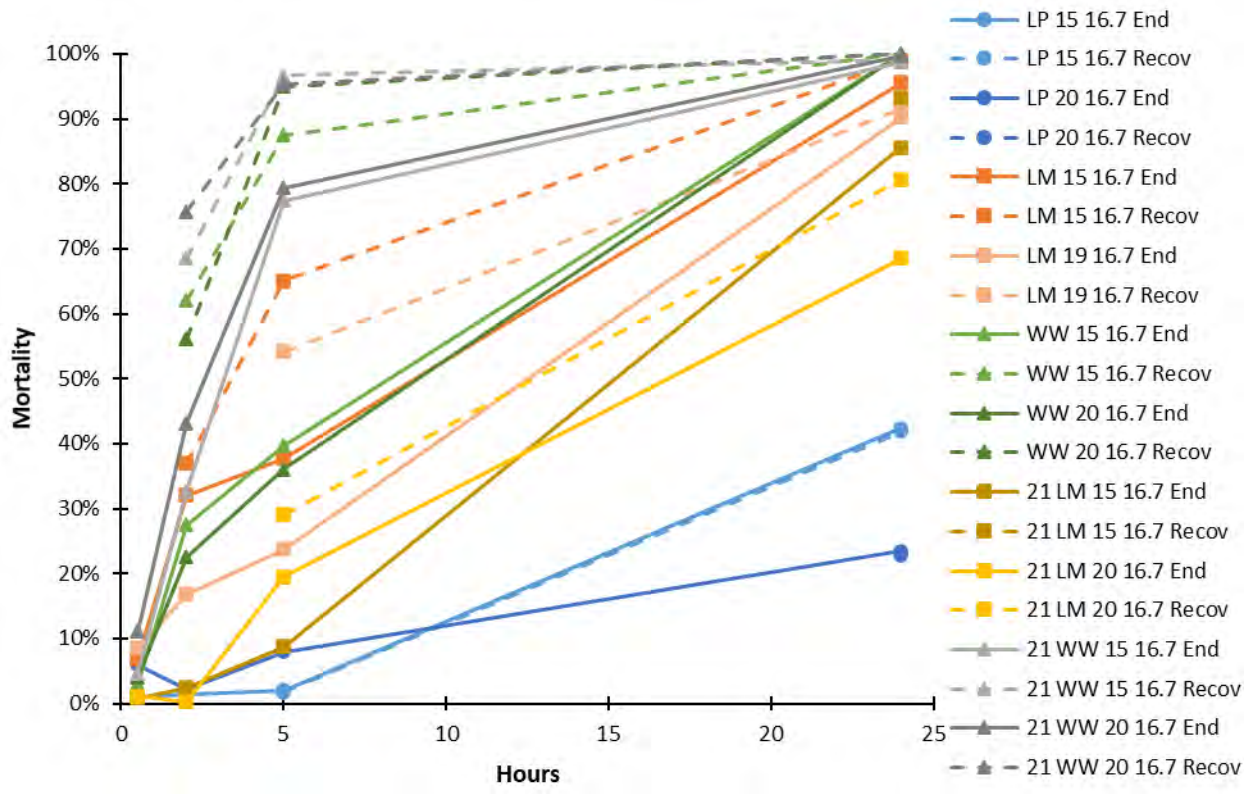

Figure L3. Veliger mortality of the EarthTec QZ 16.7 µl/L treatment at the end of each exposure and recovery period at 15 & 20°C for Lake Piru (LP), 2020 and 2021 Lake Mathews (LM), and 2020 and 2021 Weymouth WTP (WW) trials

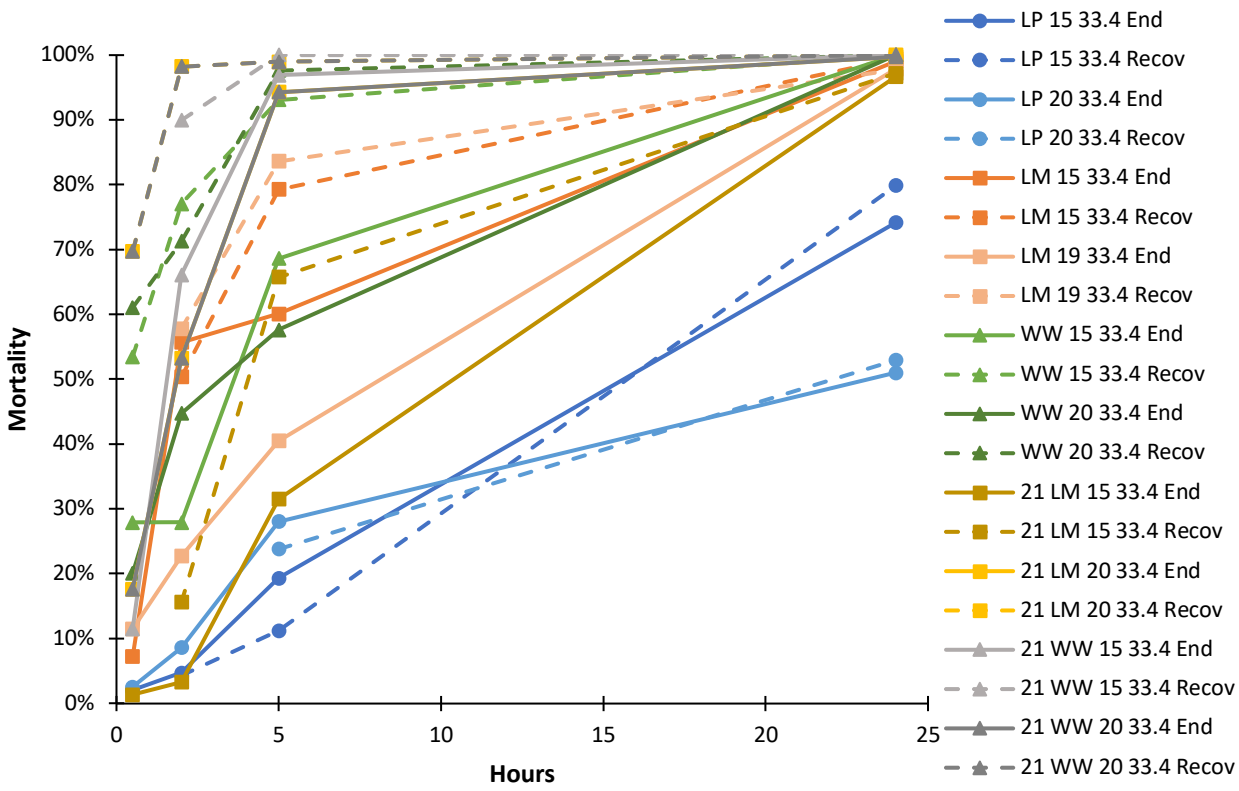

Figure L4. Veliger mortality of the EarthTec QZ 33.4 µl/L treatment at the end of each exposure and recovery period at 15 & 20°C for Lake Piru (LP), 2020 and 2021 Lake Mathews (LM), and 2020 and 2021 Weymouth WTP (WW) trials

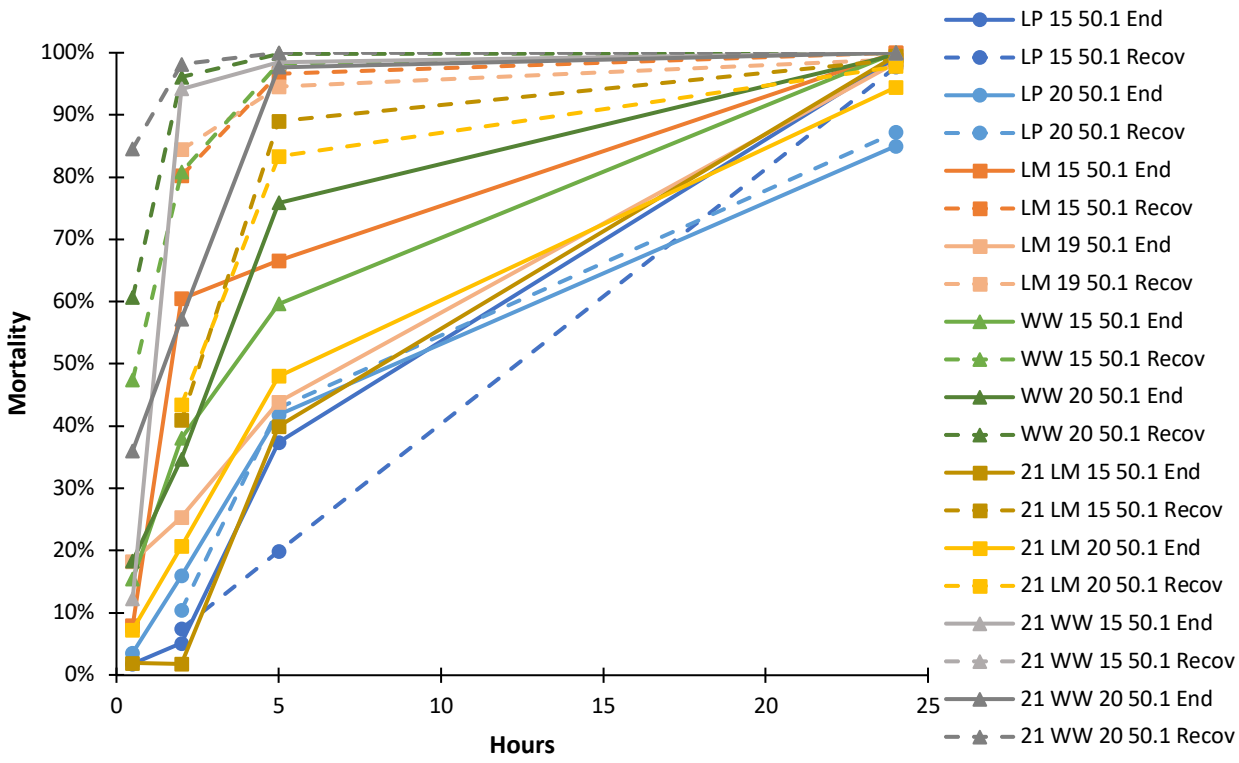

Figure L5. Veliger mortality of the EarthTec QZ 50.1 µl/L treatment at the end of each exposure and recovery period at 15 & 20°C for Lake Piru (LP), 2020 and 2021 Lake Mathews (LM), and 2020 and 2021 Weymouth WTP (WW) trials

Appendix B. Technical Memorandum 2 Toxicity Assessment

TECHNICAL MEMORANDUM 2

Toxicity Assessment

Final Date: December 20, 2021
Draft Date: September 29, 2021

Prepared for: Metropolitan Water District of Southern California
City of Santa Ana
Orange County Water District

Copies to: Water Replenishment District of Southern California
United Water Conservation District
Main San Gabriel Basin Watermaster
Central Basin Municipal Water District
California Department of Fish and Wildlife
California Department of Water Resources

Authors: Emily Owens-Bennett, P.E., Trussell
Liana Olivas, Trussell
Chao-Chun Yang, Trussell
Kelly Stockton-Fiti, KASF Consulting

Reviewers: David Hokanson, Ph.D., P.E., BCEE, Trussell
Adam Hutchinson, PG, CHG, OCWD
Christine Pham, OCWD
Tessa Lenz, UWCD

Subject: Technical Memorandum 2

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1 Introduction

1.1 Background

Since their discovery in Lake Mead within the Colorado River system in January 2007, Quagga mussels (*Dreissena bugensis*) have rapidly spread through the Metropolitan Water District of Southern California's (MWD's) Colorado River Aqueduct (CRA) and have been detected at many sites in California. The presence of this invasive species presents a barrier to the use of imported water and storm water for groundwater replenishment. Considering these challenges, the City of Santa Ana proposed a study entitled "Restoration of Local Recharge Sources from Invasive Dreissenid Mussels" (Study) to MWD's Future Supply Actions Funding Program. The Study was conducted by Trussell Technologies (Trussell), KASF Consulting, and Orange County Water District (OCWD), with support from project partners including the Water Replenishment

District of Southern California (WRD), the United Water Conservation District (UWCD), the Main San Gabriel Basin Watermaster, and the Central Basin Municipal Water District. Although not directly contributing financially to the study, the California Department of Fish and Wildlife (CDFW) and California Department of Water Resources are supporting the Study by providing technical review and assistance.

1.2 Study Description

The purpose of this Study is to evaluate a potential treatment method, other than desiccation, for use in addressing quagga mussel infestations and preventing the spread of this invasive species.

Trussell led the study in partnership with KASF Consulting, a firm specializing in quagga mussel sampling, identification, and testing. The Study evaluated the effectiveness of EarthTec QZ™[1] for control of quagga mussel veligers (the mussel's final larval stage). The testing locations included: (1) Lake Piru, (2) Lake Mathews, and (3) MWD's Weymouth Water Treatment Plant (WTP). A fourth location, OCWD's OC-28 turnout at Anaheim Lake, was originally planned but later eliminated based on scheduling issues and identification of a surrogate location. Lake Piru contains mostly imported water from the State Water Project (SWP), supplemented with local runoff. Lake Mathews is part of the CRA system. Water from Lake Mathews is chlorinated at the inlet to a pipeline that supplies Weymouth WTP; thus, these two locations represent the same water, with and without chlorine treatment. The Study included testing with samples collected from multiple locations, to allow the project team to evaluate the impact of site-specific water quality on the performance of EarthTec QZ.

The following objectives were evaluated at each testing location:

1. Veliger dose-response testing of EarthTec QZ
2. Toxicity assessment of non-target aquatic species

The purpose of this technical memorandum (TM) is to summarize the results of **Objective #2** for the study.

2 Schedule Summary

Veliger dose-response testing and associated toxicity assessments for non-target species were completed according to the schedule summarized in Table 2-1.

[1] EarthTec QZ™ is registered with the United States Environmental Protection Agency (USEPA) as a molluscicide for the control of quagga and zebra mussels. It is also registered with the California Department of Pesticide Regulation as a molluscicide for use in lakes, streams, rivers, waterways, canals, ponds, reservoirs, irrigation systems, and catch basins.

Table 2-1. Study status at each sampling location

Location	Study Status	Testing Date
Lake Piru	Completed	December 2 – 6, 2019
Lake Mathews 2020	Completed	September 9 – 11, 2020 September 18 – 20, 2020
Weymouth WTP Influent 2020	Completed	September 14 – 18, 2020
OC-28 Turnout	N/A[2]	
Weymouth WTP Influent 2021	Completed	June 28 – 30, 2021 July 12 – 14, 2021
Lake Mathews 2021	Completed	June 30 – July 2, 2021 July 14 – 16, 2021

2.1 Exemption of OC-28 Turnout

Testing was completed for each of the study testing locations (Table 2-1), with the exception of the OC-28 turnout. The study was designed with the intention of correlating the dose-response efficacy of EarthTec QZ at Lake Mathews with that of two downstream locations. Water delivered from Lake Mathews is chlorinated prior to entering transmission pipelines supplying both OC-28 and Weymouth WTP. Considering the connection of these water supplies, the Study aimed to correlate testing from Lake Mathews with the two downstream locations – OC-28 turnout and Weymouth WTP influent. Testing these three locations in a series would allow for comparison of the effects of EarthTec QZ with and without chlorine exposure. However, due to prior budgeting, OCWD only had access to Lake Mathews water at the OC-28 turnout through June 2020, and not during the 2021 fiscal year (July 2020 – June 2021). Covid-19 restrictions precluded the project team from sampling at Lake Mathews until September 2020. For these reasons, it was not possible to complete sampling from the OC-28 turnout in conjunction with the sampling and testing at Lake Mathews and Weymouth WTP influent.

The project team met with MWD staff in August 2020 to discuss alternative sampling locations for the OC-28 turnout. Although already included as a sampling location for the Study, Weymouth WTP was identified as the most appropriate surrogate monitoring location for the OC-28 turnout, as both sites can receive CRA water from Lake Mathews that is chlorinated and transmitted within a pipeline for a similar travel time and therefore, similar contact time with chlorine. Thus, sampling at OC-28 was eliminated as a sampling location for the Study. The project team proceeded with testing at Lake Mathews and Weymouth WTP during September 2020 and completed follow up testing at both of these locations in 2021.

[2] Sampling at OC-28 is no longer applicable given the similarity with that of Weymouth WTP, as is later discussed.

2.2 Repetition of Lake Mathews and Weymouth WTP

The project team found value in repeating paired veliger and toxicity testing at Lake Mathews and Weymouth WTP (2020 and 2021). Advantages of repeating this testing included:

1. Repeating toxicity testing for Weymouth WTP influent water samples after inconsistent acute toxicity test results from the 2020 Weymouth WTP samples (discussed in Section 3);
2. Assessing the same acute toxicity doses on all three non-target species with both Lake Mathews and Weymouth WTP sample water;
3. Capturing seasonal variability by testing water samples from these locations collected during different months in the year;
4. Potentially capturing a larger veliger size-distribution and assessing the effects of EarthTec QZ on larger quagga mussel veligers; and
5. Comparing the effect that EarthTec QZ has on veligers with no significant exposure to chlorine (Lake Mathews) to those which have been exposed to chlorine (Weymouth WTP influent).

The project team proceeded with testing at Weymouth WTP and Lake Mathews in June & July 2021.

3 Study Results

Objective #2 of the Study was to assess acute toxicity of EarthTec QZ for non-target aquatic species at the four Study locations. Testing was completed as defined in the 'Test Plan for Toxicity Assessment of Non-Target Aquatic Species,' provided in the Semi-Annual Progress Report 2019 No. 1.

Trussell collected large volume samples from each test location and delivered these to Aquatic Bioassay and Consulting Laboratories (ABC Labs) in Ventura, CA to conduct acute toxicity testing on three non-targeted indicator species: fathead minnow (*Pimephales promelas)*, water flea (*Ceriodaphnia dubia*), and rainbow trout (*Oncorhynchus mykiss)*. This testing was completed in parallel with the dose-response veliger testing. Each round of toxicity testing utilized the same site-specific water and EarthTec QZ stock that was used during the veliger dose-response testing (findings from Objective #1 are reported in TM 1). ABC Labs evaluated 96-hour acute toxicity for either two or three doses of EarthTec QZ per water sample. The EarthTec QZ doses were determined by Trussell and KASF Consulting based on the most effective dose identified from preliminary veliger dose-response data from each test location.

The results from the acute toxicity testing for non-target indicators are discussed in the following subsections by sampling event: Lake Piru, Lake Mathews 2020, Weymouth WTP influent 2020, Lake Mathews 2021, and Weymouth WTP influent 2021. Additional plots of the results are provided in Appendix A and the individual test reports for the acute toxicity testing from each location are provided in Appendices B-G.

3.1 Lake Piru

In December 2019, toxicity testing was conducted with Lake Piru water. The preliminary data from the Lake Piru 15ºC veliger dose-response tests suggested that veligers were only experiencing significant mortality (>50%) at the two highest doses of EarthTec QZ: 33.4 and 50.1 µL/L. Trussell and KASF Consulting selected the EarthTec QZ dose with the highest veliger mortality (50.1 µL/L or 3.0 mg/L as Cu), and the lowest dose that resulted in mortality greater than control mortality (16.7 µL/L or 1.0 mg/L as Cu) as the targets for acute toxicity testing on the three indicator species. The results are shown in Table 3-1 and presented graphically in Appendix A. The full toxicity test reports for Lake Piru are included as Appendix B.

Table 3-1. 96-hour acute toxicity results of non-target indicator species in Lake Piru sample water

	Fathead Minnow			Water Flea			Rainbow Trout		
EarthTec QZ Dose	**0 µL/L**	**16.7 µL/L**	**50.1 µL/L**	**0 µL/L**	**16.7 µL/L**	**50.1 µL/L**	**0 µL/L**	**16.7 µL/L**	**50.1 µL/L**
Average 96-hour Survival Rate	100%	57.5%	0%	100%	0%	0%	100%	0%	0%
TU(a) *	-	0.96	>1.00	-	>1.00	>1.00	-	>1.00	>1.00

**The acute toxicity unit [TU(a)] is calculated as: log (% mortality)/1.7*

As shown in Table 3-1, the average survival rate for the fathead minnow was 57.5% after 96 hours of exposure to 16.7 µL/L EarthTec QZ. This same dose and exposure time resulted in 0% survival for water fleas and rainbow trout. None of the tested species showed survival after 96 hours of exposure to 50.1 µL/L EarthTec QZ. Acute toxicity results are typically presented relative to the effective concentration (EC) required to reach a biological endpoint (mortality, in this case). One toxic unit (TU) is defined for the standard acute toxicity analysis as EC50, or the effective concentration at which 50% of the test species reaches mortality by the end of the 96-hour exposure time. Both of the EarthTec QZ doses evaluated with the Lake Piru water samples (16.7 and 50.1 µL/L) yielded acute toxicity values of greater than 1.0 TU(a).

The Test Plan included provisions for evaluating chronic toxicity for the non-target aquatic indicator species pending Round 1 acute toxicity testing results. Because Round 1 acute toxicity testing results indicated very limited survival rates (i.e., high levels of mortality) and chronic toxicity testing would entail longer exposure times, it is expected that chronic toxicity testing would not provide additional value to the study. For these reasons, toxicity testing focused on acute toxicity response for the non-target aquatic indicator species with subsequent rounds of testing at Lake Mathews and Weymouth WTP influent.

3.2 Lake Mathews 2020

In September 2020, toxicity testing was conducted with Lake Mathews water. The preliminary data from the Lake Mathews 20ºC veliger dose-response testing suggested that veligers were more susceptible to lower EarthTec QZ doses when compared to Lake Piru. Veliger mortality was higher than 90% at 16.7 µL/L EarthTec QZ (1.0 mg/L as Cu) after 24 hours of exposure. Based on these veliger mortality results, Trussell and KASF Consulting selected 8.35 µL/L (50% of 16.7 µL/L or 0.5 mg/L as Cu) and 16.7 µL/L EarthTec QZ as the target test doses for acute toxicity assessment. The results are summarized in Table 3-2 and graphed in Appendix A. The full toxicity test reports for the Lake Mathews 2020 samples are included as Appendix C.

Table 3-2. 96-hour acute toxicity results of non-target indicator species in Lake Mathews 2020 water

	Fathead Minnow			Water Flea			Rainbow Trout		
EarthTec QZ Dose	**0 µL/L**	**8.35 µL/L**	**16.7 µL/L**	**0 µL/L**	**8.35 µL/L**	**16.7 µL/L**	**0 µL/L**	**8.35 µL/L**	**16.7 µL/L**
Average 96-hour Survival Rate	100%	97.5%	92.5%	100%	0%	0%	100%	0%	0%
TU(a)	-	0.23	0.51	-	>1.00	>1.00	-	>1.00	>1.00

As shown in Table 3-2 the average survival rate for fathead minnow was 97.5% (0.23 TU(a)) and 92.5% (0.51 TU(a)) after 96 hours of exposure to 8.35 µL/L and 16.7 µL/L EarthTec QZ, respectively. However, 96-hour exposure to both tested doses resulted in 0% survival for water fleas and rainbow trout, which translates to acute toxicity values of greater than 1.0 TU(a) for both EarthTec QZ doses.

3.3 Weymouth Water Treatment Plant Influent 2020

In September 2020, toxicity testing was conducted with Weymouth WTP influent. Preliminary data from the 20ºC veliger dose-response testing in Weymouth WTP influent indicated similar or higher efficacy of EarthTec QZ as was observed with Lake Mathews water. After 24 hours of exposure to 16.7 µL/L EarthTec QZ (1.0 mg/L as Cu), veliger mortality was 93%. ABC Labs completed the acute toxicity assessment with the Weymouth WTP influent using the same doses as those tested with Lake Mathews water: 8.35 µL/L and 16.7 µL/L EarthTec QZ.

The final veliger mortality results from the Weymouth WTP influent indicated significantly higher mortality (78-89% at 24 hours exposure) at the lowest EarthTec QZ dose of 3.0 µL/L (0.18 mg/L as Cu) when compared with the same dose at Lake Mathews (48-65% at 24 hours exposure). Therefore, Trussell and KASF Consulting requested that ABC Labs analyze acute toxicity for the Weymouth WTP influent with an additional dose of 3.0 µL/L EarthTec QZ. This additional test condition required re-

sampling Weymouth WTP influent water, which took place two weeks after the original testing. The results from both rounds of acute toxicity testing with Weymouth WTP influent are summarized in Table 3-3 and graphed in Appendix A. The full toxicity test reports for the original Weymouth WTP influent 2020 samples are included as Appendix D. The full toxicity test reports for the Weymouth WTP 2020 re-sample are included as Appendix E.

Table 3-3. 96-hour acute toxicity results of non-target indicator species in Weymouth WTP 2020 sample water

	Fathead Minnow				Water Flea				Rainbow Trout			
EarthTec QZ Dose	**0 µL/L**	**3 µL/L**	**8.35 µL/L**	**16.7 µL/L**	**0 µL/L**	**3 µL/L**	**8.35 µL/L**	**16.7 µL/L**	**0 µL/L**	**3 µL/L**	**8.35 µL/L**	**16.7 µL/L**
Average 96-hour Survival Rate	97.5%	100%	100%	100%	100%	0%	100%	100%	100%	65%	0%	0%
TU(a)	0.23	-	-	-	-	>1.00	-	-	-	0.91	>1.00	>1.00

As shown in Table 3-3, the fathead minnow survival rate was 100% after 96 hours of exposure to all three tested doses. An average of 65% of the rainbow trout survived after 96 hours of exposure to 3.0 µL/L EarthTec QZ, which translates to an acute toxicity value of 0.91 TU(a). The same exposure to the two higher doses of 8.35 µL/L and 16.7 µL/L EarthTec QZ both resulted in 0% survival and acute toxicity values greater than 1.0 TU(a).

Water fleas survived at a rate of 100% at the two higher doses of 8.35 µL/L and 16.7 µL/L EarthTec QZ. However, 0% of water fleas survived the lower dose of 3 µL/L EarthTec QZ, which translates to an acute toxicity value of 1.0 TU(a).

It is unclear why no water fleas survived at the lowest EarthTec QZ dose, whereas 100% survival was observed with the two higher doses. As previously mentioned, the addition of the third and lower dose condition (3.0 µL/L EarthTec QZ) required a separate sampling event two weeks after the initial Weymouth WTP influent sample was collected for the higher two doses (8.35 µL/L and 16 µL/L EarthTec QZ). The results indicate limited change in the water quality of the two Weymouth WTP samples. A slightly higher chlorine residual (0.05 mg/L) was measured in the second sample, used for the 3.0 µL/L EarthTec QZ condition, compared to the original water sample (0.02 mg/L). ABC Labs used the same batch of diluted EarthTec QZ for dosing the tested conditions for both rounds of toxicity testing. ABC Labs conducted a quality control audit of the results and found no obvious explanation for the inconsistent results.

As previously discussed in Section 2, these inconsistent results were one of the motivations to repeat testing at both Lake Mathews and Weymouth WTP.

3.4 Weymouth Water Treatment Plant Influent 2021

In July 2021, a repeat round of toxicity testing was conducted with Weymouth WTP water. As discussed in the previous section, the acute toxicity tests with the Weymouth WTP 2020 sample water resulted in inconsistent water flea survival data. To provide more clarity on the effects that EarthTec QZ has on the water flea, the same three doses that were used in the 2020 Weymouth WTP toxicity tests were selected for the follow up testing in 2021: 3, 8.35 & 16.7 µL/L as EarthTec QZ, or 0.18, 0.5 and 1.0 mg/L as Cu, respectively. The results are presented in Table 3-4 and graphed in Appendix A. The full toxicity test reports for Lake Mathews 2021 are included as Appendix F.

Table 3-4. 96-hour acute toxicity results of non-target indicator species in Weymouth WTP 2021 sample water

	Fathead Minnow				Water Flea				Rainbow Trout			
EarthTec QZ Dose	**0 µL/L**	**3 µL/L**	**8.35 µL/L**	**16.7 µL/L**	**0 µL/L**	**3 µL/L**	**8.35 µL/L**	**16.7 µL/L**	**0 µL/L**	**3 µL/L**	**8.35 µL/L**	**16.7 µL/L**
Average 96-hour Survival Rate	100%	100%	97.5%	97.5%	100%	0%	0%	0%	100%	100%	0%	0%
TU(a)	-	-	0.23	0.23	-	>1.00	>1.00	>1.00	-	-	>1.00	>1.00

As shown in Table 3-4, the average survival rate for fathead minnow and rainbow trout was 100% after 96 hours of exposure to the lowest tested dose 3 µL/L EarthTec QZ. However, the average survival rate for water flea was 0% after 96 hours of exposure to this same lowest dose. Furthermore, the average survival rate for fathead minnow was 97.5% (0.23 TU(a)) after 96 hours of exposure to both 8.35 µL/L and 16.7 µL/L EarthTec QZ. However, 96-hour exposure to 8.35 µL/L and 16.7 µL/L EarthTec QZ resulted in 0% survival for water fleas and rainbow trout, which translates to acute toxicity values of greater than 1.0 TU(a) for both EarthTec QZ doses.

3.5 Lake Mathews 2021

In July 2021, a repeat round of toxicity testing was conducted with Lake Mathews sample water. As discussed in the previous section, the original acute toxicity tests with Weymouth WTP sample water resulted in inconsistent water flea survival data. To obtain consistent data with the Weymouth WTP, the same three doses that were used for the repeated Weymouth WTP toxicity testing (3, 8.35 & 16.7 µL/L as EarthTec QZ, or 0.18, 0.5 and 1.0 mg/L as Cu, respectively) were selected for this location as well. The results are presented in Table 3-5 and graphed in Appendix A. The full toxicity test reports for Lake Mathews are included as Appendix G.

Table 3-5. 96-hour acute toxicity results of non-target indicator species in Lake Mathews 2021 sample water

	Fathead Minnow				Water Flea				Rainbow Trout			
EarthTec QZ Dose	**0 µL/L**	**3 µL/L**	**8.35 µL/L**	**16.7 µL/L**	**0 µL/L**	**3 µL/L**	**8.35 µL/L**	**16.7 µL/L**	**0 µL/L**	**3 µL/L**	**8.35 µL/L**	**16.7 µL/L**
Average 96-hour Survival Rate	100%	100%	100%	97.5%	100%	15%	0%	0%	100%	100%	0%	0%
TU(a)	-	-	-	0.23	-	>1.00	>1.00	>1.00	-	-	>1.00	>1.00

As shown in Table 3-5, the average survival rate for fathead minnow and rainbow trout was 100% after 96 hours of exposure to the lowest tested dose 3 µL/L EarthTec QZ. However, the average survival rate for water flea was only 15% (>1.0 TU(a)) after 96 hours of exposure to this same lowest dose. The average fathead minnow survival rates after 96 hours of exposure to 8.35 µL/L and 16.7 µL/L EarthTec QZ were also high at 100% and 97.5% (0.23 TU(a)), respectively. However, the same exposure to 8.35 µL/L and 16.7 µL/L EarthTec QZ resulted in 0% survival for water fleas and rainbow trout, or acute toxicity values of greater than 1.0 TU(a).

4 Analysis

This section includes a high-level analysis of the results presented in Section 3. Further discussion, correlations, statistical analysis, integrated analyses and recommendations will be provided in the Final Report.

4.1 Experimental Setup

The toxicity assessment for non-target organisms was based on the standard acute toxicity test method, which exposes each of the three indicator species to the selected EarthTec QZ dose for 96 hours. This is 72 hours longer than the longest exposure time for the veliger dose-response tests at 24 hours (an additional 24-hour recovery period was assessed after exposure to fresh sample water without EarthTec QZ). Therefore, it should be noted that the non-target indicator species experienced significantly longer exposure times to EarthTec QZ compared to the veligers in the corresponding dose-response tests. The final report compares the acute toxicity results with the veliger mortality on the basis of total exposure to EarthTec QZ, using a time-dose variable (the product of the copper concentration and exposure time of each sample).

Another notable difference in the veliger dose-response testing conditions compared with the acute toxicity test procedure is the ambient temperature. The veliger dose-response testing included two temperature conditions, 15 ºC and 20ºC. ABC Labs followed the standard method for acute toxicity testing and maintained a temperature of approximately 24ºC for the fathead minnow and water flea, and a temperature of

approximately 13ºC for rainbow trout. These temperature conditions were selected as they provide constant and acceptable temperature conditions for the non-target indicator species, so that the control may yield high survival and varying temperature does not add an additional factor that could cause non-chemical related mortality.

4.2 Water Quality Comparison

The water quality associated with acute toxicity testing is shown in Table 4-1. Raw water quality values include the sample water that the project team collected and analyzed during veliger testing, along with the water quality analysis provided by ABC Labs for the same sample water.[3] The results in Table 4-1 indicated as being analyzed by ABC Labs include both raw water quality and measurements from the sample containing EarthTec QZ. While copper was not reported by ABC Labs, it was measured to confirm accurate dosing of EarthTec QZ during testing. The full laboratory reports provided by ABC Labs for each of the tests are included in **Appendices B through G**.

The water quality for Lake Mathews and Weymouth WTP was mostly consistent, since these two locations represent the same CRA water, with the addition of chlorine in the pipeline upstream of Weymouth WTP. Lake Piru represents mostly imported water from the SWP and had higher DOC, hardness, and alkalinity, but lower conductivity compared with Lake Mathews and Weymouth WTP. Similar pH values were measured from all three locations.

[3] ABC Labs used both the sample raw water and their own lab-made water for the control conditions during acute toxicity testing. The control conditions with the sample water served as a quality control method to rule out mortality to the indicator species caused by the water sample, and not the EarthTec QZ. All the control conditions with sample water yielded 100% survival, confirming that none of the mortality was attributed to the sample source. The survival results reported in **Section 3** for 0 µL/L EarthTec QZ reflect the test results run with the lab-made control water. The water quality titled 'ABC Labs: Raw Water' in **Table 4-1** reflects the sample water quality measured by ABC Labs.

Table 4-1. Water Quality of Acute Toxicity Test Samples with and without EarthTec QZ

Location	Year	Analysis	Test Condition	Dissolved Oxygen mg/L	Conductivity µS/cm	pH pH unit	Temperature °C	Alkalinity mg/L as CaCO3	Hardness mg/L as CaCO3	Total Copper mg/L	Free Copper mg/L	Total Chlorine mg/L	ORP mV	Chemical Oxygen Demand mg/L	Dissolved Organic Carbon mg/L
Lake Piru	**Raw Water Quality**														
	2019	Field	20ºC Tests	8.88	913	8.25	14.8	154	316	0.16	0.16	0.12	192	5.65	5.05
		ABC Labs	Raw Water	7.50	776	7.70	24.0	98	315	-	-	-	-	-	-
			16.7 µL/L EarthTec (1.0 mg/L as Cu)	8.37	947	8.83	24.1	-	-	-	-	-	-	-	-
			50.1 µL/L EarthTec (3.0 mg/L as Cu)	8.25	990	8.10	24.1	160	161	-	-	-	-	-	-
	Statistical Analysis														
	All Sample Collections		Mean	8.19	845	7.98	19.4	126	316	0.16	0.16	0.12	192	5.65	5.05
			Standard Deviation	0.98	97	0.39	6.5	40	0.71	-	-	-	-	-	-
Lake Mathews	**Raw Water Quality**														
	2020	Field	15ºC Tests	7.57	946	8.18	25.1	115	138	0.02	0.02	0.02	213	4.67	3.01
		ABC Labs	Raw Water	7.90	788	8.30	23.9	113	288	-	-	-	-	-	-
			8.35 µL/L EarthTec (0.5 mg/L as Cu)	7.60	1129	7.90	24.1	100	350	-	-	-	-	-	-
			16.7 µL/L EarthTec (1.0 mg/L as Cu)	7.46	1097	7.87	24.1	-	-	-	-	-	-	-	-
	2021	Field	15ºC Tests	8.46	950	8.24	24.6	138	281	0.00	0.00	0.01	186	4.65	2.98
		ABC Labs	Raw Water	7.83	1044	7.53	24.1	120	250	-	-	-	-	-	-
			3 µL/L EarthTec (0.15 mg/L as Cu)	8.00	1013	7.53	24.1	-	-	-	-	-	-	-	-
			8.35 µL/L EarthTec (0.5 mg/L as Cu)	7.87	1013	7.53	24.1	-	-	-	-	-	-	-	-
			16.7 µL/L EarthTec (1.0 mg/L as Cu)	7.87	1023	7.53	24.1	125	330	-	-	-	-	-	-
	Statistical Analysis														
	2020 Raw Samples		Mean	7.74	867	8.24	24.5	114	213	0.02	0.02	0.02	213	4.67	3.01
			Standard Deviation	0.23	112	0.08	0.8	1	106	-	-	-	-	-	-
	2021 Raw Samples		Mean	8.15	997.0	7.89	24.34	129	266	0.00	0.00	0.01	186	4.65	2.98
			Standard Deviation	0.44	66.47	0.50	0.37	13	22	-	-	-	-	-	-
Weymouth WTP	**Raw Water Qualty**														
	2020	Field	15ºC Tests	8.27	944	8.02	25.6	119	150	0.01	0.01	0.04	214	8.50	4.09
		ABC Labs	Raw Water	8.10	1056	8.00	24.1	111	362	-	-	-	-	-	-
			8.35 µL/L EarthTec (0.5 mg/L as Cu)	7.67	1107	7.90	24.1	108	315	-	-	-	-	-	-
			16.7 µL/L EarthTec (1.0 mg/L as Cu)	7.50	1101	7.87	24.2	-	-	-	-	-	-	-	-
		Field	Additional Sampling *	8.19	955	8.12	26.8	119	145	0.02	0.02	0.05	211	7.88	4.05
		ABC Labs	Raw Water	8.30	1022	8.10	24.0	103	311	-	-	-	-	-	-
			3 µL/L EarthTec (0.15 mg/L as Cu)	7.40	1025	7.70	24.0	113	238	-	-	-	-	-	-
	2021	Field	15ºC Tests	8.69	969	8.18	21.9	137	275	0.02	0.03	0.10	196	4.46	3.01
		ABC Labs	Raw Water	7.90	1009	7.67	24.1	123	295	-	-	-	-	-	-
			3 µL/L EarthTec (0.15 mg/L as Cu)	7.93	1011	7.70	24.1	-	-	-	-	-	-	-	-
			8.35 µL/L EarthTec (0.5 mg/L as Cu)	7.90	105	7.63	24.1	-	-	-	-	-	-	-	-
			16.7 µL/L EarthTec (1.0 mg/L as Cu)	7.87	1012	7.63	24.1	129	330	-	-	-	-	-	-
	Statistical Analysis														
	2020 Raw Samples		Mean	8.19	1000	8.01	24.9	115	256	0.01	0.01	0.04	214	8.50	4.09
			Standard Deviation	0.12	79	0.01	1.1	6	150	-	-	-	-	-	-
	2020 Raw Samples (Additional Sampling)		Mean	8.25	989	8.11	25.4	111	228	0.02	0.02	0.05	211	7.88	4.05
			Standard Deviation	0.08	47	0.01	2.0	11	117	-	-	-	-	-	-
	2021 Raw Samples		Mean	8.30	989	7.92	23.0	130	285	0.02	0.03	0.10	196	4.46	3.01
			Standard Deviation	0.56	28	0.36	1.6	10	14	-	-	-	-	-	-

* Re-sample of Weymouth WTP influent for acute toxicity testing with 3 µL/L EarthTec QZ. This water was not used for veliger testing.

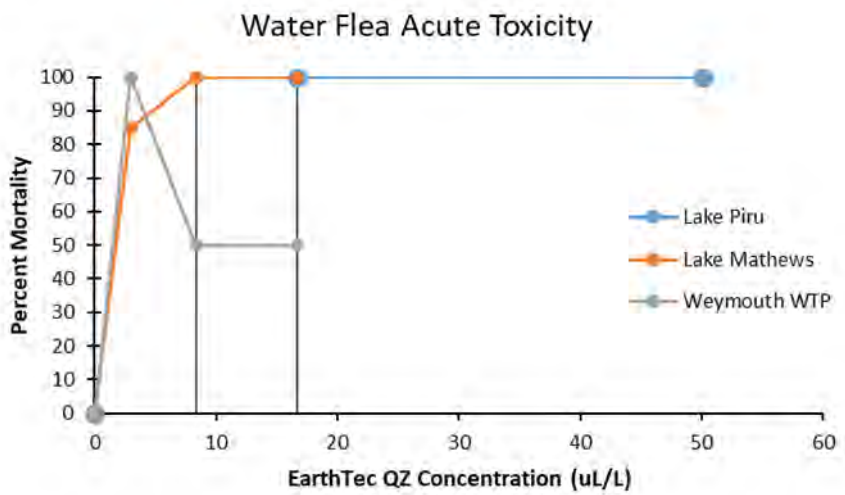
4.3 General Trends

The toxicity response of non-target organisms to EarthTec QZ were tested on three species – fathead minnow, water flea, and rainbow trout. The mortality response of the tested non-target organisms is summarized in Figure 4-1.

Both water flea and rainbow trout were highly sensitive to EarthTez QZ and exhibited acute mortality response at the lower concentrations tested in this Study. Water flea and rainbow trout acute mortality was observed to be 100% at a dose greater than or equal to 8.35 µL/L EarthTec QZ which corresponds to 0.5 mg/L as copper.

Meanwhile, fathead minnow showed a mixed response to EarthTec QZ. Lake Mathews and Weymouth WTP water test conditions showed little to no mortality of fathead minnows for all EarthTec QZ doses tested up to 16.7 µL/L which corresponds to 1 mg/L as copper. However, the Lake Piru water test conditions had 42.5% mortality at 16.7 µL/L. The difference in mortality observed for fathead minnows is thought to be the result of differences in water quality between the test locations. The discussion on water quality and mortality can be found in Section 4.4.

A.



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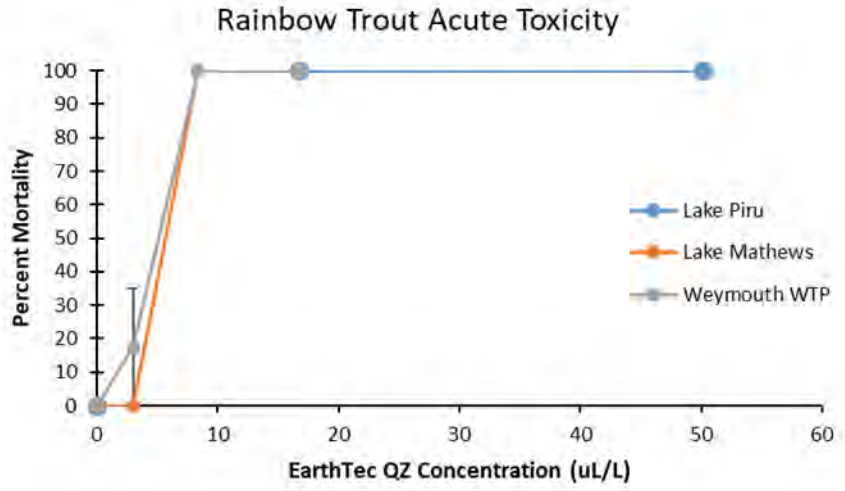


C.

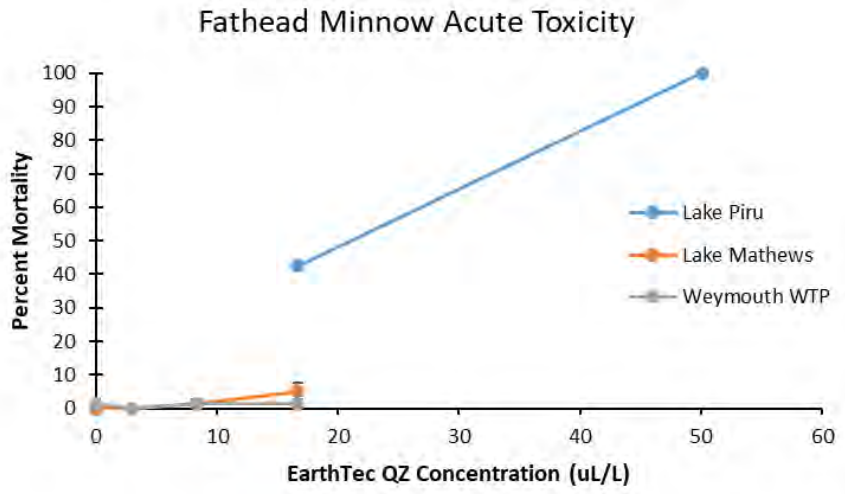


Figure 4-1 Average acute toxicity with standard deviations bars
(A) Water Flea; (B) Rainbow Trout; and (C) Fathead Minnow for each location with EarthTec QZ

4.4 Water Quality Impact on EarthTec QZ Performance

Water quality affects the toxicity of EarthTec QZ. The following information is listed on the EarthTec QZ label:

> *"Certain water conditions including low pH (≤6.5), low dissolved organic carbon (DOC) levels (3.0 mg/L or lower) and "soft" waters (i.e., alkalinity less than 50 mg/L) increases the potential acute toxicity to non-target aquatic organisms. The application rates on this label are appropriate for water with alkalinity greater than 50 mg/L. Do not use these application rates for water with less than 50 ppm alkalinity (e.g., soft or acid waters) because trout and other species of fish may be killed under such conditions."*

The raw water quality for each test condition is summarized in Table 4-1 and discussed briefly in Section 4.2. While the alkalinity, pH, and DOC are within the conditions specified by the EarthTec QZ label, it is expected that water quality still plays a role in the toxicity response. The influence of water quality on toxicity response can be explained using the biotic ligand model as described in Section 4.4.1.

4.4.1 Copper Bioavailability and Oxidants on Mortality

Copper toxicity in fish is a function of bioavailable copper as well as competitive ions in the water. Typically, the biotic ligand model is used to predict copper toxicity by accounting for bioavailability of copper and ion competition through modeling using a multitude of water quality parameters. The water quality parameters used by the model are pH, DOC, alkalinity, ions, and temperature (USEPA 2016). These water quality parameters affect the chemical composition of copper in the water and determine the bioavailability. For example, alkalinity, pH, hardness, and anions affect the saturation indices for copper and the percentage of total copper existing in dissolved form. It has been demonstrated that copper toxicity decreases when pH and hardness increase (Welsh et al. 1995). Additionally, DOC can also form organic complexes with copper that decrease the fraction of bioavailable copper existing in the dissolved fraction. Meanwhile, other cations, especially sodium, can compete with free copper ions for binding sites in the gills of fish (Nelson et al., 1986; Welsh et al. 1995; Parametrix and HydroQual 2006; USEPA 2016). Therefore, different waters can exhibit a wide range of mortality response at the same given dose of EarthTec QZ due to site-specific water quality differences that affect copper toxicity.

The water quality of the three test locations varied for the influential parameters related to the biotic ligand model, which likely explained why there was no clear trend in the toxicity of EarthTec QZ to the three non-target species based on location. Lake Piru had higher DOC, hardness, and alkalinity compared with Lake Mathews and Weymouth WTP; these parameters were more correlated with lowering copper toxicity. Similar pH values were measured from all three locations. The full suite of water quality for predicting toxicity with the biotic ligand model was not measured for these tests. In prior studies conducted by KASF Consulting, sodium levels in Lake Piru in 2019 were

lower (65 mg/L) than those measured in Colorado River water (100 mg/L) in 2015. While sodium measurements were not included in this study, the higher sodium concentration for Lake Mathews and Weymouth WTP should have provided more protection from the copper toxicity for fish species. In general, water quality clearly affects copper toxicity and could be the reason for the difference in Lake Piru fathead minnow mortality.

In addition to bioavailability of copper, other toxic constituents in water are expected to increase overall mortality. While not apparent in the non-target organisms in this Study, total chlorine residual is another environmental stressor and is thought to compound copper toxicity. An increase in toxicity response is seen in veligers as seen from the results of Objective 1 of this Study.

References

Nelson, Henry, Duane Benoit, Russ Erickson, Vince Mattson, and Jim Lindberg. 1986. *The Effects of Variable Hardness, pH, and Alkalinity, and Humics on the Chemical Speciation and Aquatic Toxicity of Copper.* USEPA (Duluth, MN 55804).

Parametrix and HydroQual. 2006. Evaluation of the Reliability of Biotic Ligand Model Predictions for Copper Toxicity in Waters Characteristic of the Ardi West. Final Report for Arid West Water Quality Research Project prepared for Pima County Wastewater Management. Available: https://webcms.pima.gov/UserFiles/Servers/Server_6/File/Government/Wastewater%20Reclamation/AAWQRP/BioticLigandModel.pdf

USEPA. 2016. *Water Quality Standards Academy: Biotic Ligand Model and Copper Criteria.*

Welsh, Paul, Joanne Parrott, D. G. Dixon, Peter Hodson, Doug Spry, and G. Mierle. 2011. "Estimating acute copper toxicity to larval fathead minnow (Pimephales promelas) in soft water from measurements of dissolved organic carbon, calcium, and pH." *Canadian Journal of Fisheries and Aquatic Sciences* 53: 1263-1271.

Appendix List

APPENDIX A – PLOTS OF ACUTE TOXICITY RESULTS FROM LAKE PIRU, LAKE MATHEWS 2020, WEYMOUTH WTP 2020, WEYMOUTH WTP 2021, AND LAKE MATHEWS 2021

APPENDIX B – LAKE PIRU TOXICITY TEST REPORT

APPENDIX C – LAKE MATHEWS 2020 TOXICITY TEST REPORT

APPENDIX D – WEYMOUTH WTP INFLUENT 2020 TOXICITY TEST REPORT

APPENDIX E – WEYMOUTH WTP INFLUENT 2020 (RE-SAMPLE) TOXICITY TEST REPORT

APPENDIX F – WEYMOUTH WTP INFLUENT 2021 TOXICITY TEST REPORT

APPENDIX G – LAKE MATHEWS 2021 TOXICITY TEST REPORT

Appendix A –

Plots of Acute Toxicity Results from Lake Piru, Lake Mathews 2020, Weymouth WTP 2020, Weymouth WTP 2021, and Lake Mathews 2021

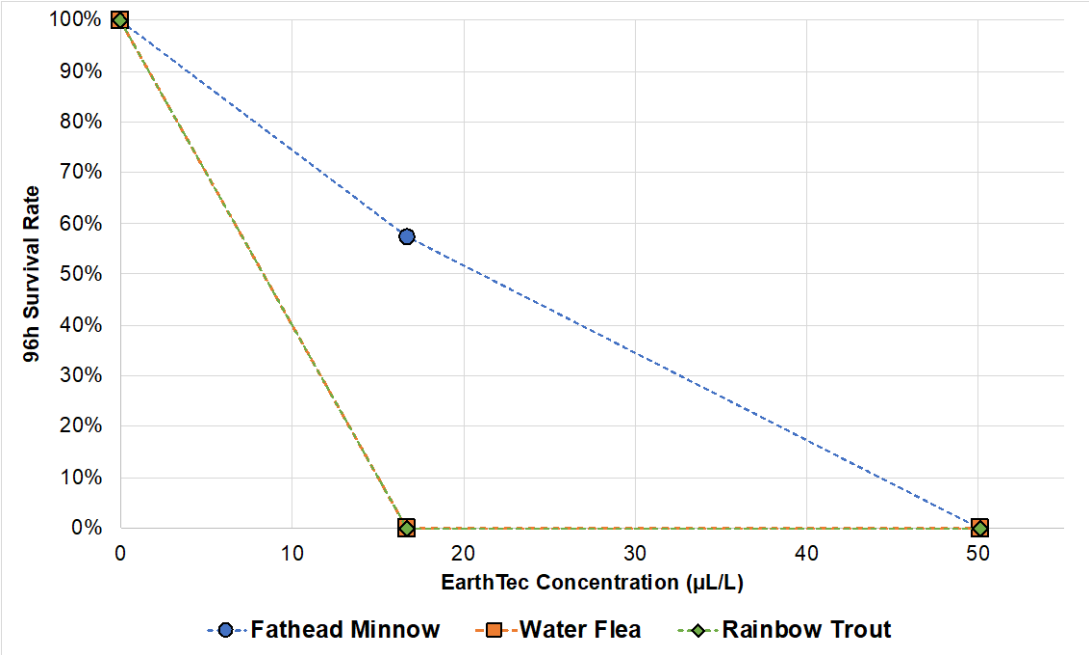


Figure A1. 96-hour acute toxicity results of non-target indicator species in Lake Piru sample water

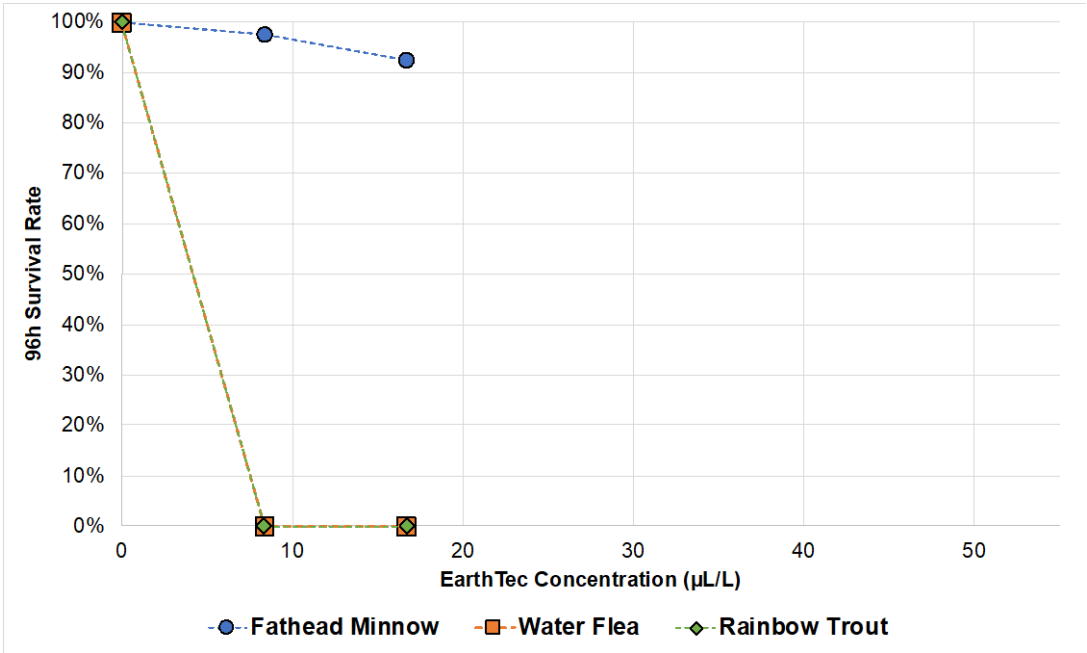


Figure A2. 96-hour acute toxicity results of non-target indicator species in Lake Mathews 2020 sample water

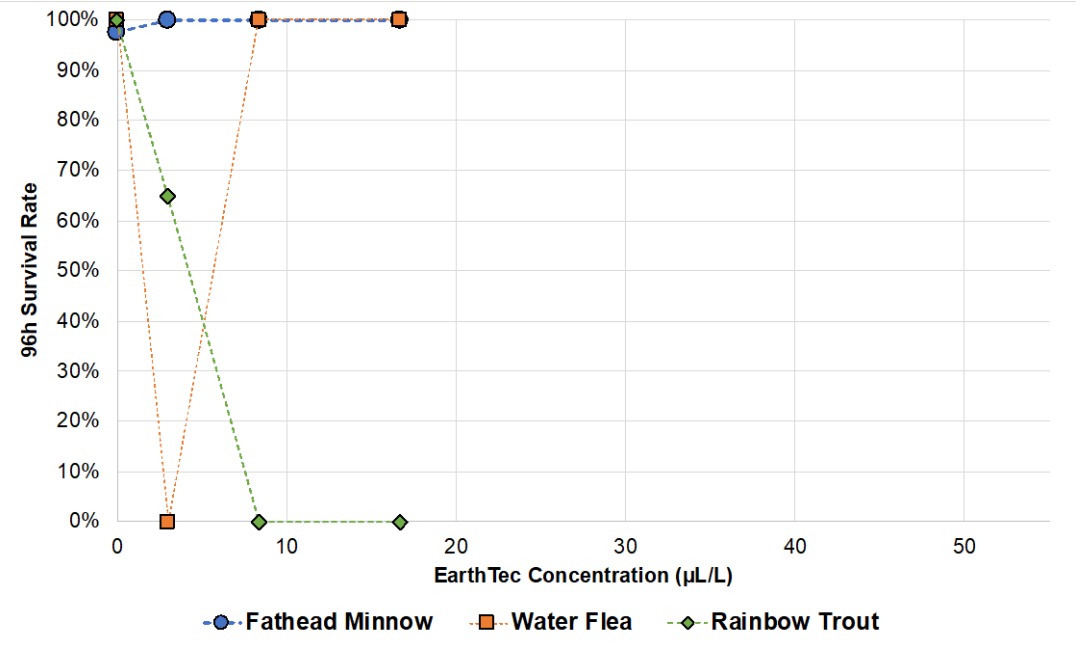


Figure A3. 96-hour acute toxicity results of non-target indicator species in Weymouth WTP influent 2020 sample water

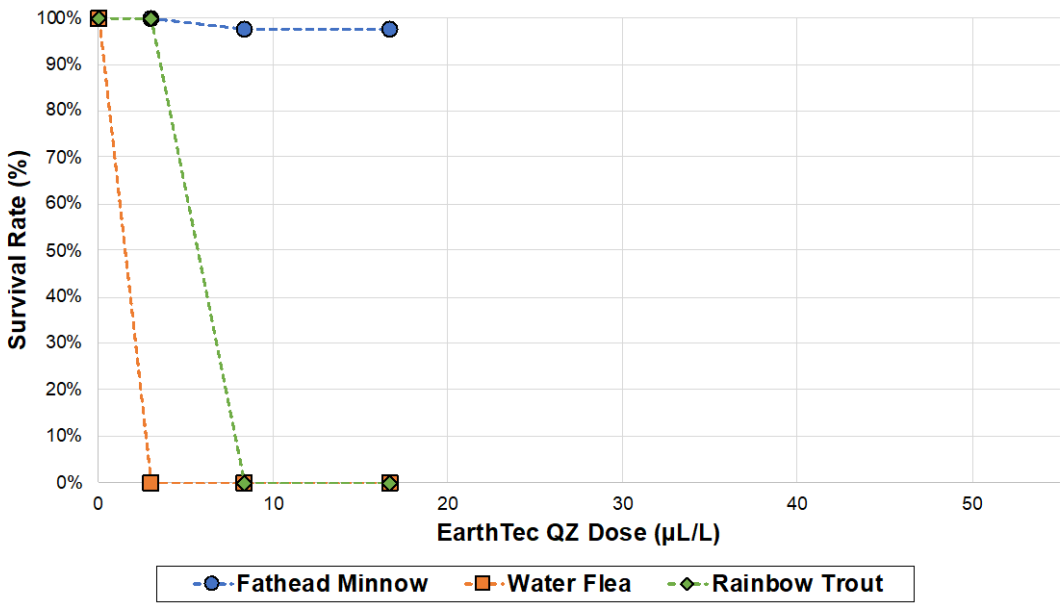


Figure A4. 96-hour acute toxicity results of non-target indicator species in Weymouth WTP influent 2021 sample water

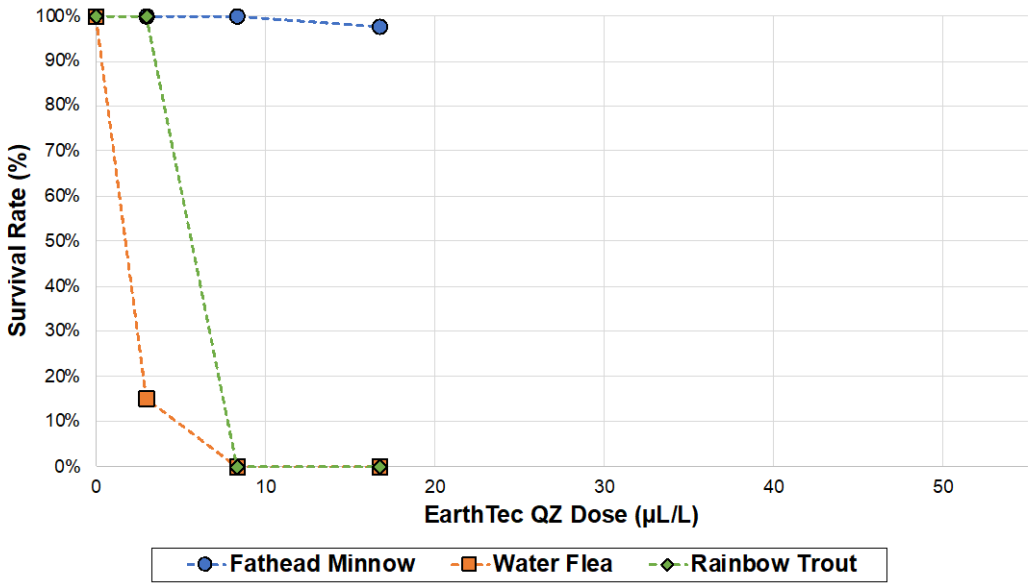


Figure A5. 96-hour acute toxicity results of non-target indicator species in Lake Mathews 2021 sample water

Appendix B –
Lake Piru Toxicity Test Report

aquatic bioassay & consulting laboratories, inc

December 23, 2019

Mr. David Hokanson
Trussell Technologies
232 N. Lake Avenue
Pasadena, CA 91101

Dear Mr. Hokanson:

We are pleased to present the enclosed bioassay report. The test was conducted under guidelines prescribed in *Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms EPA-821-R-02-012.* "All acceptability criteria were met and the concentration-response was normal. This is a valid test." Results were as follows:

CLIENT:	Trussell Technologies
SAMPLE ID.:	Lake Piru Raw
DATE RECEIVED:	6 Dec – 19
ABC LAB NO.:	EUR1219.056

ACUTE FATHEAD MINNOW SURVIVAL BIOASSAY

% Survival = 0 % Survival in 50.1 mg/l Sample
*TU(a) = >1.00 @ 16.7 mg/l
*TU(a) = >1.00 @ 50.1 mg/l
* TU(a) Is calculated by: log (% Mortality)/1.7

Yours very truly,

Scott Johnson
Laboratory Director

CETIS Summary Report

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	07-7352-6059	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	06 Dec-19 16:05	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water
Ending Date:	10 Dec-19 14:10	**Species:**	Pimephales promelas	**Brine:**	Not Applicable
Test Length:	94h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO **Age:**
Sample ID:	20-2611-5180	**Code:**	TRU1219.056afml	**Project:**	
Sample Date:	04 Dec-19 09:00	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	06 Dec-19 13:00	**CAS (PC):**		**Station:**	Lake Piru Raw
Sample Age:	55h (6.5 °C)	**Client:**	Trussell Technologies		

Single Comparison Summary

Analysis ID	Endpoint	Comparison Method	P-Value	Comparison Result	S
11-9941-2527	96h Survival Rate	Steel Many-One Rank Sum Test	0.0105	16.7mg/L failed 96h survival rate	1

Point Estimate Summary

Analysis ID	Endpoint	Point Estimate Method	✓	Level	mg/L	95% LCL	95% UCL	TU	S
05-9379-4708	96h Survival Rate	Linear Interpolation (ICPIN)		EC5	1.965	1.048	4.165		1
				EC10	3.929	2.096	8.33		
				EC15	5.894	3.144	12.5		
				EC20	7.859	4.191	16.66		
				EC25	9.824	5.239	20.83		
				EC40	15.72	8.383	27.98		
				EC50	21.06	9.633	31.9		

Test Acceptability

				TAC Limits			
Analysis ID	**Endpoint**	**Attribute**	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
05-9379-4708	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria
11-9941-2527	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria

96h Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	N	4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	0.00%	0.00%
16.7		4	0.5750	0.2470	0.9030	0.4000	0.8000	0.1031	0.2062	35.85%	42.50%
50.1		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000		100.00%

96h Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
16.7		0.8000	0.7000	0.4000	0.4000
50.1		0.0000	0.0000	0.0000	0.0000

96h Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	10/10	10/10	10/10	10/10
16.7		8/10	7/10	4/10	4/10
50.1		0/10	0/10	0/10	0/10

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	11-9941-2527	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.5
Analyzed:	19 Dec-19 12:47	**Analysis:**	Nonparametric-Control vs Treatments	**Status Level:**	1
Batch ID:	07-7352-6059	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	06 Dec-19 16:05	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water
Ending Date:	10 Dec-19 14:10	**Species:**	Pimephales promelas	**Brine:**	Not Applicable
Test Length:	94h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO **Age:**
Sample ID:	20-2611-5180	**Code:**	TRU1219.056afml	**Project:**	
Sample Date:	04 Dec-19 09:00	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	06 Dec-19 13:00	**CAS (PC):**		**Station:**	Lake Piru Raw
Sample Age:	55h (6.5 °C)	**Client:**	Trussell Technologies		

Data Transform	Alt Hyp	Comparison Result	PMSD
Angular (Corrected)	C > T	16.7mg/L failed 96h survival rate	12.96%

Steel Many-One Rank Sum Test

Control	vs	Control II	Test Stat	Critical	Ties	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		16.7*	10	12	0	6	CDF	0.0105	Significant Effect

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.594225	0.594225	1	25.55	0.0023	Significant Effect
Error	0.139539	0.0232565	6			
Total	0.733764		7			

ANOVA Assumptions Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variance	Levene Equality of Variance Test	59.23	13.75	2.5E-04	Unequal Variances
	Mod Levene Equality of Variance Test	39.49	13.75	7.6E-04	Unequal Variances
Distribution	Anderson-Darling A2 Normality Test	0.5621	3.878	0.1497	Normal Distribution
	Kolmogorov-Smirnov D Test	0.25	0.3313	0.1599	Normal Distribution
	Shapiro-Wilk W Normality Test	0.8906	0.6451	0.2370	Normal Distribution

96h Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%
16.7		4	0.5750	0.2470	0.9030	0.5500	0.4000	0.8000	0.1031	35.85%	42.50%
50.1		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000		100.00%

Angular (Corrected) Transformed Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	1.412	1.412	1.412	1.412	1.412	1.412	0	0.00%	0.00%
16.7		4	0.8669	0.5238	1.21	0.8379	0.6847	1.107	0.1078	24.88%	38.60%
50.1		4	0.1588	0.1588	0.1588	0.1588	0.1588	0.1588	0	0.00%	88.76%

96h Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
16.7		0.8000	0.7000	0.4000	0.4000
50.1		0.0000	0.0000	0.0000	0.0000

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	11-9941-2527	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.5
Analyzed:	19 Dec-19 12:47	**Analysis:**	Nonparametric-Control vs Treatments	**Status Level:**	1

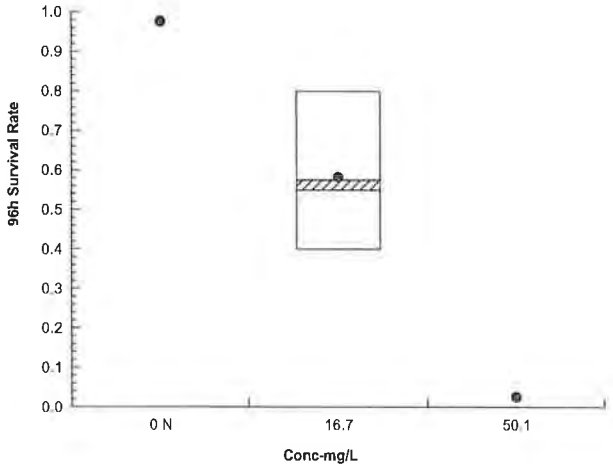
Angular (Corrected) Transformed Detail

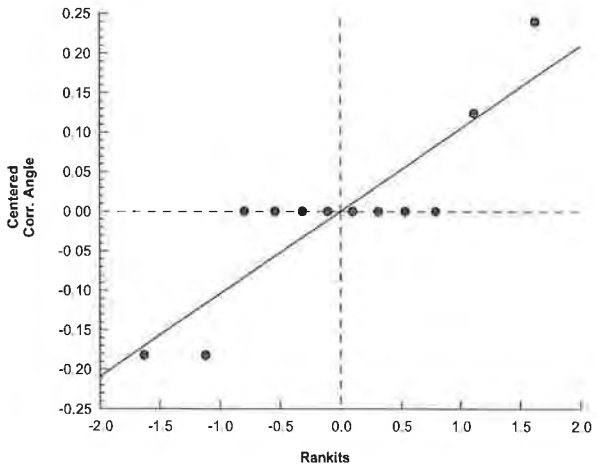
Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.412	1.412	1.412	1.412
16.7		1.107	0.9912	0.6847	0.6847
50.1		0.1588	0.1588	0.1588	0.1588

96h Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	10/10	10/10	10/10	10/10
16.7		8/10	7/10	4/10	4/10
50.1		0/10	0/10	0/10	0/10

Graphics





Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	05-9379-4708	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.5
Analyzed:	19 Dec-19 12:48	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1
Batch ID:	07-7352-6059	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	06 Dec-19 16:05	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water
Ending Date:	10 Dec-19 14:10	**Species:**	Pimephales promelas	**Brine:**	Not Applicable
Test Length:	94h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO **Age:**
Sample ID:	20-2611-5180	**Code:**	TRU1219.056afml	**Project:**	
Sample Date:	04 Dec-19 09:00	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	06 Dec-19 13:00	**CAS (PC):**		**Station:**	Lake Piru Raw
Sample Age:	55h (6.5 °C)	**Client:**	Trussell Technologies		

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	0	280	Yes	Two-Point Interpolation

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

Point Estimates

Level	mg/L	95% LCL	95% UCL
EC5	1.965	1.048	4.165
EC10	3.929	2.096	8.33
EC15	5.894	3.144	12.5
EC20	7.859	4.191	16.66
EC25	9.824	5.239	20.83
EC40	15.72	8.383	27.98
EC50	21.06	9.633	31.9

96h Survival Rate Summary

			Calculated Variate(A/B)							Isotonic Variate	
Conc-mg/L	**Code**	**Count**	**Mean**	**Min**	**Max**	**Std Dev**	**CV%**	**%Effect**	**A/B**	**Mean**	**%Effect**
0	N	4	1.0000	1.0000	1.0000	0.0000	0.00%	0.0%	40/40	1	0.0%
16.7		4	0.5750	0.4000	0.8000	0.2062	35.85%	42.5%	23/40	0.575	42.5%
50.1		4	0.0000	0.0000	0.0000	0.0000		100.0%	0/40	0	100.0%

96h Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
16.7		0.8000	0.7000	0.4000	0.4000
50.1		0.0000	0.0000	0.0000	0.0000

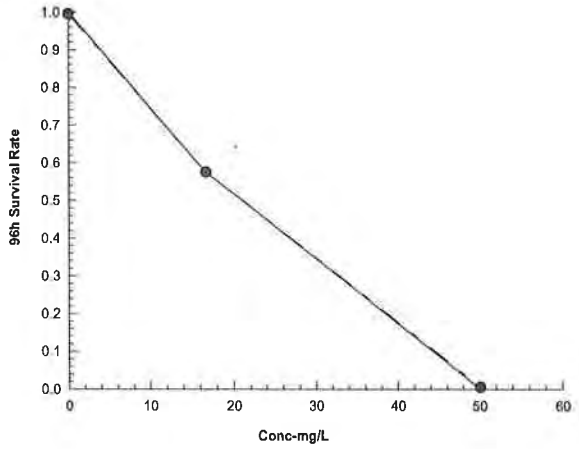
96h Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	10/10	10/10	10/10	10/10
16.7		8/10	7/10	4/10	4/10
50.1		0/10	0/10	0/10	0/10

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	05-9379-4708	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.5
Analyzed:	19 Dec-19 12:48	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1

Graphics



Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	07-7352-6059	**Test Type:**	Survival (96h)	**Analyst:**		
Start Date:	06 Dec-19 16:05	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water	
Ending Date:	10 Dec-19 14:10	**Species:**	Pimephales promelas	**Brine:**	Not Applicable	
Test Length:	94h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO	**Age:**

Sample ID:	20-2611-5180	**Code:**	TRU1219.056afml	**Project:**	
Sample Date:	04 Dec-19 09:00	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	06 Dec-19 13:00	**CAS (PC):**		**Station:**	Lake Piru Raw
Sample Age:	55h (6.5 °C)	**Client:**	Trussell Technologies		

Alkalinity (CaCO3)-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	60	60	60	60	60	0	0	0.0%	0
50.1		3	160	160	160	160	160	0	0	0.0%	0
Overall		6	110	52.52	167.5	60	160	22.36	54.77	49.79%	0 (0%)

Conductivity-µmhos

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	336.3	296.1	376.6	326	355	9.351	16.2	4.82%	0
16.7		3	946.7	920.8	972.5	935	955	6.009	10.41	1.1%	0
50.1		2	990	939.2	1041	986	994	4	5.657	0.57%	0
Overall		8	728.6	456.5	1001	326	994	115.1	325.5	44.67%	0 (0%)

Dissolved Oxygen-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	7.9	7.896	7.904	7.9	7.9	0	0	0.0%	0
16.7		3	8.367	5.878	10.85	7.4	9.4	0.5783	1.002	11.97%	0
50.1		2	8.25	-6.362	22.86	7.1	9.4	1.15	1.626	19.71%	0
Overall		8	8.162	7.456	8.869	7.1	9.4	0.2988	0.845	10.35%	0 (0%)

Hardness (CaCO3)-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	94	94	94	94	94	0	0	0.0%	0
50.1		3	161	161	161	161	161	0	0	0.0%	0
Overall		6	127.5	88.99	166	94	161	14.98	36.7	28.78%	0 (0%)

pH-Units

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	7.667	7.38	7.954	7.6	7.8	0.06667	0.1155	1.51%	0
16.7		3	7.833	7.69	7.977	7.8	7.9	0.03333	0.05774	0.74%	0
50.1		2	8.1	6.829	9.371	8	8.2	0.1	0.1414	1.75%	0
Overall		8	7.838	7.671	8.004	7.6	8.2	0.07055	0.1996	2.55%	0 (0%)

Temperature-°C

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	24	24	24	24	24	0	0	0.0%	0
16.7		3	24.07	23.92	24.21	24	24.1	0.03323	0.05755	0.24%	0
50.1		2	24.05	23.42	24.68	24	24.1	0.0499	0.07056	0.29%	0
Overall		8	24.04	23.99	24.08	24	24.1	0.0183	0.05176	0.22%	0 (0%)

Fathead Minnow 96-h Acute Survival Test — Aquatic Bioassay & Consulting Labs, Inc.

Alkalinity (CaCO3)-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		60					
50.1				160					
0	N	2		60					
50.1				160					
0	N	3		60					
50.1				160					

Conductivity-µmhos

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		326					
16.7				955					
50.1				994					
0	N	2		328					
16.7				950					
50.1				986					
0	N	3		355					
16.7				935					

Dissolved Oxygen-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		7.9					
16.7				9.4					
50.1				9.4					
0	N	2		7.9					
16.7				7.4					
50.1				7.1					
0	N	3		7.9					
16.7				8.3					

Hardness (CaCO3)-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		94					
50.1				161					
0	N	2		94					
50.1				161					
0	N	3		94					
50.1				161					

pH-Units

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		7.6					
16.7				7.9					
50.1				8.2					
0	N	2		7.8					
16.7				7.8					
50.1				8					
0	N	3		7.6					
16.7				7.8					

Fathead Minnow 96-h Acute Survival Test — Aquatic Bioassay & Consulting Labs, Inc.

Temperature-°C

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		24					
16.7				24.1					
50.1				24.1					
0	N	2		24					
16.7				24.1					
50.1				24					
0	N	3		24					
16.7				24					

aquatic bioassay & consulting laboratories, inc

December 23, 2019

Mr. David Hokanson
Trussell Technologies
232 N. Lake Avenue
Pasadena, CA 91101

Dear Mr. Hokanson:

We are pleased to present the enclosed bioassay report. The test was conducted under guidelines prescribed in *Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms EPA-821-R-02-012*. "All acceptability criteria were met and the concentration-response was normal. This is a valid test." Results were as follows:

CLIENT:	Trussell Technologies
SAMPLE ID.:	Lake Piru Raw
DATE RECEIVED:	6 Dec – 19
ABC LAB NO.:	EUR1219.056

ACUTE CERIODAPHNIA SURVIVAL BIOASSAY

% Survival	= 0 % Survival in 50.1mg/l Sample
*TU(a)	= >1.00 @ 16.7 mg/l
*TU(a)	= >1.00 @ 50.1 mg/l

* TU(a) Is calculated by: log (% Mortality)/1.7

Yours very truly,

Scott Johnson
Laboratory Director

CETIS Summary Report

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	18-1458-7538	**Test Type:**	Survival (96h)	**Analyst:**			
Start Date:	06 Dec-19 16:05	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water		
Ending Date:	10 Dec-19 14:10	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable		
Test Length:	94h	**Taxon:**	Branchiopoda	**Source:**	Aquatic Biosystems, CO	**Age:**	
Sample ID:	09-3098-8150	**Code:**	TRU1219.056acer	**Project:**			
Sample Date:	04 Dec-19 09:00	**Material:**	Sample Water	**Source:**	Bioassay Report		
Receipt Date:	06 Dec-19 13:00	**CAS (PC):**		**Station:**	Lake Piru Raw		
Sample Age:	55h (6.5 °C)	**Client:**	Trussell Technologies				

Point Estimate Summary

Analysis ID	Endpoint	Point Estimate Method	✓	Level	mg/L	95% LCL	95% UCL	TU	S
18-7406-0446	96h Survival Rate	Linear Interpolation (ICPIN)		EC5	0.835	0.835	0.835		1
				EC10	1.67	1.67	1.67		
				EC15	2.505	2.505	2.505		
				EC20	3.34	3.34	3.34		
				EC25	4.175	4.175	4.175		
				EC40	6.68	6.68	6.68		
				EC50	8.35	8.35	8.35		

Test Acceptability

				TAC Limits			
Analysis ID	**Endpoint**	**Attribute**	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
18-7406-0446	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria
21-2716-7875	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria

96h Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	N	4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	0.00%	0.00%
16.7		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000		100.00%
50.1		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000		100.00%

96h Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
16.7		0.0000	0.0000	0.0000	0.0000
50.1		0.0000	0.0000	0.0000	0.0000

96h Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	5/5	5/5	5/5	5/5
16.7		0/5	0/5	0/5	0/5
50.1		0/5	0/5	0/5	0/5

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	21-2716-7875	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.5
Analyzed:	19 Dec-19 15:28	**Analysis:**	Nonparametric-Two Sample	**Status Level:**	1
Batch ID:	18-1458-7538	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	06 Dec-19 16:05	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water
Ending Date:	10 Dec-19 14:10	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable
Test Length:	94h	**Taxon:**	Branchiopoda	**Source:**	Aquatic Biosystems, CO **Age:**
Sample ID:	09-3098-8150	**Code:**	TRU1219.056acer	**Project:**	
Sample Date:	04 Dec-19 09:00	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	06 Dec-19 13:00	**CAS (PC):**		**Station:**	Lake Piru Raw
Sample Age:	55h (6.5 °C)	**Client:**	Trussell Technologies		

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

96h Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%
16.7		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000		100.00%
50.1		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000		100.00%

Angular (Corrected) Transformed Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	1.345	1.345	1.346	1.345	1.345	1.345	0	0.00%	0.00%
16.7		4	0.2255	0.2255	0.2256	0.2255	0.2255	0.2255	0	0.00%	83.24%
50.1		4	0.2255	0.2255	0.2256	0.2255	0.2255	0.2255	0	0.00%	83.24%

96h Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
16.7		0.0000	0.0000	0.0000	0.0000
50.1		0.0000	0.0000	0.0000	0.0000

Angular (Corrected) Transformed Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.345	1.345	1.345	1.345
16.7		0.2255	0.2255	0.2255	0.2255
50.1		0.2255	0.2255	0.2255	0.2255

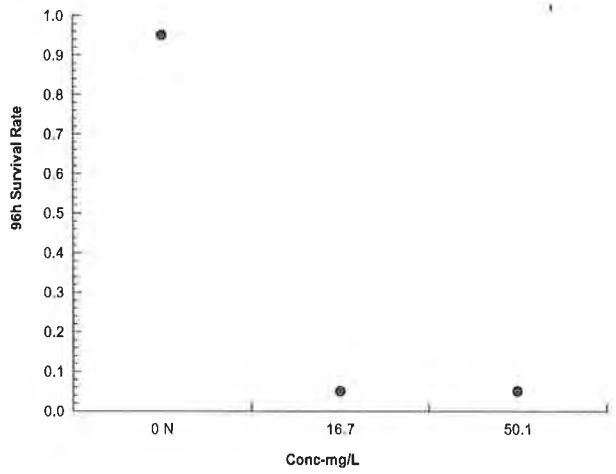
96h Survival Rate Binomials

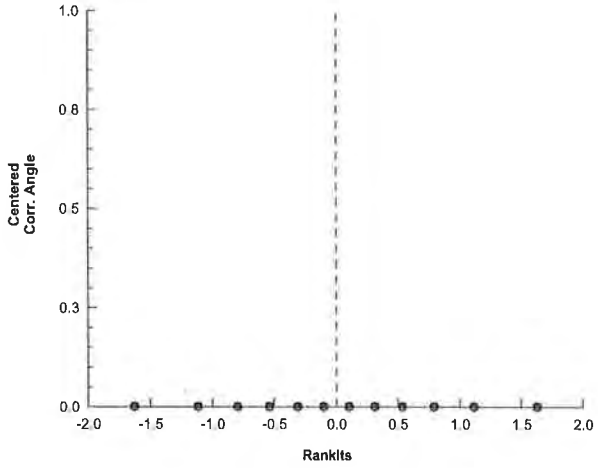
Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	5/5	5/5	5/5	5/5
16.7		0/5	0/5	0/5	0/5
50.1		0/5	0/5	0/5	0/5

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	21-2716-7875	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.5
Analyzed:	19 Dec-19 15:28	**Analysis:**	Nonparametric-Two Sample	**Status Level:**	1

Graphics





Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	18-7406-0446	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.5
Analyzed:	19 Dec-19 15:28	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1
Batch ID:	18-1458-7538	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	06 Dec-19 16:05	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water
Ending Date:	10 Dec-19 14:10	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable
Test Length:	94h	**Taxon:**	Branchiopoda	**Source:**	Aquatic Biosystems, CO **Age:**
Sample ID:	09-3098-8150	**Code:**	TRU1219.056acer	**Project:**	
Sample Date:	04 Dec-19 09:00	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	06 Dec-19 13:00	**CAS (PC):**		**Station:**	Lake Piru Raw
Sample Age:	55h (6.5 °C)	**Client:**	Trussell Technologies		

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	0	280	Yes	Two-Point Interpolation

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

Point Estimates

Level	mg/L	95% LCL	95% UCL
EC5	0.835	0.835	0.835
EC10	1.67	1.67	1.67
EC15	2.505	2.505	2.505
EC20	3.34	3.34	3.34
EC25	4.175	4.175	4.175
EC40	6.68	6.68	6.68
EC50	8.35	8.35	8.35

96h Survival Rate Summary

			Calculated Variate(A/B)							Isotonic Variate	
Conc-mg/L	**Code**	**Count**	**Mean**	**Min**	**Max**	**Std Dev**	**CV%**	**%Effect**	**A/B**	**Mean**	**%Effect**
0	N	4	1.0000	1.0000	1.0000	0.0000	0.00%	0.0%	20/20	1	0.0%
16.7		4	0.0000	0.0000	0.0000	0.0000		100.0%	0/20	0	100.0%
50.1		4	0.0000	0.0000	0.0000	0.0000		100.0%	0/20	0	100.0%

96h Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
16.7		0.0000	0.0000	0.0000	0.0000
50.1		0.0000	0.0000	0.0000	0.0000

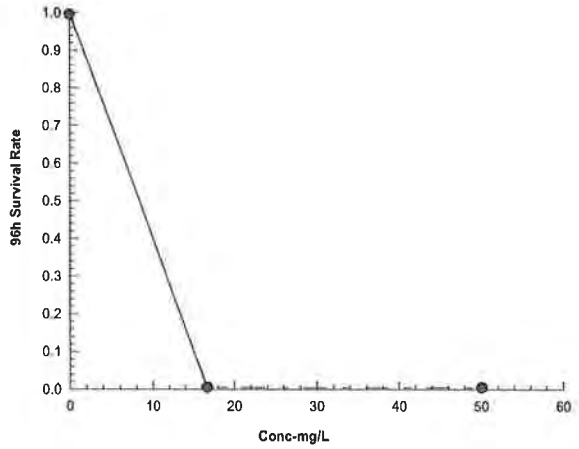
96h Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	5/5	5/5	5/5	5/5
16.7		0/5	0/5	0/5	0/5
50.1		0/5	0/5	0/5	0/5

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	18-7406-0446	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.5
Analyzed:	19 Dec-19 15:28	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1

Graphics



Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	18-1458-7538	**Test Type:**	Survival (96h)	**Analyst:**		
Start Date:	06 Dec-19 16:05	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water	
Ending Date:	10 Dec-19 14:10	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable	
Test Length:	94h	**Taxon:**	Branchiopoda	**Source:**	Aquatic Biosystems, CO	**Age:**

Sample ID:	09-3098-8150	**Code:**	TRU1219.056acer	**Project:**	
Sample Date:	04 Dec-19 09:00	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	06 Dec-19 13:00	**CAS (PC):**		**Station:**	Lake Piru Raw
Sample Age:	55h (6.5 °C)	**Client:**	Trussell Technologies		

Alkalinity (CaCO3)-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	60	60	60	60	60	0	0	0.0%	0
50.1		2	160	160	160	160	160	0	0	0.0%	0
Overall		5	100	31.99	168	60	160	24.49	54.77	54.77%	0 (0%)

Conductivity-µmhos

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	336.3	296.1	376.6	326	355	9.351	16.2	4.82%	0
16.7		3	946.7	920.8	972.5	935	955	6.009	10.41	1.1%	0
50.1		2	990	939.2	1041	986	994	4	5.657	0.57%	0
Overall		8	728.6	456.5	1001	326	994	115.1	325.5	44.67%	0 (0%)

Dissolved Oxygen-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	7.9	7.896	7.904	7.9	7.9	0	0	0.0%	0
16.7		3	8.367	5.878	10.85	7.4	9.4	0.5783	1.002	11.97%	0
50.1		2	8.25	-6.362	22.86	7.1	9.4	1.15	1.626	19.71%	0
Overall		8	8.162	7.456	8.869	7.1	9.4	0.2988	0.845	10.35%	0 (0%)

Hardness (CaCO3)-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	94	94	94	94	94	0	0	0.0%	0
50.1		2	161	161	161	161	161	0	0	0.0%	0
Overall		5	120.8	75.23	166.4	94	161	16.41	36.7	30.38%	0 (0%)

pH-Units

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	7.667	7.38	7.954	7.6	7.8	0.06667	0.1155	1.51%	0
16.7		3	7.833	7.69	7.977	7.8	7.9	0.03333	0.05774	0.74%	0
50.1		2	8.1	6.829	9.371	8	8.2	0.1	0.1414	1.75%	0
Overall		8	7.838	7.671	8.004	7.6	8.2	0.07055	0.1996	2.55%	0 (0%)

Temperature-°C

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	24	24	24	24	24	0	0	0.0%	0
16.7		3	24.07	23.92	24.21	24	24.1	0.03323	0.05755	0.24%	0
50.1		2	24.05	23.42	24.68	24	24.1	0.0499	0.07056	0.29%	0
Overall		8	24.04	23.99	24.08	24	24.1	0.0183	0.05176	0.22%	0 (0%)

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Alkalinity (CaCO3)-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		60					
50.1				160					
0	N	2		60					
50.1				160					
0	N	3		60					

Conductivity-µmhos

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		326					
16.7				955					
50.1				994					
0	N	2		328					
16.7				950					
50.1				986					
0	N	3		355					
16.7				935					

Dissolved Oxygen-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		7.9					
16.7				9.4					
50.1				9.4					
0	N	2		7.9					
16.7				7.4					
50.1				7.1					
0	N	3		7.9					
16.7				8.3					

Hardness (CaCO3)-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		94					
50.1				161					
0	N	2		94					
50.1				161					
0	N	3		94					

pH-Units

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		7.6					
16.7				7.9					
50.1				8.2					
0	N	2		7.8					
16.7				7.8					
50.1				8					
0	N	3		7.6					
16.7				7.8					

Ceriodaphnia 96-h Acute Survival Test — Aquatic Bioassay & Consulting Labs, Inc.

Temperature-°C

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		24					
16.7				24.1					
50.1				24.1					
0	N	2		24					
16.7				24.1					
50.1				24					
0	N	3		24					
16.7				24					

aquatic bioassay & consulting laboratories, inc

December 19, 2019

Mr. David Hokanson
Trussell Technologies
232 N. Lake Ave
Pasadena, CA 91101

Dear Mr. Hokanson:

We are pleased to present the enclosed acute bioassay report. The test was conducted under the guidelines prescribed in "Methods for Measuring the Acute Toxicity of Effluents and Receiving water to Freshwater and Marine Organisms" (Fourth Edition) EPA/600/ 4-90/027F, August 1993. The results were as follows:

CLIENT:	Trussell Technologies
SAMPLE I.D.:	Lake Piru Raw (16.7mg/l)
DATE RECEIVED:	12/06/19
ABC LAB. NO.:	TRU1219.056

NPDES PERCENT SURVIVAL BIOASSAY
RAINBOW TROUT

PERCENT SURVIVAL = 0% Survival in 16.7mg/l Sample

TUa = >1.00 @ 16.7mg/l

Yours very truly,

Scott Johnson
Laboratory Director

AQUATIC BIOASSAY AND CONSULTING LABORATORIES, INC.
29 North Olive Street
Ventura, CA 93001
(805) 643-5621

NPDES Percent Survival Bioassay for Effluents and Stormwater

SAMPLE INFORMATION

CLIENT:	Trussell Technologies	Date:	12/06/19
SAMPLE I.D.:	Lake Piru Raw (16.7mg/l)	LAB #	TRU1219.056

WATER QUALITY

DILUTION WATER: Reconst. Fresh	AERATION: Single Bubble Air
CONTROL HARDNESS	CONTROL ALKALINITY
Beg: 90 mg/l End: 100 mg/l	Beg: 60 mg/l End: 63 mg/l
SAMPLE HARDNESS	SAMPLE ALKALINITY
Beg: 300 mg/l End: 310 mg/l	Beg: 150 mg/l End: 154 mg/l

ORGANISM INFORMATION

SPECIES:	Oncorhynchus mykiss	DATE REC'D:	11/24/19
COMMON NAME:	Rainbow Trout		
SOURCE:	Thomas Fish Co.		
CARRIER:	California Overnight	NO. FISH / TANK	10

TEST DATA

	INITIAL				24 HOURS				48 HOURS				72 HOURS				96 HOURS			
DATE:	12/06/19				12/07/19				12/08/19				12/09/19				12/10/19			
TIME:	1450				1100				1530				1200				1450			
CONC.	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp dg.C	pH	#Fish
0 (Con.)	9.8	11.8	7.9	10	6.9	11.0	7.9	10	7.0	12.1	7.1	10	7.0	12.0	7.1	10	11.0	11.0	8.0	10
0 (Con.)	10.0	11.8	8.0	10	7.0	11.0	8.0	10	7.0	12.0	7.1	10	7.1	12.0	7.0	10	10.9	11.0	8.1	10
16.7mg/l	9.2	13.9	7.5	10	10.9	11.0	7.8	0				0				0				0
16.7mg/l	9.3	13.9	7.5	10	10.9	11.0	7.8	0				0				0				0

FINAL DATA

TOTAL	FISH SURVIVAL
0 (Con.)	10
0 (Con.)	10
16.7mg/l	0
16.7mg/l	0

FINAL RESULTS

PERCENT SURVIVAL =	0% Survival in 16.7mg/l Sample
TUa =	>1.00 @ 16.7mg/l

Joe Freas, Senior Toxicologist

Date 12/27/19

aquatic bioassay & consulting laboratories, inc

December 19, 2019

Mr. David Hokanson
Trussell Technologies
232 N. Lake Ave
Pasadena, CA 91101

Dear Mr. Hokanson:

We are pleased to present the enclosed acute bioassay report. The test was conducted under the guidelines prescribed in "Methods for Measuring the Acute Toxicity of Effluents and Receiving water to Freshwater and Marine Organisms" (Fourth Edition) EPA/600/ 4-90/027F, August 1993. The results were as follows:

CLIENT:	Trussell Technologies
SAMPLE I.D.:	Lake Piru Raw (50.1mg/l)
DATE RECEIVED:	12/06/19
ABC LAB. NO.:	TRU1219.056

NPDES PERCENT SURVIVAL BIOASSAY
RAINBOW TROUT

PERCENT SURVIVAL = 0% Survival in 50.1/mg/l Sample

TUa = >1.00 @ 50.1mg/l

Yours very truly,

Scott Johnson
Laboratory Director

AQUATIC BIOASSAY AND CONSULTING LABORATORIES, INC.
29 North Olive Street
Ventura, CA 93001
(805) 643-5621

NPDES Percent Survival Bioassay for Effluents and Stormwater

SAMPLE INFORMATION

CLIENT:	Trussell Technologies	Date:	12/06/19
SAMPLE I.D.:	Lake Piru Raw (50.1mg/l)	LAB #	TRU1219.056

WATER QUALITY

DILUTION WATER: Reconst. Fresh	AERATION: Single Bubble Air
CONTROL HARDNESS	CONTROL ALKALINITY
Beg: 90 mg/l End: 100 mg/l	Beg: 60 mg/l End: 63 mg/l
SAMPLE HARDNESS	SAMPLE ALKALINITY
Beg: 320 mg/l End: 325 mg/l	Beg: 161 mg/l End: 162 mg/l

ORGANISM INFORMATION

SPECIES:	Oncorhynchus mykiss	DATE REC'D:	11/24/19
COMMON NAME:	Rainbow Trout		
SOURCE:	Thomas Fish Co.		
CARRIER:	California Overnight	NO. FISH / TANK	10

TEST DATA

	INITIAL				24 HOURS				48 HOURS				72 HOURS				96 HOURS			
DATE:	12/06/19				12/07/19				12/08/19				12/09/19				12/10/19			
TIME:	1450				1100				1530				1200				1450			
CONC.	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp dg.C	pH	#Fish
0 (Con.)	9.8	11.8	7.9	10	6.9	11.0	7.9	10	7.0	12.1	7.1	10	7.0	12.0	7.1	10	11.0	11.0	8.0	10
0 (Con.)	10.0	11.8	8.0	10	7.0	11.0	8.0	10	7.0	12.0	7.1	10	7.1	12.0	7.0	10	10.9	11.0	8.1	10
50.1mg/l	9.2	13.9	7.3	10	10.7	11.0	7.7	0				0				0				0
50.1mg/l	8.9	13.9	7.0	10	10.6	11.0	7.7	0				0				0				0

FINAL DATA

TOTAL	FISH SURVIVAL
0 (Con.)	10
0 (Con.)	10
50.1mg/l	0
50.1mg/l	0

FINAL RESULTS

PERCENT SURVIVAL =	0% Survival in 50.1mg/l Sample
TUa =	>1.00 @ 50.1mg/l

Joe Freas, Senior Toxicologist

Date 12/27/19

CHAIN OF CUSTODY RECORD

Client:	Project Name/Number:	Analysis
Trussell Technologies	Liana Olivas (626) 375-8150	
Address: 232 N. Lake Ave Pasadena, CA 91101	Project Mgr. David Hokanson	
Phone Number: (626) 375-8150	P.O. #	
	Sampled By (signature)	

Date	Time	Comp	Grab	Matrix	Sample ID	Volume/ Number	Acute Toxicity Fathead minnow	Acute Toxicity o. mykiss	Acute Toxicity ceriodaphnia								Comments
12/4/19	9:00		✓	Liquid	Lake Piru Raw	28 L	2	2	2								Two Doses Each (16.7 mg/L + 50.1 mg/L)
					.05												.05
					Temp. deg. C = 6.5 C			=										
					Chlorine (mg/L) =	<0.1		=										
					NH3 (mg/L) =	<0.1		=										

Relinquished By: (signature)	Date: Time:	Relinquished By: (signature)	Date: Time:
Liana Olivas	12/6/19 13:00		
Received By: (signature)	Date: Time:	Received By: (signature)	Date: Time:
	12-6-19 13:00		
		Temp. upon sample receipt: _____ °C	

Aquatic Bioassay and Consulting Laboratories
29 N. Olive Street Ventura, CA 93001 Phone: (805) 643-5621 Fax: (805) 643-2930

Appendix C –

Lake Mathews 2020 Toxicity Test Report

aquatic bioassay & consulting laboratories, inc

October 22, 2020

Mr. David Hokanson
Trussell Technologies
232 N. Lake Avenue
Pasadena, CA 91101

Dear Mr. Hokanson:

We are pleased to present the enclosed bioassay report. The test was conducted under guidelines prescribed in *Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms EPA-821-R-02-012*. "All acceptability criteria were met and the concentration-response was normal. This is a valid test." Results were as follows:

CLIENT:	Trussell Technologies
SAMPLE ID.:	Lake Mathews Raw with EarthTec
DATE RECEIVED:	10 Sept – 20
ABC LAB NO.:	TRU0920.134

ACUTE FATHEAD MINNOW SURVIVAL BIOASSAY

% Survival = 92.5 % Survival in 16.7 mg/l Sample

EC50 = >16.7 mg/l

Yours very truly,

Scott Johnson
Laboratory Director

CETIS Summary Report

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	03-3365-6779	**Test Type:**	Survival (96h)	**Analyst:**			
Start Date:	22 Sep-20 13:25	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water		
Ending Date:	26 Sep-20 13:30	**Species:**	Pimephales promelas	**Brine:**	Not Applicable		
Test Length:	4d 0h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO	**Age:**	
Sample ID:	05-3885-0428	**Code:**	TRU0920.134afml	**Project:**			
Sample Date:	10 Sep-20 09:50	**Material:**	Sample Water	**Source:**	Bioassay Report		
Receipt Date:	10 Sep-20 09:50	**CAS (PC):**		**Station:**	Lake Mathews Raw with Earth Tec		
Sample Age:	12d 4h (9.3 °C)	**Client:**	Trussell Technologies				

Multiple Comparison Summary

Analysis ID	Endpoint	Comparison Method	✓	NOEL	LOEL	TOEL	PMSD	S
07-4137-4317	96h Survival Rate	Steel Many-One Rank Sum Test		16.7	>16.7	---	9.21%	1

Point Estimate Summary

Analysis ID	Endpoint	Point Estimate Method	✓	Level	mg/L	95% LCL	95% UCL	S
09-5562-0350	96h Survival Rate	Linear Interpolation (ICPIN)		EC10	>16.7	---	---	1
				EC15	>16.7	---	---	
				EC20	>16.7	---	---	
				EC25	>16.7	---	---	
				EC40	>16.7	---	---	
				EC50	>16.7	---	---	

Test Acceptability

				TAC Limits			
Analysis ID	Endpoint	Attribute	Test Stat	Lower	Upper	Overlap	Decision
07-4137-4317	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria
09-5562-0350	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria

96h Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	N	4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	---	0.00%
8.35		4	0.9750	0.8954	1.0550	0.9000	1.0000	0.0250	0.0500	5.13%	2.50%
16.7		4	0.9250	0.7727	1.0770	0.8000	1.0000	0.0479	0.0957	10.35%	7.50%

96h Survival Rate Detail — MD5: B7E7C8A5E8F5272CD29973742D21B3F9

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
8.35		1.0000	1.0000	1.0000	0.9000
16.7		0.8000	0.9000	1.0000	1.0000

96h Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	10/10	10/10	10/10	10/10
8.35		10/10	10/10	10/10	9/10
16.7		8/10	9/10	10/10	10/10

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	07-4137-4317	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	21 Oct-20 15:14	**Analysis:**	Nonparametric-Control vs Treatments	**Status Level:**	1
Edit Date:	21 Oct-20 15:13	**MD5 Hash:**	B7E7C8A5E8F5272CD29973742D21B3F9	**Editor ID:**	000-189-126-0
Batch ID:	03-3365-6779	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	22 Sep-20 13:25	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water
Ending Date:	26 Sep-20 13:30	**Species:**	Pimephales promelas	**Brine:**	Not Applicable
Test Length:	4d 0h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO **Age:**
Sample ID:	05-3885-0428	**Code:**	TRU0920.134afml	**Project:**	
Sample Date:	10 Sep-20 09:50	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	10 Sep-20 09:50	**CAS (PC):**		**Station:**	Lake Mathews Raw with Earth Tec
Sample Age:	12d 4h (9.3 °C)	**Client:**	Trussell Technologies		

Data Transform	Alt Hyp	NOEL	LOEL	TOEL	TU	MSDu	PMSD
Angular (Corrected)	C > T	16.7	>16.7	---	---	0.09209	9.21%

Steel Many-One Rank Sum Test

Control	vs	Conc-mg/L	Test Stat	Critical	Ties	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		8.35	16	11	1	6	CDF	0.4206	Non-Significant Effect
		16.7	14	11	1	6	CDF	0.2042	Non-Significant Effect

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.028198	0.014099	2	1.498	0.2744	Non-Significant Effect
Error	0.0847049	0.0094117	9			
Total	0.112903		11			

ANOVA Assumptions Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variance	Bartlett Equality of Variance Test				Indeterminate
	Levene Equality of Variance Test	8.187	8.022	0.0094	Unequal Variances
	Mod Levene Equality of Variance Test	3.623	8.022	0.0701	Equal Variances
Distribution	Anderson-Darling A2 Test	0.6319	3.878	0.1002	Normal Distribution
	D'Agostino Skewness Test	1.332	2.576	0.1827	Normal Distribution
	Kolmogorov-Smirnov D Test	0.25	0.2801	0.0369	Normal Distribution
	Shapiro-Wilk W Normality Test	0.906	0.8025	0.1897	Normal Distribution

96h Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%
8.35		4	0.9750	0.8954	1.0000	1.0000	0.9000	1.0000	0.0250	5.13%	2.50%
16.7		4	0.9250	0.7727	1.0000	0.9500	0.8000	1.0000	0.0479	10.35%	7.50%

Angular (Corrected) Transformed Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	1.4120	1.4120	1.4120	1.4120	1.4120	1.4120	0.0000	0.00%	0.00%
8.35		4	1.3710	1.2420	1.5010	1.4120	1.2490	1.4120	0.0407	5.94%	2.89%
16.7		4	1.2950	1.0610	1.5290	1.3310	1.1070	1.4120	0.0735	11.35%	8.28%

96h Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
8.35		1.0000	1.0000	1.0000	0.9000
16.7		0.8000	0.9000	1.0000	1.0000

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	07-4137-4317	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	21 Oct-20 15:14	**Analysis:**	Nonparametric-Control vs Treatments	**Status Level:**	1
Edit Date:	21 Oct-20 15:13	**MD5 Hash:**	B7E7C8A5E8F5272CD29973742D21B3F9	**Editor ID:**	000-189-126-0

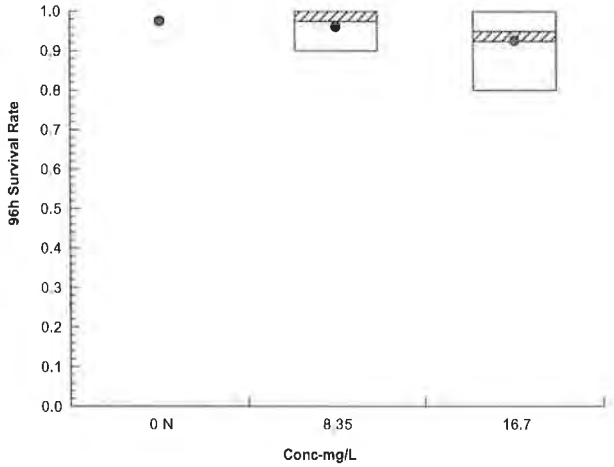
Angular (Corrected) Transformed Detail

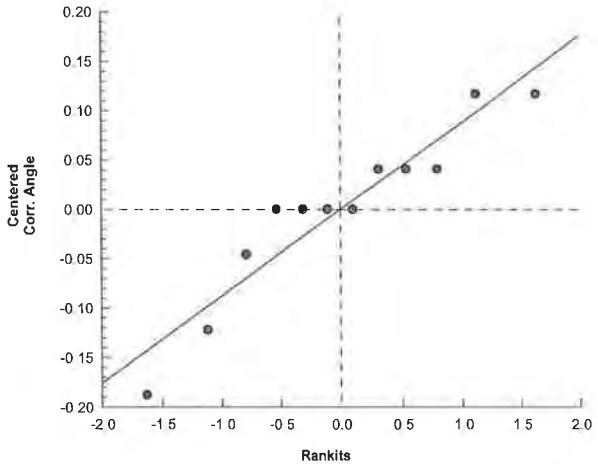
Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.4120	1.4120	1.4120	1.4120
8.35		1.4120	1.4120	1.4120	1.2490
16.7		1.1070	1.2490	1.4120	1.4120

96h Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	10/10	10/10	10/10	10/10
8.35		10/10	10/10	10/10	9/10
16.7		8/10	9/10	10/10	10/10

Graphics





Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	09-5562-0350	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	21 Oct-20 15:14	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1
Edit Date:	21 Oct-20 15:13	**MD5 Hash:**	B7E7C8A5E8F5272CD29973742D21B3F9	**Editor ID:**	000-189-126-0
Batch ID:	03-3365-6779	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	22 Sep-20 13:25	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water
Ending Date:	26 Sep-20 13:30	**Species:**	Pimephales promelas	**Brine:**	Not Applicable
Test Length:	4d 0h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO **Age:**
Sample ID:	05-3885-0428	**Code:**	TRU0920.134afml	**Project:**	
Sample Date:	10 Sep-20 09:50	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	10 Sep-20 09:50	**CAS (PC):**		**Station:**	Lake Mathews Raw with Earth Tec
Sample Age:	12d 4h (9.3 °C)	**Client:**	Trussell Technologies		

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	0	280	Yes	Two-Point Interpolation

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

Point Estimates

Level	mg/L	95% LCL	95% UCL
EC10	>16.7	---	---
EC15	>16.7	---	---
EC20	>16.7	---	---
EC25	>16.7	---	---
EC40	>16.7	---	---
EC50	>16.7	---	---

96h Survival Rate Summary

			Calculated Variate(A/B)							Isotonic Variate	
Conc-mg/L	**Code**	**Count**	**Mean**	**Median**	**Min**	**Max**	**CV%**	**%Effect**	**A/B**	**Mean**	**%Effect**
0	N	4	1.0000	1.0000	1.0000	1.0000	0.00%	0.00%	40/40	1.0000	0.00%
8.35		4	0.9750	1.0000	0.9000	1.0000	5.13%	2.50%	39/40	0.9750	2.50%
16.7		4	0.9250	0.9500	0.8000	1.0000	10.35%	7.50%	37/40	0.9250	7.50%

96h Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
8.35		1.0000	1.0000	1.0000	0.9000
16.7		0.8000	0.9000	1.0000	1.0000

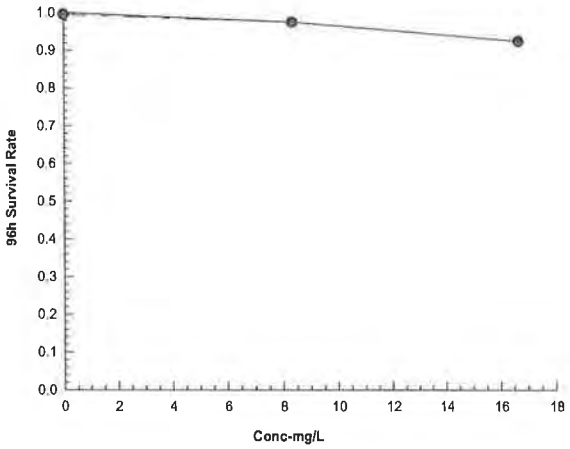
96h Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	10/10	10/10	10/10	10/10
8.35		10/10	10/10	10/10	9/10
16.7		8/10	9/10	10/10	10/10

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	09-5562-0350	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	21 Oct-20 15:14	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1
Edit Date:	21 Oct-20 15:13	**MD5 Hash:**	B7E7C8A5E8F5272CD29973742D21B3F9	**Editor ID:**	000-189-126-0

Graphics



Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	03-3365-6779	**Test Type:**	Survival (96h)	**Analyst:**			
Start Date:	22 Sep-20 13:25	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water		
Ending Date:	26 Sep-20 13:30	**Species:**	Pimephales promelas	**Brine:**	Not Applicable		
Test Length:	4d 0h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO	**Age:**	
Sample ID:	05-3885-0428	**Code:**	TRU0920.134afml	**Project:**			
Sample Date:	10 Sep-20 09:50	**Material:**	Sample Water	**Source:**	Bioassay Report		
Receipt Date:	10 Sep-20 09:50	**CAS (PC):**		**Station:**	Lake Mathews Raw with Earth Tec		
Sample Age:	12d 4h (9.3 °C)	**Client:**	Trussell Technologies				

Alkalinity (CaCO3)-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	60	60	60	60	60	0	0	0.00%	0
8.35		3	100	100	100	100	100	0	0	0.00%	0
Overall		6	80	57.01	103	60	100	8.944	21.91	27.39%	0 (0%)

Conductivity-µmhos

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	337.7	332.5	342.8	336	340	0.6939	2.082	0.62%	0
8.35		3	1129	1067	1191	1102	1151	8.293	24.88	2.20%	0
16.7		3	1097	1090	1103	1094	1099	0.8389	2.517	0.23%	0
Overall		9	854.4	556.2	1153	336	1151	129.3	388	45.41%	0 (0%)

Dissolved Oxygen-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	7.4	6.655	8.145	7.1	7.7	0.1	0.3	4.05%	0
8.35		3	7.6	6.11	9.09	7	8.2	0.2	0.6	7.89%	0
16.7		3	7.467	6.463	8.471	7	7.7	0.1347	0.4041	5.41%	0
Overall		9	7.489	7.18	7.797	7	8.2	0.1338	0.4014	5.36%	0 (0%)

Hardness (CaCO3)-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	85	85	85	85	85	0	0	0.00%	0
8.35		3	350	350	350	350	350	0	0	0.00%	0
Overall		6	217.5	65.18	369.8	85	350	59.26	145.1	66.73%	0 (0%)

pH-Units

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	8	7.57	8.43	7.9	8.2	0.05773	0.1732	2.17%	0
8.35		3	7.9	7.652	8.148	7.8	8	0.03333	0.1	1.27%	0
16.7		3	7.867	7.723	8.01	7.8	7.9	0.01924	0.05772	0.73%	0
Overall		9	7.922	7.83	8.015	7.8	8.2	0.04006	0.1202	1.52%	0 (0%)

Temperature-°C

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	24	24	24	24	24	0	0	0.00%	0
8.35		3	24.1	23.67	24.53	24	24.3	0.05775	0.1732	0.72%	0
16.7		3	24.13	23.56	24.71	24	24.4	0.07698	0.2309	0.96%	0
Overall		9	24.08	23.96	24.2	24	24.4	0.05212	0.1563	0.65%	0 (0%)

Fathead Minnow 96-h Acute Survival Test

Aquatic Bioassay & Consulting Labs, Inc.

Alkalinity (CaCO3)-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		60					
8.35				100					
0	N	2		60					
8.35				100					
0	N	3		60					
8.35				100					

Conductivity-µmhos

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		337					
8.35				1102					
16.7				1094					
0	N	2		336					
8.35				1134					
16.7				1097					
0	N	3		340					
8.35				1151					
16.7				1099					

Dissolved Oxygen-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		7.7					
8.35				8.2					
16.7				7.7					
0	N	2		7.4					
8.35				7.6					
16.7				7.7					
0	N	3		7.1					
8.35				7					
16.7				7					

Hardness (CaCO3)-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		85					
8.35				350					
0	N	2		85					
8.35				350					
0	N	3		85					
8.35				350					

pH-Units

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		8.2					
8.35				7.8					
16.7				7.8					
0	N	2		7.9					
8.35				8					
16.7				7.9					
0	N	3		7.9					
8.35				7.9					
16.7				7.9					

Fathead Minnow 96-h Acute Survival Test **Aquatic Bioassay & Consulting Labs, Inc.**

Temperature-°C

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		24					
8.35				24					
16.7				24					
0	N	2		24					
8.35				24.3					
16.7				24.4					
0	N	3		24					
8.35				24					
16.7				24					

aquatic bioassay & consulting laboratories, inc

October 22, 2020

Mr. David Hokanson
Trussell Technologies
232 N. Lake Avenue
Pasadena, CA 91101

Dear Mr. Hokanson:

We are pleased to present the enclosed bioassay report. The test was conducted under guidelines prescribed in *Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms EPA-821-R-02-012*. "All acceptability criteria were met and the concentration-response was normal. This is a valid test." Results were as follows:

CLIENT:	Trussell Technologies
SAMPLE ID.:	Lake Mathews Raw with EarthTec
DATE RECEIVED:	10 Sept – 20
ABC LAB NO.:	TRU0920.134

ACUTE CERIODAPHNIA SURVIVAL BIOASSAY

% Survival = 0 % Survival in 8.35mg/l & 16.7mg/l Sample

EC50 = 4.175mg/l

Yours very truly,

Scott Johnson
Laboratory Director

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	16-7661-4185	**Test Type:**	Survival (96h)	**Analyst:**			
Start Date:	22 Sep-20 13:25	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water		
Ending Date:	26 Sep-20 13:30	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable		
Test Length:	4d 0h	**Taxon:**	Branchiopoda	**Source:**	Aquatic Biosystems, CO	**Age:**	
Sample ID:	12-0106-0394	**Code:**	TRU0920.134acer	**Project:**			
Sample Date:	10 Sep-20 09:50	**Material:**	Sample Water	**Source:**	Bioassay Report		
Receipt Date:	10 Sep-20 09:50	**CAS (PC):**		**Station:**	Lake Mathews Raw with Earth Tec		
Sample Age:	12d 4h (9.3 °C)	**Client:**	Trussell Technologies				

Point Estimate Summary

Analysis ID	Endpoint	Point Estimate Method	✓	Level	mg/L	95% LCL	95% UCL	S
14-9409-9402	96h Survival Rate	Linear Interpolation (ICPIN)		EC10	0.835	0.835	0.835	1
				EC15	1.253	1.253	1.253	
				EC20	1.67	1.67	1.67	
				EC25	2.088	2.088	2.088	
				EC40	3.34	3.34	3.34	
				EC50	4.175	4.175	4.175	

Test Acceptability

				TAC Limits			
Analysis ID	**Endpoint**	**Attribute**	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
14-9409-9402	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria
17-2439-4970	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria

96h Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	N	4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	---	0.00%
8.35		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	---	100.00%
16.7		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	---	100.00%

96h Survival Rate Detail — MD5: 79242C0938E02BA8B3CB4769263E98D9

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
8.35		0.0000	0.0000	0.0000	0.0000
16.7		0.0000	0.0000	0.0000	0.0000

96h Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	5/5	5/5	5/5	5/5
8.35		0/5	0/5	0/5	0/5
16.7		0/5	0/5	0/5	0/5

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	17-2439-4970	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	21 Oct-20 15:23	**Analysis:**	Parametric-Two Sample	**Status Level:**	1
Edit Date:	21 Oct-20 15:21	**MD5 Hash:**	79242C0938E02BA8B3CB4769263E98D9	**Editor ID:**	000-189-126-0
Batch ID:	16-7661-4185	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	22 Sep-20 13:25	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water
Ending Date:	26 Sep-20 13:30	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable
Test Length:	4d 0h	**Taxon:**	Branchiopoda	**Source:**	Aquatic Biosystems, CO **Age:**
Sample ID:	12-0106-0394	**Code:**	TRU0920.134acer	**Project:**	
Sample Date:	10 Sep-20 09:50	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	10 Sep-20 09:50	**CAS (PC):**		**Station:**	Lake Mathews Raw with Earth Tec
Sample Age:	12d 4h (9.3 °C)	**Client:**	Trussell Technologies		

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

ANOVA Assumptions Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Distribution	Shapiro-Wilk W Normality Test				Indeterminate

96h Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%
8.35		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	---	100.00%
16.7		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	---	100.00%

Angular (Corrected) Transformed Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	1.3450	1.3450	1.3460	1.3450	1.3450	1.3450	0.0000	0.00%	0.00%
8.35		4	0.2255	0.2255	0.2256	0.2255	0.2255	0.2255	0.0000	0.00%	83.24%
16.7		4	0.2255	0.2255	0.2256	0.2255	0.2255	0.2255	0.0000	0.00%	83.24%

96h Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
8.35		0.0000	0.0000	0.0000	0.0000
16.7		0.0000	0.0000	0.0000	0.0000

Angular (Corrected) Transformed Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.3450	1.3450	1.3450	1.3450
8.35		0.2255	0.2255	0.2255	0.2255
16.7		0.2255	0.2255	0.2255	0.2255

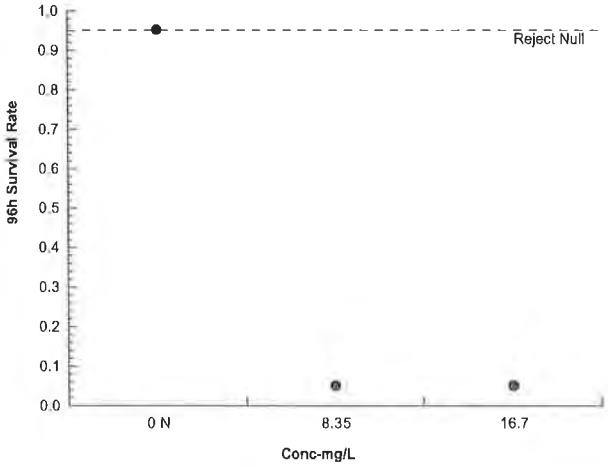
96h Survival Rate Binomials

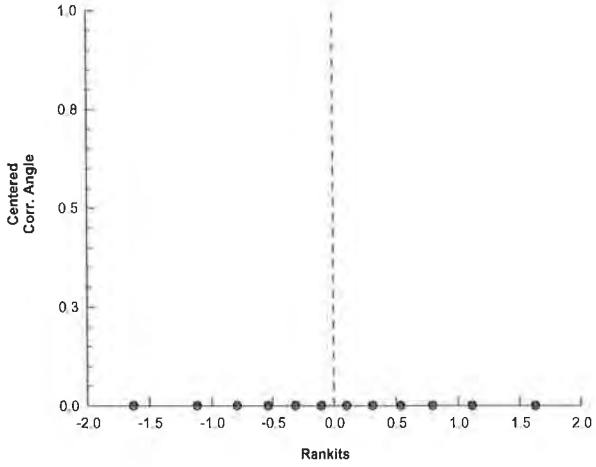
Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	5/5	5/5	5/5	5/5
8.35		0/5	0/5	0/5	0/5
16.7		0/5	0/5	0/5	0/5

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	17-2439-4970	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	21 Oct-20 15:23	**Analysis:**	Parametric-Two Sample	**Status Level:**	1
Edit Date:	21 Oct-20 15:21	**MD5 Hash:**	79242C0938E02BA8B3CB4769263E98D9	**Editor ID:**	000-189-126-0

Graphics





Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	14-9409-9402	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	21 Oct-20 15:23	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1
Edit Date:	21 Oct-20 15:21	**MD5 Hash:**	79242C0938E02BA8B3CB4769263E98D9	**Editor ID:**	000-189-126-0
Batch ID:	16-7661-4185	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	22 Sep-20 13:25	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water
Ending Date:	26 Sep-20 13:30	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable
Test Length:	4d 0h	**Taxon:**	Branchiopoda	**Source:**	Aquatic Biosystems, CO **Age:**
Sample ID:	12-0106-0394	**Code:**	TRU0920.134acer	**Project:**	
Sample Date:	10 Sep-20 09:50	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	10 Sep-20 09:50	**CAS (PC):**		**Station:**	Lake Mathews Raw with Earth Tec
Sample Age:	12d 4h (9.3 °C)	**Client:**	Trussell Technologies		

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	0	280	Yes	Two-Point Interpolation

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

Point Estimates

Level	mg/L	95% LCL	95% UCL
EC10	0.835	0.835	0.835
EC15	1.253	1.253	1.253
EC20	1.67	1.67	1.67
EC25	2.088	2.088	2.088
EC40	3.34	3.34	3.34
EC50	4.175	4.175	4.175

96h Survival Rate Summary

			Calculated Variate(A/B)							Isotonic Variate	
Conc-mg/L	**Code**	**Count**	**Mean**	**Median**	**Min**	**Max**	**CV%**	**%Effect**	**A/B**	**Mean**	**%Effect**
0	N	4	1.0000	1.0000	1.0000	1.0000	0.00%	0.00%	20/20	1.0000	0.00%
8.35		4	0.0000	0.0000	0.0000	0.0000	---	100.00%	0/20	0.0000	100.00%
16.7		4	0.0000	0.0000	0.0000	0.0000	---	100.00%	0/20	0.0000	100.00%

96h Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
8.35		0.0000	0.0000	0.0000	0.0000
16.7		0.0000	0.0000	0.0000	0.0000

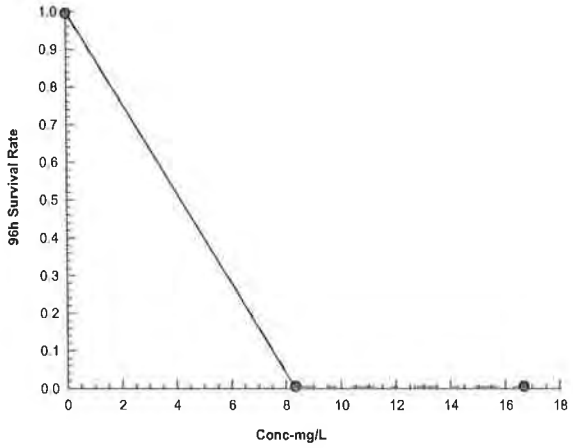
96h Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	5/5	5/5	5/5	5/5
8.35		0/5	0/5	0/5	0/5
16.7		0/5	0/5	0/5	0/5

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	14-9409-9402	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	21 Oct-20 15:23	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1
Edit Date:	21 Oct-20 15:21	**MD5 Hash:**	79242C0938E02BA8B3CB4769263E98D9	**Editor ID:**	000-189-126-0

Graphics



Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	16-7661-4185	**Test Type:**	Survival (96h)	**Analyst:**			
Start Date:	22 Sep-20 13:25	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water		
Ending Date:	26 Sep-20 13:30	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable		
Test Length:	4d 0h	**Taxon:**	Branchiopoda	**Source:**	Aquatic Biosystems, CO	**Age:**	
Sample ID:	12-0106-0394	**Code:**	TRU0920.134acer	**Project:**			
Sample Date:	10 Sep-20 09:50	**Material:**	Sample Water	**Source:**	Bioassay Report		
Receipt Date:	10 Sep-20 09:50	**CAS (PC):**		**Station:**	Lake Mathews Raw with Earth Tec		
Sample Age:	12d 4h (9.3 °C)	**Client:**	Trussell Technologies				

Alkalinity (CaCO3)-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	60	60	60	60	60	0	0	0.00%	0
8.35		3	100	100	100	100	100	0	0	0.00%	0
Overall		6	80	57.01	103	60	100	8.944	21.91	27.39%	0 (0%)

Conductivity-µmhos

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	337.7	332.5	342.8	336	340	0.6939	2.082	0.62%	0
8.35		3	1129	1067	1191	1102	1151	8.293	24.88	2.20%	0
16.7		3	1097	1090	1103	1094	1099	0.8389	2.517	0.23%	0
Overall		9	854.4	556.2	1153	336	1151	129.3	388	45.41%	0 (0%)

Dissolved Oxygen-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	7.4	6.655	8.145	7.1	7.7	0.1	0.3	4.05%	0
8.35		3	7.6	6.11	9.09	7	8.2	0.2	0.6	7.89%	0
16.7		3	7.467	6.463	8.471	7	7.7	0.1347	0.4041	5.41%	0
Overall		9	7.489	7.18	7.797	7	8.2	0.1338	0.4014	5.36%	0 (0%)

Hardness (CaCO3)-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	85	85	85	85	85	0	0	0.00%	0
8.35		3	350	350	350	350	350	0	0	0.00%	0
Overall		6	217.5	65.18	369.8	85	350	59.26	145.1	66.73%	0 (0%)

pH-Units

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	8	7.57	8.43	7.9	8.2	0.05773	0.1732	2.17%	0
8.35		3	7.9	7.652	8.148	7.8	8	0.03333	0.1	1.27%	0
16.7		3	7.867	7.723	8.01	7.8	7.9	0.01924	0.05772	0.73%	0
Overall		9	7.922	7.83	8.015	7.8	8.2	0.04006	0.1202	1.52%	0 (0%)

Temperature-°C

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	24	24	24	24	24	0	0	0.00%	0
8.35		3	24.1	23.67	24.53	24	24.3	0.05775	0.1732	0.72%	0
16.7		3	24.13	23.56	24.71	24	24.4	0.07698	0.2309	0.96%	0
Overall		9	24.08	23.96	24.2	24	24.4	0.05212	0.1563	0.65%	0 (0%)

Ceriodaphnia 96-h Acute Survival Test

Aquatic Bioassay & Consulting Labs, Inc.

Alkalinity (CaCO3)-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		60					
8.35				100					
0	N	2		60					
8.35				100					
0	N	3		60					
8.35				100					

Conductivity-µmhos

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		337					
8.35				1102					
16.7				1094					
0	N	2		336					
8.35				1134					
16.7				1097					
0	N	3		340					
8.35				1151					
16.7				1099					

Dissolved Oxygen-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		7.7					
8.35				8.2					
16.7				7.7					
0	N	2		7.4					
8.35				7.6					
16.7				7.7					
0	N	3		7.1					
8.35				7					
16.7				7					

Hardness (CaCO3)-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		85					
8.35				350					
0	N	2		85					
8.35				350					
0	N	3		85					
8.35				350					

pH-Units

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		8.2					
8.35				7.8					
16.7				7.8					
0	N	2		7.9					
8.35				8					
16.7				7.9					
0	N	3		7.9					
8.35				7.9					
16.7				7.9					

Ceriodaphnia 96-h Acute Survival Test **Aquatic Bioassay & Consulting Labs, Inc.**

Temperature-°C

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		24					
8.35				24					
16.7				24					
0	N	2		24					
8.35				24.3					
16.7				24.4					
0	N	3		24					
8.35				24					
16.7				24					

aquatic bioassay & consulting laboratories, inc

October 21, 2020

Mr. David Hokanson
Trussell Technologies
232 N. Lake Ave
Pasadena, CA 91101

Dear Mr. Hokanson:

We are pleased to present the enclosed acute bioassay report. The test was conducted under the guidelines prescribed in "Methods for Measuring the Acute Toxicity of Effluents and Receiving water to Freshwater and Marine Organisms" (Fourth Edition) EPA/600/ 4-90/027F, August 1993. The results were as follows:

CLIENT:	Trussell Technologies
SAMPLE I.D.:	Lake Mathews Raw Earthtec 8.35mg/l
DATE RECEIVED:	09/10/20
ABC LAB. NO.:	TRU0920.134

NPDES PERCENT SURVIVAL BIOASSAY
RAINBOW TROUT

PERCENT SURVIVAL = 0% Survival in 8.35/mg/l Sample

TUa = >1.00 @ 8.35mg/l

Yours very truly,

Scott Johnson
Laboratory Director

AQUATIC BIOASSAY AND CONSULTING LABORATORIES, INC.
29 North Olive Street
Ventura, CA 93001
(805) 643-5621

NPDES Percent Survival Bioassay for Effluents and Stormwater

SAMPLE INFORMATION

CLIENT:	Trussell Technologies Date: 09/10/20
SAMPLE I.D.:	Lake Mathews Raw Eartht LAB # TRU0920.134

WATER QUALITY

DILUTION WATI Reconst. Fresh	AERATION Single Bubble Air
CONTROL HARDNESS	CONTROL ALKALINITY
Beg: 94 mg/l End: 101 mg/l	Beg: 63 mg/l End: 68 mg/l
SAMPLE HARDNESS	SAMPLE ALKALINITY
Beg: 330 mg/l End: 330 mg/l	Beg: 117 mg/l End: 122 mg/l

ORGANISM INFORMATION

SPECIES:	Oncorhynchus mykiss	DATE REC'D:	09/15/20
COMMON NAME:	Rainbow Trout		
SOURCE:	Thomas Fish Co.		
CARRIER:	California Overnight	NO. FISH / TANK	10

TEST DATA

	INITIAL				24 HOURS				48 HOURS				72 HOURS				96 HOURS			
DATE:	09/22/20				09/23/20				09/24/20				09/25/20				09/26/20			
TIME:	1500				1450				1550				1500				1550			
CONC.	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish
0 (Con.)	10.0	13.3	7.4	10	10.6	13.8	6.9	10	10.5	13.8	6.9	10	10.5	13.8	6.8	10	10.3	13.8	6.9	10
0 (Con.)	10.1	13.3	7.4	10	10.9	13.8	6.9	10	10.4	13.8	6.9	10	10.6	13.8	6.9	10	10.4	13.8	6.9	10
50.1mg/l	10.6	13.6	7.5	10	10.6	13.8	7.3	0				0				0				0
50.1mg/l	10.7	13.6	7.4	10	10.5	13.8	7.2	0				0				0				0

FINAL DATA

TOTAL	FISH SURVIVAL
0 (Con.)	10
0 (Con.)	10
50.1mg/l	0
50.1mg/l	0

FINAL RESULTS

PERCENT SURVIVAL =	0% Survival in 8.35/mg/l Sample
TUa =	>1.00 @ 8.35mg/l

Joe Freas, Senior Toxicologist

Date 10/22/20

aquatic bioassay & consulting laboratories, inc

October 21, 2020

Mr. David Hokanson
Trussell Technologies
232 N. Lake Ave
Pasadena, CA 91101

Dear Mr. Hokanson:

We are pleased to present the enclosed acute bioassay report. The test was conducted under the guidelines prescribed in "Methods for Measuring the Acute Toxicity of Effluents and Receiving water to Freshwater and Marine Organisms" (Fourth Edition) EPA/600/ 4-90/027F, August 1993. The results were as follows:

CLIENT:	Trussell Technologies
SAMPLE I.D.:	Lake Mathews Raw Earthtec 16.7mg/l
DATE RECEIVED:	09/10/20
ABC LAB. NO.:	TRU0920.134

NPDES PERCENT SURVIVAL BIOASSAY
RAINBOW TROUT

PERCENT SURVIVAL = 0% Survival in 16.7/mg/l Sample

TUa = >1.00 @ 16.7mg/l

Yours very truly,

Scott Johnson
Laboratory Director

AQUATIC BIOASSAY AND CONSULTING LABORATORIES, INC.
29 North Olive Street
Ventura, CA 93001
(805) 643-5621

NPDES Percent Survival Bioassay for Effluents and Stormwater

SAMPLE INFORMATION

CLIENT:	Trussell Technologies Date: 09/10/20
SAMPLE I.D.:	Lake Mathews Raw Eartht LAB # TRU0920.134

WATER QUALITY

DILUTION WATI Reconst. Fresh	AERATION Single Bubble Air
CONTROL HARDNESS	CONTROL ALKALINITY
Beg: 94 mg/l End: 101 mg/l	Beg: 63 mg/l End: 68 mg/l
SAMPLE HARDNESS	SAMPLE ALKALINITY
Beg: 350 mg/l End: 360 mg/l	Beg: 100 mg/l End: 117 mg/l

ORGANISM INFORMATION

SPECIES:	Oncorhynchus mykiss	DATE REC'D:	09/15/20
COMMON NAME:	Rainbow Trout		
SOURCE:	Thomas Fish Co.		
CARRIER:	California Overnight	NO. FISH / TANK	10

TEST DATA

	INITIAL				24 HOURS				48 HOURS				72 HOURS				96 HOURS			
DATE:	09/22/20				09/23/20				09/24/20				09/25/20				09/26/20			
TIME:	1500				1450				1550				1500				1550			
CONC.	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish
0 (Con.)	10.0	13.3	7.4	10	10.6	13.8	6.9	10	10.5	13.8	6.9	10	10.5	13.8	6.8	10	10.3	13.8	6.9	10
0 (Con.)	10.1	13.3	7.4	10	10.9	13.8	6.9	10	10.4	13.8	6.9	10	10.6	13.8	6.9	10	10.4	13.8	6.9	10
50.1mg/l	10.4	13.5	7.4	10	10.6	13.8	7.3	0				0				0				0
50.1mg/l	10.3	13.5	7.4	10	10.5	13.8	7.3	0				0				0				0

FINAL DATA

TOTAL	FISH SURVIVAL
0 (Con.)	10
0 (Con.)	10
50.1mg/l	0
50.1mg/l	0

FINAL RESULTS

PERCENT SURVIVAL =	0% Survival in 16.7/mg/l Sample
TUa =	>1.00 @ 16.7mg/l

Joe Freas, Senior Toxicologist

Date 10/12/20

CHAIN OF CUSTODY RECORD

Client:	Project Name/Number:	Analysis
Trussell Technologies	Liana Olivas 626 - 375 - 8150	
Address 232 N. Lake Ave. Pasadena, CA 91101	**Project Mgr.** David Hokanson	
Phone Number:	**P.O. #**	
	Sampled By *(signature)* Jacob Newman	

Date	Time	Comp.	Grab	Matrix	Sample ID	Volume/ Number	Acute Toxicity Fathead Minnow	Acute Toxicity O. mykiss	Acute Toxicity Ceriodaphnia						Comments
9/10/20	9:50a		✓	Liquid	Lake Mathews Raw Earth Tec (Fr, S, Lake)	28 L	2	2	2						Two Earthtec doses each

Relinquished By: *(signature)*	Date: Time:	Relinquished By: *(signature)*	Date: Time:
Jacob Newman Jacob Newman	9/10/20 9:50 am		
Received By: *(signature)*	**Date: Time:**	**Received By:** *(signature)*	**Date: Time:**
	9/10/20 9:50		

Upon sample reciept record the following results:

Temp (°C) 9.3 °C NH_3 (mg/L) _____ Cl (mg/L): _____

Aquatic Bioassay and Consulting Laboratories
29 N. Olive Street Ventura, CA 93001 Phone: (805) 643-5621 Fax: (805) 643-2930

Appendix D –

Weymouth WTP Influent 2020 Toxicity Test Report

aquatic bioassay & consulting laboratories, inc

October 22, 2020

Mr. David Hokanson
Trussell Technologies
232 N. Lake Avenue
Pasadena, CA 91101

Dear Mr. Hokanson:

We are pleased to present the enclosed bioassay report. The test was conducted under guidelines prescribed in *Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms EPA-821-R-02-012.* "All acceptability criteria were met and the concentration-response was normal. This is a valid test." Results were as follows:

CLIENT:	Trussell Technologies
SAMPLE ID.:	Lake Weymouth Raw with EarthTec
DATE RECEIVED:	17 Sept – 20
ABC LAB NO.:	TRU0920.262

ACUTE FATHEAD MINNOW SURVIVAL BIOASSAY

% Survival = 100 % Survival in 16.7 mg/l Sample

EC50 = >16.7 mg/l

Yours very truly,

Scott Johnson
Laboratory Director

CETIS Summary Report

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	13-6522-3425	**Test Type:**	Survival (96h)	**Analyst:**			
Start Date:	22 Sep-20 13:22	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water		
Ending Date:	26 Sep-20 13:35	**Species:**	Pimephales promelas	**Brine:**	Not Applicable		
Test Length:	4d 0h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO	**Age:**	
Sample ID:	02-8796-6967	**Code:**	TRU0920.262afml	**Project:**			
Sample Date:	17 Sep-20 09:44	**Material:**	Sample Water	**Source:**	Bioassay Report		
Receipt Date:	17 Sep-20 09:44	**CAS (PC):**		**Station:**	Lake Weymouth Raw with Earth Tec		
Sample Age:	5d 4h (20.8 °C)	**Client:**	Trussell Technologies				

Multiple Comparison Summary

Analysis ID	Endpoint	Comparison Method	✓	NOEL	LOEL	TOEL	PMSD	S
09-5347-6586	96h Survival Rate	Steel Many-One Rank Sum Test		16.7	>16.7	---	4.84%	1

Point Estimate Summary

Analysis ID	Endpoint	Point Estimate Method	✓	Level	mg/L	95% LCL	95% UCL	S
09-1766-0348	96h Survival Rate	Linear Interpolation (ICPIN)		EC10	>16.7	---	---	1
				EC15	>16.7	---	---	
				EC20	>16.7	---	---	
				EC25	>16.7	---	---	
				EC40	>16.7	---	---	
				EC50	>16.7	---	---	

Test Acceptability

				TAC Limits			
Analysis ID	Endpoint	Attribute	Test Stat	Lower	Upper	Overlap	Decision
09-1766-0348	96h Survival Rate	Control Resp	0.975	0.9	>>	Yes	Passes Criteria
09-5347-6586	96h Survival Rate	Control Resp	0.975	0.9	>>	Yes	Passes Criteria

96h Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	N	4	0.9750	0.8954	1.0550	0.9000	1.0000	0.0250	0.0500	5.13%	0.00%
8.35		4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	---	-2.56%
16.7		4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	---	-2.56%

96h Survival Rate Detail — MD5: 4D4EE565771C4BA0CE57BE0E4B88EAF3

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	0.9000	1.0000	1.0000	1.0000
8.35		1.0000	1.0000	1.0000	1.0000
16.7		1.0000	1.0000	1.0000	1.0000

96h Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	9/10	10/10	10/10	10/10
8.35		10/10	10/10	10/10	10/10
16.7		10/10	10/10	10/10	10/10

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	09-5347-6586	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	21 Oct-20 15:32	**Analysis:**	Nonparametric-Control vs Treatments	**Status Level:**	1
Edit Date:	21 Oct-20 15:30	**MD5 Hash:**	4D4EE565771C4BA0CE57BE0E4B88EAF	**Editor ID:**	000-189-126-0
Batch ID:	13-6522-3425	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	22 Sep-20 13:22	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water
Ending Date:	26 Sep-20 13:35	**Species:**	Pimephales promelas	**Brine:**	Not Applicable
Test Length:	4d 0h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO **Age:**
Sample ID:	02-8796-6967	**Code:**	TRU0920.262afml	**Project:**	
Sample Date:	17 Sep-20 09:44	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	17 Sep-20 09:44	**CAS (PC):**		**Station:**	Lake Weymouth Raw with Earth Tec
Sample Age:	5d 4h (20.8 °C)	**Client:**	Trussell Technologies		

Data Transform	Alt Hyp	NOEL	LOEL	TOEL	TU	MSDu	PMSD
Angular (Corrected)	C > T	16.7	>16.7	---	---	0.0472	4.84%

Steel Many-One Rank Sum Test

Control	vs	Conc-mg/L	Test Stat	Critical	Ties	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		8.35	20	11	1	6	CDF	0.8569	Non-Significant Effect
		16.7	20	11	1	6	CDF	0.8569	Non-Significant Effect

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	0.975	0.9	>>	Yes	Passes Criteria

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.0044266	0.0022133	2	1	0.4053	Non-Significant Effect
Error	0.0199195	0.0022133	9			
Total	0.0243461		11			

ANOVA Assumptions Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variance	Bartlett Equality of Variance Test				Indeterminate
	Levene Equality of Variance Test	9	8.022	0.0071	Unequal Variances
	Mod Levene Equality of Variance Test	1	8.022	0.4053	Equal Variances
Distribution	Anderson-Darling A2 Test	2.222	3.878	<1.0E-05	Non-Normal Distribution
	D'Agostino Skewness Test	3.23	2.576	0.0012	Non-Normal Distribution
	Kolmogorov-Smirnov D Test	0.4167	0.2801	<1.0E-05	Non-Normal Distribution
	Shapiro-Wilk W Normality Test	0.6334	0.8025	0.0002	Non-Normal Distribution

96h Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	0.9750	0.8954	1.0000	1.0000	0.9000	1.0000	0.0250	5.13%	0.00%
8.35		4	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	-2.56%
16.7		4	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	-2.56%

Angular (Corrected) Transformed Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	1.3710	1.2420	1.5010	1.4120	1.2490	1.4120	0.0407	5.94%	0.00%
8.35		4	1.4120	1.4120	1.4120	1.4120	1.4120	1.4120	0.0000	0.00%	-2.97%
16.7		4	1.4120	1.4120	1.4120	1.4120	1.4120	1.4120	0.0000	0.00%	-2.97%

96h Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	0.9000	1.0000	1.0000	1.0000
8.35		1.0000	1.0000	1.0000	1.0000
16.7		1.0000	1.0000	1.0000	1.0000

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	09-5347-6586	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	21 Oct-20 15:32	**Analysis:**	Nonparametric-Control vs Treatments	**Status Level:**	1
Edit Date:	21 Oct-20 15:30	**MD5 Hash:**	4D4EE565771C4BA0CE57BE0E4B88EAF	**Editor ID:**	000-189-126-0

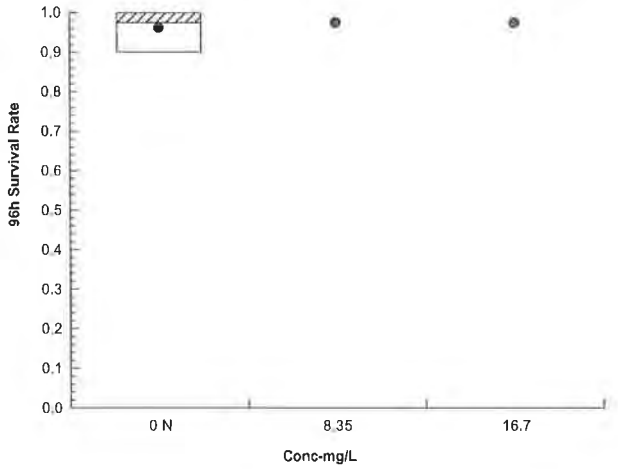
Angular (Corrected) Transformed Detail

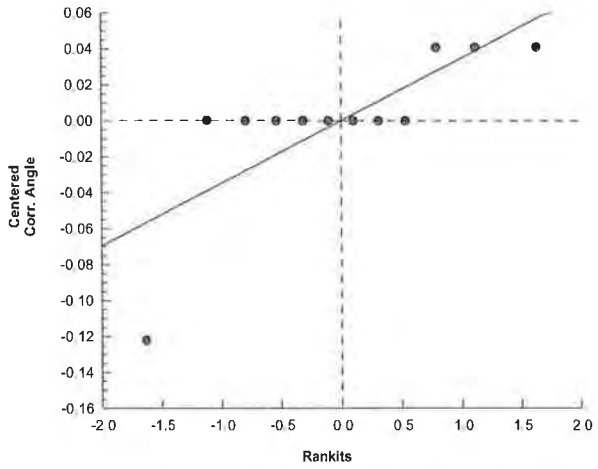
Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.2490	1.4120	1.4120	1.4120
8.35		1.4120	1.4120	1.4120	1.4120
16.7		1.4120	1.4120	1.4120	1.4120

96h Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	9/10	10/10	10/10	10/10
8.35		10/10	10/10	10/10	10/10
16.7		10/10	10/10	10/10	10/10

Graphics





Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	09-1766-0348	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	21 Oct-20 15:32	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1
Edit Date:	21 Oct-20 15:30	**MD5 Hash:**	4D4EE565771C4BA0CE57BE0E4B88EAF	**Editor ID:**	000-189-126-0
Batch ID:	13-6522-3425	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	22 Sep-20 13:22	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water
Ending Date:	26 Sep-20 13:35	**Species:**	Pimephales promelas	**Brine:**	Not Applicable
Test Length:	4d 0h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO **Age:**
Sample ID:	02-8796-6967	**Code:**	TRU0920.262afml	**Project:**	
Sample Date:	17 Sep-20 09:44	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	17 Sep-20 09:44	**CAS (PC):**		**Station:**	Lake Weymouth Raw with Earth Tec
Sample Age:	5d 4h (20.8 °C)	**Client:**	Trussell Technologies		

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	0	280	Yes	Two-Point Interpolation

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	0.975	0.9	>>	Yes	Passes Criteria

Point Estimates

Level	mg/L	95% LCL	95% UCL
EC10	>16.7	---	---
EC15	>16.7	---	---
EC20	>16.7	---	---
EC25	>16.7	---	---
EC40	>16.7	---	---
EC50	>16.7	---	---

96h Survival Rate Summary

			Calculated Variate(A/B)							Isotonic Variate	
Conc-mg/L	**Code**	**Count**	**Mean**	**Median**	**Min**	**Max**	**CV%**	**%Effect**	**A/B**	**Mean**	**%Effect**
0	N	4	0.9750	1.0000	0.9000	1.0000	5.13%	0.00%	39/40	0.9917	0.00%
8.35		4	1.0000	1.0000	1.0000	1.0000	0.00%	-2.56%	40/40	0.9917	0.00%
16.7		4	1.0000	1.0000	1.0000	1.0000	0.00%	-2.56%	40/40	0.9917	0.00%

96h Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	0.9000	1.0000	1.0000	1.0000
8.35		1.0000	1.0000	1.0000	1.0000
16.7		1.0000	1.0000	1.0000	1.0000

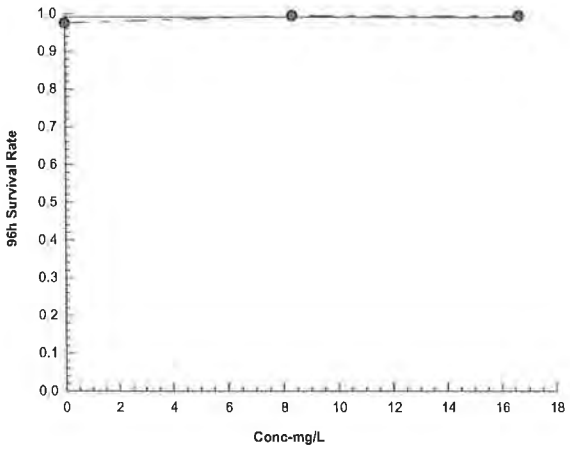
96h Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	9/10	10/10	10/10	10/10
8.35		10/10	10/10	10/10	10/10
16.7		10/10	10/10	10/10	10/10

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	09-1766-0348	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	21 Oct-20 15:32	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1
Edit Date:	21 Oct-20 15:30	**MD5 Hash:**	4D4EE565771C4BA0CE57BE0E4B88EAF	**Editor ID:**	000-189-126-0

Graphics



Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	13-6522-3425	**Test Type:**	Survival (96h)	**Analyst:**			
Start Date:	22 Sep-20 13:22	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water		
Ending Date:	26 Sep-20 13:35	**Species:**	Pimephales promelas	**Brine:**	Not Applicable		
Test Length:	4d 0h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO	**Age:**	
Sample ID:	02-8796-6967	**Code:**	TRU0920.262afml	**Project:**			
Sample Date:	17 Sep-20 09:44	**Material:**	Sample Water	**Source:**	Bioassay Report		
Receipt Date:	17 Sep-20 09:44	**CAS (PC):**		**Station:**	Lake Weymouth Raw with Earth Tec		
Sample Age:	5d 4h (20.8 °C)	**Client:**	Trussell Technologies				

Alkalinity (CaCO3)-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	60	60	60	60	60	0	0	0.00%	0
8.35		3	108	108	108	108	108	0	0	0.00%	0
Overall		6	84	56.41	111.6	60	108	10.73	26.29	31.30%	0 (0%)

Conductivity-µmhos

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	337.7	332.5	342.8	336	340	0.6939	2.082	0.62%	0
8.35		3	1107	1098	1117	1103	1110	1.262	3.786	0.34%	0
16.7		3	1101	1099	1103	1100	1102	0.3333	1	0.09%	0
Overall		9	848.7	554.1	1143	336	1110	127.8	383.3	45.16%	0 (0%)

Dissolved Oxygen-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	7.4	6.655	8.145	7.1	7.7	0.1	0.3	4.05%	0
8.35		3	7.667	6.441	8.892	7.1	8	0.1644	0.4933	6.43%	0
16.7		3	7.5	6.639	8.361	7.1	7.7	0.1155	0.3464	4.62%	0
Overall		9	7.522	7.248	7.796	7.1	8	0.1188	0.3563	4.74%	0 (0%)

Hardness (CaCO3)-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	85	85	85	85	85	0	0	0.00%	0
8.35		3	315	315	315	315	315	0	0	0.00%	0
Overall		6	200	67.8	332.2	85	315	51.43	126	62.99%	0 (0%)

pH-Units

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	8	7.57	8.43	7.9	8.2	0.05773	0.1732	2.17%	0
8.35		3	7.9	7.652	8.148	7.8	8	0.03333	0.1	1.27%	0
16.7		3	7.867	7.723	8.01	7.8	7.9	0.01924	0.05772	0.73%	0
Overall		9	7.922	7.83	8.015	7.8	8.2	0.04006	0.1202	1.52%	0 (0%)

Temperature-°C

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	24	24	24	24	24	0	0	0.00%	0
8.35		3	24.13	23.56	24.71	24	24.4	0.07698	0.2309	0.96%	0
16.7		3	24.17	23.45	24.88	24	24.5	0.09623	0.2887	1.19%	0
Overall		9	24.1	23.95	24.25	24	24.5	0.06667	0.2	0.83%	0 (0%)

Fathead Minnow 96-h Acute Survival Test

Aquatic Bioassay & Consulting Labs, Inc.

Alkalinity (CaCO3)-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		60					
8.35				108					
0	N	2		60					
8.35				108					
0	N	3		60					
8.35				108					

Conductivity-µmhos

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		337					
8.35				1103					
16.7				1102					
0	N	2		336					
8.35				1109					
16.7				1101					
0	N	3		340					
8.35				1110					
16.7				1100					

Dissolved Oxygen-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		7.7					
8.35				7.9					
16.7				7.7					
0	N	2		7.4					
8.35				8					
16.7				7.7					
0	N	3		7.1					
8.35				7.1					
16.7				7.1					

Hardness (CaCO3)-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		85					
8.35				315					
0	N	2		85					
8.35				315					
0	N	3		85					
8.35				315					

pH-Units

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		8.2					
8.35				7.8					
16.7				7.8					
0	N	2		7.9					
8.35				8					
16.7				7.9					
0	N	3		7.9					
8.35				7.9					
16.7				7.9					

Fathead Minnow 96-h Acute Survival Test **Aquatic Bioassay & Consulting Labs, Inc.**

Temperature-°C

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		24					
8.35				24					
16.7				24					
0	N	2		24					
8.35				24.4					
16.7				24.5					
0	N	3		24					
8.35				24					
16.7				24					

aquatic bioassay & consulting laboratories, inc

October 22, 2020

Mr. David Hokanson
Trussell Technologies
232 N. Lake Avenue
Pasadena, CA 91101

Dear Mr. Hokanson:

We are pleased to present the enclosed bioassay report. The test was conducted under guidelines prescribed in *Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms EPA-821-R-02-012*. "All acceptability criteria were met and the concentration-response was normal. This is a valid test." Results were as follows:

CLIENT:	Trussell Technologies
SAMPLE ID.:	Lake Weymouth Raw with EarthTec
DATE RECEIVED:	17 Sept – 20
ABC LAB NO.:	TRU0920.262

ACUTE CERIODAPHNIA SURVIVAL BIOASSAY

% Survival = 100 % Survival in 16.7mg/l Sample

EC50 = >16.7 mg/l

Yours very truly,

Scott Johnson
Laboratory Director

CETIS Summary Report

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	17-4635-5680	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	22 Sep-20 13:22	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water
Ending Date:	26 Sep-20 13:35	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable
Test Length:	4d 0h	**Taxon:**	Branchiopoda	**Source:**	Aquatic Biosystems, CO **Age:**
Sample ID:	16-1980-9739	**Code:**	TRU0920.262acer	**Project:**	
Sample Date:	17 Sep-20 09:44	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	17 Sep-20 09:44	**CAS (PC):**		**Station:**	Lake Weymouth Raw with Earth Tec
Sample Age:	5d 4h (20.8 °C)	**Client:**	Trussell Technologies		

Multiple Comparison Summary

Analysis ID	Endpoint	Comparison Method	✓	NOEL	LOEL	TOEL	PMSD	S
19-3074-8622	96h Survival Rate	Steel Many-One Rank Sum Test		16.7	>16.7	---	---	1

Point Estimate Summary

Analysis ID	Endpoint	Point Estimate Method	✓	Level	mg/L	95% LCL	95% UCL	S
18-6039-6023	96h Survival Rate	Linear Interpolation (ICPIN)		EC10	>16.7	---	---	1
				EC15	>16.7	---	---	
				EC20	>16.7	---	---	
				EC25	>16.7	---	---	
				EC40	>16.7	---	---	
				EC50	>16.7	---	---	

Test Acceptability

				TAC Limits			
Analysis ID	Endpoint	Attribute	Test Stat	Lower	Upper	Overlap	Decision
18-6039-6023	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria
19-3074-8622	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria

96h Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	N	4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	---	0.00%
8.35		4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	---	0.00%
16.7		4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	---	0.00%

96h Survival Rate Detail — MD5: 9E2E5D7DCCA34813F07FFE0218C5D2D0

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
8.35		1.0000	1.0000	1.0000	1.0000
16.7		1.0000	1.0000	1.0000	1.0000

96h Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	5/5	5/5	5/5	5/5
8.35		5/5	5/5	5/5	5/5
16.7		5/5	5/5	5/5	5/5

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	19-3074-8622	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	21 Oct-20 15:36	**Analysis:**	Nonparametric-Control vs Treatments	**Status Level:**	1
Edit Date:	21 Oct-20 15:36	**MD5 Hash:**	9E2E5D7DCCA34813F07FFE0218C5D2D0	**Editor ID:**	000-189-126-0

Batch ID:	17-4635-5680	**Test Type:**	Survival (96h)	**Analyst:**		
Start Date:	22 Sep-20 13:22	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water	
Ending Date:	26 Sep-20 13:35	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable	
Test Length:	4d 0h	**Taxon:**	Branchiopoda	**Source:**	Aquatic Biosystems, CO	**Age:**

Sample ID:	16-1980-9739	**Code:**	TRU0920.262acer	**Project:**	
Sample Date:	17 Sep-20 09:44	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	17 Sep-20 09:44	**CAS (PC):**		**Station:**	Lake Weymouth Raw with Earth Tec
Sample Age:	5d 4h (20.8 °C)	**Client:**	Trussell Technologies		

Data Transform	Alt Hyp	NOEL	LOEL	TOEL	TU
Angular (Corrected)	C > T	16.7	>16.7	---	---

Steel Many-One Rank Sum Test

Control	vs	Conc-mg/L	Test Stat	Critical	Ties	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		8.35	18	11	1	6	CDF	0.6667	Non-Significant Effect
		16.7	18	11	1	6	CDF	0.6667	Non-Significant Effect

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0	0	2			Indeterminate
Error	0	0	9			
Total	0		11			

ANOVA Assumptions Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variance	Bartlett Equality of Variance Test				Indeterminate
Distribution	Shapiro-Wilk W Normality Test				Indeterminate

96h Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%
8.35		4	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%
16.7		4	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%

Angular (Corrected) Transformed Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	1.3450	1.3450	1.3460	1.3450	1.3450	1.3450	0.0000	0.00%	0.00%
8.35		4	1.3450	1.3450	1.3460	1.3450	1.3450	1.3450	0.0000	0.00%	0.00%
16.7		4	1.3450	1.3450	1.3460	1.3450	1.3450	1.3450	0.0000	0.00%	0.00%

96h Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
8.35		1.0000	1.0000	1.0000	1.0000
16.7		1.0000	1.0000	1.0000	1.0000

Angular (Corrected) Transformed Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.3450	1.3450	1.3450	1.3450
8.35		1.3450	1.3450	1.3450	1.3450
16.7		1.3450	1.3450	1.3450	1.3450

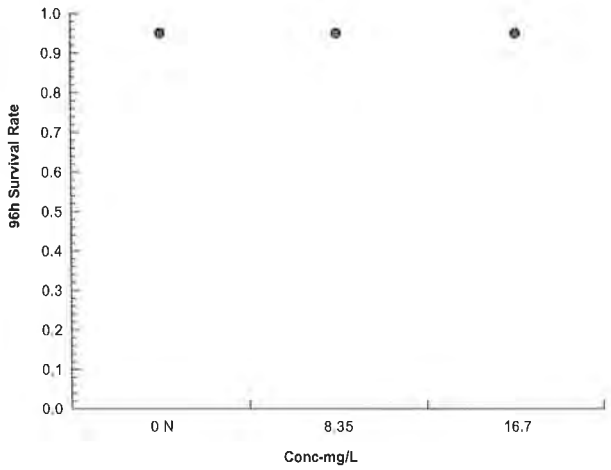
Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

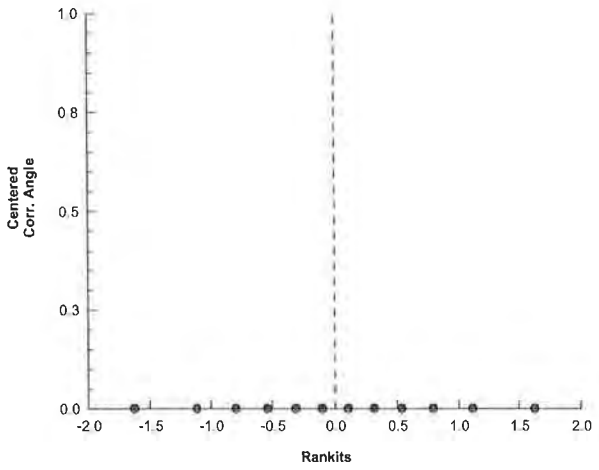
Analysis ID:	19-3074-8622	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	21 Oct-20 15:36	**Analysis:**	Nonparametric-Control vs Treatments	**Status Level:**	1
Edit Date:	21 Oct-20 15:36	**MD5 Hash:**	9E2E5D7DCCA34813F07FFE0218C5D2D0	**Editor ID:**	000-189-126-0

96h Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	5/5	5/5	5/5	5/5
8.35		5/5	5/5	5/5	5/5
16.7		5/5	5/5	5/5	5/5

Graphics





Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	18-6039-6023	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	21 Oct-20 15:36	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1
Edit Date:	21 Oct-20 15:36	**MD5 Hash:**	9E2E5D7DCCA34813F07FFE0218C5D2D0	**Editor ID:**	000-189-126-0
Batch ID:	17-4635-5680	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	22 Sep-20 13:22	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water
Ending Date:	26 Sep-20 13:35	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable
Test Length:	4d 0h	**Taxon:**	Branchiopoda	**Source:**	Aquatic Biosystems, CO **Age:**
Sample ID:	16-1980-9739	**Code:**	TRU0920.262acer	**Project:**	
Sample Date:	17 Sep-20 09:44	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	17 Sep-20 09:44	**CAS (PC):**		**Station:**	Lake Weymouth Raw with Earth Tec
Sample Age:	5d 4h (20.8 °C)	**Client:**	Trussell Technologies		

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	0	280	Yes	Two-Point Interpolation

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

Point Estimates

Level	mg/L	95% LCL	95% UCL
EC10	>16.7	---	---
EC15	>16.7	---	---
EC20	>16.7	---	---
EC25	>16.7	---	---
EC40	>16.7	---	---
EC50	>16.7	---	---

96h Survival Rate Summary

			Calculated Variate(A/B)							Isotonic Variate	
Conc-mg/L	**Code**	**Count**	**Mean**	**Median**	**Min**	**Max**	**CV%**	**%Effect**	**A/B**	**Mean**	**%Effect**
0	N	4	1.0000	1.0000	1.0000	1.0000	0.00%	0.00%	20/20	1.0000	0.00%
8.35		4	1.0000	1.0000	1.0000	1.0000	0.00%	0.00%	20/20	1.0000	0.00%
16.7		4	1.0000	1.0000	1.0000	1.0000	0.00%	0.00%	20/20	1.0000	0.00%

96h Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
8.35		1.0000	1.0000	1.0000	1.0000
16.7		1.0000	1.0000	1.0000	1.0000

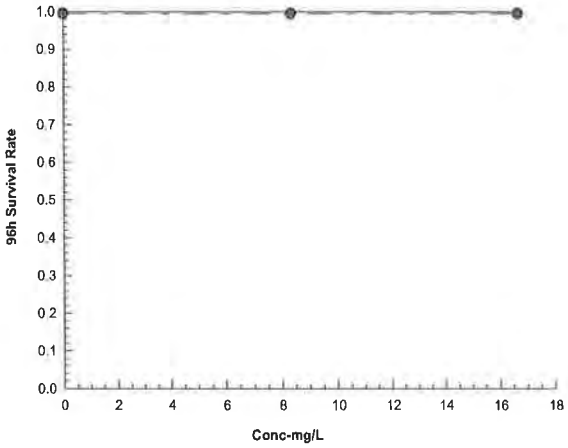
96h Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	5/5	5/5	5/5	5/5
8.35		5/5	5/5	5/5	5/5
16.7		5/5	5/5	5/5	5/5

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	18-6039-6023	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	21 Oct-20 15:36	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1
Edit Date:	21 Oct-20 15:36	**MD5 Hash:**	9E2E5D7DCCA34813F07FFE0218C5D2D0	**Editor ID:**	000-189-126-0

Graphics



Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	17-4635-5680	**Test Type:**	Survival (96h)	**Analyst:**			
Start Date:	22 Sep-20 13:22	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water		
Ending Date:	26 Sep-20 13:35	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable		
Test Length:	4d 0h	**Taxon:**	Branchiopoda	**Source:**	Aquatic Biosystems, CO	**Age:**	
Sample ID:	16-1980-9739	**Code:**	TRU0920.262acer	**Project:**			
Sample Date:	17 Sep-20 09:44	**Material:**	Sample Water	**Source:**	Bioassay Report		
Receipt Date:	17 Sep-20 09:44	**CAS (PC):**		**Station:**	Lake Weymouth Raw with Earth Tec		
Sample Age:	5d 4h (20.8 °C)	**Client:**	Trussell Technologies				

Alkalinity (CaCO3)-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	60	60	60	60	60	0	0	0.00%	0
8.35		3	108	108	108	108	108	0	0	0.00%	0
Overall		6	84	56.41	111.6	60	108	10.73	26.29	31.30%	0 (0%)

Conductivity-µmhos

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	337.7	332.5	342.8	336	340	0.6939	2.082	0.62%	0
8.35		3	1107	1098	1117	1103	1110	1.262	3.786	0.34%	0
16.7		3	1101	1099	1103	1100	1102	0.3333	1	0.09%	0
Overall		9	848.7	554.1	1143	336	1110	127.8	383.3	45.16%	0 (0%)

Dissolved Oxygen-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	7.4	6.655	8.145	7.1	7.7	0.1	0.3	4.05%	0
8.35		3	7.667	6.441	8.892	7.1	8	0.1644	0.4933	6.43%	0
16.7		3	7.5	6.639	8.361	7.1	7.7	0.1155	0.3464	4.62%	0
Overall		9	7.522	7.248	7.796	7.1	8	0.1188	0.3563	4.74%	0 (0%)

Hardness (CaCO3)-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	85	85	85	85	85	0	0	0.00%	0
8.35		3	315	315	315	315	315	0	0	0.00%	0
Overall		6	200	67.8	332.2	85	315	51.43	126	62.99%	0 (0%)

pH-Units

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	8	7.57	8.43	7.9	8.2	0.05773	0.1732	2.17%	0
8.35		3	7.9	7.652	8.148	7.8	8	0.03333	0.1	1.27%	0
16.7		3	7.867	7.723	8.01	7.8	7.9	0.01924	0.05772	0.73%	0
Overall		9	7.922	7.83	8.015	7.8	8.2	0.04006	0.1202	1.52%	0 (0%)

Temperature-°C

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	24	24	24	24	24	0	0	0.00%	0
8.35		3	24.13	23.56	24.71	24	24.4	0.07698	0.2309	0.96%	0
16.7		3	24.17	23.45	24.88	24	24.5	0.09623	0.2887	1.19%	0
Overall		9	24.1	23.95	24.25	24	24.5	0.06667	0.2	0.83%	0 (0%)

Ceriodaphnia 96-h Acute Survival Test

Aquatic Bioassay & Consulting Labs, Inc.

Alkalinity (CaCO3)-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		60					
8.35				108					
0	N	2		60					
8.35				108					
0	N	3		60					
8.35				108					

Conductivity-µmhos

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		337					
8.35				1103					
16.7				1102					
0	N	2		336					
8.35				1109					
16.7				1101					
0	N	3		340					
8.35				1110					
16.7				1100					

Dissolved Oxygen-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		7.7					
8.35				7.9					
16.7				7.7					
0	N	2		7.4					
8.35				8					
16.7				7.7					
0	N	3		7.1					
8.35				7.1					
16.7				7.1					

Hardness (CaCO3)-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		85					
8.35				315					
0	N	2		85					
8.35				315					
0	N	3		85					
8.35				315					

pH-Units

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		8.2					
8.35				7.8					
16.7				7.8					
0	N	2		7.9					
8.35				8					
16.7				7.9					
0	N	3		7.9					
8.35				7.9					
16.7				7.9					

Ceriodaphnia 96-h Acute Survival Test **Aquatic Bioassay & Consulting Labs, Inc.**

Temperature-°C

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		24					
8.35				24					
16.7				24					
0	N	2		24					
8.35				24.4					
16.7				24.5					
0	N	3		24					
8.35				24					
16.7				24					

aquatic bioassay & consulting laboratories, inc

October 21, 2020

Mr. David Hokanson
Trussell Technologies
232 N. Lake Ave
Pasadena, CA 91101

Dear Mr. Hokanson:

We are pleased to present the enclosed acute bioassay report. The test was conducted under the guidelines prescribed in "Methods for Measuring the Acute Toxicity of Effluents and Receiving water to Freshwater and Marine Organisms" (Fourth Edition) EPA/600/ 4-90/027F, August 1993. The results were as follows:

CLIENT:	Trussell Technologies
SAMPLE I.D.:	Lake Weymouth Raw Earthtec 8.35mg/l
DATE RECEIVED:	09/10/20
ABC LAB. NO.:	TRU0920.262

NPDES PERCENT SURVIVAL BIOASSAY
RAINBOW TROUT

PERCENT SURVIVAL = 0% Survival in 8.35/mg/l Sample

TUa = >1.00 @ 8.35mg/l

Yours very truly,

Scott Johnson
Laboratory Director

AQUATIC BIOASSAY AND CONSULTING LABORATORIES, INC.
29 North Olive Street
Ventura, CA 93001
(805) 643-5621

NPDES Percent Survival Bioassay for Effluents and Stormwater

SAMPLE INFORMATION

CLIENT:	Trussell Technologies Date: 09/10/20
SAMPLE I.D.:	Lake Weymouth Raw Eart LAB # TRU0920.262

WATER QUALITY

DILUTION WATI Reconst. Fresh	AERATION Single Bubble Air
CONTROL HARDNESS	CONTROL ALKALINITY
Beg: 94 mg/l End: 101 mg/l	Beg: 63 mg/l End: 68 mg/l
SAMPLE HARDNESS	SAMPLE ALKALINITY
Beg: 370 mg/l End: 380 mg/l	Beg: 116 mg/l End: 126 mg/l

ORGANISM INFORMATION

SPECIES:	Oncorhynchus mykiss	DATE REC'D:	09/15/20
COMMON NAME:	Rainbow Trout		
SOURCE:	Thomas Fish Co.		
CARRIER:	California Overnight	NO. FISH / TANK	10

TEST DATA

	INITIAL				24 HOURS				48 HOURS				72 HOURS				96 HOURS			
DATE:	09/22/20				09/23/20				09/24/20				09/25/20				09/26/20			
TIME:	1500				1450				1550				1500				1550			
CONC.	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish
0 (Con.)	10.0	13.3	7.4	10	10.6	13.8	6.9	10	10.5	13.8	6.9	10	10.5	13.8	6.8	10	10.3	13.8	6.9	10
0 (Con.)	10.1	13.3	7.4	10	10.9	13.8	6.9	10	10.4	13.8	6.9	10	10.6	13.8	6.9	10	10.4	13.8	6.9	10
50.1mg/l	10.2	13.5	7.4	10	10.6	13.8	7.4	0				0				0				0
50.1mg/l	10.1	13.5	7.4	10	10.5	13.9	7.5	0				0				0				0

FINAL DATA

TOTAL	FISH SURVIVAL
0 (Con.)	10
0 (Con.)	10
50.1mg/l	0
50.1mg/l	0

FINAL RESULTS

PERCENT SURVIVAL =	0% Survival in 8.35/mg/l Sample
TUa =	>1.00 @ 8.35mg/l

Joe Freas, Senior Toxicologist

Date 10/22/20

aquatic bioassay & consulting laboratories, inc

October 21, 2020

Mr. David Hokanson
Trussell Technologies
232 N. Lake Ave
Pasadena, CA 91101

Dear Mr. Hokanson:

We are pleased to present the enclosed acute bioassay report. The test was conducted under the guidelines prescribed in "Methods for Measuring the Acute Toxicity of Effluents and Receiving water to Freshwater and Marine Organisms" (Fourth Edition) EPA/600/ 4-90/027F, August 1993. The results were as follows:

CLIENT:	Trussell Technologies
SAMPLE I.D.:	Lake Weymouth Raw Earthtec 16.7mg/l
DATE RECEIVED:	09/10/20
ABC LAB. NO.:	TRU0920.262

NPDES PERCENT SURVIVAL BIOASSAY
RAINBOW TROUT

PERCENT SURVIVAL = 0% Survival in 16.7/mg/l Sample

TUa = >1.00 @ 16.7mg/l

Yours very truly,

Scott Johnson
Laboratory Director

AQUATIC BIOASSAY AND CONSULTING LABORATORIES, INC.
29 North Olive Street
Ventura, CA 93001
(805) 643-5621

NPDES Percent Survival Bioassay for Effluents and Stormwater

SAMPLE INFORMATION

CLIENT:	Trussell Technologies Date: 09/10/20
SAMPLE I.D.:	Lake Weymouth Raw Eart LAB # TRU0920.262

WATER QUALITY

DILUTION WAT Reconst. Fresh	AERATION Single Bubble Air
CONTROL HARDNESS	CONTROL ALKALINITY
Beg: 94 mg/l End: 101 mg/l	Beg: 63 mg/l End: 68 mg/l
SAMPLE HARDNESS	SAMPLE ALKALINITY
Beg: 315 mg/l End: 300 mg/l	Beg: 108 mg/l End: 115 mg/l

ORGANISM INFORMATION

SPECIES:	Oncorhynchus mykiss	DATE REC'D:	09/15/20
COMMON NAME:	Rainbow Trout		
SOURCE:	Thomas Fish Co.		
CARRIER:	California Overnight	NO. FISH / TANK	10

TEST DATA

	INITIAL				24 HOURS				48 HOURS				72 HOURS				96 HOURS			
DATE:	09/22/20				09/23/20				09/24/20				09/25/20				09/26/20			
TIME:	1500				1450				1550				1500				1550			
CONC.	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish
0 (Con.)	10.0	13.3	7.4	10	10.6	13.8	6.9	10	10.5	13.8	6.9	10	10.5	13.8	6.8	10	10.3	13.8	6.9	10
0 (Con.)	10.1	13.3	7.4	10	10.9	13.8	6.9	10	10.4	13.8	6.9	10	10.6	13.8	6.9	10	10.4	13.8	6.9	10
50.1mg/l	8.9	13.4	7.4	10	10.6	13.7	7.5	0				0				0				0
50.1mg/l	9.8	13.4	7.4	10	10.5	13.8	7.5	0				0				0				0

FINAL DATA

TOTAL	FISH SURVIVAL
0 (Con.)	10
0 (Con.)	10
50.1mg/l	0
50.1mg/l	0

FINAL RESULTS

PERCENT SURVIVAL =	0% Survival in 16.7/mg/l Sample
TUa =	>1.00 @ 16.7mg/l

Joe Freas, Senior Toxicologist

Date 10/22/20

CHAIN OF CUSTODY RECORD

Client:	Project Name/Number:
Trussell Technologies	Liana Olivas
Address 232 N. Lake Ave. Pasadena, CA 91101	**Project Mgr.** David Hokanson
	P.O. #
Phone Number: (626) 375 - 8150	**Sampled By** *(signature)* Jacob Neumann

Date	Time	Comp.	Grab	Matrix	Sample ID	Volume/ Number	Analysis: Acute Toxicity Fathead Minnow	Acute Toxicity O. Nykiss	Acute Toxicity Ceriodaphia							Comments
9/17/20	9:44		✓	Liquid	Lake Weymouth Raw	28 L	2	2	2							2 EarthTec doses each

Relinquised By:*(signature)* Jacob Neumann — Date: Time: 9/17/20 9:44

Received By:*(signature)* [signature] — Date: Time: 9-17-20 9:44

Relinquised By:*(signature)* — Date: Time:

Received By:*(signature)* — Date: Time:

Upon sample reciept record the following results:

Temp (°C) 20.8°C NH_3 (mg/L) <0.1 Cl (mg/L): <0.1

Aquatic Bioassay and Consulting Laboratories
29 N. Olive Street Ventura, CA 93001 Phone: (805) 643-5621 Fax: (805) 643-2930

Appendix E –

Weymouth WTP Influent 2020 (Re-sample) Toxicity Test Report

aquatic bioassay & consulting laboratories, inc

October 28, 2020

Mr. David Hokanson
Trussell Technologies
232 N. Lake Avenue
Pasadena, CA 91101

Dear Mr. Hokanson:

We are pleased to present the enclosed bioassay report. The test was conducted under guidelines prescribed in *Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms EPA-821-R-02-012*. "All acceptability criteria were met and the concentration-response was normal. This is a valid test." Results were as follows:

CLIENT:	Trussell Technologies
SAMPLE ID.:	Weymouth Resample with EarthTec
DATE RECEIVED:	1 Oct – 20
ABC LAB NO.:	TRU1020.001

ACUTE FATHEAD MINNOW SURVIVAL BIOASSAY

% Survival = 100 % Survival in 3.0 mg/l Sample

EC50 = >3.0 mg/l

Yours very truly,

Scott Johnson
Laboratory Director

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	03-5918-3368	**Test Type:**	Survival (96h)	**Analyst:**			
Start Date:	13 Oct-20 14:10	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water		
Ending Date:	17 Oct-20 12:55	**Species:**	Pimephales promelas	**Brine:**	Not Applicable		
Test Length:	95h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO	**Age:**	
Sample ID:	18-2122-7453	**Code:**	TRU1020.001afml	**Project:**			
Sample Date:	30 Sep-20 15:00	**Material:**	Sample Water	**Source:**	Bioassay Report		
Receipt Date:	01 Oct-20 10:28	**CAS (PC):**		**Station:**	Weymouth Resample with Earth Tec		
Sample Age:	12d 23h (7.3 °C)	**Client:**	Trussell Technologies				

Single Comparison Summary

Analysis ID	Endpoint	Comparison Method	P-Value	Comparison Result	S
12-8029-7781	96h Survival Rate	Wilcoxon Rank Sum Two-Sample Test	1.0000	3mg/L passed 96h survival rate	1

Test Acceptability

				TAC Limits			
Analysis ID	**Endpoint**	**Attribute**	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
12-8029-7781	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria

96h Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	N	4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	---	0.00%
3		4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	---	0.00%

96h Survival Rate Detail — MD5: 7FE761365B732DC2AD0B61FEAB25F3F5

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
3		1.0000	1.0000	1.0000	1.0000

96h Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	10/10	10/10	10/10	10/10
3		10/10	10/10	10/10	10/10

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	12-8029-7781	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	26 Oct-20 15:16	**Analysis:**	Nonparametric-Two Sample	**Status Level:**	1
Edit Date:	24 Oct-20 20:54	**MD5 Hash:**	7FE761365B732DC2AD0B61FEAB25F3F5	**Editor ID:**	000-189-126-0
Batch ID:	03-5918-3368	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	13 Oct-20 14:10	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water
Ending Date:	17 Oct-20 12:55	**Species:**	Pimephales promelas	**Brine:**	Not Applicable
Test Length:	95h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO **Age:**
Sample ID:	18-2122-7453	**Code:**	TRU1020.001afml	**Project:**	
Sample Date:	30 Sep-20 15:00	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	01 Oct-20 10:28	**CAS (PC):**		**Station:**	Weymouth Resample with Earth Tec
Sample Age:	12d 23h (7.3 °C)	**Client:**	Trussell Technologies		

Data Transform	Alt Hyp	Comparison Result
Angular (Corrected)	C > T	3mg/L passed 96h survival rate endpoint

Wilcoxon Rank Sum Two-Sample Test

Control	vs	Conc-mg/L	Test Stat	Critical	Ties	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		3	18	---	1	6	Exact	1.0000	Non-Significant Effect

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0	0	1			Indeterminate
Error	0	0	6			
Total	0		7			

ANOVA Assumptions Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variance	Variance Ratio F Test				Indeterminate
Distribution	Shapiro-Wilk W Normality Test				Indeterminate

96h Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%
3		4	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%

Angular (Corrected) Transformed Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	1.4120	1.4120	1.4120	1.4120	1.4120	1.4120	0.0000	0.00%	0.00%
3		4	1.4120	1.4120	1.4120	1.4120	1.4120	1.4120	0.0000	0.00%	0.00%

96h Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
3		1.0000	1.0000	1.0000	1.0000

Angular (Corrected) Transformed Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.4120	1.4120	1.4120	1.4120
3		1.4120	1.4120	1.4120	1.4120

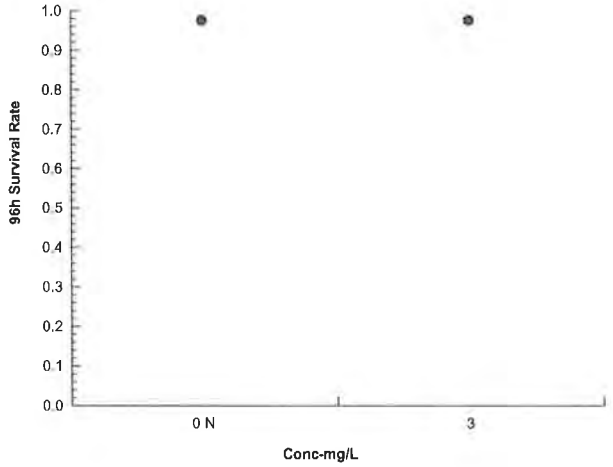
96h Survival Rate Binomials

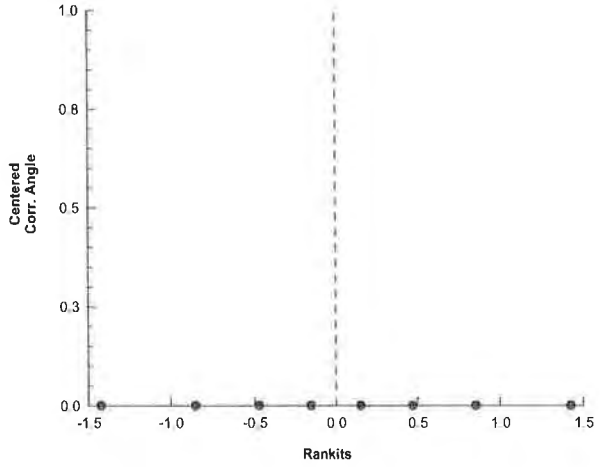
Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	10/10	10/10	10/10	10/10
3		10/10	10/10	10/10	10/10

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	12-8029-7781	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	26 Oct-20 15:16	**Analysis:**	Nonparametric-Two Sample	**Status Level:**	1
Edit Date:	24 Oct-20 20:54	**MD5 Hash:**	7FE761365B732DC2AD0B61FEAB25F3F5	**Editor ID:**	000-189-126-0

Graphics





Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	03-5918-3368	**Test Type:**	Survival (96h)	**Analyst:**		
Start Date:	13 Oct-20 14:10	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water	
Ending Date:	17 Oct-20 12:55	**Species:**	Pimephales promelas	**Brine:**	Not Applicable	
Test Length:	95h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO	**Age:**
Sample ID:	18-2122-7453	**Code:**	TRU1020.001afml	**Project:**		
Sample Date:	30 Sep-20 15:00	**Material:**	Sample Water	**Source:**	Bioassay Report	
Receipt Date:	01 Oct-20 10:28	**CAS (PC):**		**Station:**	Weymouth Resample with Earth Tec	
Sample Age:	12d 23h (7.3 °C)	**Client:**	Trussell Technologies			

Alkalinity (CaCO3)-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	60	60	60	60	60	0	0	0.00%	0
3		3	113	113	113	113	113	0	0	0.00%	0
Overall		6	86.5	56.04	117	60	113	11.85	29.03	33.56%	0 (0%)

Conductivity-µmhos

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	352	321.8	382.2	338	360	4.055	12.17	3.46%	0
3		3	1025	1017	1032	1022	1028	1.018	3.055	0.30%	0
Overall		6	688.3	301.6	1075	338	1028	150.4	368.5	53.54%	0 (0%)

Dissolved Oxygen-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	7.633	7.49	7.777	7.6	7.7	0.01924	0.05773	0.76%	0
3		3	7.4	6.743	8.057	7.2	7.7	0.08819	0.2646	3.58%	0
Overall		6	7.517	7.292	7.741	7.2	7.7	0.08724	0.2137	2.84%	0 (0%)

Hardness (CaCO3)-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	85	85	85	85	85	0	0	0.00%	0
3		3	238	238	238	238	238	0	0	0.00%	0
Overall		6	161.5	73.56	249.4	85	238	34.21	83.8	51.89%	0 (0%)

pH-Units

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	7.7	7.699	7.701	7.7	7.7	0	0	0.00%	0
3		3	7.7	7.699	7.701	7.7	7.7	0	0	0.00%	0
Overall		6	7.7	7.7	7.7	7.7	7.7	0	0	0.00%	0 (0%)

Temperature-°C

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	24	24	24	24	24	0	0	0.00%	0
3		3	24	24	24	24	24	0	0	0.00%	0
Overall		6	24	24	24	24	24	0	0	0.00%	0 (0%)

Fathead Minnow 96-h Acute Survival Test

Aquatic Bioassay & Consulting Labs, Inc.

Alkalinity (CaCO3)-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		60					
3				113					
0	N	2		60					
3				113					
0	N	3		60					
3				113					

Conductivity-µmhos

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		358					
3				1024					
0	N	2		360					
3				1028					
0	N	3		338					
3				1022					

Dissolved Oxygen-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		7.6					
3				7.7					
0	N	2		7.7					
3				7.3					
0	N	3		7.6					
3				7.2					

Hardness (CaCO3)-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		85					
3				238					
0	N	2		85					
3				238					
0	N	3		85					
3				238					

pH-Units

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		7.7					
3				7.7					
0	N	2		7.7					
3				7.7					
0	N	3		7.7					
3				7.7					

Temperature-°C

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		24					
3				24					
0	N	2		24					
3				24					
0	N	3		24					
3				24					

aquatic bioassay & consulting laboratories, inc

October 28, 2020

Mr. David Hokanson
Trussell Technologies
232 N. Lake Avenue
Pasadena, CA 91101

Dear Mr. Hokanson:

We are pleased to present the enclosed bioassay report. The test was conducted under guidelines prescribed in *Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms EPA-821-R-02-012.* "All acceptability criteria were met and the concentration-response was normal. This is a valid test." Results were as follows:

CLIENT:	Trussell Technologies
SAMPLE ID.:	Weymouth Resample with EarthTec
DATE RECEIVED:	1 Oct – 20
ABC LAB NO.:	TRU1020.001

ACUTE CERIODAPHNIA SURVIVAL BIOASSAY

% Survival = 0 % Survival in 3.0 mg/l Sample

EC50 = < 3.0 mg/l

Yours very truly,

Scott Johnson
Laboratory Director

CETIS Summary Report

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	00-9934-3961	**Test Type:**	Survival (96h)	**Analyst:**			
Start Date:	13 Oct-20 14:10	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water		
Ending Date:	17 Oct-20 12:55	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable		
Test Length:	95h	**Taxon:**	Branchiopoda	**Source:**	Aquatic Biosystems, CO	**Age:**	
Sample ID:	00-5310-5516	**Code:**	TRU1020.001acer	**Project:**			
Sample Date:	30 Sep-20 15:00	**Material:**	Sample Water	**Source:**	Bioassay Report		
Receipt Date:	01 Oct-20 10:28	**CAS (PC):**		**Station:**	Weymouth Resample with Earth Tec		
Sample Age:	12d 23h (7.3 °C)	**Client:**	Trussell Technologies				

Single Comparison Summary

Analysis ID	Endpoint	Comparison Method	P-Value	Comparison Result	S
10-1520-9557	96h Survival Rate	Wilcoxon Rank Sum Two-Sample Test	0.0143	3mg/L failed 96h survival rate	1

Test Acceptability

				TAC Limits			
Analysis ID	**Endpoint**	**Attribute**	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
10-1520-9557	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria

96h Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	N	4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	---	0.00%
3		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	---	100.00%

96h Survival Rate Detail — MD5: A855D6A5B6135CE1F9C8F16971C8225F

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
3		0.0000	0.0000	0.0000	0.0000

96h Survival Rate Binomials

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	5/5	5/5	5/5	5/5
3		0/5	0/5	0/5	0/5

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	10-1520-9557	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	24 Oct-20 21:00	**Analysis:**	Nonparametric-Two Sample	**Status Level:**	1
Edit Date:	24 Oct-20 20:59	**MD5 Hash:**	A855D6A5B6135CE1F9C8F16971C8225F	**Editor ID:**	000-189-126-0
Batch ID:	00-9934-3961	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	13 Oct-20 14:10	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water
Ending Date:	17 Oct-20 12:55	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable
Test Length:	95h	**Taxon:**	Branchiopoda	**Source:**	Aquatic Biosystems, CO **Age:**
Sample ID:	00-5310-5516	**Code:**	TRU1020.001acer	**Project:**	
Sample Date:	30 Sep-20 15:00	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	01 Oct-20 10:28	**CAS (PC):**		**Station:**	Weymouth Resample with Earth Tec
Sample Age:	12d 23h (7.3 °C)	**Client:**	Trussell Technologies		

Data Transform	Alt Hyp	Comparison Result
Angular (Corrected)	C > T	3mg/L failed 96h survival rate endpoint

Wilcoxon Rank Sum Two-Sample Test

Control	vs	Conc-mg/L	Test Stat	Critical	Ties	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		3*	10	---	0	6	Exact	0.0143	Significant Effect

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	2.50777	2.50777	1			Indeterminate
Error	0	0	6			
Total	2.50777		7			

ANOVA Assumptions Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variance	Variance Ratio F Test				Indeterminate
Distribution	Shapiro-Wilk W Normality Test				Indeterminate

96h Survival Rate Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%
3		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	---	100.00%

Angular (Corrected) Transformed Summary

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	1.3450	1.3450	1.3460	1.3450	1.3450	1.3450	0.0000	0.00%	0.00%
3		4	0.2255	0.2255	0.2256	0.2255	0.2255	0.2255	0.0000	0.00%	83.24%

96h Survival Rate Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
3		0.0000	0.0000	0.0000	0.0000

Angular (Corrected) Transformed Detail

Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.3450	1.3450	1.3450	1.3450
3		0.2255	0.2255	0.2255	0.2255

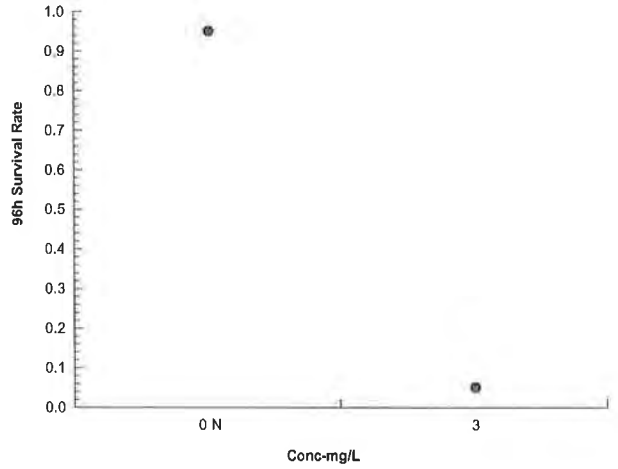
96h Survival Rate Binomials

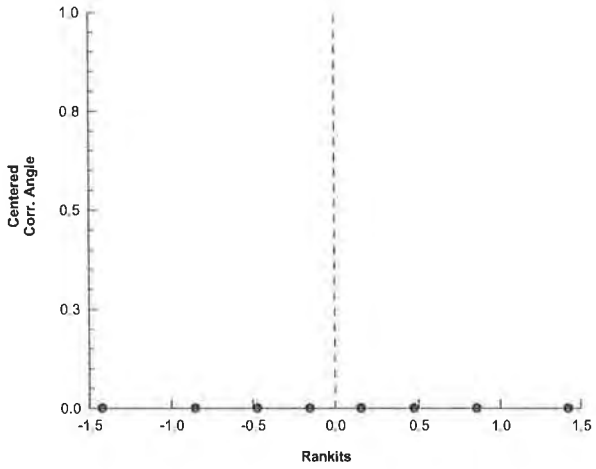
Conc-mg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	5/5	5/5	5/5	5/5
3		0/5	0/5	0/5	0/5

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	10-1520-9557	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	24 Oct-20 21:00	**Analysis:**	Nonparametric-Two Sample	**Status Level:**	1
Edit Date:	24 Oct-20 20:59	**MD5 Hash:**	A855D6A5B6135CE1F9C8F16971C8225F	**Editor ID:**	000-189-126-0

Graphics





Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	00-9934-3961	**Test Type:**	Survival (96h)	**Analyst:**			
Start Date:	13 Oct-20 14:10	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water		
Ending Date:	17 Oct-20 12:55	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable		
Test Length:	95h	**Taxon:**	Branchiopoda	**Source:**	Aquatic Biosystems, CO	**Age:**	
Sample ID:	00-5310-5516	**Code:**	TRU1020.001acer	**Project:**			
Sample Date:	30 Sep-20 15:00	**Material:**	Sample Water	**Source:**	Bioassay Report		
Receipt Date:	01 Oct-20 10:28	**CAS (PC):**		**Station:**	Weymouth Resample with Earth Tec		
Sample Age:	12d 23h (7.3 °C)	**Client:**	Trussell Technologies				

Alkalinity (CaCO3)-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	60	60	60	60	60	0	0	0.00%	0
3		3	113	113	113	113	113	0	0	0.00%	0
Overall		6	86.5	56.04	117	60	113	11.85	29.03	33.56%	0 (0%)

Conductivity-µmhos

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	352	321.8	382.2	338	360	4.055	12.17	3.46%	0
3		3	1025	1017	1032	1022	1028	1.018	3.055	0.30%	0
Overall		6	688.3	301.6	1075	338	1028	150.4	368.5	53.54%	0 (0%)

Dissolved Oxygen-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	7.633	7.49	7.777	7.6	7.7	0.01924	0.05773	0.76%	0
3		3	7.4	6.743	8.057	7.2	7.7	0.08819	0.2646	3.58%	0
Overall		6	7.517	7.292	7.741	7.2	7.7	0.08724	0.2137	2.84%	0 (0%)

Hardness (CaCO3)-mg/L

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	85	85	85	85	85	0	0	0.00%	0
3		3	238	238	238	238	238	0	0	0.00%	0
Overall		6	161.5	73.56	249.4	85	238	34.21	83.8	51.89%	0 (0%)

pH-Units

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	7.7	7.699	7.701	7.7	7.7	0	0	0.00%	0
3		3	7.7	7.699	7.701	7.7	7.7	0	0	0.00%	0
Overall		6	7.7	7.7	7.7	7.7	7.7	0	0	0.00%	0 (0%)

Temperature-°C

Conc-mg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	3	24	24	24	24	24	0	0	0.00%	0
3		3	24	24	24	24	24	0	0	0.00%	0
Overall		6	24	24	24	24	24	0	0	0.00%	0 (0%)

Ceriodaphnia 96-h Acute Survival Test

Aquatic Bioassay & Consulting Labs, Inc.

Alkalinity (CaCO3)-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		60					
3				113					
0	N	2		60					
3				113					
0	N	3		60					
3				113					

Conductivity-µmhos

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		358					
3				1024					
0	N	2		360					
3				1028					
0	N	3		338					
3				1022					

Dissolved Oxygen-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		7.6					
3				7.7					
0	N	2		7.7					
3				7.3					
0	N	3		7.6					
3				7.2					

Hardness (CaCO3)-mg/L

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		85					
3				238					
0	N	2		85					
3				238					
0	N	3		85					
3				238					

pH-Units

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		7.7					
3				7.7					
0	N	2		7.7					
3				7.7					
0	N	3		7.7					
3				7.7					

Temperature-°C

Conc-mg/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	N	1		24					
3				24					
0	N	2		24					
3				24					
0	N	3		24					
3				24					

aquatic bioassay & consulting laboratories, inc

October 27, 2020

Mr. David Hokanson
Trussell Technologies
232 N. Lake Ave
Pasadena, CA 91101

Dear Mr. Hokanson:

We are pleased to present the enclosed acute bioassay report. The test was conducted under the guidelines prescribed in "Methods for Measuring the Acute Toxicity of Effluents and Receiving water to Freshwater and Marine Organisms" (Fourth Edition) EPA/600/ 4-90/027F, August 1993. The results were as follows:

CLIENT:	Trussell Technologies
SAMPLE I.D.:	Weymouth Besample Earthtec 3.0mg/l
DATE RECEIVED:	09/10/20
ABC LAB. NO.:	TRU1020.001

NPDES PERCENT SURVIVAL BIOASSAY
RAINBOW TROUT

PERCENT SURVIVAL = 65% Survival in 3.0/mg/l Sample

TUa = 0.91 @ 3.0mg/l

Yours very truly,

Scott Johnson
Laboratory Director

AQUATIC BIOASSAY AND CONSULTING LABORATORIES, INC.
29 North Olive Street
Ventura, CA 93001
(805) 643-5621

NPDES Percent Survival Bioassay for Effluents and Stormwater

SAMPLE INFORMATION

CLIENT:	Trussell Technologies	Date:	09/10/20
SAMPLE I.D.:	Weymouth Besample Eartl	LAB #	TRU1020.001

WATER QUALITY

DILUTION WATI Reconst. Fresh	AERATION Single Bubble Air
CONTROL HARDNESS	CONTROL ALKALINITY
Beg: 94 mg/l End: 100 mg/l	Beg: 63 mg/l End: 66 mg/l
SAMPLE HARDNESS	SAMPLE ALKALINITY
Beg: 238 mg/l End: 250 mg/l	Beg: 113 mg/l End: 116 mg/l

ORGANISM INFORMATION

SPECIES:	Oncorhynchus mykiss	DATE REC'D:	10/08/20
COMMON NAME:	Rainbow Trout		
SOURCE:	Thomas Fish Co.		
CARRIER:	California Overnight	NO. FISH / TANK	10

TEST DATA

	INITIAL				24 HOURS				48 HOURS				72 HOURS				96 HOURS			
DATE:	10/15/20				10/16/20				10/17/20				10/18/20				10/19/20			
TIME:	1400				1500				1450				1400				1400			
CONC.	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish	Dis. Oxy.	Temp. dg.C	pH	#Fish
0 (Con.)	9.9	13.8	6.8	10	10.1	13.9	6.5	10	9.9	13.8	6.5	10	9.9	13.7	6.5	10	9.9	13.6	6.7	10
0 (Con.)	9.8	13.8	6.8	10	9.9	13.8	6.7	10	9.8	13.8	6.6	10	9.9	13.6	6.7	10	9.9	13.7	6.8	10
50.1mg/l	9.8	13.6	7.0	10	10.1	13.7	6.7	10	10.0	13.8	6.4	6	10.0	13.6	6.2	6	9.9	13.7	6.7	6
50.1mg/l	9.6	13.6	7.0	10	9.9	13.7	6.6	10	10.1	13.8	6.5	7	10.0	13.6	6.4	7	10.0	13.7	6.5	7

FINAL DATA

TOTAL	FISH SURVIVAL
0 (Con.)	10
0 (Con.)	10
50.1mg/l	6
50.1mg/l	7

FINAL RESULTS

PERCENT SURVIVAL =	65% Survival in 3.0/mg/l Sample
TUa =	0.91 @ 3.0mg/l

Joe Freas, Senior Toxicologist

Date 10/20/20

CHAIN OF CUSTODY RECORD

Client:	Project Name/Number:	Analysis
Trussell Technologies	Liam. Clivas 626 375 8150	
Address 232 N Lake Ave. Pasadena CA 91101	**Project Mgr.** David Hokanson	
Phone Number: (626) 375-8150	**P.O. #**	
	Sampled By *(signature)*	

Date	Time	Comp.	Grab	Matrix	Sample ID	Volume/ Number	Acute Toxicity Fathead Minnow	Acute Toxicity O. Mykiss	Acute Toxicity Ceriodaphnia	Comments
9/30/20	15:00		✓	Liquid	Weymouth Resample	5 gal	1	1	1	Test 3.0 mg/L EarthTec QZ ↑ = 0.18 mg/L Cu

Relinquished By:*(signature)*	Date: Time: 10/1/20 10:28
Received By:*(signature)* Victor Myers	Date: Time: 10/1/20 1028
Relinquished By:*(signature)*	Date: Time:
Received By:*(signature)*	Date: Time:

Upon sample reciept record the following results: .001

Temp (°C) 7.3 NH_3 (mg/L) <0.1 Cl (mg/L): 0.1

Aquatic Bioassay and Consulting Laboratories
29 N. Olive Street Ventura, CA 93001 Phone: (805) 643-5621 Fax: (805) 643-2930

Appendix F –

Weymouth WTP Influent 2021 Toxicity Test Report

aquatic bioassay & consulting laboratories, inc

September 8, 2021

Mr. David Hokanson
Trussell Technologies
232 N. Lake Avenue
Pasadena, CA 91101

Dear Mr. Hokanson:

We are pleased to present the enclosed bioassay report. The test was conducted under guidelines prescribed in *Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms EPA-821-R-02-012*. "All acceptability criteria were met and the concentration-response was normal. This is a valid test." Results were as follows:

CLIENT:	Trussell Technologies
SAMPLE ID.:	Weymouth WTP Infl. with EarthTec QZ
DATE RECEIVED:	1 July – 21
ABC LAB NO.:	TRU0721.006

RAINBOW TROUT SURVIVAL BIOASSAY

% Survival = CON = 100.00%
3.0uL/L = 100.00%
8.35uL/L = 0.00%
16.7uL/L = 0.00%

EC50 = 5.675 ul/l

Yours very truly,

Scott Johnson
Laboratory Director

CETIS Summary Report

Fish 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	19-9127-3820	**Test Type:**	Survival (96h)	**Analyst:**	Joe Freas		
Start Date:	07 Jul-21 16:00	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water		
Ending Date:	11 Jul-21 14:00	**Species:**	Oncorhynchus mykiss	**Brine:**	Not Applicable		
Test Length:	94h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO	**Age:**	
Sample ID:	07-2433-7579	**Code:**	TRU0721.006	**Project:**			
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report		
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Weymouth WTP Influent		
Sample Age:	6d 4h	**Client:**	Trussell Technologies				

SIngle Comparison Summary

Analysis ID	Endpoint	Comparison Method	P-Value	Comparison Result	S
01-7403-9603	96h Survival Rate	Steel Many-One Rank Sum Test	0.5000	3µL/L passed 96h survival rate	1

Point Estimate Summary

Analysis ID	Endpoint	Point Estimate Method	✓	Level	µL/L	95% LCL	95% UCL	S
19-6588-7535	96h Survival Rate	Linear Interpolation (ICPIN)		EC10	3.535	3.535	3.535	1
				EC15	3.803	3.803	3.803	
				EC20	4.07	4.07	4.07	
				EC25	4.338	4.338	4.338	
				EC40	5.14	5.14	5.14	
				EC50	5.675	5.675	5.675	

Test Acceptability

				TAC Limits			
Analysis ID	Endpoint	Attribute	Test Stat	Lower	Upper	Overlap	Decision
01-7403-9603	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria
19-6588-7535	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria

96h Survival Rate Summary

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	N	4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	---	0.00%
3		4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	---	0.00%
8.35		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	---	100.00%
16.7		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	---	100.00%

96h Survival Rate Detail

MD5: 3A0D5E0FE40D419F8BDE03F5112603FC

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
3		1.0000	1.0000	1.0000	1.0000
8.35		0.0000	0.0000	0.0000	0.0000
16.7		0.0000	0.0000	0.0000	0.0000

96h Survival Rate Binomials

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	10/10	10/10	10/10	10/10
3		10/10	10/10	10/10	10/10
8.35		0/10	0/10	0/10	0/10
16.7		0/10	0/10	0/10	0/10

CETIS Analytical Report

Fish 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	01-7403-9603	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	08 Sep-21 13:18	**Analysis:**	Nonparametric-Control vs Treatments	**Status Level:**	1
Edit Date:	08 Sep-21 13:07	**MD5 Hash:**	3A0D5E0FE40D419F8BDE03F5112603FC	**Editor ID:**	007-979-628-1
Batch ID:	19-9127-3820	**Test Type:**	Survival (96h)	**Analyst:**	Joe Freas
Start Date:	07 Jul-21 16:00	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water
Ending Date:	11 Jul-21 14:00	**Species:**	Oncorhynchus mykiss	**Brine:**	Not Applicable
Test Length:	94h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO **Age:**
Sample ID:	07-2433-7579	**Code:**	TRU0721.006	**Project:**	
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Weymouth WTP Influent
Sample Age:	6d 4h	**Client:**	Trussell Technologies		

Data Transform	Alt Hyp	Comparison Result
Angular (Corrected)	C > T	3µL/L passed 96h survival rate endpoint

Steel Many-One Rank Sum Test

Control	vs	Conc-µL/L	Test Stat	Critical	Ties	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		3	18	12	1	6	CDF	0.5000	Non-Significant Effect

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0	0	1			Indeterminate
Error	0	0	6			
Total	0		7			

ANOVA Assumptions Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variance	Variance Ratio F Test				Indeterminate
Distribution	Shapiro-Wilk W Normality Test				Indeterminate

96h Survival Rate Summary

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	1.0000	1.0000	1.0000		1.0000	1.0000	0.0000	0.00%	0.00%
3		4	1.0000	1.0000	1.0000		1.0000	1.0000	0.0000	0.00%	0.00%
8.35		4	0.0000	0.0000	0.0000		0.0000	0.0000	0.0000	---	100.00%
16.7		4	0.0000	0.0000	0.0000		0.0000	0.0000	0.0000	---	100.00%

Angular (Corrected) Transformed Summary

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	1.4120	1.4120	1.4120		1.4120	1.4120	0.0000	0.00%	0.00%
3		4	1.4120	1.4120	1.4120		1.4120	1.4120	0.0000	0.00%	0.00%
8.35		4	0.1588	0.1588	0.1588		0.1588	0.1588	0.0000	0.00%	88.76%
16.7		4	0.1588	0.1588	0.1588		0.1588	0.1588	0.0000	0.00%	88.76%

96h Survival Rate Detail

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
3		1.0000	1.0000	1.0000	1.0000
8.35		0.0000	0.0000	0.0000	0.0000
16.7		0.0000	0.0000	0.0000	0.0000

Fish 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	01-7403-9603	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	08 Sep-21 13:18	**Analysis:**	Nonparametric-Control vs Treatments	**Status Level:**	1
Edit Date:	08 Sep-21 13:07	**MD5 Hash:**	3A0D5E0FE40D419F8BDE03F5112603FC	**Editor ID:**	007-979-628-1

Angular (Corrected) Transformed Detail

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.4120	1.4120	1.4120	1.4120
3		1.4120	1.4120	1.4120	1.4120
8.35		0.1588	0.1588	0.1588	0.1588
16.7		0.1588	0.1588	0.1588	0.1588

CETIS Analytical Report

Fish 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	19-6588-7535	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	08 Sep-21 13:18	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1
Edit Date:	08 Sep-21 13:07	**MD5 Hash:**	3A0D5E0FE40D419F8BDE03F5112603FC	**Editor ID:**	007-979-628-1
Batch ID:	19-9127-3820	**Test Type:**	Survival (96h)	**Analyst:**	Joe Freas
Start Date:	07 Jul-21 16:00	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water
Ending Date:	11 Jul-21 14:00	**Species:**	Oncorhynchus mykiss	**Brine:**	Not Applicable
Test Length:	94h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO **Age:**
Sample ID:	07-2433-7579	**Code:**	TRU0721.006	**Project:**	
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Weymouth WTP Influent
Sample Age:	6d 4h	**Client:**	Trussell Technologies		

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	0	280	Yes	Two-Point Interpolation

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

Point Estimates

Level	µL/L	95% LCL	95% UCL
EC10	3.535	3.535	3.535
EC15	3.803	3.803	3.803
EC20	4.07	4.07	4.07
EC25	4.338	4.338	4.338
EC40	5.14	5.14	5.14
EC50	5.675	5.675	5.675

96h Survival Rate Summary

			Calculated Variate(A/B)							Isotonic Variate	
Conc-µL/L	**Code**	**Count**	**Mean**	**Median**	**Min**	**Max**	**CV%**	**%Effect**	**A/B**	**Mean**	**%Effect**
0	N	4	1.0000	1.0000	1.0000	1.0000	0.00%	0.00%	40/40	1.0000	0.00%
3		4	1.0000	1.0000	1.0000	1.0000	0.00%	0.00%	40/40	1.0000	0.00%
8.35		4	0.0000	0.0000	0.0000	0.0000	---	100.00%	0/40	0.0000	100.00%
16.7		4	0.0000	0.0000	0.0000	0.0000	---	100.00%	0/40	0.0000	100.00%

96h Survival Rate Detail

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
3		1.0000	1.0000	1.0000	1.0000
8.35		0.0000	0.0000	0.0000	0.0000
16.7		0.0000	0.0000	0.0000	0.0000

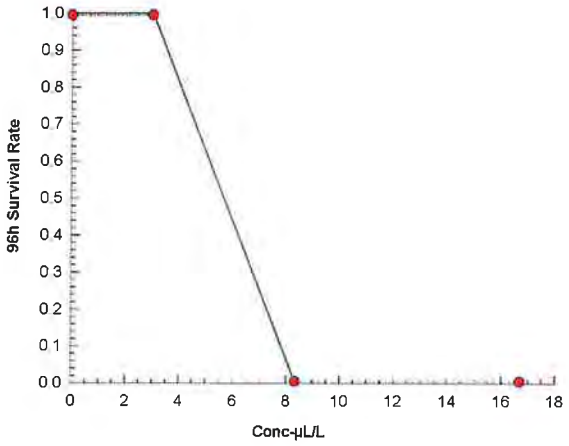
96h Survival Rate Binomials

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	10/10	10/10	10/10	10/10
3		10/10	10/10	10/10	10/10
8.35		0/10	0/10	0/10	0/10
16.7		0/10	0/10	0/10	0/10

Fish 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	19-6588-7535	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	08 Sep-21 13:18	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1
Edit Date:	08 Sep-21 13:07	**MD5 Hash:**	3A0D5E0FE40D419F8BDE03F5112603FC	**Editor ID:**	007-979-628-1

Graphics



CETIS Measurement Report

Fish 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	19-9127-3820	**Test Type:**	Survival (96h)	**Analyst:**	Joe Freas	
Start Date:	07 Jul-21 16:00	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water	
Ending Date:	11 Jul-21 14:00	**Species:**	Oncorhynchus mykiss	**Brine:**	Not Applicable	
Test Length:	94h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO	**Age:**
Sample ID:	07-2433-7579	**Code:**	TRU0721.006	**Project:**		
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report	
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Weymouth WTP Influent	
Sample Age:	6d 4h	**Client:**	Trussell Technologies			

Alkalinity (CaCO3)-mg/L

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	1	125	---	---	125	125	---	0	---	0
3		1	125	---	---	125	125	---	0	---	0
8.35		1	125	---	---	125	125	---	0	---	0
16.7		1	125	---	---	125	125	---	0	---	0
Overall		4	125	125	125	125	125	0	0	0.00%	0 (0%)

Dissolved Oxygen-mg/L

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	5	10.22	9.875	10.56	9.9	10.5	0.0555	0.2775	2.72%	0
3		5	10.2	10.08	10.32	10.1	10.3	0.02	0.1	0.98%	0
8.35		2	10.05	8.144	11.96	9.9	10.2	0.1061	0.2121	2.11%	0
16.7		2	10.05	8.144	11.96	9.9	10.2	0.1061	0.2121	2.11%	0
Overall		14	10.16	10.05	10.28	9.9	10.5	0.05305	0.1985	1.95%	0 (0%)

Hardness (CaCO3)-mg/L

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	1	300	---	---	300	300	---	0	---	0
3		1	330	---	---	330	330	---	0	---	0
16.7		1	323	---	---	323	323	---	0	---	0
Overall		3	317.7	278.7	356.7	300	330	9.062	15.7	4.94%	0 (0%)

pH-Units

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	5	7.98	7.635	8.325	7.5	8.2	0.0555	0.2775	3.48%	0
3		5	7.96	7.562	8.358	7.4	8.2	0.06419	0.3209	4.03%	0
8.35		2	7.7	3.888	11.51	7.4	8	0.2121	0.4243	5.51%	0
16.7		2	7.65	3.203	12.1	7.3	8	0.2475	0.495	6.47%	0
Overall		14	7.886	7.696	8.075	7.3	8.2	0.08762	0.3278	4.16%	0 (0%)

Temperature-°C

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	5	12.06	10.02	14.1	10.2	14	0.3282	1.641	13.61%	0
3		5	13.08	12.5	13.66	12.8	13.9	0.09317	0.4658	3.56%	0
8.35		2	13.4	5.776	21.02	12.8	14	0.4243	0.8485	6.33%	0
16.7		2	13.4	5.776	21.02	12.8	14	0.4243	0.8485	6.33%	0
Overall		14	12.81	12.13	13.48	10.2	14	0.3114	1.165	9.10%	0 (0%)

aquatic bioassay & consulting laboratories, inc

July 21, 2021

Mr. David Hokanson
Trussell Technologies
232 N. Lake Avenue
Pasadena, CA 91101

Dear Mr. Hokanson:

We are pleased to present the enclosed bioassay report. The test was conducted under guidelines prescribed in *Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms EPA-821-R-02-012*. "All acceptability criteria were met and the concentration-response was normal. This is a valid test." Results were as follows:

CLIENT:	Trussell Technologies
SAMPLE ID.:	Weymouth WTP Infl. with EarthTec QZ
DATE RECEIVED:	1 July – 21
ABC LAB NO.:	TRU0721.006

ACUTE FATHEAD MINNOW SURVIVAL BIOASSAY

% Survival = 97.50% Survival in 16.7 ul/l Sample

EC50 = >16.7 ul/l

Yours very truly,

Scott Johnson
Laboratory Director

CETIS Summary Report

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	02-4273-4213	**Test Type:**	Survival (96h)	**Analyst:**			
Start Date:	07 Jul-21 14:50	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Receiving Water		
Ending Date:	11 Jul-21 14:10	**Species:**	Pimephales promelas	**Brine:**	Not Applicable		
Test Length:	95h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO	**Age:**	<24
Sample ID:	20-8538-1072	**Code:**	TRU0721.006afml	**Project:**			
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report		
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Weymouth WTP Infl. with Earth Tec Q		
Sample Age:	6d 3h (12 °C)	**Client:**	Trussell Technologies				

Multiple Comparison Summary

Analysis ID	Endpoint	Comparison Method	✓	NOEL	LOEL	TOEL	PMSD	S
15-1204-8978	96h Survival Rate	Steel Many-One Rank Sum Test		16.7	>16.7	---	6.22%	1

Point Estimate Summary

Analysis ID	Endpoint	Point Estimate Method	✓	Level	µL/L	95% LCL	95% UCL	S
04-0318-1598	96h Survival Rate	Linear Interpolation (ICPIN)		EC10	>16.7	---	---	1
				EC15	>16.7	---	---	
				EC20	>16.7	---	---	
				EC25	>16.7	---	---	
				EC40	>16.7	---	---	
				EC50	>16.7	---	---	

Test Acceptability

				TAC Limits			
Analysis ID	Endpoint	Attribute	Test Stat	Lower	Upper	Overlap	Decision
04-0318-1598	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria
15-1204-8978	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria

96h Survival Rate Summary

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	D	4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	---	0.00%
3		4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	---	0.00%
8.35		4	0.9750	0.8954	1.0550	0.9000	1.0000	0.0250	0.0500	5.13%	2.50%
16.7		4	0.9750	0.8954	1.0550	0.9000	1.0000	0.0250	0.0500	5.13%	2.50%

96h Survival Rate Detail — MD5: 766BA0F6A592363228F6858710ABB13B

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	1.0000	1.0000	1.0000	1.0000
3		1.0000	1.0000	1.0000	1.0000
8.35		1.0000	1.0000	0.9000	1.0000
16.7		1.0000	0.9000	1.0000	1.0000

96h Survival Rate Binomials

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	10/10	10/10	10/10	10/10
3		10/10	10/10	10/10	10/10
8.35		10/10	10/10	9/10	10/10
16.7		10/10	9/10	10/10	10/10

CETIS Analytical Report

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	15-1204-8978	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	20 Jul-21 15:08	**Analysis:**	Nonparametric-Control vs Treatments	**Status Level:**	1
Edit Date:	20 Jul-21 14:49	**MD5 Hash:**	766BA0F6A592363228F6858710ABB13B	**Editor ID:**	000-189-126-0
Batch ID:	02-4273-4213	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	07 Jul-21 14:50	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Receiving Water
Ending Date:	11 Jul-21 14:10	**Species:**	Pimephales promelas	**Brine:**	Not Applicable
Test Length:	95h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO **Age:** <24
Sample ID:	20-8538-1072	**Code:**	TRU0721.006afml	**Project:**	
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Weymouth WTP Infl. with Earth Tec Q
Sample Age:	6d 3h (12 °C)	**Client:**	Trussell Technologies		

Data Transform	Alt Hyp	NOEL	LOEL	TOEL	TU	MSDu	PMSD
Angular (Corrected)	C > T	16.7	>16.7	---	---	0.06216	6.22%

Steel Many-One Rank Sum Test

Control	vs	Conc-µL/L	Test Stat	Critical	Ties	DF	P-Type	P-Value	Decision(α:5%)
Dilution Water		3	18	10	1	6	CDF	0.7500	Non-Significant Effect
		8.35	16	10	1	6	CDF	0.5065	Non-Significant Effect
		16.7	16	10	1	6	CDF	0.5065	Non-Significant Effect

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.0066398	0.0022133	3	0.6667	0.5885	Non-Significant Effect
Error	0.039839	0.0033199	12			
Total	0.0464788		15			

ANOVA Assumptions Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variance	Bartlett Equality of Variance Test				Indeterminate
	Levene Equality of Variance Test	6	5.953	0.0097	Unequal Variances
	Mod Levene Equality of Variance Test	0.6667	5.953	0.5885	Equal Variances
Distribution	Anderson-Darling A2 Test	2.447	3.878	<1.0E-05	Non-Normal Distribution
	D'Agostino Skewness Test	2.906	2.576	0.0037	Non-Normal Distribution
	Kolmogorov-Smirnov D Test	0.375	0.2471	<1.0E-05	Non-Normal Distribution
	Shapiro-Wilk W Normality Test	0.677	0.8408	9.5E-05	Non-Normal Distribution

96h Survival Rate Summary

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	D	4	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%
3		4	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%
8.35		4	0.9750	0.8954	1.0000	1.0000	0.9000	1.0000	0.0250	5.13%	2.50%
16.7		4	0.9750	0.8954	1.0000	1.0000	0.9000	1.0000	0.0250	5.13%	2.50%

Angular (Corrected) Transformed Summary

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	D	4	1.4120	1.4120	1.4120	1.4120	1.4120	1.4120	0.0000	0.00%	0.00%
3		4	1.4120	1.4120	1.4120	1.4120	1.4120	1.4120	0.0000	0.00%	0.00%
8.35		4	1.3710	1.2420	1.5010	1.4120	1.2490	1.4120	0.0407	5.94%	2.89%
16.7		4	1.3710	1.2420	1.5010	1.4120	1.2490	1.4120	0.0407	5.94%	2.89%

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	15-1204-8978	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	20 Jul-21 15:08	**Analysis:**	Nonparametric-Control vs Treatments	**Status Level:**	1
Edit Date:	20 Jul-21 14:49	**MD5 Hash:**	766BA0F6A592363228F6858710ABB13B	**Editor ID:**	000-189-126-0

96h Survival Rate Detail

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	1.0000	1.0000	1.0000	1.0000
3		1.0000	1.0000	1.0000	1.0000
8.35		1.0000	1.0000	0.9000	1.0000
16.7		1.0000	0.9000	1.0000	1.0000

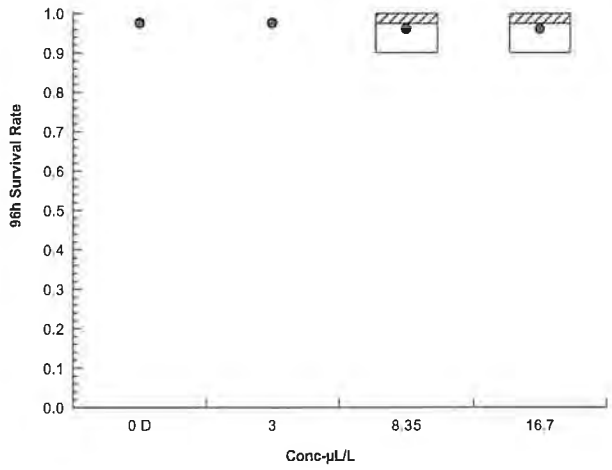
Angular (Corrected) Transformed Detail

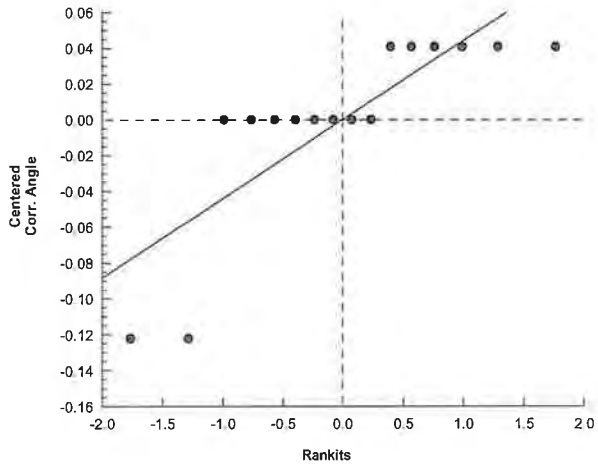
Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	1.4120	1.4120	1.4120	1.4120
3		1.4120	1.4120	1.4120	1.4120
8.35		1.4120	1.4120	1.2490	1.4120
16.7		1.4120	1.2490	1.4120	1.4120

96h Survival Rate Binomials

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	10/10	10/10	10/10	10/10
3		10/10	10/10	10/10	10/10
8.35		10/10	10/10	9/10	10/10
16.7		10/10	9/10	10/10	10/10

Graphics





CETIS Analytical Report

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	04-0318-1598	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	20 Jul-21 15:08	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1
Edit Date:	20 Jul-21 14:49	**MD5 Hash:**	766BA0F6A592363228F6858710ABB13B	**Editor ID:**	000-189-126-0
Batch ID:	02-4273-4213	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	07 Jul-21 14:50	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Receiving Water
Ending Date:	11 Jul-21 14:10	**Species:**	Pimephales promelas	**Brine:**	Not Applicable
Test Length:	95h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO **Age:** <24
Sample ID:	20-8538-1072	**Code:**	TRU0721.006afml	**Project:**	
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Weymouth WTP Infl. with Earth Tec Q
Sample Age:	6d 3h (12 °C)	**Client:**	Trussell Technologies		

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	0	280	Yes	Two-Point Interpolation

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

Point Estimates

Level	µL/L	95% LCL	95% UCL
EC10	>16.7	---	---
EC15	>16.7	---	---
EC20	>16.7	---	---
EC25	>16.7	---	---
EC40	>16.7	---	---
EC50	>16.7	---	---

96h Survival Rate Summary

			Calculated Variate(A/B)							Isotonic Variate	
Conc-µL/L	**Code**	**Count**	**Mean**	**Median**	**Min**	**Max**	**CV%**	**%Effect**	**A/B**	**Mean**	**%Effect**
0	D	4	1.0000	1.0000	1.0000	1.0000	0.00%	0.00%	40/40	1.0000	0.00%
3		4	1.0000	1.0000	1.0000	1.0000	0.00%	0.00%	40/40	1.0000	0.00%
8.35		4	0.9750	1.0000	0.9000	1.0000	5.13%	2.50%	39/40	0.9750	2.50%
16.7		4	0.9750	1.0000	0.9000	1.0000	5.13%	2.50%	39/40	0.9750	2.50%

96h Survival Rate Detail

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	1.0000	1.0000	1.0000	1.0000
3		1.0000	1.0000	1.0000	1.0000
8.35		1.0000	1.0000	0.9000	1.0000
16.7		1.0000	0.9000	1.0000	1.0000

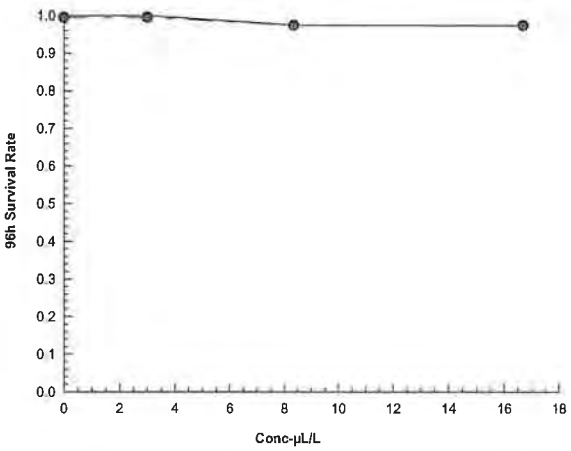
96h Survival Rate Binomials

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	10/10	10/10	10/10	10/10
3		10/10	10/10	10/10	10/10
8.35		10/10	10/10	9/10	10/10
16.7		10/10	9/10	10/10	10/10

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	04-0318-1598	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	20 Jul-21 15:08	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1
Edit Date:	20 Jul-21 14:49	**MD5 Hash:**	766BA0F6A592363228F6858710ABB13B	**Editor ID:**	000-189-126-0

Graphics



Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	02-4273-4213	**Test Type:**	Survival (96h)	**Analyst:**			
Start Date:	07 Jul-21 14:50	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Receiving Water		
Ending Date:	11 Jul-21 14:10	**Species:**	Pimephales promelas	**Brine:**	Not Applicable		
Test Length:	95h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO	**Age:**	<24

Sample ID:	20-8538-1072	**Code:**	TRU0721.006afml	**Project:**			
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report		
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Weymouth WTP Infl. with Earth Tec Q		
Sample Age:	6d 3h (12 °C)	**Client:**	Trussell Technologies				

Alkalinity (CaCO3)-mg/L

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	120	120	120	120	120	0	0	0.00%	0
16.7		3	125	125	125	125	125	0	0	0.00%	0
Overall		6	122.5	119.6	125.4	120	125	1.118	2.739	2.24%	0 (0%)

Conductivity-µmhos

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	1044	1036	1052	1042	1048	1.072	3.215	0.31%	0
3		3	1013	1004	1022	1010	1017	1.202	3.606	0.36%	0
8.35		3	1013	1009	1017	1012	1015	0.5774	1.732	0.17%	0
16.7		3	1023	1010	1036	1019	1029	1.764	5.292	0.52%	0
Overall		12	1023	1015	1032	1010	1048	3.963	13.73	1.34%	0 (0%)

Dissolved Oxygen-mg/L

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	7.833	6.659	9.007	7.3	8.2	0.1575	0.4726	6.03%	0
3		3	8	6.917	9.083	7.5	8.3	0.1453	0.4359	5.45%	0
8.35		3	7.867	6.832	8.901	7.4	8.2	0.1388	0.4163	5.29%	0
16.7		3	7.867	6.832	8.901	7.4	8.2	0.1388	0.4163	5.29%	0
Overall		12	7.892	7.652	8.132	7.3	8.3	0.109	0.3777	4.79%	0 (0%)

Hardness (CaCO3)-mg/L

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	250	250	250	250	250	0	0	0.00%	0
16.7		3	330	330	330	330	330	0	0	0.00%	0
Overall		6	290	244	336	250	330	17.89	43.82	15.11%	0 (0%)

pH-Units

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	7.533	6.96	8.107	7.4	7.8	0.07698	0.2309	3.07%	0
3		3	7.533	7.154	7.913	7.4	7.7	0.05092	0.1528	2.03%	0
8.35		3	7.533	7.39	7.677	7.5	7.6	0.01924	0.05773	0.77%	0
16.7		3	7.567	7.423	7.71	7.5	7.6	0.01924	0.05773	0.76%	0
Overall		12	7.542	7.463	7.62	7.4	7.8	0.0358	0.124	1.64%	0 (0%)

Temperature-°C

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	24.07	23.78	24.35	24	24.2	0.03847	0.1154	0.48%	0
3		3	24.07	23.92	24.21	24	24.1	0.01918	0.05755	0.24%	0
8.35		3	24.1	23.85	24.35	24	24.2	0.0333	0.09989	0.41%	0
16.7		3	24.13	23.75	24.51	24	24.3	0.05092	0.1528	0.63%	0
Overall		12	24.09	24.03	24.15	24	24.3	0.02876	0.09962	0.41%	0 (0%)

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Alkalinity (CaCO3)-mg/L

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		120					
16.7				125					
0	D	2		120					
16.7				125					
0	D	3		120					
16.7				125					

Conductivity-µmhos

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		1043					
3				1017					
8.35				1012					
16.7				1029					
0	D	2		1042					
3				1010					
8.35				1012					
16.7				1019					
0	D	3		1048					
3				1012					
8.35				1015					
16.7				1021					

Dissolved Oxygen-mg/L

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		8.2					
3				8.3					
8.35				8					
16.7				8					
0	D	2		8					
3				8.2					
8.35				8.2					
16.7				8.2					
0	D	3		7.3					
3				7.5					
8.35				7.4					
16.7				7.4					

Hardness (CaCO3)-mg/L

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		250					
16.7				330					
0	D	2		250					
16.7				330					
0	D	3		250					
16.7				330					

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

pH-Units

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		7.4					
3				7.4					
8.35				7.5					
16.7				7.6					
0	D	2		7.4					
3				7.5					
8.35				7.5					
16.7				7.6					
0	D	3		7.8					
3				7.7					
8.35				7.6					
16.7				7.5					

Temperature-°C

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		24					
3				24.1					
8.35				24.1					
16.7				24.1					
0	D	2		24.2					
3				24.1					
8.35				24.2					
16.7				24.3					
0	D	3		24					
3				24					
8.35				24					
16.7				24					

aquatic bioassay & consulting laboratories, inc

July 21, 2021

Mr. David Hokanson
Trussell Technologies
232 N. Lake Avenue
Pasadena, CA 91101

Dear Mr. Hokanson:

We are pleased to present the enclosed bioassay report. The test was conducted under guidelines prescribed in *Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms EPA-821-R-02-012.* "All acceptability criteria were met and the concentration-response was normal. This is a valid test." Results were as follows:

CLIENT:	Trussell Technologies
SAMPLE ID.:	Weymouth WTP Infl. with EarthTec QZ
DATE RECEIVED:	1 July – 21
ABC LAB NO.:	TRU0721.006

ACUTE CERIODAPHNIA SURVIVAL BIOASSAY

% Survival = 0 % Survival in 3.0 ul/l Sample

EC50 = 1.5 ul/l

Yours very truly,

Scott Johnson
Laboratory Director

CETIS Summary Report

Test Code/ID: TRU0721.006acer / 01-9970-4374

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	11-4125-0527	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	07 Jul-21 14:50	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Receiving Water
Ending Date:	11 Jul-21 14:10	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable
Test Length:	95h	**Taxon:**	Branchiopoda	**Source:**	Aquatic Biosystems, CO **Age:** <24
Sample ID:	09-4593-1718	**Code:**	TRU0721.006acer	**Project:**	
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Weymouth WTP Infl. with Earth Tec Q
Sample Age:	6d 3h (12 °C)	**Client:**	Trussell Technologies		

Point Estimate Summary

Analysis ID	Endpoint	Point Estimate Method	✓	Level	µL/L	95% LCL	95% UCL	S
01-5908-9405	96h Survival Rate	Linear Interpolation (ICPIN)		EC10	0.3	0.3	0.3	1
				EC15	0.45	0.45	0.45	
				EC20	0.6	0.6	0.6	
				EC25	0.75	0.75	0.75	
				EC40	1.2	1.2	1.2	
				EC50	1.5	1.5	1.5	

Test Acceptability

				TAC Limits			
Analysis ID	**Endpoint**	**Attribute**	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
01-5908-9405	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria
12-5132-4588	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria

96h Survival Rate Summary

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	D	4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	---	0.00%
3		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	---	100.00%
8.35		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	---	100.00%
16.7		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	---	100.00%

96h Survival Rate Detail — MD5: 03CCEF1D141244C83E77C1C00B25A1DE

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	1.0000	1.0000	1.0000	1.0000
3		0.0000	0.0000	0.0000	0.0000
8.35		0.0000	0.0000	0.0000	0.0000
16.7		0.0000	0.0000	0.0000	0.0000

96h Survival Rate Binomials

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	5/5	5/5	5/5	5/5
3		0/5	0/5	0/5	0/5
8.35		0/5	0/5	0/5	0/5
16.7		0/5	0/5	0/5	0/5

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	12-5132-4588	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	20 Jul-21 15:10	**Analysis:**	Parametric-Two Sample	**Status Level:**	1
Edit Date:	20 Jul-21 14:52	**MD5 Hash:**	03CCEF1D141244C83E77C1C00B25A1DE	**Editor ID:**	000-189-126-0
Batch ID:	11-4125-0527	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	07 Jul-21 14:50	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Receiving Water
Ending Date:	11 Jul-21 14:10	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable
Test Length:	95h	**Taxon:**	Branchiopoda	**Source:**	Aquatic Biosystems, CO **Age:** <24
Sample ID:	09-4593-1718	**Code:**	TRU0721.006acer	**Project:**	
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Weymouth WTP Infl. with Earth Tec Q
Sample Age:	6d 3h (12 °C)	**Client:**	Trussell Technologies		

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

ANOVA Assumptions Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Distribution	Shapiro-Wilk W Normality Test				Indeterminate

96h Survival Rate Summary

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	D	4	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%
3		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	---	100.00%
8.35		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	---	100.00%
16.7		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	---	100.00%

Angular (Corrected) Transformed Summary

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	D	4	1.3450	1.3450	1.3460	1.3450	1.3450	1.3450	0.0000	0.00%	0.00%
3		4	0.2255	0.2255	0.2256	0.2255	0.2255	0.2255	0.0000	0.00%	83.24%
8.35		4	0.2255	0.2255	0.2256	0.2255	0.2255	0.2255	0.0000	0.00%	83.24%
16.7		4	0.2255	0.2255	0.2256	0.2255	0.2255	0.2255	0.0000	0.00%	83.24%

96h Survival Rate Detail

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	1.0000	1.0000	1.0000	1.0000
3		0.0000	0.0000	0.0000	0.0000
8.35		0.0000	0.0000	0.0000	0.0000
16.7		0.0000	0.0000	0.0000	0.0000

Angular (Corrected) Transformed Detail

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	1.3450	1.3450	1.3450	1.3450
3		0.2255	0.2255	0.2255	0.2255
8.35		0.2255	0.2255	0.2255	0.2255
16.7		0.2255	0.2255	0.2255	0.2255

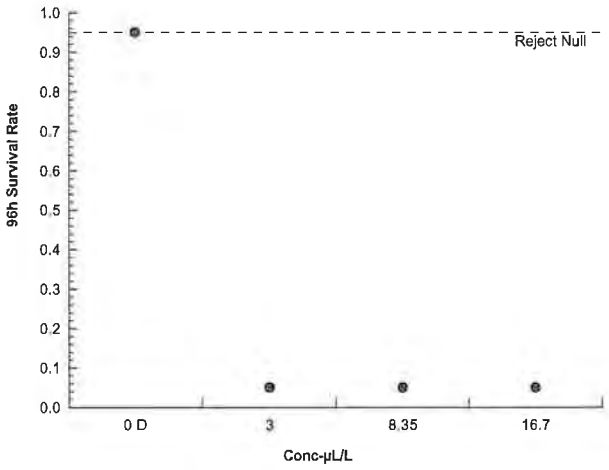
96h Survival Rate Binomials

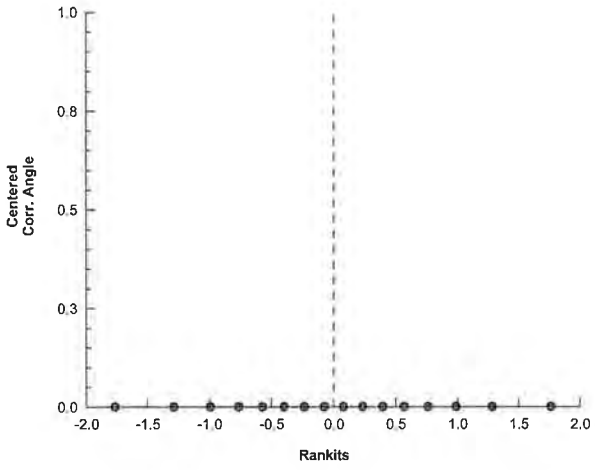
Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	5/5	5/5	5/5	5/5
3		0/5	0/5	0/5	0/5
8.35		0/5	0/5	0/5	0/5
16.7		0/5	0/5	0/5	0/5

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	12-5132-4588	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	20 Jul-21 15:10	**Analysis:**	Parametric-Two Sample	**Status Level:**	1
Edit Date:	20 Jul-21 14:52	**MD5 Hash:**	03CCEF1D141244C83E77C1C00B25A1DE	**Editor ID:**	000-189-126-0

Graphics





CETIS Analytical Report

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	01-5908-9405	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	20 Jul-21 15:10	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1
Edit Date:	20 Jul-21 14:52	**MD5 Hash:**	03CCEF1D141244C83E77C1C00B25A1DE	**Editor ID:**	000-189-126-0
Batch ID:	11-4125-0527	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	07 Jul-21 14:50	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Receiving Water
Ending Date:	11 Jul-21 14:10	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable
Test Length:	95h	**Taxon:**	Branchiopoda	**Source:**	Aquatic Biosystems, CO **Age:** <24
Sample ID:	09-4593-1718	**Code:**	TRU0721.006acer	**Project:**	
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Weymouth WTP Infl. with Earth Tec Q
Sample Age:	6d 3h (12 °C)	**Client:**	Trussell Technologies		

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	0	280	Yes	Two-Point Interpolation

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

Point Estimates

Level	µL/L	95% LCL	95% UCL
EC10	0.3	0.3	0.3
EC15	0.45	0.45	0.45
EC20	0.6	0.6	0.6
EC25	0.75	0.75	0.75
EC40	1.2	1.2	1.2
EC50	1.5	1.5	1.5

96h Survival Rate Summary

			Calculated Variate(A/B)							Isotonic Variate	
Conc-µL/L	**Code**	**Count**	**Mean**	**Median**	**Min**	**Max**	**CV%**	**%Effect**	**A/B**	**Mean**	**%Effect**
0	D	4	1.0000	1.0000	1.0000	1.0000	0.00%	0.00%	20/20	1.0000	0.00%
3		4	0.0000	0.0000	0.0000	0.0000	---	100.00%	0/20	0.0000	100.00%
8.35		4	0.0000	0.0000	0.0000	0.0000	---	100.00%	0/20	0.0000	100.00%
16.7		4	0.0000	0.0000	0.0000	0.0000	---	100.00%	0/20	0.0000	100.00%

96h Survival Rate Detail

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	1.0000	1.0000	1.0000	1.0000
3		0.0000	0.0000	0.0000	0.0000
8.35		0.0000	0.0000	0.0000	0.0000
16.7		0.0000	0.0000	0.0000	0.0000

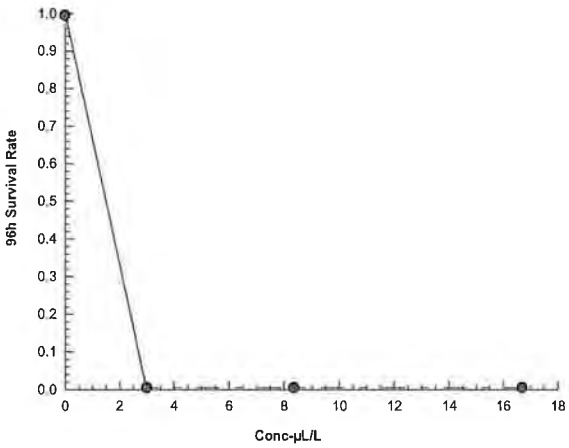
96h Survival Rate Binomials

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	5/5	5/5	5/5	5/5
3		0/5	0/5	0/5	0/5
8.35		0/5	0/5	0/5	0/5
16.7		0/5	0/5	0/5	0/5

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	01-5908-9405	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	20 Jul-21 15:10	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1
Edit Date:	20 Jul-21 14:52	**MD5 Hash:**	03CCEF1D141244C83E77C1C00B25A1DE	**Editor ID:**	000-189-126-0

Graphics



Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	11-4125-0527	**Test Type:**	Survival (96h)	**Analyst:**			
Start Date:	07 Jul-21 14:50	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Receiving Water		
Ending Date:	11 Jul-21 14:10	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable		
Test Length:	95h	**Taxon:**	Branchiopoda	**Source:**	Aquatic Biosystems, CO	**Age:**	<24
Sample ID:	09-4593-1718	**Code:**	TRU0721.006acer	**Project:**			
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report		
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Weymouth WTP Infl. with Earth Tec Q		
Sample Age:	6d 3h (12 °C)	**Client:**	Trussell Technologies				

Alkalinity (CaCO3)-mg/L

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	120	120	120	120	120	0	0	0.00%	0
16.7		3	125	125	125	125	125	0	0	0.00%	0
Overall		6	122.5	119.6	125.4	120	125	1.118	2.739	2.24%	0 (0%)

Conductivity-µmhos

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	1044	1036	1052	1042	1048	1.072	3.215	0.31%	0
3		3	1013	1004	1022	1010	1017	1.202	3.606	0.36%	0
8.35		3	1013	1009	1017	1012	1015	0.5774	1.732	0.17%	0
16.7		3	1023	1010	1036	1019	1029	1.764	5.292	0.52%	0
Overall		12	1023	1015	1032	1010	1048	3.963	13.73	1.34%	0 (0%)

Dissolved Oxygen-mg/L

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	7.833	6.659	9.007	7.3	8.2	0.1575	0.4726	6.03%	0
3		3	8	6.917	9.083	7.5	8.3	0.1453	0.4359	5.45%	0
8.35		3	7.867	6.832	8.901	7.4	8.2	0.1388	0.4163	5.29%	0
16.7		3	7.867	6.832	8.901	7.4	8.2	0.1388	0.4163	5.29%	0
Overall		12	7.892	7.652	8.132	7.3	8.3	0.109	0.3777	4.79%	0 (0%)

Hardness (CaCO3)-mg/L

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	250	250	250	250	250	0	0	0.00%	0
16.7		3	330	330	330	330	330	0	0	0.00%	0
Overall		6	290	244	336	250	330	17.89	43.82	15.11%	0 (0%)

pH-Units

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	7.533	6.96	8.107	7.4	7.8	0.07698	0.2309	3.07%	0
3		3	7.533	7.154	7.913	7.4	7.7	0.05092	0.1528	2.03%	0
8.35		3	7.533	7.39	7.677	7.5	7.6	0.01924	0.05773	0.77%	0
16.7		3	7.567	7.423	7.71	7.5	7.6	0.01924	0.05773	0.76%	0
Overall		12	7.542	7.463	7.62	7.4	7.8	0.0358	0.124	1.64%	0 (0%)

Temperature-°C

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	24.07	23.78	24.35	24	24.2	0.03847	0.1154	0.48%	0
3		3	24.07	23.92	24.21	24	24.1	0.01918	0.05755	0.24%	0
8.35		3	24.1	23.85	24.35	24	24.2	0.0333	0.09989	0.41%	0
16.7		3	24.13	23.75	24.51	24	24.3	0.05092	0.1528	0.63%	0
Overall		12	24.09	24.03	24.15	24	24.3	0.02876	0.09962	0.41%	0 (0%)

Alkalinity (CaCO3)-mg/L

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		120					
16.7				125					
0	D	2		120					
16.7				125					
0	D	3		120					
16.7				125					

Conductivity-µmhos

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		1043					
3				1017					
8.35				1012					
16.7				1029					
0	D	2		1042					
3				1010					
8.35				1012					
16.7				1019					
0	D	3		1048					
3				1012					
8.35				1015					
16.7				1021					

Dissolved Oxygen-mg/L

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		8.2					
3				8.3					
8.35				8					
16.7				8					
0	D	2		8					
3				8.2					
8.35				8.2					
16.7				8.2					
0	D	3		7.3					
3				7.5					
8.35				7.4					
16.7				7.4					

Hardness (CaCO3)-mg/L

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		250					
16.7				330					
0	D	2		250					
16.7				330					
0	D	3		250					
16.7				330					

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

pH-Units

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		7.4					
3				7.4					
8.35				7.5					
16.7				7.6					
0	D	2		7.4					
3				7.5					
8.35				7.5					
16.7				7.6					
0	D	3		7.8					
3				7.7					
8.35				7.6					
16.7				7.5					

Temperature-°C

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		24					
3				24.1					
8.35				24.1					
16.7				24.1					
0	D	2		24.2					
3				24.1					
8.35				24.2					
16.7				24.3					
0	D	3		24					
3				24					
8.35				24					
16.7				24					

Appendix G –

Lake Mathews 2021 Toxicity Test Report

aquatic bioassay & consulting laboratories, inc

September 8, 2021

Mr. David Hokanson
Trussell Technologies
232 N. Lake Avenue
Pasadena, CA 91101

Dear Mr. Hokanson:

We are pleased to present the enclosed bioassay report. The test was conducted under guidelines prescribed in *Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms EPA-821-R-02-012*. "All acceptability criteria were met and the concentration-response was normal. This is a valid test." Results were as follows:

CLIENT:	Trussell Technologies
SAMPLE ID.:	Lake Mathews with EarthTec QZ
DATE RECEIVED:	1 July – 21
ABC LAB NO.:	TRU0721.007

RAINBOW TROUT SURVIVAL BIOASSAY

% Survival = CON = 100.00%
3.0uL/L = 100.00%
8.35uL/L = 0.00%
16.7uL/L = 0.00%

EC50 = 5.675 ul/l

Yours very truly,

Scott Johnson
Laboratory Director

CETIS Summary Report

Fish 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	15-2797-9235	**Test Type:**	Survival (96h)	**Analyst:**	Joe Freas		
Start Date:	07 Jul-21 16:01	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water		
Ending Date:	11 Jul-21 14:01	**Species:**	Oncorhynchus mykiss	**Brine:**	Not Applicable		
Test Length:	94h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO	**Age:**	
Sample ID:	01-7395-3681	**Code:**	TRU0721.007	**Project:**			
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report		
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Lake Mathews		
Sample Age:	6d 5h	**Client:**	Trussell Technologies				

SIngle Comparison Summary

Analysis ID	Endpoint	Comparison Method	P-Value	Comparison Result	S
14-8891-5003	96h Survival Rate	Steel Many-One Rank Sum Test	0.5000	3µL/L passed 96h survival rate	1

Point Estimate Summary

Analysis ID	Endpoint	Point Estimate Method	✓	Level	µL/L	95% LCL	95% UCL	S
04-3346-7794	96h Survival Rate	Linear Interpolation (ICPIN)		EC10	3.535	3.535	3.535	1
				EC15	3.803	3.803	3.803	
				EC20	4.07	4.07	4.07	
				EC25	4.338	4.338	4.338	
				EC40	5.14	5.14	5.14	
				EC50	5.675	5.675	5.675	

Test Acceptability

				TAC Limits			
Analysis ID	Endpoint	Attribute	Test Stat	Lower	Upper	Overlap	Decision
04-3346-7794	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria
14-8891-5003	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria

96h Survival Rate Summary

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	N	4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	---	0.00%
3		4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	---	0.00%
8.35		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	---	100.00%
16.7		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	---	100.00%

96h Survival Rate Detail

MD5: 3A0D5E0FE40D419F8BDE03F5112603FC

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
3		1.0000	1.0000	1.0000	1.0000
8.35		0.0000	0.0000	0.0000	0.0000
16.7		0.0000	0.0000	0.0000	0.0000

96h Survival Rate Binomials

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	10/10	10/10	10/10	10/10
3		10/10	10/10	10/10	10/10
8.35		0/10	0/10	0/10	0/10
16.7		0/10	0/10	0/10	0/10

CETIS Analytical Report

Fish 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	14-8891-5003	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	08 Sep-21 13:30	**Analysis:**	Nonparametric-Control vs Treatments	**Status Level:**	1
Edit Date:	08 Sep-21 13:26	**MD5 Hash:**	3A0D5E0FE40D419F8BDE03F5112603FC	**Editor ID:**	007-979-628-1
Batch ID:	15-2797-9235	**Test Type:**	Survival (96h)	**Analyst:**	Joe Freas
Start Date:	07 Jul-21 16:01	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water
Ending Date:	11 Jul-21 14:01	**Species:**	Oncorhynchus mykiss	**Brine:**	Not Applicable
Test Length:	94h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO **Age:**
Sample ID:	01-7395-3681	**Code:**	TRU0721.007	**Project:**	
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Lake Mathews
Sample Age:	6d 5h	**Client:**	Trussell Technologies		

Data Transform	Alt Hyp	Comparison Result
Angular (Corrected)	C > T	3µL/L passed 96h survival rate endpoint

Steel Many-One Rank Sum Test

Control	vs	Conc-µL/L	Test Stat	Critical	Ties	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		3	18	12	1	6	CDF	0.5000	Non-Significant Effect

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0	0	1			Indeterminate
Error	0	0	6			
Total	0		7			

ANOVA Assumptions Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variance	Variance Ratio F Test				Indeterminate
Distribution	Shapiro-Wilk W Normality Test				Indeterminate

96h Survival Rate Summary

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%
3		4	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%
8.35		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	---	100.00%
16.7		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	---	100.00%

Angular (Corrected) Transformed Summary

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	4	1.4120	1.4120	1.4120	1.4120	1.4120	1.4120	0.0000	0.00%	0.00%
3		4	1.4120	1.4120	1.4120	1.4120	1.4120	1.4120	0.0000	0.00%	0.00%
8.35		4	0.1588	0.1588	0.1588	0.1588	0.1588	0.1588	0.0000	0.00%	88.76%
16.7		4	0.1588	0.1588	0.1588	0.1588	0.1588	0.1588	0.0000	0.00%	88.76%

96h Survival Rate Detail

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
3		1.0000	1.0000	1.0000	1.0000
8.35		0.0000	0.0000	0.0000	0.0000
16.7		0.0000	0.0000	0.0000	0.0000

Fish 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	14-8891-5003	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	08 Sep-21 13:30	**Analysis:**	Nonparametric-Control vs Treatments	**Status Level:**	1
Edit Date:	08 Sep-21 13:26	**MD5 Hash:**	3A0D5E0FE40D419F8BDE03F5112603FC	**Editor ID:**	007-979-628-1

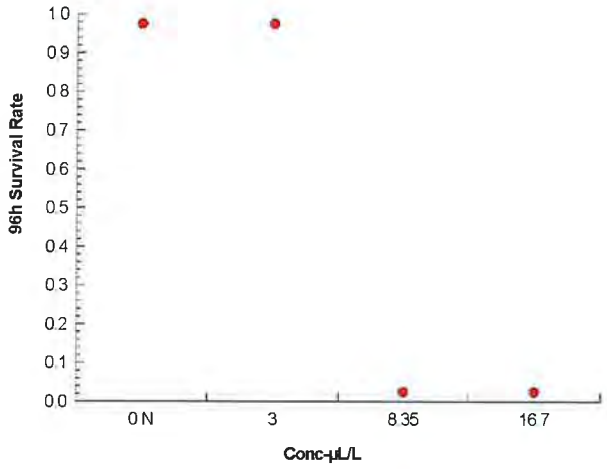
Angular (Corrected) Transformed Detail

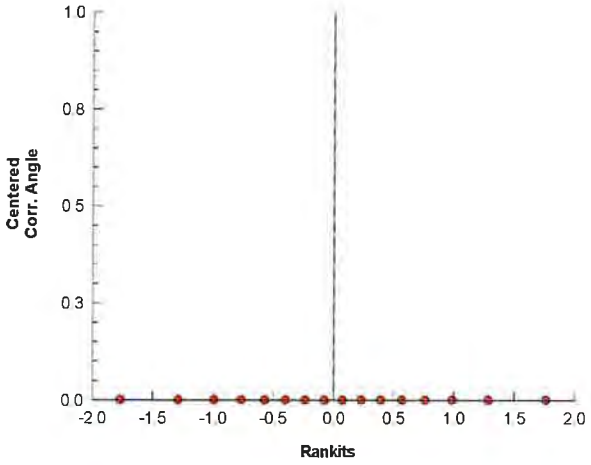
Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.4120	1.4120	1.4120	1.4120
3		1.4120	1.4120	1.4120	1.4120
8.35		0.1588	0.1588	0.1588	0.1588
16.7		0.1588	0.1588	0.1588	0.1588

96h Survival Rate Binomials

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	10/10	10/10	10/10	10/10
3		10/10	10/10	10/10	10/10
8.35		0/10	0/10	0/10	0/10
16.7		0/10	0/10	0/10	0/10

Graphics





CETIS Analytical Report

Fish 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	04-3346-7794	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	08 Sep-21 13:30	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1
Edit Date:	08 Sep-21 13:26	**MD5 Hash:**	3A0D5E0FE40D419F8BDE03F5112603FC	**Editor ID:**	007-979-628-1
Batch ID:	15-2797-9235	**Test Type:**	Survival (96h)	**Analyst:**	Joe Freas
Start Date:	07 Jul-21 16:01	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water
Ending Date:	11 Jul-21 14:01	**Species:**	Oncorhynchus mykiss	**Brine:**	Not Applicable
Test Length:	94h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO **Age:**
Sample ID:	01-7395-3681	**Code:**	TRU0721.007	**Project:**	
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Lake Mathews
Sample Age:	6d 5h	**Client:**	Trussell Technologies		

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	0	280	Yes	Two-Point Interpolation

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

Point Estimates

Level	µL/L	95% LCL	95% UCL
EC10	3.535	3.535	3.535
EC15	3.803	3.803	3.803
EC20	4.07	4.07	4.07
EC25	4.338	4.338	4.338
EC40	5.14	5.14	5.14
EC50	5.675	5.675	5.675

96h Survival Rate Summary

			Calculated Variate(A/B)							Isotonic Variate	
Conc-µL/L	**Code**	**Count**	**Mean**	**Median**	**Min**	**Max**	**CV%**	**%Effect**	**A/B**	**Mean**	**%Effect**
0	N	4	1.0000	1.0000	1.0000	1.0000	0.00%	0.00%	40/40	1.0000	0.00%
3		4	1.0000	1.0000	1.0000	1.0000	0.00%	0.00%	40/40	1.0000	0.00%
8.35		4	0.0000	0.0000	0.0000	0.0000	---	100.00%	0/40	0.0000	100.00%
16.7		4	0.0000	0.0000	0.0000	0.0000	---	100.00%	0/40	0.0000	100.00%

96h Survival Rate Detail

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	1.0000	1.0000	1.0000	1.0000
3		1.0000	1.0000	1.0000	1.0000
8.35		0.0000	0.0000	0.0000	0.0000
16.7		0.0000	0.0000	0.0000	0.0000

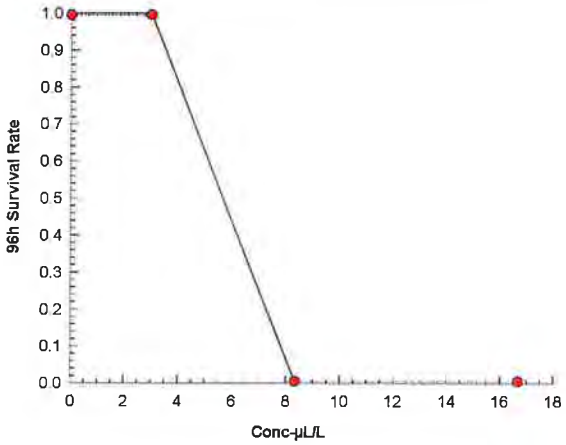
96h Survival Rate Binomials

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	N	10/10	10/10	10/10	10/10
3		10/10	10/10	10/10	10/10
8.35		0/10	0/10	0/10	0/10
16.7		0/10	0/10	0/10	0/10

Fish 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	04-3346-7794	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	08 Sep-21 13:30	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1
Edit Date:	08 Sep-21 13:26	**MD5 Hash:**	3A0D5E0FE40D419F8BDE03F5112603FC	**Editor ID:**	007-979-628-1

Graphics



CETIS Measurement Report

Fish 96-h Acute Survival Test

Aquatic Bioassay & Consulting Labs, Inc.

Batch ID:	15-2797-9235	**Test Type:**	Survival (96h)	**Analyst:**	Joe Freas		
Start Date:	07 Jul-21 16:01	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water		
Ending Date:	11 Jul-21 14:01	**Species:**	Oncorhynchus mykiss	**Brine:**	Not Applicable		
Test Length:	94h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO	**Age:**	
Sample ID:	01-7395-3681	**Code:**	TRU0721.007	**Project:**			
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report		
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Lake Mathews		
Sample Age:	6d 5h	**Client:**	Trussell Technologies				

Alkalinity (CaCO3)-mg/L

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	2	125.5	119.1	131.9	125	126	0.3536	0.7071	0.56%	0
3		2	133.5	127.1	139.9	133	134	0.3536	0.7071	0.53%	0
8.35		2	136.5	130.1	142.9	136	137	0.3536	0.7071	0.52%	0
16.7		2	129.5	123.1	135.9	129	130	0.3536	0.7071	0.55%	0
Overall		8	131.2	127.5	135	125	137	1.578	4.464	3.40%	0 (0%)

Dissolved Oxygen-mg/L

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	5	10.18	9.996	10.36	10	10.4	0.02966	0.1483	1.46%	0
3		5	10.04	9.57	10.51	9.4	10.4	0.07563	0.3782	3.77%	0
8.35		2	9.75	6.573	12.93	9.5	10	0.1768	0.3536	3.63%	0
16.7		2	9.7	5.888	13.51	9.4	10	0.2121	0.4243	4.37%	0
Overall		14	10	9.808	10.19	9.4	10.4	0.08895	0.3328	3.33%	0 (0%)

Hardness (CaCO3)-mg/L

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	2	125.5	119.1	131.9	125	126	0.3536	0.7071	0.56%	0
3		2	292.5	260.7	324.3	290	295	1.768	3.536	1.21%	0
8.35		2	327.5	295.7	359.3	325	330	1.768	3.536	1.08%	0
Overall		6	248.5	147.1	349.9	125	330	39.43	96.58	38.86%	0 (0%)

pH-Units

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	5	7.9	7.549	8.251	7.4	8.1	0.05657	0.2828	3.58%	0
3		5	7.86	7.536	8.184	7.4	8	0.05215	0.2608	3.32%	0
8.35		2	7.7	5.159	10.24	7.5	7.9	0.1414	0.2828	3.67%	0
16.7		2	7.65	4.473	10.83	7.4	7.9	0.1768	0.3536	4.62%	0
Overall		14	7.821	7.668	7.975	7.4	8.1	0.07124	0.2665	3.41%	0 (0%)

Temperature-°C

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	5	13.1	12.47	13.73	12.8	14	0.102	0.5099	3.89%	0
3		5	13.06	12.36	13.76	12.5	14	0.1128	0.5639	4.32%	0
8.35		2	13.3	5.676	20.92	12.7	13.9	0.4243	0.8485	6.38%	0
16.7		2	13.3	6.947	19.65	12.8	13.8	0.3536	0.7071	5.32%	0
Overall		14	13.14	12.84	13.45	12.5	14	0.1421	0.5316	4.05%	0 (0%)

aquatic bioassay & consulting laboratories, inc

July 21, 2021

Mr. David Hokanson
Trussell Technologies
232 N. Lake Avenue
Pasadena, CA 91101

Dear Mr. Hokanson:

We are pleased to present the enclosed bioassay report. The test was conducted under guidelines prescribed in *Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms EPA-821-R-02-012*. "All acceptability criteria were met and the concentration-response was normal. This is a valid test." Results were as follows:

CLIENT:	Trussell Technologies
SAMPLE ID.:	Lake Matthew Raw with EarthTec QZ
DATE RECEIVED:	1 July – 21
ABC LAB NO.:	TRU0721.007

ACUTE FATHEAD MINNOW SURVIVAL BIOASSAY

% Survival = 97.50% Survival in 16.7 ul/l Sample

EC50 = >16.7 ul/l

Yours very truly,

Scott Johnson
Laboratory Director

CETIS Summary Report

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	10-3180-3556	**Test Type:**	Survival (96h)	**Analyst:**			
Start Date:	07 Jul-21 15:01	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Receiving Water		
Ending Date:	11 Jul-21 14:15	**Species:**	Pimephales promelas	**Brine:**	Not Applicable		
Test Length:	95h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO	**Age:**	<24
Sample ID:	13-0172-2416	**Code:**	TRU0721.007afml	**Project:**			
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report		
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Lake Matthew Raw with Earth Tec QZ		
Sample Age:	6d 4h (11 °C)	**Client:**	Trussell Technologies				

Multiple Comparison Summary

Analysis ID	Endpoint	Comparison Method	✓	NOEL	LOEL	TOEL	PMSD	S
07-1275-9365	96h Survival Rate	Steel Many-One Rank Sum Test		16.7	>16.7	---	4.96%	1

Point Estimate Summary

Analysis ID	Endpoint	Point Estimate Method	✓	Level	µL/L	95% LCL	95% UCL	S
07-7480-8197	96h Survival Rate	Linear Interpolation (ICPIN)		EC10	>16.7	---	---	1
				EC15	>16.7	---	---	
				EC20	>16.7	---	---	
				EC25	>16.7	---	---	
				EC40	>16.7	---	---	
				EC50	>16.7	---	---	

Test Acceptability

				TAC Limits			
Analysis ID	**Endpoint**	**Attribute**	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
07-1275-9365	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria
07-7480-8197	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria

96h Survival Rate Summary

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	D	4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	---	0.00%
3		4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	---	0.00%
8.35		4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	---	0.00%
16.7		4	0.9750	0.8954	1.0550	0.9000	1.0000	0.0250	0.0500	5.13%	2.50%

96h Survival Rate Detail — MD5: 64CD5B1D74951B1BFA48CE8575DEE9E9

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	1.0000	1.0000	1.0000	1.0000
3		1.0000	1.0000	1.0000	1.0000
8.35		1.0000	1.0000	1.0000	1.0000
16.7		0.9000	1.0000	1.0000	1.0000

96h Survival Rate Binomials

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	10/10	10/10	10/10	10/10
3		10/10	10/10	10/10	10/10
8.35		10/10	10/10	10/10	10/10
16.7		9/10	10/10	10/10	10/10

CETIS Analytical Report

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	07-1275-9365	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	20 Jul-21 15:15	**Analysis:**	Nonparametric-Control vs Treatments	**Status Level:**	1
Edit Date:	20 Jul-21 15:12	**MD5 Hash:**	64CD5B1D74951B1BFA48CE8575DEE9E9	**Editor ID:**	000-189-126-0
Batch ID:	10-3180-3556	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	07 Jul-21 15:01	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Receiving Water
Ending Date:	11 Jul-21 14:15	**Species:**	Pimephales promelas	**Brine:**	Not Applicable
Test Length:	95h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO **Age:** <24
Sample ID:	13-0172-2416	**Code:**	TRU0721.007afml	**Project:**	
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Lake Matthew Raw with Earth Tec QZ
Sample Age:	6d 4h (11 °C)	**Client:**	Trussell Technologies		

Data Transform	Alt Hyp	NOEL	LOEL	TOEL	TU	MSDu	PMSD
Angular (Corrected)	C > T	16.7	>16.7	---	---	0.04964	4.96%

Steel Many-One Rank Sum Test

Control	vs	Conc-µL/L	Test Stat	Critical	Ties	DF	P-Type	P-Value	Decision(α:5%)
Dilution Water		3	18	10	1	6	CDF	0.7500	Non-Significant Effect
		8.35	18	10	1	6	CDF	0.7500	Non-Significant Effect
		16.7	16	10	1	6	CDF	0.5065	Non-Significant Effect

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.0049799	0.00166	3	1	0.4262	Non-Significant Effect
Error	0.0199195	0.00166	12			
Total	0.0248994		15			

ANOVA Assumptions Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variance	Bartlett Equality of Variance Test				Indeterminate
	Levene Equality of Variance Test	9	5.953	0.0021	Unequal Variances
	Mod Levene Equality of Variance Test	1	5.953	0.4262	Equal Variances
Distribution	Anderson-Darling A2 Test	3.496	3.878	<1.0E-05	Non-Normal Distribution
	D'Agostino Skewness Test	3.733	2.576	0.0002	Non-Normal Distribution
	Kolmogorov-Smirnov D Test	0.4375	0.2471	<1.0E-05	Non-Normal Distribution
	Shapiro-Wilk W Normality Test	0.5647	0.8408	<1.0E-05	Non-Normal Distribution

96h Survival Rate Summary

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	D	4	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%
3		4	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%
8.35		4	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%
16.7		4	0.9750	0.8954	1.0000	1.0000	0.9000	1.0000	0.0250	5.13%	2.50%

Angular (Corrected) Transformed Summary

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	D	4	1.4120	1.4120	1.4120	1.4120	1.4120	1.4120	0.0000	0.00%	0.00%
3		4	1.4120	1.4120	1.4120	1.4120	1.4120	1.4120	0.0000	0.00%	0.00%
8.35		4	1.4120	1.4120	1.4120	1.4120	1.4120	1.4120	0.0000	0.00%	0.00%
16.7		4	1.3710	1.2420	1.5010	1.4120	1.2490	1.4120	0.0407	5.94%	2.89%

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	07-1275-9365	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	20 Jul-21 15:15	**Analysis:**	Nonparametric-Control vs Treatments	**Status Level:**	1
Edit Date:	20 Jul-21 15:12	**MD5 Hash:**	64CD5B1D74951B1BFA48CE8575DEE9E9	**Editor ID:**	000-189-126-0

96h Survival Rate Detail

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	1.0000	1.0000	1.0000	1.0000
3		1.0000	1.0000	1.0000	1.0000
8.35		1.0000	1.0000	1.0000	1.0000
16.7		0.9000	1.0000	1.0000	1.0000

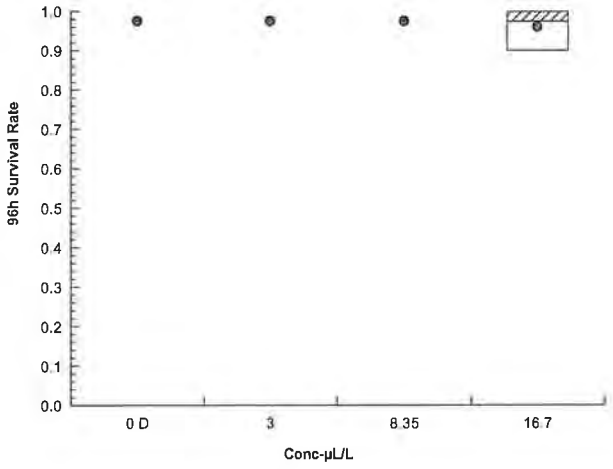
Angular (Corrected) Transformed Detail

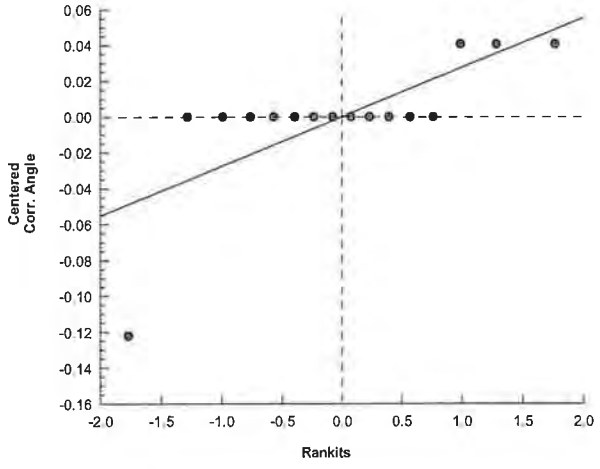
Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	1.4120	1.4120	1.4120	1.4120
3		1.4120	1.4120	1.4120	1.4120
8.35		1.4120	1.4120	1.4120	1.4120
16.7		1.2490	1.4120	1.4120	1.4120

96h Survival Rate Binomials

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	10/10	10/10	10/10	10/10
3		10/10	10/10	10/10	10/10
8.35		10/10	10/10	10/10	10/10
16.7		9/10	10/10	10/10	10/10

Graphics





Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	07-7480-8197	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	20 Jul-21 15:15	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1
Edit Date:	20 Jul-21 15:12	**MD5 Hash:**	64CD5B1D74951B1BFA48CE8575DEE9E9	**Editor ID:**	000-189-126-0
Batch ID:	10-3180-3556	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	07 Jul-21 15:01	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Receiving Water
Ending Date:	11 Jul-21 14:15	**Species:**	Pimephales promelas	**Brine:**	Not Applicable
Test Length:	95h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO **Age:** <24
Sample ID:	13-0172-2416	**Code:**	TRU0721.007afml	**Project:**	
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Lake Matthew Raw with Earth Tec QZ
Sample Age:	6d 4h (11 °C)	**Client:**	Trussell Technologies		

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	0	280	Yes	Two-Point Interpolation

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

Point Estimates

Level	µL/L	95% LCL	95% UCL
EC10	>16.7	---	---
EC15	>16.7	---	---
EC20	>16.7	---	---
EC25	>16.7	---	---
EC40	>16.7	---	---
EC50	>16.7	---	---

96h Survival Rate Summary

			Calculated Variate(A/B)							Isotonic Variate	
Conc-µL/L	**Code**	**Count**	**Mean**	**Median**	**Min**	**Max**	**CV%**	**%Effect**	**A/B**	**Mean**	**%Effect**
0	D	4	1.0000	1.0000	1.0000	1.0000	0.00%	0.00%	40/40	1.0000	0.00%
3		4	1.0000	1.0000	1.0000	1.0000	0.00%	0.00%	40/40	1.0000	0.00%
8.35		4	1.0000	1.0000	1.0000	1.0000	0.00%	0.00%	40/40	1.0000	0.00%
16.7		4	0.9750	1.0000	0.9000	1.0000	5.13%	2.50%	39/40	0.9750	2.50%

96h Survival Rate Detail

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	1.0000	1.0000	1.0000	1.0000
3		1.0000	1.0000	1.0000	1.0000
8.35		1.0000	1.0000	1.0000	1.0000
16.7		0.9000	1.0000	1.0000	1.0000

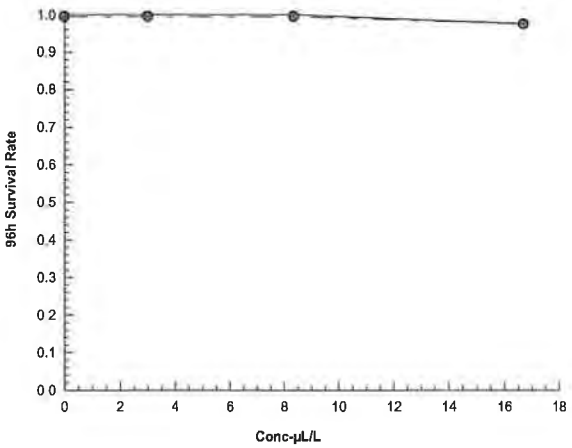
96h Survival Rate Binomials

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	10/10	10/10	10/10	10/10
3		10/10	10/10	10/10	10/10
8.35		10/10	10/10	10/10	10/10
16.7		9/10	10/10	10/10	10/10

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	07-7480-8197	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	20 Jul-21 15:15	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1
Edit Date:	20 Jul-21 15:12	**MD5 Hash:**	64CD5B1D74951B1BFA48CE8575DEE9E9	**Editor ID:**	000-189-126-0

Graphics



Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	10-3180-3556	**Test Type:**	Survival (96h)	**Analyst:**			
Start Date:	07 Jul-21 15:01	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Receiving Water		
Ending Date:	11 Jul-21 14:15	**Species:**	Pimephales promelas	**Brine:**	Not Applicable		
Test Length:	95h	**Taxon:**	Actinopterygii	**Source:**	Aquatic Biosystems, CO	**Age:**	<24
Sample ID:	13-0172-2416	**Code:**	TRU0721.007afml	**Project:**			
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report		
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Lake Matthew Raw with Earth Tec QZ		
Sample Age:	6d 4h (11 °C)	**Client:**	Trussell Technologies				

Alkalinity (CaCO3)-mg/L

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	123	123	123	123	123	0	0	0.00%	0
16.7		3	129	129	129	129	129	0	0	0.00%	0
Overall		6	126	122.6	129.4	123	129	1.342	3.286	2.61%	0 (0%)

Conductivity-µmhos

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	1009	1002	1016	1007	1012	0.8819	2.646	0.26%	0
3		3	1011	1008	1014	1010	1012	0.3849	1.155	0.11%	0
8.35		3	1005	996	1014	1001	1008	1.202	3.606	0.36%	0
16.7		3	1012	1011	1014	1012	1013	0.1925	0.5774	0.06%	0
Overall		12	1009	1007	1011	1001	1013	1.001	3.467	0.34%	0 (0%)

Dissolved Oxygen-mg/L

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	7.9	6.586	9.214	7.3	8.3	0.1764	0.5291	6.70%	0
3		3	7.933	6.929	8.937	7.5	8.3	0.1347	0.4041	5.09%	0
8.35		3	7.9	6.817	8.983	7.4	8.2	0.1453	0.4359	5.52%	0
16.7		3	7.867	6.832	8.901	7.4	8.2	0.1388	0.4163	5.29%	0
Overall		12	7.9	7.656	8.144	7.3	8.3	0.1108	0.3838	4.86%	0 (0%)

Hardness (CaCO3)-mg/L

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	295	295	295	295	295	0	0	0.00%	0
16.7		3	330	330	330	330	330	0	0	0.00%	0
Overall		6	312.5	292.4	332.6	295	330	7.826	19.17	6.13%	0 (0%)

pH-Units

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	7.667	7.38	7.954	7.6	7.8	0.03849	0.1155	1.51%	0
3		3	7.7	7.452	7.948	7.6	7.8	0.03333	0.1	1.30%	0
8.35		3	7.633	7.49	7.777	7.6	7.7	0.01924	0.05773	0.76%	0
16.7		3	7.633	7.49	7.777	7.6	7.7	0.01924	0.05773	0.76%	0
Overall		12	7.658	7.608	7.709	7.6	7.8	0.02289	0.0793	1.04%	0 (0%)

Temperature-°C

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	24.1	23.85	24.35	24	24.2	0.0333	0.09989	0.41%	0
3		3	24.1	23.85	24.35	24	24.2	0.0333	0.09989	0.41%	0
8.35		3	24.1	23.85	24.35	24	24.2	0.0333	0.09989	0.41%	0
16.7		3	24.1	23.85	24.35	24	24.2	0.0333	0.09989	0.41%	0
Overall		12	24.1	24.05	24.15	24	24.2	0.02462	0.08528	0.35%	0 (0%)

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Alkalinity (CaCO3)-mg/L

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		123					
16.7				129					
0	D	2		123					
16.7				129					
0	D	3		123					
16.7				129					

Conductivity-µmhos

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		1012					
3				1010					
8.35				1008					
16.7				1012					
0	D	2		1007					
3				1010					
8.35				1006					
16.7				1012					
0	D	3		1008					
3				1012					
8.35				1001					
16.7				1013					

Dissolved Oxygen-mg/L

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		8.3					
3				8.3					
8.35				8.2					
16.7				8.2					
0	D	2		8.1					
3				8					
8.35				8.1					
16.7				8					
0	D	3		7.3					
3				7.5					
8.35				7.4					
16.7				7.4					

Hardness (CaCO3)-mg/L

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		295					
16.7				330					
0	D	2		295					
16.7				330					
0	D	3		295					
16.7				330					

Fathead Minnow 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

pH-Units

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		7.6					
3				7.6					
8.35				7.6					
16.7				7.6					
0	D	2		7.6					
3				7.7					
8.35				7.6					
16.7				7.6					
0	D	3		7.8					
3				7.8					
8.35				7.7					
16.7				7.7					

Temperature-°C

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		24.2					
3				24.2					
8.35				24.2					
16.7				24.2					
0	D	2		24.1					
3				24.1					
8.35				24.1					
16.7				24.1					
0	D	3		24					
3				24					
8.35				24					
16.7				24					

aquatic bioassay & consulting laboratories, inc

July 21, 2021

Mr. David Hokanson
Trussell Technologies
232 N. Lake Avenue
Pasadena, CA 91101

Dear Mr. Hokanson:

We are pleased to present the enclosed bioassay report. The test was conducted under guidelines prescribed in *Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms EPA-821-R-02-012*. "All acceptability criteria were met and the concentration-response was normal. This is a valid test." Results were as follows:

CLIENT:	Trussell Technologies
SAMPLE ID.:	Lake Matthew Raw with EarthTec QZ
DATE RECEIVED:	1 July – 21
ABC LAB NO.:	TRU0721.007

ACUTE CERIODAPHNIA SURVIVAL BIOASSAY

% Survival = 15 % Survival in 3.0 ul/l Sample

EC50 = 1.765 ul/l

Yours very truly,

Scott Johnson
Laboratory Director

CETIS Summary Report

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	00-9683-5858	**Test Type:**	Survival (96h)	**Analyst:**			
Start Date:	07 Jul-21 15:01	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water		
Ending Date:	11 Jul-21 14:15	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable		
Test Length:	95h	**Taxon:**	Branchiopoda	**Source:**	Aquatic Biosystems, CO	**Age:**	
Sample ID:	16-7326-1851	**Code:**	TRU0721.007acer	**Project:**			
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report		
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Lake Matthew Raw with Earth Tec QZ		
Sample Age:	6d 4h (11 °C)	**Client:**	Trussell Technologies				

SIngle Comparison Summary

Analysis ID	Endpoint	Comparison Method	P-Value	Comparison Result	S
12-0540-3137	96h Survival Rate	Steel Many-One Rank Sum Test	0.0105	3µL/L failed 96h survival rate	1

Point Estimate Summary

Analysis ID	Endpoint	Point Estimate Method	✓	Level	µL/L	95% LCL	95% UCL	S
00-7855-5265	96h Survival Rate	Linear Interpolation (ICPIN)		EC10	0.3529	0.2682	0.4739	1
				EC15	0.5294	0.4024	0.7109	
				EC20	0.7059	0.5365	0.9479	
				EC25	0.8824	0.6706	1.185	
				EC40	1.412	1.073	1.896	
				EC50	1.765	1.341	2.37	

Test Acceptability

				TAC Limits			
Analysis ID	**Endpoint**	**Attribute**	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
00-7855-5265	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria
12-0540-3137	96h Survival Rate	Control Resp	1	0.9	>>	Yes	Passes Criteria

96h Survival Rate Summary

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	D	4	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	---	0.00%
3		4	0.1500	-0.1547	0.4547	0.0000	0.4000	0.0957	0.1915	127.66%	85.00%
8.35		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	---	100.00%
16.7		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	---	100.00%

96h Survival Rate Detail — MD5: 1FDA7366E4C0792CC7290D2E2907677A

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	1.0000	1.0000	1.0000	1.0000
3		0.4000	0.0000	0.0000	0.2000
8.35		0.0000	0.0000	0.0000	0.0000
16.7		0.0000	0.0000	0.0000	0.0000

96h Survival Rate Binomials

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	5/5	5/5	5/5	5/5
3		2/5	0/5	0/5	1/5
8.35		0/5	0/5	0/5	0/5
16.7		0/5	0/5	0/5	0/5

CETIS Analytical Report

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	12-0540-3137	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	20 Jul-21 15:16	**Analysis:**	Nonparametric-Control vs Treatments	**Status Level:**	1
Edit Date:	20 Jul-21 15:16	**MD5 Hash:**	1FDA7366E4C0792CC7290D2E2907677A	**Editor ID:**	000-189-126-0
Batch ID:	00-9683-5858	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	07 Jul-21 15:01	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water
Ending Date:	11 Jul-21 14:15	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable
Test Length:	95h	**Taxon:**	Branchiopoda	**Source:**	Aquatic Biosystems, CO **Age:**
Sample ID:	16-7326-1851	**Code:**	TRU0721.007acer	**Project:**	
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Lake Matthew Raw with Earth Tec QZ
Sample Age:	6d 4h (11 °C)	**Client:**	Trussell Technologies		

Data Transform	Alt Hyp	Comparison Result	PMSD
Angular (Corrected)	C > T	3µL/L failed 96h survival rate endpoint	18.13%

Steel Many-One Rank Sum Test

Control	vs	Conc-µL/L	Test Stat	Critical	Ties	DF	P-Type	P-Value	Decision(α:5%)
Dilution Water		3*	10	12	0	6	CDF	0.0105	Significant Effect

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	1.78769	1.78769	1	73.46	0.0001	Significant Effect
Error	0.146007	0.0243345	6			
Total	1.9337		7			

ANOVA Assumptions Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variance	Levene Equality of Variance Test	14.93	13.75	0.0083	Unequal Variances
	Mod Levene Equality of Variance Test	9.95	13.75	0.0197	Equal Variances
	Variance Ratio F Test				Indeterminate
Distribution	Anderson-Darling A2 Test	0.7003	3.878	0.0674	Normal Distribution
	Kolmogorov-Smirnov D Test	0.25	0.3313	0.1599	Normal Distribution
	Shapiro-Wilk W Normality Test	0.8543	0.6451	0.1052	Normal Distribution

96h Survival Rate Summary

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	D	4	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.00%	0.00%
3		4	0.1500	0.0000	0.4547	0.1000	0.0000	0.4000	0.0957	127.66%	85.00%
8.35		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	---	100.00%
16.7		4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	---	100.00%

Angular (Corrected) Transformed Summary

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	D	4	1.3450	1.3450	1.3460	1.3450	1.3450	1.3450	0.0000	0.00%	0.00%
3		4	0.3998	0.0488	0.7509	0.3446	0.2255	0.6847	0.1103	55.17%	70.28%
8.35		4	0.2255	0.2255	0.2256	0.2255	0.2255	0.2255	0.0000	0.00%	83.24%
16.7		4	0.2255	0.2255	0.2256	0.2255	0.2255	0.2255	0.0000	0.00%	83.24%

96h Survival Rate Detail

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	1.0000	1.0000	1.0000	1.0000
3		0.4000	0.0000	0.0000	0.2000
8.35		0.0000	0.0000	0.0000	0.0000
16.7		0.0000	0.0000	0.0000	0.0000

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	12-0540-3137	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	20 Jul-21 15:16	**Analysis:**	Nonparametric-Control vs Treatments	**Status Level:**	1
Edit Date:	20 Jul-21 15:16	**MD5 Hash:**	1FDA7366E4C0792CC7290D2E2907677A	**Editor ID:**	000-189-126-0

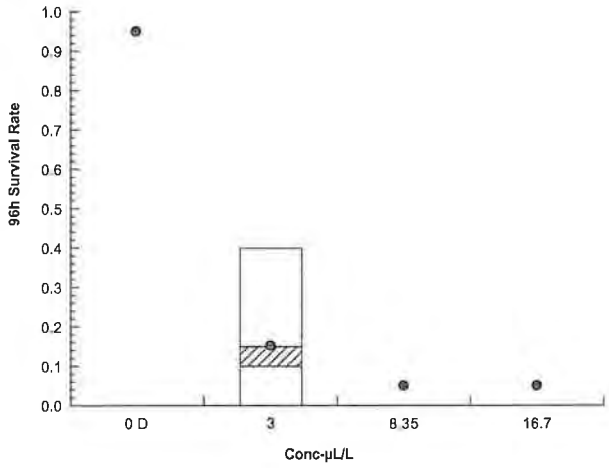
Angular (Corrected) Transformed Detail

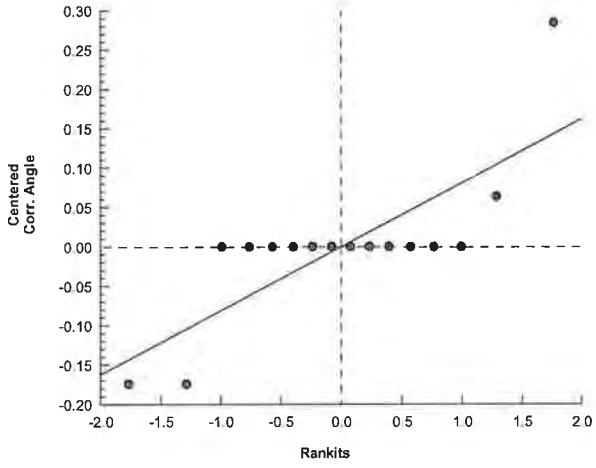
Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	1.3450	1.3450	1.3450	1.3450
3		0.6847	0.2255	0.2255	0.4636
8.35		0.2255	0.2255	0.2255	0.2255
16.7		0.2255	0.2255	0.2255	0.2255

96h Survival Rate Binomials

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	5/5	5/5	5/5	5/5
3		2/5	0/5	0/5	1/5
8.35		0/5	0/5	0/5	0/5
16.7		0/5	0/5	0/5	0/5

Graphics





CETIS Analytical Report

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	00-7855-5265	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	20 Jul-21 15:16	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1
Edit Date:	20 Jul-21 15:16	**MD5 Hash:**	1FDA7366E4C0792CC7290D2E2907677A	**Editor ID:**	000-189-126-0
Batch ID:	00-9683-5858	**Test Type:**	Survival (96h)	**Analyst:**	
Start Date:	07 Jul-21 15:01	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water
Ending Date:	11 Jul-21 14:15	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable
Test Length:	95h	**Taxon:**	Branchiopoda	**Source:**	Aquatic Biosystems, CO **Age:**
Sample ID:	16-7326-1851	**Code:**	TRU0721.007acer	**Project:**	
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Lake Matthew Raw with Earth Tec QZ
Sample Age:	6d 4h (11 °C)	**Client:**	Trussell Technologies		

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	0	280	Yes	Two-Point Interpolation

Test Acceptability Criteria

		TAC Limits			
Attribute	**Test Stat**	**Lower**	**Upper**	**Overlap**	**Decision**
Control Resp	1	0.9	>>	Yes	Passes Criteria

Point Estimates

Level	µL/L	95% LCL	95% UCL
EC10	0.3529	0.2682	0.4739
EC15	0.5294	0.4024	0.7109
EC20	0.7059	0.5365	0.9479
EC25	0.8824	0.6706	1.185
EC40	1.412	1.073	1.896
EC50	1.765	1.341	2.37

96h Survival Rate Summary

			Calculated Variate(A/B)							Isotonic Variate	
Conc-µL/L	**Code**	**Count**	**Mean**	**Median**	**Min**	**Max**	**CV%**	**%Effect**	**A/B**	**Mean**	**%Effect**
0	D	4	1.0000	1.0000	1.0000	1.0000	0.00%	0.00%	20/20	1.0000	0.00%
3		4	0.1500	0.1000	0.0000	0.4000	127.66%	85.00%	3/20	0.1500	85.00%
8.35		4	0.0000	0.0000	0.0000	0.0000	---	100.00%	0/20	0.0000	100.00%
16.7		4	0.0000	0.0000	0.0000	0.0000	---	100.00%	0/20	0.0000	100.00%

96h Survival Rate Detail

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	1.0000	1.0000	1.0000	1.0000
3		0.4000	0.0000	0.0000	0.2000
8.35		0.0000	0.0000	0.0000	0.0000
16.7		0.0000	0.0000	0.0000	0.0000

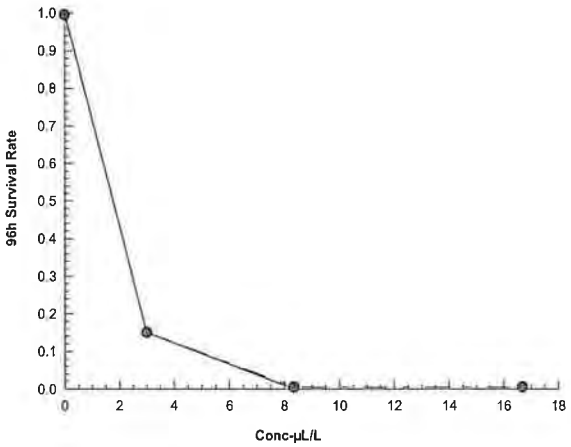
96h Survival Rate Binomials

Conc-µL/L	Code	Rep 1	Rep 2	Rep 3	Rep 4
0	D	5/5	5/5	5/5	5/5
3		2/5	0/5	0/5	1/5
8.35		0/5	0/5	0/5	0/5
16.7		0/5	0/5	0/5	0/5

Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Analysis ID:	00-7855-5265	**Endpoint:**	96h Survival Rate	**CETIS Version:**	CETISv1.9.7
Analyzed:	20 Jul-21 15:16	**Analysis:**	Linear Interpolation (ICPIN)	**Status Level:**	1
Edit Date:	20 Jul-21 15:16	**MD5 Hash:**	1FDA7366E4C0792CC7290D2E2907677A	**Editor ID:**	000-189-126-0

Graphics



Ceriodaphnia 96-h Acute Survival Test — **Aquatic Bioassay & Consulting Labs, Inc.**

Batch ID:	00-9683-5858	**Test Type:**	Survival (96h)	**Analyst:**			
Start Date:	07 Jul-21 15:01	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Laboratory Water		
Ending Date:	11 Jul-21 14:15	**Species:**	Ceriodaphnia dubia	**Brine:**	Not Applicable		
Test Length:	95h	**Taxon:**	Branchiopoda	**Source:**	Aquatic Biosystems, CO	**Age:**	
Sample ID:	16-7326-1851	**Code:**	TRU0721.007acer	**Project:**			
Sample Date:	01 Jul-21 11:30	**Material:**	Sample Water	**Source:**	Bioassay Report		
Receipt Date:	01 Jul-21 11:30	**CAS (PC):**		**Station:**	Lake Matthew Raw with Earth Tec QZ		
Sample Age:	6d 4h (11 °C)	**Client:**	Trussell Technologies				

Alkalinity (CaCO3)-mg/L

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	123	123	123	123	123	0	0	0.00%	0
16.7		3	129	129	129	129	129	0	0	0.00%	0
Overall		6	126	122.6	129.4	123	129	1.342	3.286	2.61%	0 (0%)

Conductivity-µmhos

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	1009	1002	1016	1007	1012	0.8819	2.646	0.26%	0
3		3	1011	1008	1014	1010	1012	0.3849	1.155	0.11%	0
8.35		3	1005	996	1014	1001	1008	1.202	3.606	0.36%	0
16.7		3	1012	1011	1014	1012	1013	0.1925	0.5774	0.06%	0
Overall		12	1009	1007	1011	1001	1013	1.001	3.467	0.34%	0 (0%)

Dissolved Oxygen-mg/L

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	7.9	6.586	9.214	7.3	8.3	0.1764	0.5291	6.70%	0
3		3	7.933	6.929	8.937	7.5	8.3	0.1347	0.4041	5.09%	0
8.35		3	7.9	6.817	8.983	7.4	8.2	0.1453	0.4359	5.52%	0
16.7		3	7.867	6.832	8.901	7.4	8.2	0.1388	0.4163	5.29%	0
Overall		12	7.9	7.656	8.144	7.3	8.3	0.1108	0.3838	4.86%	0 (0%)

Hardness (CaCO3)-mg/L

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	295	295	295	295	295	0	0	0.00%	0
16.7		3	330	330	330	330	330	0	0	0.00%	0
Overall		6	312.5	292.4	332.6	295	330	7.826	19.17	6.13%	0 (0%)

pH-Units

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	7.667	7.38	7.954	7.6	7.8	0.03849	0.1155	1.51%	0
3		3	7.7	7.452	7.948	7.6	7.8	0.03333	0.1	1.30%	0
8.35		3	7.633	7.49	7.777	7.6	7.7	0.01924	0.05773	0.76%	0
16.7		3	7.633	7.49	7.777	7.6	7.7	0.01924	0.05773	0.76%	0
Overall		12	7.658	7.608	7.709	7.6	7.8	0.02289	0.0793	1.04%	0 (0%)

Temperature-°C

Conc-µL/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	D	3	24.1	23.85	24.35	24	24.2	0.0333	0.09989	0.41%	0
3		3	24.1	23.85	24.35	24	24.2	0.0333	0.09989	0.41%	0
8.35		3	24.1	23.85	24.35	24	24.2	0.0333	0.09989	0.41%	0
16.7		3	24.1	23.85	24.35	24	24.2	0.0333	0.09989	0.41%	0
Overall		12	24.1	24.05	24.15	24	24.2	0.02462	0.08528	0.35%	0 (0%)

Ceriodaphnia 96-h Acute Survival Test **Aquatic Bioassay & Consulting Labs, Inc.**

Alkalinity (CaCO3)-mg/L

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		123					
16.7				129					
0	D	2		123					
16.7				129					
0	D	3		123					
16.7				129					

Conductivity-µmhos

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		1012					
3				1010					
8.35				1008					
16.7				1012					
0	D	2		1007					
3				1010					
8.35				1006					
16.7				1012					
0	D	3		1008					
3				1012					
8.35				1001					
16.7				1013					

Dissolved Oxygen-mg/L

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		8.3					
3				8.3					
8.35				8.2					
16.7				8.2					
0	D	2		8.1					
3				8					
8.35				8.1					
16.7				8					
0	D	3		7.3					
3				7.5					
8.35				7.4					
16.7				7.4					

Hardness (CaCO3)-mg/L

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		295					
16.7				330					
0	D	2		295					
16.7				330					
0	D	3		295					
16.7				330					

pH-Units

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		7.6					
3				7.6					
8.35				7.6					
16.7				7.6					
0	D	2		7.6					
3				7.7					
8.35				7.6					
16.7				7.6					
0	D	3		7.8					
3				7.8					
8.35				7.7					
16.7				7.7					

Temperature-°C

Conc-µL/L	Code	Read	Time	Measure	QA	Diff-%	Inst ID	Analyst	Notes
0	D	1		24.2					
3				24.2					
8.35				24.2					
16.7				24.2					
0	D	2		24.1					
3				24.1					
8.35				24.1					
16.7				24.1					
0	D	3		24					
3				24					
8.35				24					
16.7				24					

CHAIN OF CUSTODY RECORD

Client:	Project Name/Number:
Trussell Technologies	Andrew Rudchenko (626) 390-3638
Address: 232 N. Lake Ave Pasadena, CA 91101	Project Mgr.: David Hokanson
	P.O. #:
Phone Number: (626) 390 - 3638	Sampled By (signature):

Date	Time	Comp.	Grab	Matrix	Sample ID	Volume/ Number	Acute Toxicity Fathead minnow	Acute Toxicity/ Rainbow Trout	Acute Toxicity Cerodaphnia									Comments
7/1/21	11:30		✓	Liquid	Weymouth WTP Infl.	15 gal	3	3	3									, 006
7/1/21	11:30		✓	Liquid	Lake Matthews Raw	15 gal	3	3	3									, 007
					EarthTec QZ	~250 mL												
																		WEYMOUTH - LAKE MATTHEWS
										Temp. deg. C =								12.0°C - 11.0°C
										Chlorine (mg/L) =								<0.1 - <0.1
										NH3 (mg/L) =								<0.1 - <0.1

Relinquished By: (signature)	Date: Time:	Relinquished By: (signature)	Date: Time:
[signature]	7/1/2021 11:30		
Received By: (signature)	Date: Time:	Received By: (signature)	Date: Time:
Victor Magy	7/1/21 1130		
		Temp. upon sample receipt: _____ °C	

Aquatic Bioassay and Consulting Laboratories
29 N. Olive Street Ventura, CA 93001 Phone: (805) 643-5621 Fax: (805) 643-2930

SAFETY DATA SHEET
According to OSHA Hazard Communication Standard 29 CFR 1910.1200 (GHS)

EARTH SCIENCE LABORATORIES, INC.
113 SE 22nd Street, Suite 105
Rogers, AR 72756
earthsciencelabs.com

Emergency Phone Number: 1-800-535-5053 (Infotrac)
Information Phone Number: 1-800-962-1492

Material Name: *EarthTec QZ™*

Issue Date: 01/14
Revision Date: 01/2020

Section 1 –IDENTIFICATION

Product Name: EarthTec QZ™

EPA Reg. No. 64962-1

Manufactured by: Earth Science Laboratories, Inc.
903 N 47th St., Suite 105
Rogers, AR 72756

Certified to: NSF/ANSI Standard 60. Do not exceed 19 mg/L.

Section 2 –HAZARDS IDENTIFICATION

NFPA Health = 2, Fire = 0, Reactivity = 1
HMIS III: H2///F0/PH1

GHS Signal Word:
Warning
GHS Hazard Pictograms:

GHS Classifications:
Physical, Corrosive to Metals, 1
Health, Acute Toxicity, 4 Oral
Health, Harmful if absorbed through skin. Avoid contact with skin, 2
Health, Causes substantial but temporary eye injury. Do not get in eyes, 2
Environmental, Hazards to the aquatic environment - Chronic, 1

GHS Phrases:
H290 – May be corrosive to metal
H302 – Harmful if swallowed
H315 – Causes skin irritation
H319 – Causes serious eye irritation
H410 - Very toxic to aquatic life with long lasting effects

GHS Precautionary Statements:
P102 – Keep out of reach of children.
P233 – Keep container tightly closed.
P262 – Do not get in eyes, on skin or clothing.
P264 – Wash skin thoroughly after handling.
P270 – Do not eat, drink or smoke when handling this product.
P280 – Wear protective gloves/protective clothing/eye protection/face protection.
P281 – Use personal protective equipment as required.
P301+310 – IF SWALLOWED: Immediately call a POISON CENTER or doctor/physician.
P301+330+331 – IF SWALLOWED: Rinse mouth. DO NOT induce vomiting.
P303+361+353 – IF ON SKIN (or hair): Remove/Take off immediately all contaminated clothing. Rinse skin with water/shower.
P304+340 – IF INHALED: Remove victim to fresh air and keep at rest in a position comfortable for breathing.
P305+351+338 – IF IN EYES: Rinse continuously with water for several minutes. Remove contact lenses if present and easy to do. Continue rinsing.
P403+233 – Store in a well ventilated place. Keep container tightly closed
P501 – Dispose of contents /container according to State and Federal laws.

Section 3 –COMPOSITION/INFORMATION ON INGREDIENT

Components	CAS#	OSHA PEL	ACGIH TLV	%
Copper sulfate pentahydrate	7758-99-8	1mg/m^3	1mg/m^3	18.25-21.75%

Section 4 – FIRST AID MEASURES

If in Eyes: Hold eye open and rinse slowly and gently with water for 20 minutes. Remove contact lenses, if present, after first 5 minutes, then continue rinsing eye. Call a poison control center or doctor for advice.
If on Skin or Clothing: Take off contaminated clothing. Rinse skin immediately with plenty of soap and water for 15 to 20 minutes. Call a poison control center or doctor for treatment.
If Swallowed: Call a poison control center or doctor immediately for treatment advice. Have a person sip a glass of water if able to swallow. Do not induce vomiting unless told to do so by a poison control center or doctor. Do not give anything to an unconscious person.
Note to Physician: Probable mucosal damage may contraindicate the use of gastric lavage.
Have the product container or label with you when calling a poison control center or doctor or going for treatment. You may also contact INFOTRAC 1-800-535-5053 for emergency treatment.

Section 5 – FIRE FIGHTING MEASURES

Flash Point: N/E **UFL:** N/E **LFL:** N/E

Hazardous Combustion Products: May react with high carbon metals to produce hydrogen gas, which can form an explosive mixture.
Fire Fighting Equipment/Instructions: Firefighters must wear MSHA/NIOSH approved positive pressure breathing apparatus (SCBA) with full face mask and full protective equipment.

NFPA Ratings:	**Fire:** 0	**Health:** 2	**Reactivity:** 1	**Other:** X
HMIS III Ratings:	**Fire:** 0	**Health:** 2	**Reactivity:** 1	**Personal Protection:** X

Section 6 – ACCIDENTAL RELEASE MEASURES

Containment Procedures: Flush with water into retaining area or container. Caution should be exercised regarding personal safety and exposure to released product.
Clean-Up Procedures: Neutralize solution with bicarbonate of soda.
Evacuation Procedures: Keep unnecessary people away; isolate hazard area and deny entry.
Special Instructions: Notify local authorities and the National Response Center, if required.

Section 7 – HANDLING AND STORAGE

Application and Handling Equipment: Application, handling or storage equipment MUST consist of fiberglass, PVC, polypropylene, viton, corrosion resistant plastics or stainless steel. Never use mild steel, nylon, brass or copper around product. Always rinse and clean equipment thoroughly each night with plenty of fresh, clean water.
Storage: Store in a safe place away from pets and keep out of the reach of children. Store away from excessive heat. Product will freeze. Always store product above 32 degrees F (Do Not Freeze). Freezing may cause product separation. Always keep container closed. Keep away from galvanized pipe, and any nylon storage or handling equipment.

Section 8 – EXPOSURE CONTROLS/PERSONAL PROTECTION

Personal Protective Equipment (PPE)
Mixers, loaders, applicators and other handlers must wear the following: long-sleeved shirt, long pants, shoes plus socks, chemical-resistant gloves made of any water proof material (Chemical Resistance Category A), and protective eyewear.

Follow manufacturer's instructions for cleaning/maintaining PPE. If no such instructions for washables exist, use detergent and hot water. Keep and wash PPE separately from other laundry. Discard clothing and other absorbent material that have been drenched or heavily contaminated with the product's concentrate. Do not reuse them.

Section 9 – PHYSICAL AND CHEMICAL PROPERTIES

Appearance: Clear blue liquid
Physical State: Liquid
pH: 0.2/0.3
Vapor Pressure: 0.1mm 68° F
Boiling Point: 220° F
Melting Point: N/A
Odor: Minimal odor
Vapor Density (Air=1): 1.0
Evaporation Rate: N/A
Solubility in Water: Complete
Specific Gravity (H_20=1): 1.188 +/- 0.05

Section 10 – STABILITY AND REACTIVITY

Chemical Stability: Stable.
Conditions to Avoid: Avoid mixing with strong bases and strong reducing agents.
Incompatibility: Incompatible with strong bases and strong reducing agents.
Hazardous Decomposition Products: Sulfur dioxide and sulfur trioxide may be produced with decomposition.
Hazardous Polymerization: Will not occur.

Section 11 - TOXICOLOGICAL INFORMATION

Acute Toxicity / Chronic Toxicity: Continued overexposure to this solution may cause systemic toxicity.
Carcinogenicity: N/A
Signs and Symptoms of Exposure: Overexposure may cause the following specific symptoms, depending on the concentration and duration of exposure: vomiting, shallow respiration and lung function changes.

Section 12 - ECOLOGICAL INFORMATION

Waters treated with this product may be hazardous to aquatic organisms.

Section 13 – DISPOSAL CONSIDERATIONS

Pesticide wastes are acutely hazardous. Improper disposal of excess product mixture or rinsate is a violation of federal law. If these wastes cannot be disposed of by use according to label instructions, contact your state pesticide or environmental control agency, or the hazardous waste representative at the nearest EPA regional office for guidance. In the event of spill, neutralize with limestone or baking soda before disposal.
May deteriorate concrete.

Section 14 – TRANSPORT INFORMATION

DOT Information
Proper Shipping Name: Corrosive liquid, acidic, inorganic, n.o.s., (contains cupric sulfate)
Hazard Class: 8
UN/NA #: UN3264
Packing Group: III

- *Packages that contain more than 5.1 US gallons are **RQ** (reportable quantity)*
- *Packages that contain less than 4.0 liters could be **ORM-D***
- *The proper shipping information is the responsibility of the shipper and this information is only guidelines.*
-

Section 15 - REGULATORY INFORMATION

This chemical is a pesticide product registered by the Environmental Protection Agency and is subject to certain labeling requirements under federal pesticide law. These requirements differ from the classification criteria and hazard information required for Safety Data Sheets, and for workplace labels of non-pesticide chemicals. Following is the hazard information as required on the pesticide label:

WARNING
Causes substantial but temporary eye injury.
Harmful if swallowed.
Harmful if absorbed through skin.

Section 16 - OTHER INFORMATION

Date of Last Revision: January 2020

The information set forth herein is furnished free of charge and is based on technical data that Earth Science Laboratories, Inc. believes to be reliable. It is intended for use by persons having technical skill and at their own discretion and risk. Since conditions of use are outside our control, we make no warranties, express or implied, and assume no liability in connection with any use of the information. Nothing herein is to be taken as a license to operate under or a recommendation to infringe any patents.