

**SELECTION OF NEGATIVE CONTROL SEDIMENTS AND
USE OF CONTROL SEDIMENTS AS REFERENCE SEDIMENTS
DMMP CLARIFICATION PAPER
SMS DRAFT TECHNICAL INFORMATION MEMORANDUM**

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INTRODUCTION

Bioassays performed for dredged material management programs in the State of Washington or for comparison to the state's Sediment Management Standards (SMS) utilize negative controls to provide an estimate of test organism general health during the test exposure period (PSDDA, 1989). A negative control sediment is utilized in the amphipod mortality and *Neanthes* growth bioassays, while a clean seawater control is used in the larval test. Negative control performance standards must be met for the test results to be considered valid.

Reference sediments are included in bioassays to address nontreatment effects from physical factors such as grain size. Performance standards also exist for reference sediments and may be based on a comparison to the negative control (all bioassays except the SMS amphipod and Microtox tests). Only reference sediments that meet the performance standard can be used for statistical comparison to test sediments.

PROBLEM IDENTIFICATION

Negative controls for the amphipod and *Neanthes* bioassays must be a sediment type in which the test organism is found and thrives in the environment. However, in some cases laboratories have used a single type of control sediment for all bioassays, even though that type of sediment may not have been appropriate as a control for one or more of the test organisms. For example, West Beach sand has been used for both the *Neanthes* growth test and for the amphipod mortality test using *Ampelisca abdita*. While *Neanthes arenaceodentata* thrives in West Beach sand, *Ampelisca abdita* favors finer-grained sediment. The result of using an inappropriate sediment is generally an elevated response in the negative control. This creates problems both for determining the general health of the test organisms and judging the performance of reference sediments.

In addition, at times reference sediments fail to meet performance standards. Under SMS, guidance is not currently provided on whether, or how, statistical comparisons can be made when a reference sediment fails to perform adequately. If a reference sediment fails to meet its performance standard there may be no way to interpret test results.

CLARIFICATION

SELECTION OF NEGATIVE CONTROL SEDIMENTS. An appropriate negative control sediment must be used for the amphipod mortality and *Neanthes* growth tests. PSEP (1995) provides the following description of native habitat for various amphipods: "*Rhepoxynius abronius* and *Eohaustorius estuarius* typically inhabit well-sorted, fine sand while *Ampelisca abdita* is a tube-dwelling amphipod found mainly in protected areas and is often abundant in sediments with a high organic content. It generally inhabits sediments from fine sand to mud and silt without shell, although it can also be found in relatively coarser sediments with a sizable fine component." The best way to ensure a good negative control is to collect the control sediment from the same location at which the test organisms are collected.

Neanthes arenaceodentata is cultured in the lab rather than field-collected. However, PSEP (1995) states that, "For the *Neanthes* bioassay, sand should be used as the control sediment". West Beach of Whidbey Island is most often used as a collection site for clean control sediment. From PSEP (1995), "*Neanthes* maintained in West Beach sand exhibited low mortality and high percentage increases in biomass during the exposure period, indicating that West Beach sand is a suitable material for a control sediment".

PSEP (1995) also states that, "All bioassays must be conducted using well-established negative (clean) controls. Such controls are clean, nontoxic seawater and/or sediment samples taken from outside each study area". For dredged material management programs in the State of Washington or for comparison to SMS, sediments proposed for use as negative controls must be approved before bioassays commence. If an area without a proven track record is proposed for collection of negative control sediment, sufficient data (such as grain size, organic carbon content, chemical data, bioassay results) must be submitted before its use can be approved by the regulatory agencies.

USE OF CONTROL SEDIMENTS AS REFERENCE SEDIMENTS. When a reference sediment fails to meet its performance standard, and more than one reference has been collected, Michelsen and Shaw (1996) provide procedures for statistical comparisons. If no reference sediments meet performance standards, or if the control sediment is closer in grain size and TOC to one or more stations being evaluated than any of the remaining reference sediments, the control sediment should be evaluated for use as a reference sediment. If the control sediment is similar in grain size and TOC to the site sediments and/or a reference sediment that failed to meet performance standards, it will be considered an acceptable substitute for the reference sediment and the data will be interpreted accordingly.

If a control sediment is substantially dissimilar to the site stations and a failed reference sediment in its physical characteristics (e.g., >25% difference in fines and a difference of 1% TOC), it may still be used as a substitute for the reference station if both the agencies/site manager and the project proponent agree that this is appropriate. Otherwise, the data will be considered uninterpretable and the bioassay(s) in question will need to be rerun.

REFERENCES

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