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## **PROPOSED DMMP/RSET Clarification Paper: Freshwater Bioassays Species, Endpoints, Methods and Interpretive Criteria**

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**NOTE: THIS PAPER UPDATES AND REPLACES THE SMARM 2014 DMMP/RSET CLARIFICATION PAPER: FRESHWATER BIOASSAYS**

### **INTRODUCTION**

The Washington State Sediment Management Standards (SMS) and DMMP/RSET dredging guidance require biological testing of dredged material when chemical testing results indicate the potential for unacceptable adverse environmental or human health effects. The response of bioassay organisms exposed to the sediment sample representing each DMMU is compared to the response of these organisms in both control and reference treatments. This comparison will determine whether the material is suitable for unconfined, open-water disposal.

In 2014, the freshwater bioassay standards were adopted as guidance by DMMP/RSET, with the SCO representing SL1 and CSL representing SL2. One and two hit guidance were also adopted at this time to be consistent with marine guidance. However, there are differences in the origins and use of the marine and freshwater guidance, which requires a different approach for freshwater. This paper outlines the proposed changes and will replace the 2014 paper.

#### Species and Endpoints

In both SMS and DMMP/RSET dredging guidance, bioassay results are considered to be more informative of potential resource impacts than exceedance of numeric chemical sediment standards (DMMP/RSET 2014). Thus bioassay results always take precedence over chemical results. This structure exists largely because chemical standards are informed by biological observations from some of the same suite of biological tests used to evaluate dredged material. The floating percentile model (FPM) used to develop the new Washington State freshwater sediment standards (WAC 173-204-563) used input from both acute (10-day *Chironomus* and *Hyalella*) and chronic (28-day *Hyalella*) tests. The standards were based on the most sensitive test, which was often the chronic 28-day *Hyalella* growth bioassay.

Prior to 2014, the Sediment Evaluation Framework for the Pacific Northwest (SEF 2009) and the DMMP User Manual (DMMP 2013) required only short-term bioassays using either *Hyalella* or *Chironomus*. Since the new freshwater SLs are often based on the results from a chronic bioassay, however, it is important that at least one of the bioassays conducted to override exceedances of the numeric standards should evaluate a chronic exposure measuring a sublethal endpoint.

#### Interpretive Criteria

Based on the updated SMS, interpretive criteria were developed for the DMMP freshwater bioassay tests (Section 9.6.7 and Table 9-5, UM 2014). These were modeled after the “one hit”/“two hit” criteria used in the DMMP marine guidelines. In the bioassay guidance, the “one hit” and “two hit” designations refer to a toxic response with a “one hit” response representing a greater level of impact than a “two hit” response.

There are, however some important differences between the DMMP's marine and freshwater interpretive criteria both in their derivation and application:

- This interpretive guidance was established prior to adoption of the SMS, and the “hit” definitions are more conservative than those used for SMS marine cleanup criteria. Therefore all sediments meeting the DMMP “one hit” or “two hit” definition also meet the marine Sediment Quality Standard (SQS), which is the sediment quality goal for protection of benthic communities for Washington State.
- In the freshwater guidance, the SL1 is equivalent to the Sediment Cleanup Objective (SCO) which is a no adverse effects level. The SL2 is equivalent to the Cleanup Screening Level (CSL) which establishes minor adverse effects.
- The SMS freshwater SCOs and CSLs were adopted prior to the development of freshwater guidance for the dredging program and were not designed as guidelines for open-water disposal, but rather for the identification of potential cleanup sites. The SCO/SL1 is the state goal for freshwater sediments for the protection of benthic communities, while the CSL/SL2 is used in defining potential cleanup sites. They are not equivalent to the marine “two hit” and “one hit” level of effects, both of which are more stringent than the state goal for marine sediments.
- In addition, the DMMP marine guidance includes more stringent standards for dispersive sites than those used for non-dispersive sites, which are actively monitored and managed. Open-water disposal sites in freshwater are almost exclusively dispersive in nature, and all disposed sediments must meet the more stringent SCO/SL1.

## PROBLEM STATEMENTS

### 1. Species and Endpoints

- a. Previous guidance required only 10-day acute endpoint bioassays to override the chemical testing results, although the freshwater benthic screening levels (FW benthic SLs) were often based on longer-term exposures and sublethal endpoints.
- b. When the new Washington State freshwater sediment chemical standards (WAC 173-204-563) were implemented in September 2013, chronic tests with sublethal endpoints for *Chironomus* and *Hyalella* were added to the list of approved SMS bioassays. Testing under SMS now requires a chronic test and a sublethal endpoint to be included in the suite of three bioassays used to identify toxicity in test sediments. DMMP/RSET guidance for conducting freshwater bioassays needs modification to be consistent with the new state freshwater sediment standards.

### 2. Interpretive Criteria

- a. In consideration of the differences between the freshwater and marine guidelines and their relationship to the Washington State Sediment Management Standards (SMS), a modification to the “one hit/two hit” approach is needed for freshwater bioassay interpretive criteria. Application of SL1/SL2 values in a manner similar to the “one hit”/“two hit” approach of the marine guidelines would result in open-water disposal of dredged material that exceeds the freshwater sediment quality goal for Washington State. Therefore, current freshwater bioassay hit definitions are inconsistent with WA State goals for freshwater sediments.

- b. Open water disposal sites in freshwater are almost exclusively dispersive in nature, thus it is important that the sediments disposed there meet the SL1 criteria or Sediment Cleanup Objective (SCO).

## PROPOSED PROGRAM MODIFICATIONS

1. **Bioassay Species, Endpoints, and Methods.** In order to address the issues identified above, the DMMP/RSET agencies propose making the freshwater bioassay species and endpoint requirements for dredged material consistent with the SMS rule (WAC 173-204-563).

The SMS rule includes the following requirements for freshwater sediment bioassays. Categories are defined in Table 1.

- Two different test species
- Three endpoints
- One chronic test; and
- One sublethal endpoint

**Table 1. Freshwater biological tests, species, and applicable endpoints**

Species, biological test, and endpoint	Acute effects biological test	Chronic effects biological test	Lethal effects biological test	Sub-lethal effects biological test
<b>Amphipod: <i>Hyalella azteca</i></b>				
10-Day mortality	X		X	
28-Day mortality		X	X	
28-Day growth		X		X
<b>Midge: <i>Chironomus dilutus</i></b>				
10-Day mortality	X		X	
10-Day growth	X			X
20-Day mortality		X	X	
20-Day growth		X		X

**Bioassay Methods:** The bioassays should follow the protocols specified below. These tests and parameters were developed based on the most updated American Society for Testing and Materials protocols.

### Acute Effects Tests

- *Hyalella azteca* 10-day mortality: ASTM E1706-05 (2010)/EPA Method 100.1 (US EPA, 2000)
- *Chironomus dilutus* 10-day mortality: ASTM E1706-05 (2010)/EPA Method 100.2 (US EPA, 2000)
- *Chironomus dilutus* 10-day growth: ASTM E1706-05 (2010)/EPA Method 100.2 (US EPA, 2000)

### Chronic Effects Tests

- *Hyalella azteca* 28-day mortality: EPA Method 100.4 (US EPA, 2000)
- *Hyalella azteca* 28-day growth: EPA Method 100.4 (US EPA, 2000)
- *Chironomus dilutus* 20-day mortality: EPA Method 100.5 (US EPA, 2000)
- *Chironomus dilutus* 20-day growth: EPA Method 100.5 (US EPA, 2000)

- 2. Bioassay Interpretative Criteria.** In order to address the issues identified above, the DMMP/RSET agencies propose that for dispersive disposal the “one hit” and “two hit” designations be dropped for freshwater bioassays. Failure to meet the SL1/SCO would result in material not being suitable for open-water dispersive disposal (Table 2). Failure of SL2/CSL (WAC 173-204-563 Table VII) would still be considered for antidegradation evaluation (see clarification paper “Quality of Post-Dredge Sediment Surfaces”, SMARM 2008) or non-dispersive disposal cases, but would be applied in a case-by-case manner depending on project specifics.

**Table 2. Performance standards and interpretive criteria for freshwater bioassays.**

Biological Test/ Endpoint <sup>a</sup>	Performance Standard <sup>b</sup>		Screening Level 1 (SL1) or Sediment Cleanup Objective (SCO)	Screening Level 2 (SL2) or Cleanup Screening Level (CSL)
	Control <sup>c</sup>	Reference		
<b><i>Hyalella azteca</i></b>				
10-day mortality	$M_C \leq 20\%$	$M_R \leq 25\%$	$M_T - M_C > 15\%$ and $M_T$ vs $M_C$ SD ( $p \leq 0.05$ )	$M_T - M_C > 25\%$ and $M_T$ vs $M_C$ SD ( $p \leq 0.05$ )
28-day mortality	$M_C \leq 20\%$	$M_R \leq 30\%$	$M_T - M_C > 10\%$ and $M_T$ vs $M_C$ SD ( $p \leq 0.05$ )	$M_T - M_C > 25\%$ and $M_T$ vs $M_C$ SD ( $p \leq 0.05$ )
28-day growth	$MIG_C \geq 0.15$ mg/ind	$MIG_R \geq 0.15$ mg/ind	$(MIG_C - MIG_T)/MIG_C > 0.25$ and $MIG_T$ vs $MIG_C$ SD ( $p \leq 0.05$ )	$(MIG_C - MIG_T)/MIG_C > 0.40$ and $MIG_T$ vs $MIG_C$ SD ( $p \leq 0.05$ )
<b><i>Chironomus dilutus</i></b>				
10-day mortality	$M_C \leq 30\%$	$M_R \leq 30\%$	$M_T - M_C > 20\%$ and $M_T$ vs $M_C$ SD ( $p \leq 0.05$ )	$M_T - M_C > 30\%$ And $M_T$ vs $M_C$ SD ( $p \leq 0.05$ )
10-day growth	$MIG_C \geq 0.48$ mg/ind	$MIG_R/MIG_C \geq 0.8$	$(MIG_C - MIG_T)/MIG_C > 0.20$ and $MIG_T$ vs $MIG_C$ SD ( $p \leq 0.05$ )	$(MIG_C - MIG_T)/MIG_C > 0.30$ and $MIG_T$ vs $MIG_C$ SD ( $p \leq 0.05$ )
20-day mortality	$M_C \leq 32\%$	$M_R \leq 35\%$	$M_T - M_C > 15\%$ and $M_T$ vs $M_C$ SD ( $p \leq 0.05$ )	$M_T - M_C > 25\%$ and $M_T$ vs $M_C$ SD ( $p \leq 0.05$ )
20-day growth	$MIG_C \geq 0.60$ mg/ind	$MIG_R/MIG_C \geq 0.8$	$(MIG_C - MIG_T)/MIG_C > 0.25$ and $MIG_T$ vs $MIG_C$ SD ( $p \leq 0.05$ )	$(MIG_C - MIG_T)/MIG_C > 0.40$ and $MIG_T$ vs $MIG_C$ SD ( $p \leq 0.05$ )

**Notes:**

M = Mortality; C = Control; R = Reference; T = Test; F = Final; MIG = Mean Individual Growth at time final; mg = milligrams.

Reference performance standards are provided for sites where the agencies have approved a freshwater reference sediment site(s). For reference interpretive criteria, reference results will be substituted for control in the interpretive criteria formula. When a reference sediment is used, and control, reference and test sediments all have similar physical characteristics, a hit is defined when (1) the test sediment response relative to both negative control and reference exceeds the bioassay specific guidelines; and (2) the difference between test and both control and reference is statistically significant.

Bioassay failure requires statistical significance between test and control/reference at  $p = 0.05$ .

The control performance standard for the 20 day test (0.60 mg/individual) is more stringent than for the 10 day test and the agencies may consider, on a case-by-case basis, a 20 day control has met QA/QC requirements if the mean individual growth is at least 0.48 mg/individual.