

**NACWA White Paper:  
Whole Effluent Toxicity Permit  
Testing and Limitations  
For Clean Water Agencies**

**Appendix B**

**REGION 3**  
**MONITORING**

## D. Toxics Management Program

### 1. Biological Monitoring:

In accordance with the schedule in 2. below, the permittee shall conduct annual chronic toxicity tests for the duration of the permit. The permittee should collect 24-hour flow-proportioned composite samples of final effluent from outfall 001.

b. NOEC Chronic Tests: The chronic tests shall include the following:

Chronic 3-Brood Static Renewal Survival and Reproduction Test using *Ceriodaphnia dubia*

Chronic 7-Day Static Renewal Survival and Growth Test using *Pimephales promelas*

These chronic tests shall be conducted in such a manner and at sufficient dilutions (minimum of five dilutions, derived geometrically) to determine the "No Observed Effect Concentration" (NOEC) for survival and reproduction or growth. The test dilutions should be able to determine compliance with the following end point:

Chronic NOEC of 69% is equivalent to a TUC of 1.46

Results which cannot be determined (i.e., a "less than" NOEC value) are not acceptable, and a retest will have to be performed. Express the test NOEC as  $TU_c$  (Chronic Toxic Units) for DMR reporting, where  $TUC = 100/NOEC$ . Report the  $LC_{50}$  at 48 hours and the  $IC_{25}$  with the NOEC's in the test report.

- c. The permittee may provide additional samples to address data variability during the period of initial data generation. These data shall be reported and may be included in the evaluation of effluent toxicity. Test procedures and reporting shall be in accordance with the WET testing methods cited in 40 CFR - Part 136.3
- d. The test data will be evaluated by DEQ for reasonable potential at the conclusion of the test period. The data may be evaluated sooner if requested by the permittee, or if toxicity has been noted. Should evaluation of the data indicate that a limit is needed, a WET limit and compliance schedule will be required and the toxicity tests listed in 1.A above may be discontinued.

### 2. Reporting Schedule:

The permittee shall report the results on the DMR and supply two (2) copies of the toxicity test reports specified in this Toxics Management Program in accordance with the following schedule:

Period	Compliance Sampling Period	DMR/Report Submission Date
1 <sup>st</sup> Annual Analysis	By July 31, 2002	August 10 <sup>th</sup> 2002
2 <sup>nd</sup> Annual Analysis	By July 31, 2003	August 10 <sup>th</sup> 2003
3 <sup>rd</sup> Annual Analysis	By July 31, 2004	August 10 <sup>th</sup> 2004
4 <sup>th</sup> Annual Analysis	By July 31, 2005	August 10 <sup>th</sup> 2005
5 <sup>th</sup> Annual Analysis	By July 31, 2006	August 10 <sup>th</sup> 2006

**REGION 4**  
**MONITORING**

#### D. BIOMONITORING REQUIREMENTS, ACUTE

The permittee shall conduct a 48-hour static acute toxicity test on two test species on samples of final effluent from Outfall 001 during the permit term as is necessary to comply with the EPA Form 2A permit application requirements. The test species to be used are Water Fleas (*Ceriodaphnia dubia*) and Fathead Minnows (*Pimephales promelas*).

The measured endpoint for toxicity will be the concentration causing 50% lethality (LC<sub>50</sub>) of the test organisms. The LC<sub>50</sub> shall be determined based on a 50% lethality as compared to the controls, and as derived from linear interpolation.

Test shall be conducted and its results reported based on appropriate replicates of a total of five serial dilutions and a control, using the percent effluent dilutions as presented in the following table:

Serial Dilutions for Whole Effluent Toxicity (WET) Testing					
4 X PL	2 X PL	Permit Limit (PL)	0.50 X PL	0.25 X PL	Control
% effluent					
34	17	8.5	4.3	2.13	0

The dilution/control water used will be moderately hard water as described in Methods for Measuring the Acute Toxicity of Effluents to Freshwater and Marine Organisms, EPA/600/4-90/027(F) (or the most current edition). An acute standard reference toxicant quality assurance test shall be conducted with each species used in the toxicity tests and the results submitted with the discharge monitoring report.

All tests will be conducted using four separate grab samples of final effluent, to be used in four separate tests, and shall be collected at evenly spaced (6-hour) intervals over a 24-hour period. If in any control, more than 10% of the test organisms die in 48 hours, the test (control and effluent) is considered invalid and the test shall be repeated within two (2) weeks. Furthermore, if the results do not meet the acceptability criteria as defined in Methods for Measuring the Acute Toxicity of Effluents to Freshwater and Marine Organisms, EPA/600/4-90/027(F), that test shall be repeated. Any test initiated but terminated before completion must also be reported along with a complete explanation for the termination.

The toxicity tests specified herein shall be conducted either annually for four (4) years or quarterly for one (1) year.

Test procedures, quality assurance practices and determination of effluent lethality values will be made in accordance with Methods for Measuring the Acute Toxicity of Effluents to Freshwater and Marine Organisms, EPA/600/4-90/027(F) or the most current edition.

Results of all tests, reference toxicant information, copies of raw data sheets, statistical analysis and chemical analysis shall be compiled in a report. The report shall be written in accordance with Methods for Measuring the Acute Toxicity of Effluents to Freshwater and Marine Organisms, EPA/600/4-90/027(F) or the most current edition.

Two copies of biomonitoring reports (including follow-up reports) shall be submitted to the Division. One copy of the report shall be submitted along with the discharge monitoring report (DMR). The second copy shall be submitted to the local Division of Water Pollution Control office address (see table below):

## **REGION 6**

### **MONITORING**

## BIOMONITORING REQUIREMENTS

### CHRONIC BIOMONITORING REQUIREMENTS: FRESHWATER

The provisions of this Section apply to Outfall 001 for whole effluent toxicity testing (biomonitoring).

#### 1. Scope, Frequency and Methodology

- a. The permittee shall test the effluent for toxicity in accordance with the provisions below. Such testing will determine if an appropriately dilute effluent sample adversely affects the survival, reproduction, or growth of the test organisms.
- b. The permittee shall conduct the following toxicity tests utilizing the test organisms, procedures and quality assurance requirements specified in this Part of the permit and in accordance with "Short-Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Freshwater Organisms, Fourth Edition" (EPA-821-R-02-013), or the most recent update:
  - 1) Chronic static renewal survival and reproduction test using the water flea (*Ceriodaphnia dubia*) (Method 1002.0 or the most recent update). This test should be terminated when 60% of the surviving adults in the control produce three broods or at the end of eight days, whichever comes first. This test shall be conducted once per quarter.
  - 2) Chronic static renewal 7-day larval survival and growth test using the fathead minnow (*Pimephales promelas*) (Method 1000.0 or the most recent update). A minimum of five replicates with eight organisms per replicate shall be used in the control and in each dilution. This test shall be conducted once per quarter.

The permittee must perform and report a valid test for each test species during the prescribed reporting period. An invalid test must be repeated during the same reporting period. An invalid test is herein defined as any test failing to satisfy the test acceptability criteria, including Percent Minimum Significant Difference (PMSD) boundary requirements, procedures, and quality assurance requirements specified in the test methods and permit. All test results, valid or invalid, must be submitted as described below.

- c. The permittee shall use five effluent dilution concentrations and a control in each toxicity test. These additional effluent concentrations are 30%, 40%, 53%, 71%, and 95% effluent. The critical dilution, defined as 71% effluent, is the effluent concentration representative of the proportion of effluent in the receiving water during critical low flow or critical mixing conditions.
- d. This permit may be amended to require a Whole Effluent Toxicity (WET) limit, Chemical-Specific (CS) effluent limits, a Best Management Practice (BMP), additional toxicity testing, and/or other appropriate actions to address toxicity. The permittee may be required to conduct additional biomonitoring tests and/or a Toxicity Reduction Evaluation (TRE) if biomonitoring data indicate multiple numbers of unconfirmed toxicity events.
- e. Testing Frequency Reduction
  - 1) If none of the first four consecutive quarterly fathead minnow tests demonstrates significant lethal or sub-lethal effects, the permittee may submit this information in writing and, upon approval from the Water Quality Standards Team, reduce the testing frequency to once per year.
  - 2) If one or more of the first four consecutive quarterly fathead minnow tests demonstrates significant sub-lethal effects, the permittee shall continue quarterly testing until four consecutive quarterly tests demonstrate no significant sub-lethal effects. At that time, the permittee may apply for the testing frequency reduction.

- 3) If one or more of the first four consecutive quarterly fathead minnow tests demonstrates significant lethal effects, the permittee shall continue quarterly testing until the permit is reissued. If a testing frequency reduction had been previously granted and a subsequent test demonstrates significant lethal effects, the permittee will resume a quarterly testing frequency until the permit is reissued.



## **REGION 9**

### **MONITORING**

## **S8. ACUTE TOXICITY**

### **A. Testing Requirements**

The Permittee shall test final effluent once per quarter in the year prior to submission of the application for permit renewal and in accordance with the submittal dates as listed on the Summary of Permit Report Submittals. The two species listed below shall be used on each sample and the results submitted to the Department as a part of the permit renewal application process. The Permittee shall conduct acute toxicity testing on a series of five concentrations of effluent and a control in order to be able to determine appropriate point estimates and an NOEC (no observed effect concentration). The percent survival in 100% effluent shall also be reported.

Acute toxicity tests shall be conducted with the following species and protocols:

1. Fathead minnow, *Pimephales promelas* (96-hour static-renewal test, method: EPA/800/R/02/012)

2. Daphnid, *Ceriodaphnia dubia*, *Daphnia pulex*, or *Daphnia magna* (48-hour static test, method: EPA/800/R/02/012).

B. Sampling and Reporting Requirements

1. All reports for effluent characterization or compliance monitoring shall be submitted in accordance with the most recent version of Department of Ecology Publication # WQ-R-95-80, *Laboratory Guidance and Whole Effluent Toxicity Test Review Criteria*, in regards to format and content. Reports shall contain bench sheets and reference toxicant results for test methods. If the lab provides the toxicity test data on floppy disk for electronic entry into the Department's database, then the Permittee shall send the disk to the Department along with the test report, bench sheets, and reference toxicant results.
2. Testing shall be conducted on 24-hour composite effluent samples. Samples taken for toxicity testing shall be cooled while being collected and while being delivered to the testing facility in order to meet the sample requirements of #WQ-R-95-80, *Laboratory Guidance and Whole Effluent Toxicity Test Review Criteria*. The lab shall begin toxicity testing as soon as possible but no later than 36 hours after sampling was ended.
3. All samples and test solutions for toxicity testing shall have water quality measurements as specified in Department of Ecology Publication # WQ-R-95-80, *Laboratory Guidance and Whole Effluent Toxicity Test Review Criteria*, or most recent version thereof.
4. All toxicity tests shall meet quality assurance criteria and test conditions in the most recent versions of the EPA manual listed in Subsection A and the Department of Ecology Publication # WQ-R-95-80, *Laboratory Guidance and Whole Effluent Toxicity Test Review Criteria*. If test results are determined to be invalid or anomalous by the Department, testing shall be repeated with freshly collected effluent.
5. Control water and dilution water shall be laboratory water meeting the requirements of the EPA manual listed in Subsection A or pristine natural water of sufficient quality for good control performance.
6. Effluent samples for whole effluent toxicity testing shall be collected just prior to the chlorination step in the treatment process.
7. The Permittee may choose to conduct a full dilution series test during compliance monitoring in order to determine dose response. In this case, the series must have a minimum of five (5) effluent concentrations and a control. The series of concentrations must include the acute critical effluent concentration (ACEC). The ACEC equals 3.1% effluent.

8. All whole effluent toxicity tests, effluent screening tests, and rapid screening tests that involve hypothesis testing and do not comply with the acute statistical power standard of 29% as defined in WAC 173-205-020 must be repeated on a fresh sample with an increased number of replicates to increase the power.

**S9. CHRONIC TOXICITY (**

**A. Testing Requirements**

The Permittee shall test final effluent once per quarter in the year prior to submission of the application for permit renewal and in accordance with the submittal dates as listed on the Summary of Permit Report Submittals. All of the chronic toxicity tests listed below shall be conducted on each sample. The results of this chronic toxicity testing shall be submitted to the Department as a part of the permit renewal application process.

The Permittee shall conduct chronic toxicity testing on a series of at least five (5) concentrations of effluent and a control in order to be able to determine appropriate point estimates and an NOEC. This series of dilutions shall include the ACEC. The Permittee shall compare the ACEC to the control using hypothesis testing at the 0.05 level of significance as described in Appendix H, EPA/600/4-89/001.

Chronic toxicity tests shall be conducted with the following species and the most recent version of the following protocols:

Saltwater Chronic Toxicity Test Species		Method
Topsmelt or Silverside minnow	<i>Atherinops affinis</i> or <i>Menidia beryllina</i>	EPA/600/R-95/136 or EPA/821/R/02/014
Mysid shrimp	<i>Holmesimysis costata</i> or <i>Mysidopsis bahia</i>	EPA/600/R-95/136 or EPA/821/R/02/014

The Permittee shall use the West Coast fish (topsmelt, *Atherinops affinis*) and mysid (*Holmesimysis costata*) for toxicity testing unless the lab cannot obtain a sufficient quantity of a West Coast species in good condition in which case the East Coast fish (silverside minnow, *Menidia beryllina*) or mysid (*Mysidopsis bahia*) may be substituted.

B. Sampling and Reporting Requirements

1. All reports for effluent characterization or compliance monitoring shall be submitted in accordance with the most recent version of Department of Ecology Publication # WQ-R-95-80, *Laboratory Guidance and Whole Effluent Toxicity Test Review Criteria*, in regards to format and content. Reports shall contain bench sheets and reference toxicant results for test methods. If the lab provides the toxicity test data on floppy disk for electronic entry into the Department's database, then the Permittee shall send the disk to the Department along with the test report, bench sheets, and reference toxicant results.
2. Testing shall be conducted on 24-hour composite effluent samples. Samples taken for toxicity testing shall be cooled while being collected and while being delivered to the testing facility in order to meet the sample requirements of #WQ-R-95-80, *Laboratory Guidance and Whole Effluent Toxicity Test Review Criteria*. The lab shall begin toxicity testing as soon as possible but no later than 36 hours after sampling was ended.
3. All samples and test solutions for toxicity testing shall have water quality measurements as specified in Department of Ecology Publication # WQ-R-95-80, *Laboratory Guidance and Whole Effluent Toxicity Test Review Criteria*, or most recent version thereof.
4. All toxicity tests shall meet quality assurance criteria and test conditions in the most recent versions of the EPA manual listed in Subsection A and the Department of Ecology Publication # WQ-R-95-80, *Laboratory Guidance and Whole Effluent Toxicity Test Review Criteria*. If test results are determined to be invalid or anomalous by the Department, testing shall be repeated with freshly collected effluent.
5. Control water and dilution water shall be laboratory water meeting the requirements of the EPA manual listed in Subsection A or pristine natural water of sufficient quality for good control performance.
6. Effluent samples for whole effluent toxicity testing shall be collected just prior to the chlorination step in the treatment process.
7. The Permittee may choose to conduct a full dilution series test in order to determine dose response. In this case, the series must have a minimum of five (5) effluent concentrations and a control. The series of concentrations must include the ACEC and the CCEC. The ACEC and the chronic critical effluent concentration (CCEC) may either substitute for the effluent concentration that is closest to it in the dilution series or be an extra effluent concentration. The CCEC equals 0.65% effluent.

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8. All whole effluent toxicity tests that involve hypothesis testing and do not comply with the chronic statistical power standard of 39% as defined in WAC 173-205-020 must be repeated on a fresh sample with an increased number of replicates to increase the power.