

## Section D. Test Methods

**Note: EPA published the final rule "Guidelines Establishing Test Procedures for the Analysis of Pollutants Under the Clean Water Act; Analysis and Sampling Procedures" on Federal Register, Vol. 77, No. 97, May 18, 2012. Any recent or future changes or incorporation of new testing protocol or methods in the Effluent Limitations Guideline at 40 CFR Part 435 supersede the applicable requirements in this permit.**

### 1. Samples of Wastes

If requested, the permittee shall provide EPA with a sample of any waste in a manner specified by the Agency.

### 2. Drilling Fluids Toxicity Test

The approved test method for permit compliance is identified as: Drilling Fluids Toxicity Test at 40 CFR Part 435, Subpart A, Appendix 2. Report for DMR Parameter No. 04312.

### 3. 7-Day Chronic Toxicity Testing Requirements

The approved test methods for permit compliance are identified in 40 CFR Part 136. In order to assess toxicity, the test must be conducted in accordance with the method, and any alternate test procedure requests must follow the requirements under 40- CFR §136.5.

- a) The permittee shall utilize the *Americamysis bahia* (formerly *Mysidopsis bahia* as referred to in Method 2007.0 and 1007.0, and DMRs) chronic static renewal 7-day survival and growth test using Method 1007.0. A minimum of eight (8) replicates with five (5) organisms per replicate must be used in the control and in each effluent dilution of this test.
- b) The permittee shall utilize the *Menidia beryllina* (Inland Silverside minnow) chronic static renewal 7-day larval survival and growth test using Method 1006.0. A minimum of **four (4)** replicates with **ten (10)** organisms per replicate must be used in the control and in each effluent dilution of this test.
- c) The NOEC (No Observed Effect Concentration) is defined as the greatest effluent dilution which does not result in a lethal or sub-lethal effect that is statistically different from the control (0% effluent) at the 95% confidence level. In the case of a test that exhibits a non-monotonic concentration response, determination of the NOEC will rely on the procedures described in *Method Guidance and Recommendations for Whole Effluent Toxicity (WET) Testing (40 CFR Part 136)*, July 2000, EPA 821-B-00-004.
- d) The effluent dilution series used for the toxicity test shall be based on the critical dilution, using a dilution factor of 0.5. The effluent dilution series must bracket the

critical dilution, with two effluent dilutions lower than the critical dilution and two effluent dilutions greater than the critical dilution.

e) This permit may be reopened to require chemical specific effluent limits, additional testing, and/or other appropriate actions to address toxicity.

f) Test Acceptability Criteria

The permittee shall repeat a test, including the control and all effluent dilutions, if the procedures and quality assurance requirements defined in the test methods or in this permit are not satisfied, including the following additional criteria:

Condition/Criteria	<i>Americamysis bahia</i>	<i>Menidia beryllina</i>
Test Duration	7 days	7 days
Minimum # of replicates per concentration	8	4
Minimum # of organisms per replicate	5	10
Minimum # or organisms per concentration	40	40
# of test concentrations per effluent	5 and a control	5 and a control
Holding time *	72 hours for first use	72 hours for first use
Sampling Requirement *	Minimum of 3 samples	Minimum of 3 samples
Test Acceptability Criteria	≥80% survival of all control organisms.	≥80% survival of all control organisms.
	Average dry weight per surviving organism in control ≥0.2mg.	Average dry weight per surviving unpreserved organism in the control must be ≥0.5mg when test starts with 7d old larvae, or, ≥0.43mg or greater after no more than 7days if preserved.
Coefficient of Variation**	40% or less, unless significant effects are exhibited.	40% or less, unless significant effects are exhibited.
Percent Minimum Significant Difference (PMSD range) for Sublethal Endpoint **	11 - 37	11 - 28

#### 4. 48-Hour Toxicity Testing Requirements (48-Hour Acute NOEC Marine Limits)

The approved test methods for permit compliance are identified in 40 CFR Part 136. In order to assess toxicity, the test must be conducted in accordance with the method, and any alternate test procedure requests must follow the requirements under 40- CFR §136.5.

- a) The permittee shall utilize the *Americamysis bahia* (formerly *Mysidopsis bahia* as referred to in Method 2007.0 and 1007.0, and DMRs) acute static renewal 48-hour definitive toxicity test using EPA-821-R-02-012. A minimum of **two (2)** replicates with **ten (10)** organisms per replicate must be used in the control and in each effluent dilution of this test.
- b) The permittee shall utilize the *Menidia beryllina* (Inland Silverside minnow) acute static renewal 48-hour definitive toxicity test using EPA-821-R-02-012. A minimum of **two (2)** replicates with **ten (10)** organisms per replicate must be used in the control and in each effluent dilution of this test.
- c) The NOEC is defined as the greatest effluent dilution which does not result in lethality that is statistically different from the control (0% effluent) at the 95% confidence level.
- d) The effluent dilution series used for the toxicity test shall be based on the critical dilution, using a dilution factor of 0.5. The effluent dilution series must bracket the critical dilution, with two effluent dilutions lower than the critical dilution and two effluent dilutions greater than the critical dilution.
- e) This permit may be reopened to require chemical specific effluent limits, additional testing, and/or other appropriate actions to address toxicity.
- f) Test Acceptability Criteria

The permittee shall repeat a test, including the control and all effluent dilutions, if the procedures and quality assurance requirements defined in the test methods or in this permit are not satisfied, including the following additional criteria:

Condition/Criteria	<i>Americamysis bahia</i>	<i>Menidia beryllina</i>
<b>Minimum # of replicates per concentration</b>	2	2
<b>Minimum # of organisms per replicate</b>	10	10
<b>Minimum # or organisms per concentration</b>	20	20
<b># of test concentrations per</b>	5 and a control	5 and a control

<b>effluent</b>		
<b>Holding time *</b>	72 hours for first use	72 hours for first use
<b>Test Acceptability Criteria</b>	≥90% survival of all control organisms.	≥90% survival of all control organisms.
<b>Coefficient of Variation</b>	40% or less, unless significant effects are exhibited.	40% or less unless significant effects are exhibited.

\* If the flow from the outfall(s) being tested ceases during the collection of effluent samples, the requirements for the minimum number of effluent samples and the minimum number of effluent portions are waived during that sampling period. However, the permittee must collect an effluent composite sample volume during the period of discharge that is sufficient to complete the required toxicity tests with daily renewal of effluent, and must meet the holding time between collection and first use of the sample. The effluent composite sample collection duration and the static renewal protocol associated with the abbreviated sample collection must be documented in the full report required in Item 3 of this section.

g) Statistical Interpretation

For the Mysid shrimp survival test and the Inland Silverside minnow survival test, the statistical analyses used to determine if there is a statistically significant difference between the control and the critical dilution shall be in accordance with the methods for determining the NOEC as described in EPA-821-R-02-012 or the most recent update thereof.

h) Dilution Water

Dilution water used in the toxicity tests may be receiving water collected as close to the point of discharge as possible but unaffected by the discharge, or synthetic dilution water.

i) Reporting Instructions

The permittee shall prepare a full report of the results of all tests conducted pursuant to this section in accordance with the Report Preparation Section of the method, for every valid or invalid toxicity test initiated whether carried to completion or not. The permittee shall retain each full report and submit them upon the request of the Agency.

1. Chemically Treated Miscellaneous Discharges (Toxicity Data Reporting):

Monthly reporting of toxicity data is required regardless of the testing frequency. This is to allow a space in the DMR to report data under a fluctuating frequency. If a test is not conducted every month, then the permittee must report "NODI 9" for toxicity data.