



**Whole Effluent Toxicity Test Report:
City of Everett**

May 2024

Report date: June 11, 2025

Submitted to:

City of Everett
3200 Cedar Street
Everett, WA 98201

Rainier Environmental
5013 Pacific Hwy East
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Tacoma, WA 98424

1.0 INTRODUCTION

An acute toxicity test was conducted using effluent samples collected from City of Everett's Port Gardner TF/SC System (SCE). Testing was conducted in May 2025 using the test organism *Ceriodaphnia dubia*. Testing was performed at Rainier Environmental Laboratory.

2.0 METHODS

2.1 Sample Collection and Transport

An effluent sample was collected into a LDPE cubitainer by City of Everett personnel. The sample was packed in a cooler containing ice and transported to Rainier Environmental the day of collection. Appropriate chain-of-custody procedures were employed during collection and transport.

2.2 Sample Receipt

Upon arrival at the laboratory, the cooler was opened, samples inspected, and the contents verified against information provided on the chain-of-custody forms. Receipt temperature was measured and recorded on the chain-of-custody form for each sample. Standard water quality parameters were measured and recorded on a sample check-in sheet (Appendix B). Samples were stored at 4°C in the dark until used for testing.

2.3 Test Methods

Acute toxicity tests were conducted using *C. dubia* according to procedures presented by USEPA (2002) and summarized in Table 1.

Table 1. Summary of conditions for the 48h *C. dubia* acute survival tests.

| | |
|---|--|
| Test initiation date and time | SCE: 5/22/2025; 1415h |
| Test termination date and time | SCE: 5/24/2025; 1400h |
| Test organism | <i>Ceriodaphnia dubia</i> |
| Test organism source | In-house cultures |
| Test organism age | < 24 hours |
| Test duration | 48 hours |
| Feeding | YTC:algal suspension during org. holding time. No feeding during test. |
| Test chamber and test solution volume | 30 mL plastic cup, 25 mL |
| Test temperature | 20 ± 1°C |
| Dilution water | Moderately Hard Synthetic Water |
| Test concentrations (% sample) | SCE: 100, 30, 10, 3.0, 0.64, laboratory control |
| Number of organisms/chamber | 5 |
| Number of replicates | 4 |
| Photoperiod | 16 hours light/8 hours dark |
| Aeration | None |
| Test protocol | EPA-821-R-02-012 |
| Test acceptability criterion for controls | ≥ 90% survival |
| Reference toxicant | Copper sulfate |

3.0 RESULTS

Details of standard water quality measurements conducted upon receipt of the sample are provided in Table 2.

Table 2. Sample information.

| Sample ID | SCE |
|--------------------------------------|------------------|
| Rainier Log-In No. | 25-102 |
| Collection date and time | 5/22/2025; 0700h |
| Receipt date and time | 5/22/2025; 1315h |
| Receipt temperature (°C) | 3.3 |
| Dissolved oxygen (mg/L) | 7.1 |
| pH | 7.32 |
| Conductivity (µS/cm) | 632 |
| Hardness (mg/L CaCO ₃) | 96 |
| Alkalinity (mg/L CaCO ₃) | 104 |
| Total Chlorine (mg/L) | <0.03 |
| Total Ammonia (mg/L) | 18.4 |

Survival was evaluated in the acute toxicity test after 48 hours of exposure. Results are summarized in Table 3. Mean survival in the 100 percent effluent concentration for sample SCE was 100 percent. There was no significant difference between the controls and the acute critical effluent concentration (ACEC) of 0.64 percent effluent.

Table 3. Summary of results

| Species | Concentration (%) | Survival (%) | NOEC ^a (% effluent) | LOEC ^b (% effluent) |
|---------------------------|-------------------|--------------|--------------------------------|--------------------------------|
| Sample ID: SCE | | | | |
| <i>Ceriodaphnia dubia</i> | 0.0 | 100 | 100 | >100 |
| | 0.64 | 100 | | |
| | 3.0 | 100 | | |
| | 10 | 100 | | |
| | 30 | 100 | | |
| | 100 | 100 | | |

^aNo Observed Effect Concentration, ^bLowest Observed Effect Concentration

Individual statistical summaries for all tests and copies of the laboratory bench sheets, sample check-in sheets and chain of custody forms are provided in Appendices A through C.

4.0 QA/QC

All samples were received in good condition and within the temperature range specified by WDOE (2016). The toxicity tests met all acceptability criteria for performance of control organisms. All water quality parameters remained within the ranges specified in the corresponding test methods throughout the tests.

Results for the reference toxicant test used to monitor laboratory performance and test organism sensitivity are summarized in Table 4. Results for the reference toxicant test fell within the acceptable range of mean ± two standard deviations of historical test results, indicating that the test organisms were of an appropriate degree of sensitivity. The coefficient of variation (CV) for the test is also shown in the table.

Table 4. Reference toxicant test results.

| Species | Date initiated | Endpoint | LC ₅₀ (µg/L copper) | Acceptable Range (µg/L copper) | CV (%) |
|---------------------------|----------------|--------------|--------------------------------|--------------------------------|--------|
| <i>Ceriodaphnia dubia</i> | 5/21/2025 | 96h survival | 18.3 | 8.05 - 34.0 | 43.4 |

REFERENCES

- Tidepool Scientific Software. 2000-2011. CETIS Comprehensive Environmental Toxicity Information System Software, Version 1.8.4.6.
- 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, pg. 53-54.
- WDOE. 2016. Laboratory Guidance and Whole Effluent Toxicity Test Review Criteria. Washington State Department of Ecology. Water Quality Program. Publication number: WQ-R-95-80, Revised June 2016.

Appendix A
Ceriodaphnia dubia Acute Toxicity Test
Statistical Summaries and Raw Bench Sheets

CETIS Summary Report

Report Date: 11 Jun-25 12:17 (p 1 of 1)
 Test Code: 2506-024 | 11-2841-1144

Ceriodaphnia 48-h Acute Survival Test

Rainier Environmental Laboratory

| | | |
|--------------------------------------|--|--|
| Batch ID: 14-5860-3117 | Test Type: Survival (48h) | Analyst: Eric Tollefson |
| Start Date: 22 May-25 14:15 | Protocol: EPA/821/R-02-012 (2002) | Diluent: Mod-Hard Synthetic Water |
| Ending Date: 24 May-25 14:00 | Species: Ceriodaphnia dubia | Brine: |
| Duration: 48h | Source: In-House Culture | Age: <24h |
| Sample ID: 00-1482-9678 | Code: 25-102 | Client: Everett |
| Sample Date: 22 May-25 07:00 | Material: POTW Effluent | Project: |
| Receive Date: 22 May-25 13:15 | Source: Everett (WA0024490) | |
| Sample Age: 7h (3.3 °C) | Station: SCE Outfall 100 | |

Comparison Summary

| Analysis ID | Endpoint | NOEL | LOEL | TOEL | PMSD | TU | Method |
|--------------|-------------------|------|------|------|------|----|------------------------------|
| 18-6137-7986 | 48h Survival Rate | 100 | >100 | NA | 5.0% | 1 | Steel Many-One Rank Sum Test |

Test Acceptability

| Analysis ID | Endpoint | Attribute | Test Stat | TAC Limits | Overlap | Decision |
|--------------|-------------------|--------------|-----------|------------|---------|-------------------------------|
| 18-6137-7986 | 48h Survival Rate | Control Resp | 1 | 0.9 - NL | Yes | Passes Acceptability Criteria |

48h Survival Rate Summary

| C-% | Control Type | Count | Mean | 95% LCL | 95% UCL | Min | Max | Std Err | Std Dev | CV% | %Effect |
|------|----------------|-------|------|---------|---------|-----|-----|---------|---------|------|---------|
| 0 | Dilution Water | 4 | 1 | 1 | 1 | 1 | 1 | 0 | 0 | 0.0% | 0.0% |
| 0.64 | | 4 | 1 | 1 | 1 | 1 | 1 | 0 | 0 | 0.0% | 0.0% |
| 3 | | 4 | 1 | 1 | 1 | 1 | 1 | 0 | 0 | 0.0% | 0.0% |
| 10 | | 4 | 1 | 1 | 1 | 1 | 1 | 0 | 0 | 0.0% | 0.0% |
| 30 | | 4 | 1 | 1 | 1 | 1 | 1 | 0 | 0 | 0.0% | 0.0% |
| 100 | | 4 | 1 | 1 | 1 | 1 | 1 | 0 | 0 | 0.0% | 0.0% |

48h Survival Rate Detail

| C-% | Control Type | Rep 1 | Rep 2 | Rep 3 | Rep 4 |
|------|----------------|-------|-------|-------|-------|
| 0 | Dilution Water | 1 | 1 | 1 | 1 |
| 0.64 | | 1 | 1 | 1 | 1 |
| 3 | | 1 | 1 | 1 | 1 |
| 10 | | 1 | 1 | 1 | 1 |
| 30 | | 1 | 1 | 1 | 1 |
| 100 | | 1 | 1 | 1 | 1 |

48h Survival Rate Binomials

| C-% | Control Type | Rep 1 | Rep 2 | Rep 3 | Rep 4 |
|------|----------------|-------|-------|-------|-------|
| 0 | Dilution Water | 5/5 | 5/5 | 5/5 | 5/5 |
| 0.64 | | 5/5 | 5/5 | 5/5 | 5/5 |
| 3 | | 5/5 | 5/5 | 5/5 | 5/5 |
| 10 | | 5/5 | 5/5 | 5/5 | 5/5 |
| 30 | | 5/5 | 5/5 | 5/5 | 5/5 |
| 100 | | 5/5 | 5/5 | 5/5 | 5/5 |

Freshwater Acute
48 Hour Toxicity Test Data Sheet

Client: CITY OF EVERETT
 Sample ID: SCF
 Test # 2506-024
 Log-In # 25-02

Start Date & Time: 5/22/2005 1415
 End Date & Time: 5/24/2005 1400
 Test Organism: Caecidophora dubia

| Rep. # | Conc. or % | Cont. # | Number of Live Organisms | | | Dissolved Oxygen (mg/L) | | | pH (units) | | | Cond (uhoen-cm) | | | Temperature (°C) | | | Mean Percent Survival |
|--------|------------|---------|--------------------------|----|----|-------------------------|-----|-----|------------|------|------|-----------------|-----|-----|------------------|------|------|-----------------------|
| | | | 0 | 24 | 48 | 0 | 24 | 48 | 0 | 24 | 48 | 0 | 24 | 48 | 0 | 24 | 48 | |
| 1 | CON | 9 | 5 | 5 | 5 | 7.8 | 7.5 | 7.7 | 8.17 | 8.14 | 8.01 | 319 | 320 | 318 | 20.7 | 20.1 | 19.9 | |
| 2 | | 19 | 5 | 5 | 5 | | | | | | | | | | | | | |
| 3 | | 1 | 5 | 5 | 5 | | | | | | | | | | | | | |
| 4 | | 22 | 5 | 5 | 5 | | | | | | | | | | | | | |
| 1 | 0.64 | 14 | 5 | 5 | 5 | 7.9 | 7.5 | 7.9 | 8.16 | 8.11 | 7.98 | 320 | 321 | 318 | 20.7 | 20.1 | 19.9 | |
| 2 | | 2 | 5 | 5 | 5 | | | | | | | | | | | | | |
| 3 | | 18 | 5 | 5 | 5 | | | | | | | | | | | | | |
| 4 | | 15 | 5 | 5 | 5 | | | | | | | | | | | | | |
| 1 | 3 | 23 | 5 | 5 | 5 | 8.0 | 7.8 | 7.5 | 8.12 | 8.09 | 7.98 | 320 | 321 | 317 | 20.5 | 20.0 | 19.9 | |
| 2 | | 8 | 5 | 5 | 5 | | | | | | | | | | | | | |
| 3 | | 24 | 5 | 5 | 5 | | | | | | | | | | | | | |
| 4 | | 10 | 5 | 5 | 5 | | | | | | | | | | | | | |
| 1 | 10 | 3 | 5 | 5 | 5 | 7.9 | 7.7 | 7.8 | 7.98 | 8.01 | 7.95 | 337 | 335 | 335 | 20.5 | 20.0 | 19.9 | |
| 2 | | 16 | 5 | 5 | 5 | | | | | | | | | | | | | |
| 3 | | 7 | 5 | 5 | 5 | | | | | | | | | | | | | |
| 4 | | 20 | 5 | 5 | 5 | | | | | | | | | | | | | |
| 1 | 30 | 12 | 5 | 5 | 5 | 7.9 | 7.4 | 8.0 | 7.75 | 7.84 | 7.91 | 387 | 385 | 379 | 20.9 | 20.1 | 19.9 | |
| 2 | | 6 | 5 | 5 | 5 | | | | | | | | | | | | | |
| 3 | | 17 | 5 | 5 | 5 | | | | | | | | | | | | | |
| 4 | | 11 | 5 | 5 | 5 | | | | | | | | | | | | | |
| 1 | 100 | 4 | 5 | 5 | 5 | 7.5 | 7.3 | 7.2 | 7.41 | 7.63 | 7.82 | 556 | 554 | 548 | 21.1 | 20.1 | 19.9 | |
| 2 | | 21 | 5 | 5 | 5 | | | | | | | | | | | | | |
| 3 | | 13 | 5 | 5 | 5 | | | | | | | | | | | | | |
| 4 | | 5 | 5 | 5 | 5 | | | | | | | | | | | | | |

Technician Initials: U- [Signature]

Dilution Water Batch #: MHSW059
 Test Chamber: ROOM 2

Animal Source: In House Culture
 Date Received:

Sample Description:
 QA Check: U

Comments: 0 hrs:
 24 hrs:
 48 hrs:

Appendix B
Sample Check-In Sheets

Client: CITY OF EVERETT

Tests Performed: cl-a
 Test ID No(s): 2506-0214

| Sample ID: | Log-in No. (20-xxxx): | Sample Collection Date & Time: | Sample Receipt Date & Time: | Check-in Temperature (°C) | Temperature OK? | DO (mg/L) | pH (units) | Conductivity (µS/cm) | Salinity (ppt) | Tit. Vol / Sam. Vol. / Alkalinity (mg/L)* | Tit. Vol. / Sam. Vol. / Hardness (mg/L)* | Total Chlorine (mg/L) | Total Ammonia Nitrogen (mg/L) | Technician Initials |
|------------|-----------------------|--------------------------------|-----------------------------|---------------------------|-----------------|------------|-------------|----------------------|----------------|---|--|-----------------------|-------------------------------|---------------------|
| <u>SCE</u> | <u>25-1025</u> | <u>5/22/15 0700</u> | <u>5/22/15 1315</u> | <u>3.3</u> | <u>Y</u> | <u>7.1</u> | <u>7.32</u> | <u>632</u> | <u>6.3</u> | <u>2.6 1 25 1104</u> | <u>2.4 1 25 1 96</u> | <u><0.03</u> | <u>18.4</u> | <u>ju</u> |

* = mg/L as CaCO₃; ² = Measured for freshwater samples only, NA = Not Applicable, NM = Not Measured

Freshwater Tests:

Control/Dilution Water Source: test type: cd-a 8:2 (DMW) Other: MHW -059 Alkalinity: 68 Hardness: 96
 Control/Dilution Water Source: test type: 8:2 (DMW) MHW Other: Alkalinity: Hardness:
 Additional Control? Y N = Alkalinity: Hardness:

Marine Tests:

Control/Dilution Water Source: test type: ART SW NAT SW Alkalinity: Salinity:
 Control/Dilution Water Source: test type: ART SW NAT SW Alkalinity: Salinity:
 Additional Control? Y N = Alkalinity: Salinity:
 Sample Salted w/ artificial salt? Y N If yes, what ppt? test type:
 Sample Salted w/brine? Y N If yes, what ppt? test type:

Comments: Temperature for grab sample must be 0-20°C if received within 1 hour of collection time, 0-12°C if effluent received within 4 hours of collection time, and 0-6°C for all other samples.

Sample Description:

COC Complete? Y or N
1 1 2 3

Filtration? Y N
 Pore Size:
 Organisms or Debris

Aeration? Y N
 Length of Time:
 Final DO:
 Final pH:

Hardness Adjustment? Y N
 If adjusted, please see worksheet for details.

Sub-samples for additional chemistry:

QC Check: ju

Appendix C
Chain-of-Custody Form

Sample Collection By:

Date _____ Page _____ of _____

Report to:
 Company: City of Everett
 Address: 3200 Cedar St
 City/State/Zip: Everett WA 98201
 Contact: Derek Kerlee
 Phone: 425-257-6790
 Email: D.Kerlee@everettwa.gov

Invoice To:
 Company: Same
 Address: _____
 City/State/Zip: _____
 Contact: _____
 Phone: _____
 Email: _____

| SAMPLE ID | DATE | TIME | MATRIX | CONTAINER TYPE | NO. OF CONTAINERS | COMMENTS |
|-----------|---------|------|--------|----------------|-------------------|---|
| 1 | 5-22-25 | 0700 | W/W | IDL | 1 | X Acute Daphnia WET (cerio daphnia Dobia) |
| 2 | | | | | | |
| 3 | | | | | | |
| 4 | | | | | | |
| 5 | | | | | | |
| 6 | | | | | | |
| 7 | | | | | | |
| 8 | | | | | | |
| 9 | | | | | | |
| 10 | | | | | | |

Receipt Temperature (°C)

| PROJECT INFORMATION | | SAMPLE RECEIPT | |
|---------------------|------|---------------------------|---|
| Client: | | Total No. of Containers | 1 |
| PO No.: | | Received Good Conditions? | Y |
| Shipped Via: | Just | Matches Test Schedule? | Y |

| | | | |
|--|--|---|--|
| RELINQUISHED BY (CLIENT) Signature: <i>[Signature]</i> (Time) 0900 Printed Name: Scott Ford (Date) 5-22-25 Company: COE | | RELINQUISHED BY (COURIER) Signature: _____ (Time) _____ Printed Name: _____ (Date) _____ Company: _____ | |
| RECEIVED BY (COURIER) Signature: _____ (Time) _____ Printed Name: _____ (Date) _____ Company: _____ | | RECEIVED BY (LABORATORY) Signature: <i>[Signature]</i> (Time) 1315 Printed Name: EDC TOLLEFSON (Date) 5/22/25 Log in #: 26-102 | |

SPECIAL INSTRUCTIONS/COMMENTS: