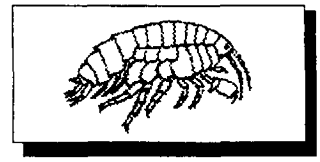

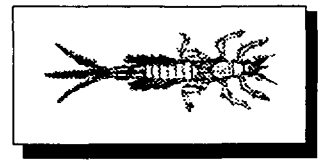

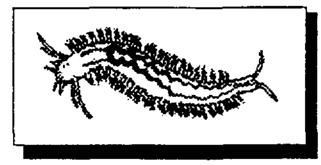

User's Guide and Technical Resource Document: Evaluation of Sediment Toxicity Tests for Biomonitoring Programs

HEALTH AND ENVIRONMENTAL SCIENCES DEPARTMENT

API PUBLICATION NUMBER 4607

STEP
Strategies for Today's Environmental Partnership

American Petroleum Institute
1220 L Street, Northwest
Washington, D.C. 20005

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OCTOBER 1994

Strategies for Today's Environmental Partnership

American
Petroleum
Institute

STEP
Strategies for Today's
Environmental Partnership

One of the most significant long-term trends affecting the future vitality of the petroleum industry is the public's concerns about the environment. Recognizing this trend, API member companies have developed a positive, forward looking strategy called STEP: Strategies for Today's Environmental Partnership. This program aims to address public concerns by improving our industry's environmental, health and safety performance; documenting performance improvements; and communicating them to the public. The foundation of STEP is the API Environmental Mission and Guiding Environmental Principles.

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The members of the American Petroleum Institute are dedicated to continuous efforts to improve the compatibility of our operations with the environment while economically developing energy resources and supplying high quality products and services to consumers. The members recognize the importance of efficiently meeting society's needs and our responsibility to work with the public, the government, and others to develop and to use natural resources in an environmentally sound manner while protecting the health and safety of our employees and the public. To meet these responsibilities, API members pledge to manage our businesses according to these principles:

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- To advise promptly, appropriate officials, employees, customers and the public of information on significant industry-related safety, health and environmental hazards, and to recommend protective measures.
- To counsel customers, transporters and others in the safe use, transportation and disposal of our raw materials, products and waste materials.
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- To extend knowledge by conducting or supporting research on the safety, health and environmental effects of our raw materials, products, processes and waste materials.
- To commit to reduce overall emission and waste generation.
- To work with others to resolve problems created by handling and disposal of hazardous substances from our operations.
- To participate with government and others in creating responsible laws, regulations and standards to safeguard the community, workplace and environment.
- To promote these principles and practices by sharing experiences and offering assistance to others who produce, handle, use, transport or dispose of similar raw materials, petroleum products and wastes.

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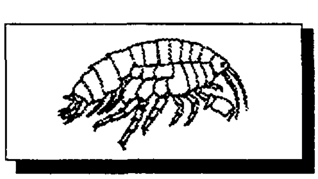
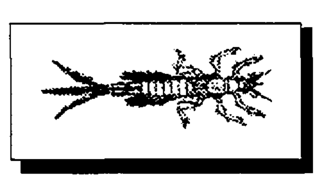
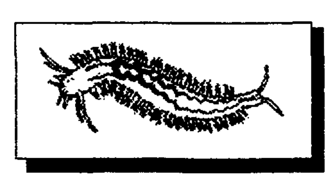
User's Guide:

Evaluation of Sediment Toxicity Tests for Biomonitoring Programs







ABSTRACT

Sediment toxicity test methods are available for marine, estuarine, and freshwater sediments and organisms. The methods can be used for a variety of purposes: for example, assessment of existing environmental conditions, monitoring changes with time, or for NPDES permit compliance. Use of inappropriate test methods or species for a given purpose can impact the toxicity results and their interpretation. This User's Guide has been prepared to assist personnel at petroleum industry facilities (refineries, marketing terminals, and production locations) in understanding sediment toxicity testing and in the selection of test methods and species which are appropriate for their needs. The general aspects of sediment toxicity testing are summarized along with technical requirements and appropriate conditions for each test type. Test methods are evaluated for their reliability, ecological relevance, exposure relevance, availability, interferences, and ability to discriminate toxicants. A companion report (Technical Resource Document) has been prepared to provide detailed technical background information on the methods.

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LIST OF FIGURES AND TABLES

INTRODUCTION

Reliable toxicity tests are currently available for testing of marine, estuarine, and freshwater sediments as part of biomonitoring programs for wastewater discharges. Sediment toxicity tests provide an integrated measure of the effects of sediment contamination that eliminates much of the uncertainty associated with predicting toxicity from sediment chemistry alone. When combined with surveys of animals living in the sediments, sediment toxicity tests can be used to assess existing conditions, rank sites for cleanup priority, and monitor changes in contaminant effects with time (Chapman *et al.* 1992). However, the use of inappropriate test methods or species and the failure to consider physical and chemical factors that can affect the results of the tests may diminish the value of biological toxicity testing (Burton 1991; Hill *et al.* 1993).

The purpose of this User's Guide is to provide information that will enable environmental personnel at petroleum facilities to select sediment toxicity tests and test methods that are scientifically valid and appropriate for a specific site. For those readers who are unfamiliar with sediment toxicity testing, this User's Guide explains general aspects of sediment toxicity testing and how to use available technical information. This document also outlines the technical requirements and appropriate conditions for using different sediment toxicity test methods. A companion document, *Evaluation of Sediment Toxicity Tests for Biomonitoring Programs* (PTI,1994) hereafter referred to as the Technical Resource Document, has been prepared to provide technical background on the test methods and the detailed rationale for the evaluations presented here. The Technical Resource Document is intended to be used as a reference tool for the test selection process and also as an information resource to support negotiations with agencies concerning the appropriateness of any recommended tests.

Sediment toxicity tests anticipated for future use in biomonitoring programs for National Pollutant Discharge Elimination System (NPDES) permit compliance are addressed in the Technical Resource Document and in this User's Guide. These documents were developed for use by petroleum industry operations (refineries, marketing terminals, and production facilities) that have discharges to surface waters. However, the Technical Resource Document and this User's Guide contain information that is applicable to other industries and could be used by any wastewater discharger.

The term *sediment toxicity test*, as used here, refers to any laboratory method that measures the adverse biological response of a group of organisms to a sample of test sediment. Some sediment toxicity tests measure lethal effects by determining the number of organisms that are killed during the exposure period. Other tests measure sublethal effects such as developmental abnormalities in juvenile stages, inhibition of reproduction, or reduced growth. Sediment toxicity tests are used in many biomonitoring programs because they integrate the effects of multiple chemicals and can be used in conjunction with chemical measurements and surveys of sediment-dwelling organisms to establish cause-effect relationships. Sediment toxicity tests are also the primary tool for any toxicity identification and evaluation program. Sediment toxicity tests are available for many different species and various life stages of some species. For example, they may be conducted on embryos, larvae, and juveniles of various fish species, as well as embryos and juveniles of invertebrates such as clams, oysters, and sea urchins. Sediment toxicity tests can also be conducted with microscopic algae and bacteria, submerged aquatic plants (e.g., water hyacinth), and wetland plants (e.g., marsh grass).

The next section presents an evaluation of available sediment toxicity tests, including descriptions of habitat type, sediment test systems, and biological endpoints. The following section provides a procedure for selection of tests at a specific site. Finally, brief summaries of sampling and data analysis issues are presented in a section on application of sediment toxicity tests.

Selected terms in this User's Guide are defined in the *Glossary*.

EVALUATION OF SEDIMENT TOXICITY TESTS

The available test methods were classified by type of habitat (marine, estuarine, and freshwater) to which each method applies and the general endpoint type (lethal or sublethal) specified for each test. This classification scheme resulted in the following six major categories of tests:

- Marine lethal
- Marine sublethal
- Estuarine lethal
- Estuarine sublethal
- Freshwater lethal
- Freshwater sublethal.

Appendix A of the Technical Resource Document presents test classification tables that contain the following information on each test: 1) organisms, including the broad biotic group, scientific name, and life stage of the species used in the test; 2) exposure medium (whole sediment, interstitial water, sediment elutriate, or sediment extract); 3) exposure duration; and 4) primary literature references for test methods. Each test was assigned a number to allow users of the Technical Resource Document to track a given test through the various evaluation tables. In many cases, several of the specified tests were actually variations of a single test method and were assigned the same test number.

KEY TEST CHARACTERISTICS

The key characteristics used to classify sediment toxicity tests are described below (see the *Test Screening Approach* section of the Technical Resource Document for details).

Habitat Type

The primary characteristic that distinguishes marine, estuarine, and freshwater habitat types is water salinity. Salinity strongly influences the distributions of most of the test organisms. In some cases, test organisms are tolerant of both marine and estuarine conditions or both estuarine and freshwater conditions. However, few test organisms tolerate both marine and freshwater conditions. For purposes of this study, habitat categories were defined as follows:

- Marine ($\geq$28 ppt)
- Estuarine ($>$0.5 ppt and $<$28 ppt)
- Freshwater ($\leq$0.5 ppt).

Because the division between habitat categories is an artificial distinction, use of a particular habitat designation for a test in this report should not necessarily preclude the application of a test to sediments in other habitats. For example, some tests that are classified as marine tests may be applied to high salinity estuarine sediments, and in some cases, adjusting the salinity of a sediment sample to allow the use of a particular test may be appropriate.

Exposure Medium

The kind of exposure medium was used to classify the various toxicity tests because each kind of exposure medium has favorable and unfavorable characteristics that can profoundly influence the toxicity test results. The four kinds of exposure media considered were as follows:

- Whole sediments
- Interstitial water
- Sediment elutriates
- Sediment extracts.

Whole Sediments—The use of whole sediments is probably the most realistic exposure scenario because it mimics the manner in which most organisms are exposed to chemicals in the environment. Whole-sediment toxicity tests integrate multiple exposure routes, including chemical intake from dermal contact with sediment particles and interstitial water as well as ingestion of sediment particles, interstitial water, and food organisms (the food uptake route applies to at least some methods in which the test species is not fed). For most whole sediment tests, the sediments are carefully placed in the exposure chamber and the chamber is then filled with clean water. Resuspended particles are allowed to settle before initiation of exposure. In whole-sediment tests, infaunal test organisms are expected to have the highest potential for exposure to chemicals because they live within the sediments.

Interstitial Water—Interstitial water as an exposure medium is prepared by removing water from the test sediments by methods such as filtration and centrifugation. The test organisms are then introduced to the interstitial water in the absence of sediments. For infaunal organisms, interstitial water is a representative exposure medium for primarily one exposure route (i.e., dermal contact with the dissolved forms of chemicals). Interstitial water is not a representative exposure medium for epifaunal, planktonic, and nektonic organisms. The degree to which the sampling of interstitial waters or the elutriation process modifies the toxicity of the sample is usually unknown.

Sediment Elutriates—Sediment elutriates are prepared by mixing sediments and test water for a fixed period of time and then removing the sediments by methods such as filtration, centrifugation, and decanting after a settling period. The test organisms are then introduced to the test water in the absence of sediments. Elutriates are useful for representing the exposure to chemicals that can occur after sediments have been resuspended into the water column or after they have passed through the water column as part of dredged material disposal operations. Although the use of a sediment elutriate as an exposure medium is realistic for planktonic and nektonic test organisms, it is unrealistic for infaunal and epibenthic organisms. The degree to which the sampling of interstitial waters or the elutriation process modifies the toxicity of the sample is usually unknown.

Sediment Extracts—Sediment extracts are prepared by mixing sediments with an organic solvent that is capable of removing specific kinds of chemicals from the sediments. After the extraction process is completed, the sediments are removed by methods such as filtration, centrifugation, and decanting after a settling period. The extractant and the extracted chemicals are diluted with water for testing. In some cases, the extracted chemicals are first exchanged with a less toxic carrier medium before the test concentrations are prepared. In either case, the test organisms are introduced to a solvent-water mixture containing the extracted chemicals. Because the test organisms are exposed to an unnatural exposure medium (organic solvent) in the absence of sediments, an extractant-prepared exposure medium is generally considered an unrealistic exposure scenario.

Endpoint Type

The major types of endpoints for most toxicity tests include the following:

- Lethal (i.e., mortality)
- Sublethal
  - Reduced growth
  - Reproductive effects
  - Developmental abnormality
  - Histopathological abnormalities.

The determination of the lethal endpoint is unambiguous and is clearly an adverse effect. The reliability of any sublethal endpoint test depends on use of experienced laboratory personnel (for details see *Endpoint Type* in the section *Classification of Available Test Methods, Classification Criteria* in the Technical Resource Document).

EVALUATION CRITERIA

A technical rating was assigned to each sediment toxicity test based on each of the following evaluation criteria:

- Reliability
  - The endpoint can be measured accurately
  - The results are repeatable
  - The negative control results generally meet quality assurance criteria
  - Intra- and interlaboratory variability studies indicate high precision
- Ecological relevance
  - The results of a test method are directly applicable to indigenous species under field conditions
  - Test organisms are species that are of commercial or ecological importance
- Exposure relevance
  - The pathway of exposure used in a test is analogous to exposure under field conditions
- Availability
  - Test organisms can be easily obtained or cultured
  - The method is standardized and well documented
  - Commercial laboratories routinely perform the test
- Interferences
  - Test methods have a low susceptibility to confounding physical or chemical factors
- Chemical discrimination
  - Test results are useful in defining gradients of sediment toxicity in the environment
  - Test methods and organisms are not overly sensitive or insensitive.

An overall technical rating was determined by summing the scores for each of the individual criteria. Because little information was available on interferences and chemical discrimination for most tests, their influence on the overall technical rating scores was moderated by use of a weighting factor (see the *Test Screening Approach*, PTI 1994, section of the Technical Resource Document).

The rating for regulatory status was based on information from regional and national EPA offices and whether a test was recommended in guidance documents for potential use in NPDES programs, cleanup assessments, baseline monitoring, and dredged material testing. The guidance documents considered as the basis for rating regulatory status included the method documents issued by the Canadian government (Environment Canada 1990a–e, 1992a–f), the dredged material testing documents issued by United States government agencies (U.S. EPA and U.S. COE 1991, 1993), and a major research and development planning document issued by EPA (U.S. EPA 1992). If a test was included in 3–4 of these document categories, it was assigned a rating of "high" for regulatory status. If a test was included in 1–2 of these document categories, it was assigned a rating of "medium." Toxicity tests that were not included in these documents and were not known to be required for use in current regulatory programs were assigned a rating of "low."

EVALUATION RESULTS

Results of the evaluation of sediment toxicity tests are presented in Tables 1 through 6. Most of the highly ranked marine and estuarine infaunal tests were based on the use of amphipods as test organisms, whereas most of the highly ranked freshwater infaunal tests were based on the use of insects (mayfly nymphs and midge larvae) as test organisms. These species groups are ecologically important, especially as key prey items for various fishes. In most cases, the highest ranking tests were the ones based on the exposure of infaunal organisms to whole sediments because: 1) exposure conditions closely mimic field conditions, 2) most of the test species are available by field collection during most of the year, and 3) many of the tests have well-developed methods.

TABLE 1. EVALUATION OF SEDIMENT TOXICITY TESTS – MARINE LETHAL

Test Number	Exposure Media	Common Name	Life Stage	Test Species	Overall Technical Rating	Reliability	Ecological Relevance	Exposure Relevance	Availability	Interferences	Chemical Responsiveness	Regulatory Status	Comments
					Technical Rating								
001	S	Amphipod	Juveniles or adult females	*Ampelisca abdita*	●	◕	◕	●	◕	◕	●	High	B,C,E
008	S	Amphipod	Adults	*Rhepoxynius abronius*	●	●	◑	●	◕	◕	●	High	C,E
003	S	Amphipod	Juveniles or young adults	*Eohaustorius washingtonianus*	●	●	◑	●	◕	◕	●	Med.	E
002	S	Amphipod	Juveniles or young adults	*Amphiporeia virginiana*	●	●	◑	●	◕	◕	◑	Med.	E
006	S	Amphipod	Juveniles or young adults	*Leptocheirus pinguis*	●	◕	◑	●	◕	◕	◑	Med.	
005	S	Amphipod	Immature	*Grandidierella japonica*	●	◕	◑	●	◕	◕	◑	Low	A,B
004	S	Amphipod	Juveniles or young adults	*Foxiphalus xiximeus*	◕	●	◑	●	◔	◕	◑	Med.	E
020	S	Littleneck clam	Juveniles	*Protothaca staminea*	◕	◕	◕	●	◑	◕	○	Low	
017	EL	Blue mussel	Embryos	*Mytilus edulis*	◕	◑	●	○	◕	●	●	Med.	B
033	S	Shrimp	Juveniles	*Pandalus* sp.	◕	◕	◕	◕	◔	●	◑	Low	
029	S	Cancer crab	Juveniles	*Cancer* sp.	◕	◕	◕	◕	◔	●	◑	Low	
027	S	Blue crab	Juveniles	*Callinectes sapidus*	◕	◕	◕	◕	◔	●	◑	Low	
034	S	Shrimp	Post-larvae	*Penaeus* sp.	◕	◕	◕	◕	◔	●	◑	Low	
071	S	Polychaete	Juveniles	*Neanthes* sp.	◕	◕	◑	●	◔	◕	◑	Med.	B
021	S	Japanese clam	Juveniles	*Tapes japonica*	◕	◕	◕	●	◔	◕	○	Low	
016	S	Blue mussel	Embryos	*Mytilus edulis*	◕	◑	●	◔	◕	○	●	Med.	B,C
011	EL	Pacific oyster	Embryos	*Crassostrea gigas*	◕	◑	●	○	◑	●	●	Med.	B,D
009	EL	Eastern oyster	Embryos	*Crassostrea virginica*	◕	◑	●	○	◑	●	●	Med.	B
055	EL	Sheepshead minnow	1-14 days old	*Cyprinodon variegatus*	◕	●	◑	○	●	◕	◔	Med.	B,E
037	S	Ridge-back prawn	Juveniles	*Sicyonia ingentis*	◕	◕	◑	◕	◔	●	◑	Low	A

Key

●	high	widely adopted
◕		
◑		
◔		
○	low	none

EL Elutriate
S Sediment

A Geographically restricted or alien to United States
B Widely distributed and/or cultured
C Potential sediment interferences
D Field validated to benthos
E Interlaboratory comparisons available

TABLE 2. EVALUATION OF SEDIMENT TOXICITY TESTS – MARINE SUBLETHAL

Test Number	Exposure Media	Common Name	Life Stage	Test Species	Technical Rating: Overall Technical Rating	Reliability	Ecological Relevance	Exposure Relevance	Availability	Interferences	Chemical Responsiveness	Regulatory Status	Comments
001	S	Amphipod	Juveniles or adult females	*Ampelisca abdita*	●	○	◕	●	◕	◕	●	Low	B,C
121	S	Polychaete	Juveniles	*Neanthes sp.*	●	◕	◑	●	◕	◕	◑	Low	B,E
109	EL	Purple sea urchin	Gametes	*Strongylocentrotus purpuratus*	●	◕	●	○	◕	●	●	Med.	E
008	S	Amphipod	Adults	*Rhepoxynius abronius*	◕	◔	◑	●	◕	◕	●	Low	C
005	S	Amphipod	Immature	*Grandidierella japonica*	◕	◔	◑	●	◕	◕	●	Low	B
017	EL	Blue mussel	Embryos	*Mytilus edulis*	◕	◑	●	○	◕	●	●	Low	B,E
111	EL	Purple sea urchin	Gametes	*Strongylocentrotus purpuratus*	◕	◕	●	○	◑	●	●	Low	E
047	EL	Purple sea urchin	Gametes	*Strongylocentrotus purpuratus*	◕	◕	●	○	◔	●	●	Med.	
102	EL	Sand dollar	Gametes	*Dendraster excentricus*	◕	◕	◑	○	◕	●	●	Med.	E
100	EL	Atlantic urchin	Gametes	*Arbacia punctulata*	◕	◕	◑	○	◕	●	●	Med.	E
115	EL	Green sea urchin	Gametes	*Strongylocentrotus droebachiensis*	◕	◕	◑	○	◕	●	●	Med.	E
015	S	Clam	Juveniles	*Mulinia lateralis*	◕	◑	◑	◕	◔	●	●	Low	
107	EL	White sea urchin	Gametes	*Lytechinus pictus*	◕	◕	◑	○	◕	●	●	Med.	A,E
112	EL	Purple sea urchin	Gametes	*Strongylocentrotus purpuratus*	◕	◑	●	○	◑	●	●	Low	
011	EL	Pacific oyster	Embryos	*Crassostrea gigas*	◕	◑	●	○	◑	●	●	Low	B,D,E
081	EL	Microtox	Cells	*Photobacterium phosphoreum*	◕	◑	◔	◑	●	◕	◕	Low	B,D
016	S	Blue mussel	Embryos	*Mytilus edulis*	◕	◔	●	◔	◕	○	●	Med.	B,C
009	EL	Eastern oyster	Embryos	*Crassostrea virginica*	◕	◔	●	○	◑	●	●	Low	B
080	INT	Microtox	Cells	*Photobacterium phosphoreum*	◕	◑	◔	◔	●	◕	◕	Low	B,D
010	S	Pacific oyster	Embryos	*Crassostrea gigas*	◕	◔	●	◔	◑	○	●	Med.	B,C,D

Key

●	high	widely adopted
◕		
◑		
◔		
○	low	none

EL Elutriate
INT Interstitial water
S Sediment

A Geographically restricted or alien to United States
B Widely distributed and/or cultured
C Potential sediment interferences
D Field validated to benthos
E Interlaboratory comparisons available

TABLE 3. EVALUATION OF SEDIMENT TOXICITY TESTS – ESTUARINE LETHAL

Test Number	Exposure Media	Common Name	Life Stage	Test Species	Overall Technical Rating	Reliability	Ecological Relevance	Exposure Relevance	Availability	Interferences	Chemical Responsiveness	Regulatory Status	Comments
					Technical Rating								
001	S	Amphipod	Juveniles or adult females	*Ampelisca abdita*	●	◕	◕	●	◕	◕	●	High	B,C,E
129	S	Amphipod	Large juveniles and adults	*Eohaustorius estuarius*	●	●	◑	●	◕	◕	●	High	C
003	S	Amphipod	Juveniles or young adults	*Eohaustorius washingtonianus*	●	●	◑	●	◕	◕	●	Med.	E
133	S	Amphipod	Juveniles	*Leptocheirus plumulosus*	●	●	◑	●	◕	◑	●	Low	B,E
135	S	Amphipod	Mixed sexes	*Leptocheirus plumulosus*	●	◕	◑	●	◕	◕	●	High	B
126	S	Amphipod	Juveniles or young adults	*Corophium volutator*	●	●	◑	◕	◕	◕	●	Med.	E
002	S	Amphipod	Juveniles or young adults	*Amphiporeia virginiana*	●	●	◑	●	◕	◕	◑	Med.	E
128	S	Amphipod	Adults	*Corophium volutator*	●	●	◑	◕	◕	◕	●	Low	E
134	S	Amphipod	Juveniles	*Leptocheirus plumulosus*	●	◕	◑	●	◕	◕	●	Low	B
131	S	Amphipod	7-14 days old	*Hyalella azteca*	●	◕	◑	◕	◑	●	●	High	B
020	S	Littleneck clam	Juveniles	*Protothaca staminea*	◕	◕	◕	●	◑	◕	○	Low	
017	EL	Blue mussel	Embryos	*Mytilus edulis*	◕	◑	●	○	◕	●	●	Med.	B
027	S	Blue crab	Juveniles	*Callinectes sapidus*	◕	◕	◕	◕	◔	●	◑	Low	
151	S	Grass shrimp	Post-hatch (1–4 days)	*Palaemonetes* sp.	◕	◕	◑	◕	◔	●	●	Low	
016	S	Blue mussel	Embryos	*Mytilus edulis*	◕	◑	●	◔	◕	○	●	Med.	B,C
009	EL	Eastern oyster	Embryos	*Crassostrea virginica*	◕	◑	●	○	◑	●	●	Med.	B
011	EL	Pacific oyster	Embryos	*Crassostrea gigas*	◕	◑	●	○	◑	●	●	Med.	B,D
030	S	Sand shrimp	Juveniles	*Crangon* sp.	◕	◕	◑	◕	◔	●	◑	Low	
153	EL	Sheepshead minnow	1–14 days old	*Cyprinodon variegatus*	◕	◕	◑	○	●	●	◔	Low	B
010	S	Pacific oyster	Embryos	*Crassostrea gigas*	◕	◑	●	◔	◑	○	●	Med.	B,C,D

Key

●	high	widely adopted
◕		
◑		
◔		
○	low	none

EL Elutriate
S Sediment

B Widely distributed and/or cultured
C Potential sediment interferences
D Field validated to benthos
E Interlaboratory comparisons available

TABLE 4. EVALUATION OF SEDIMENT TOXICITY TESTS – ESTUARINE SUBLETHAL

Test Number	Exposure Media	Common Name	Life Stage	Test Species	Overall Technical Rating	Reliability	Ecological Relevance	Exposure Relevance	Availability	Interferences	Chemical Responsiveness	Regulatory Status	Comments
					Technical Rating								
001	S	Amphipod	Juveniles or adult females	*Ampelisca abdita*	●	◕	◕	●	◕	◕	●	Low	B,C
134	S	Amphipod	Juveniles	*Leptocheirus plumulosus*	●	◑	◑	●	◕	◕	●	Low	B
135	S	Amphipod	Mixed sexes	*Leptocheirus plumulosus*	●	◑	◑	●	◕	◕	●	Low	B
129	S	Amphipod	Large juveniles and adults	*Eohaustorius estuarius*	●	◑	◑	●	◕	◕	●	Low	C
128	S	Amphipod	Adults	*Corophium volutator*	●	◕	◑	◕	◕	◕	●	Low	E
133	S	Amphipod	Juveniles	*Leptocheirus plumulosus*	●	◑	◑	●	◕	◑	●	Low	B
017	EL	Blue mussel	Embryos	*Mytilus edulis*	◕	◑	●	○	◕	●	●	Low	B,E
011	EL	Pacific oyster	Embryos	*Crassostrea gigas*	◕	◑	●	○	◑	●	●	Low	B,D,E
010	S	Pacific oyster	Embryos	*Crassostrea gigas*	◕	◑	●	◔	◑	○	●	Med.	B,C,D
016	S	Blue mussel	Embryos	*Mytilus edulis*	◕	◔	●	◔	◕	○	●	Med.	B,C
014	EL	Quahog clam	Embryos	*Mercenaria mercenaria*	◕	◔	●	○	◑	●	●	Low	B
009	EL	Eastern oyster	Embryos	*Crassostrea virginica*	◕	◔	●	○	◑	●	●	Low	B
187	EL	Sheepshead minnow	Adults	*Cyprinodon variegatus*	◕	◔	◑	○	●	●	●	Low	B
176	EX	Microtox	Cells	*Photobacterium phosphoreum*	◕	◑	◑	○	●	◔	◕	Low	B,D
191	INT	Polychaete	Females	*Dinophilus gyrociliatus*	◑	◑	◔	◔	◔	◕	●	Low	
175	S	Microtox	Cells	*Photobacterium phosphoreum*	◑	◑	◔	○	◑	○	◕	Low	B,C

Key		
●	high	widely adopted
◕		
◑		
◔		
○	low	none

EL Elutriate
EX Extract
INT Interstitial water
S Sediment

B Widely distributed and/or cultured
C Potential sediment interferences
D Field validated to benthos
E Interlaboratory comparisons available

TABLE 5. EVALUATION OF SEDIMENT TOXICITY TESTS – FRESHWATER LETHAL

Test Number	Exposure Media	Common Name	Life Stage	Test Species	Overall Technical Rating	Reliability	Ecological Relevance	Exposure Relevance	Availability	Interferences	Chemical Responsiveness	Regulatory Status	Comments
					Technical Rating								
241	S	Mayfly	Nymphs	*Hexagenia limbata*	●	◕	◕	●	◕	◕	●	Low	B,D
239	S	Mayfly	Young nymphs	*Hexagenia limbata*	●	◕	◕	●	◕	◕	●	Low	B,D
238	S	Midge	2nd instar larvae	*Chironomus tentans*	●	◕	◑	●	●	◑	●	High	B,D
237	S	Midge	Neonates or larvae	*Chironomus riparius*	●	◕	◑	●	●	◑	●	Med.	B
200	S	Amphipod	Juveniles	*Hyalella azteca*	●	◕	◑	◕	●	◕	●	High	B
242	S	Mayfly	Nymphs	*Hexagenia limbata*	●	◕	◑	●	◕	◕	●	Low	B,D
209	INT	Water flea	Neonates	*Daphnia magna*	●	●	◕	○	●	●	●	Low to Med.	B,D,E
256	S	Marsh grass	Seedlings	*Echinochloa crusgalli*	●	◕	●	●	◔	●	◑	Low	
250	S	Oligochaete	Large worms	*Lumbriculus variegatus*	●	◕	◑	●	◕	◕	◑	Low	B
249	S	Oligochaete	Mixed-age	*Lumbriculus variegatus*	●	◕	◑	●	◕	◕	◑	Low	B
212	EL	Water flea	Neonates (<24 hrs)	*Daphnia magna*	●	●	◑	○	●	●	●	Med.	B,D,E
240	INT	Mayfly	Nymphs	*Hexagenia limbata*	●	◕	◕	◑	◕	◑	●	Low	B,D
205	EL	Water flea	Neonates	*Ceriodaphnia dubia*	◕	●	◑	○	●	◕	●	Med.	B,E
213	EL	Water flea	Neonates (<24 hrs)	*Daphnia pulex*	◕	●	◑	○	●	◕	●	Med.	B,E
208	EL	Water flea	Neonates	*Daphnia magna*	◕	●	◑	○	●	◕	●	Med.	B,D,E
214	EL	Water flea	Neonates (<24 hrs)	*Daphnia pulex*	◕	◕	◑	○	●	●	●	Med.	B
202	S	Paper pondshell clam	Juveniles	*Anodonta imbecillis*	◕	◕	◑	●	◑	◕	◔	Low	
252	S	Oligochaete	Mixed age; similar size	*Pristina leidyi*	◕	◕	◑	◕	◑	●	◑	Low	B
255	S	Oligochaete	Mixed age	*Tubifex tubifex*	◕	◕	◑	◕	◑	●	◑	Low	B,D
222	EL	Bluegill	Approx. 4 days	*Lepomis macrochirus*	◕	●	◕	○	●	●	○	Low	B,E

Key

●	high	widely adopted
◕		
◑		
◔		
○	low	none

EL Elutriate
INT Interstitial water
S Sediment

B Widely distributed and/or cultured
C Potential sediment interferences
D Field validated to benthos
E Interlaboratory comparisons available

TABLE 6. EVALUATION OF SEDIMENT TOXICITY TESTS – FRESHWATER SUBLETHAL

Test Number	Exposure Media	Common Name	Life Stage	Test Species	Technical Rating: Overall Technical Rating	Reliability	Ecological Relevance	Exposure Relevance	Availability	Interferences	Chemical Responsiveness	Regulatory Status	Comments
238	S	Midge	2nd instar larvae	*Chironomus tentans*	●	◑	◑	●	●	◑	●	Low	B,D
237	S	Midge	Neonates or larvae	*Chironomus riparius*	●	◑	◑	●	●	◑	●	Low	B,D
200	S	Amphipod	Juveniles	*Hyalella azteca*	●	◑	◑	◕	●	◕	●	Low to Med.	B,D
242	S	Mayfly	Nymphs	*Hexagenia limbata*	●	◑	◑	●	◕	◕	●	Low	B
333	S	Aquatic vascular plant	Apical shoots	*Hydrilla verticillata*	●	◑	◕	●	◑	●	◑	Low	B
256	S	Marsh grass	Seedlings	*Echinochloa crusgalli*	●	◑	◕	●	◔	●	●	Low	B,D
269	S	Amphipod	Juveniles (<1 week)	*Hyalella azteca*	●	◑	◑	◕	◕	●	●	Low	B,D,E
285	EL	Water flea	Neonates	*Ceriodaphnia dubia*	◕	●	◑	○	●	◕	●	Med.	
330	S	Oligochaete	Adults	*Tubifex tubifex*	◕	◑	◑	◕	◑	●	●	Low	B
304	INT	Midge	2nd instar larvae	*Chironomus tentans*	◕	◑	◕	◑	◕	◔	●	Low to Med.	B
250	S	Oligochaete	Large worms	*Lumbriculus variegatus*	◕	◑	○	●	◕	◕	●	Low	B,D,E
323	S	Oligochaete	Juveniles and adults	*Lumbriculus variegatus*	◕	◑	◑	●	◔	◕	●	Low	B,D
324	S	Oligochaete	15 mm in length	*Lumbriculus variegatus*	◕	◑	◑	●	◔	◕	●	Low	B,E
226	EL	Fathead minnow	Larvae (<24 hrs old)	*Pimephales promelas*	◕	◕	◑	○	●	●	◔	Med.	B,E
260	EL	Green algae	Cells	*Selenastrum capricornutum*	◕	◑	◑	○	●	●	◕	Med.	B,D,E
272	INT	Microtox	Cells	*Photobacterium phosphoreum*	◕	◑	◑	○	●	●	◕	Low to Med.	B
235	S	Rainbow trout	Egg-sac stage	*Oncorhynchus mykiss*	◕	◑	◕	◔	◕	◔	●	Low	
261	INT	Green algae	Cells	*Selenastrum capricornutum*	◕	◑	◑	○	●	●	◕	Low	B
252	S	Oligochaete	Mixed age; similar size	*Pristina leidyi*	◕	◔	◑	◕	◑	●	◑	Low	B,D
176	EX	Microtox®	Cells	*Photobacterium phosphoreum*	◕	◑	◑	○	●	◑	◕	Low	B,E

Key

●	high	widely adopted
◕		
◑		
◔		
○	low	none

EL Elutriate
EX Extract
INT Interstitial water
S Sediment

B Widely distributed and/or cultured
D Field validated to benthos
E Interlaboratory comparisons available

Many of the lowest ranking toxicity tests involved exposure of planktonic organisms to whole sediments. The exposure relevance of these tests is relatively low because the test species are rarely exposed to sediments in the field and they may be sensitive to interference of suspended sediments with feeding mechanisms.

The species included in the highest ranking marine and estuarine tests for lethality include the following amphipods: *Ampelisca abdita*, *Rhepoxynius abronius*, *Grandidierella japonica*, *Eohaustorius washingtonianus*, *Eohaustorius estuarius*, *Amphiporeia virginiana*, *Foxiphalus xiximeus*, *Corophium volutator*, *Leptocheirus pinguis*, and *Leptocheirus plumulosus*. Reproductive endpoints are also well developed for the *L. plumulosus* test. Although behavioral endpoints (e.g., reburial at exposure termination) are used in many of these amphipod tests, the behavioral endpoints have generally not been field validated. The tests based on *A. abdita* and *R. abronius* are the only ones with a high regulatory status.

Taxonomic groups other than amphipods also ranked high among the marine and estuarine sublethal tests, including the polychaete (*Neanthes* sp.) growth test based on a 20–28 day exposure to whole sediments, the echinoderm (*Strongylocentrotus purpuratus*, *S. droebachiensis*, *Dendraster excentricus*, *Arbacia punctulata*, *Lytechinis pictus*) fertilization test of sediment elutriates, and the bivalve (*Mytilus edulis*, *Crassostrea gigas*, *C. virginica*) larval abnormality test of sediment elutriates. Although these elutriate tests have a lower exposure relevance than the whole sediment tests, they use sensitive life stages of ecologically important species, are widely available, and have well developed methods. Although these elutriate tests are generally reliable, their variability can be high and the negative controls fail quality assurance limits more frequently than those in the tests involving juveniles and adults of these or other species. Other high-ranking tests in the marine and estuarine sublethal category included the juvenile clam (*Mulinia lateralis*) test with whole sediments and the Microtox® (*Photobacterium phosphoreum*) test with sediment elutriates or interstitial water.

The highest ranking freshwater tests for lethal and sublethal endpoints were based on the exposure of infaunal insects (i.e., nymphs of the mayfly *Hexagenia limbata* and larvae of the midges *Chironomus riparius* and *Chironomus tentans*) and an epifaunal amphipod (*Hyalella azteca*) to whole sediments. Only the *H. azteca* and *C. tentans* lethal tests have high regulatory status. Whole sediment tests with vascular plants (*Hydrilla verticillata* and *Echinochloa crusgalli*) were among the top six ranked tests in the freshwater lethal category. These tests ranked high primarily because of their high degrees of exposure and ecological relevance and their relatively low susceptibility to interferences. The high ecological relevance of the two plant tests is based on the importance of the plants in providing habitat for other organisms. The major drawback of these two tests is their infrequent use in regulatory programs.

There is a relative lack of information on interferences and chemical discrimination for sediment toxicity tests. Further research in these areas and more comparative studies of toxicity tests with corresponding data on the bioavailability of sediment chemicals are needed.

SITE-SPECIFIC SELECTION OF SEDIMENT TOXICITY TESTS

The selection of toxicity test methods for application at a particular site involves consideration of many factors, including physical, chemical, and biological conditions at the site; regulatory requirements at federal, state, and local levels; and specific objectives for a monitoring program. Procedures for selecting sediment toxicity tests for use in biomonitoring programs are outlined in this section. First, the factors to be considered in test selection are defined. Second, the steps for selecting a test or battery of tests for application at a given site are described.

DEFINITION OF SELECTION CRITERIA

The selection of sediment toxicity tests for use in a biomonitoring program depends on site-specific characteristics, regulatory requirements, and other factors that are important in test evaluation (Table 7). Many of the decisions based on these factors may be constrained by technical specifications of a permit or monitoring program requirements.

OVERVIEW OF TEST SELECTION PROCESS

The process for selecting the most appropriate sediment test for a given study is illustrated in the decision tree shown in Figure 1. As users progress through each decision point within the tree, the number of candidate tests is reduced until the final sediment test(s) have been selected. Habitats and endpoints desired for the biomonitoring program should be matched to one of the six tables for test selection (Tables 1 through 6). Information on biotic group and geographic range for each of the tests is found in Appendix D of the Technical Resource Document. Also included in Appendix D are important comments regarding sensitivity to chemicals and interferences that, when combined with the known chemical and physical characteristics of the study site, provide critical information in the selection process. An overview of how to use the decision-making framework in selecting toxicity tests is provided in the following sections.

Site Characteristics

A review of available information on the characteristics of the discharge site to be monitored and the organisms living at the study site is the fundamental first step in the selection process. Available data on site-specific chemicals and physical properties of the sediments can be useful in selecting test species that are sensitive to the presence of the site-specific chemicals, yet have minimal interferences to other properties of the sediment (e.g., grain size, organic carbon, ammonia). Knowing what organisms live at the study site can help guide the selection of appropriate species. If, for example, polychaete worms and bivalves dominate the benthic community in a marine study area and echinoderms (sea urchins and sand dollars) are absent, it is likely that the most appropriate test would include either polychaetes or bivalves as receptors, not echinoderms. Other important information that should be assembled includes regional water quality data, sediment characteristics, habitat types, and seasonal patterns in biological or physical/chemical characteristics.

Regulatory Requirements

An equally important step in the selection of sediment toxicity tests is a thorough understanding of the applicable regulatory requirements that are driving the testing program. Regulatory programs frequently include explicit requirements that immediately limit the field of potential toxicity tests. These confining factors can include specifications for lethal or sublethal tests, exposure duration, seasons for testing, single species vs. a battery of species for testing, and data quality objectives. Guidelines for selecting toxicity tests can also be included as part of regulatory programs. Knowledge of the regulatory requirements or guidelines

TABLE 7. SELECTION OF SEDIMENT TOXICITY TESTS

Decision Factor	Alternatives	
Objectives	Single species vs. test battery	
	Season(s) for testing	
	Site-specific chemicals, receptors, and sediment types	
	Data quality objectives	
Regulatory Requirements	Various state and EPA regulations	
Geographic Zone	West Coast (north or south)	
	East Coast (north or south)	
	Gulf Coast (east or west)	
Habitat Type	Marine	
	Estuarine	
	Freshwater	
Biotic Group	Bacteria	Polychaete
	Eukaryotic cells	Oligochaete
	Algae	Mollusc
	Vascular plant	Echinoderm
	Crustacean	Amphibian
	Insect	Fish
	Nematode	
Species/Life Stage	Various species	
	Gametes	
	Embryos/Larvae	
	Juveniles	
	Adults	
Exposure Duration	Acute	
	Chronic	
Endpoint	Lethal	
	Sublethal	
Habitat Group[a]	Infauna	
	Epifauna	
	Plankton/nekton	
Exposure Medium[a]	Whole sediment	
	Sediment elutriate	
	Interstitial water	
	Sediment extract	
Potential Interferences[a]	Grain size	
	Organic carbon	
	Acid-volatile sulfides	
	Ammonia	
	Mold, pathogens	

Note: EPA - U.S. Environmental Protection Agency

[a] These decision factors were considered in ranking sediment toxicity tests. All other factors should be explicitly considered when selecting the sediment toxicity tests on a site-specific basis.

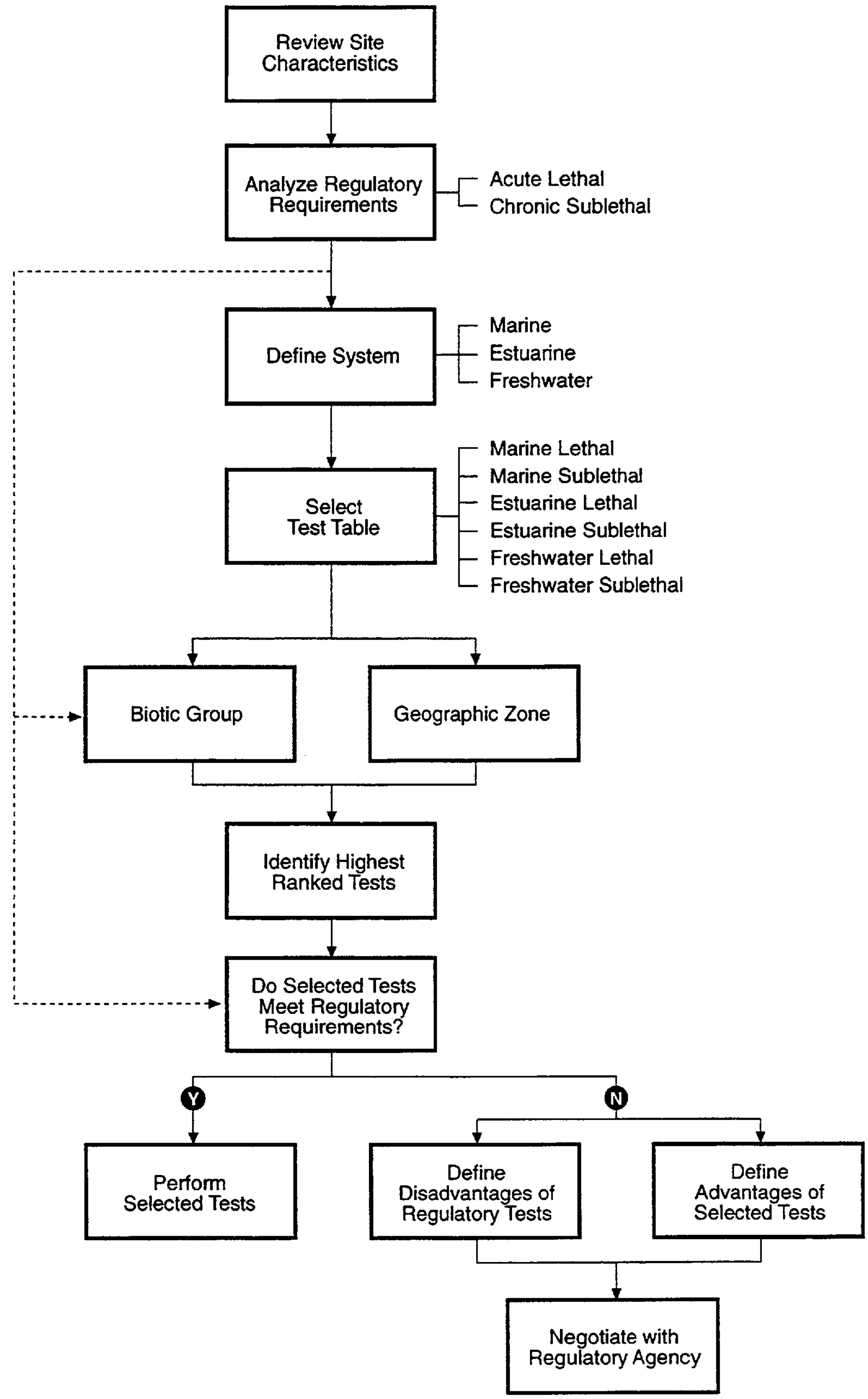

Figure 1. Approach to selection of sediment toxicity tests for a specific site.

for these or other toxicity test parameters is fundamental to the ultimate selection of the appropriate test. It is also important to have a full understanding of these regulatory requirements so that they can be evaluated in the context of the overall decision framework.

Selection of Evaluation Tables

Based on the habitat (marine, estuarine, or freshwater) and endpoint type (lethal and sublethal), one or more of the evaluation tables (Tables 1 through 6) is used to select appropriate tests. Important ancillary information relevant to each test is included in the Technical Resource Document (see especially Appendix D).

Biotic Group and Geographic Zone

A wide variety of biotic groups is represented in the listing of tests for each habitat and endpoint type. The list of candidate tests can be further reduced by deciding which organisms and which geographic zones are most relevant. The location of the study site will provide the information required to select a geographic zone. In addition, knowledge of the regulatory requirements may direct the selection of the species. If, for example, emphasis is on organisms that may be consumed by humans, then crabs, large bivalves, or fish are likely candidates for testing. If emphasis is on ecological risks, then other biotic groups such as algae, amphipods, insects, or polychaete worms become good candidate organisms.

Identify Highest Rank Tests

In the evaluation tables (Tables 1 through 6), tests are ranked from best overall candidate tests to least appropriate overall tests for each habitat/endpoint type. In most cases, the higher ranked tests may have very similar total scores. The user should select the most appropriate high-ranked test based on a consideration of site-specific factors or regulatory considerations.

Compare Selected Test(s) with Regulatory Requirements

The candidate toxicity test(s) tentatively selected should be matched with the regulatory requirements. If the test(s) meet these requirements, then the selection process is complete and the actual test(s) can be performed. If the selected toxicity test(s) do not meet the requirements of the applicable regulatory program, then low-ranked tests may need to be considered.

APPLICATION OF SEDIMENT TOXICITY TESTS

After the selected sediment toxicity tests are approved for a biomonitoring program, a sampling and analysis plan should be developed. The sampling and analysis plan specifies the study design for the field sampling program (see the *Application of Sediment Toxicity Tests* section in the Technical Resource Document), methods for implementing the toxicity tests, quality assurance procedures, and data analysis approaches. Issues related to quality assurance, sampling, and data analysis are discussed below.

METHODS AND QUALITY ASSURANCE ISSUES

The use of acceptable and well-documented laboratory methods is essential for ensuring that the results of toxicity testing are meaningful estimates of toxicity and that the tests are repeatable. Except for experimental studies, the tests that should be used for toxicity evaluations are those that have detailed, peer-reviewed methods to ensure that the testing is conducted properly and that the data will be comparable with data from other studies that use the same methods. Many of the well-standardized tests are documented in methods or guidance manuals developed by the American Society of Testing and Materials (ASTM), the U.S. Environmental Protection Agency (EPA), and Environment Canada.

It is essential that the performance of laboratory testing be monitored using quality assurance and quality control procedures to document the quality of results and determine whether the results are acceptable for their intended use (e.g., U.S. EPA 1991b; Moore et al. 1994). The major quality assurance and quality control procedures for toxicity testing are as follows:

- The use of negative controls to ensure that the test organisms are suitably healthy for testing
- The use of positive controls (i.e., reference toxicants) to ensure that the test organisms are suitably sensitive to toxic chemicals
- The monitoring of key test conditions (e.g., water temperature, dissolved oxygen) to ensure that the test results are not influenced by factors other than chemical toxicity
- The evaluation of variability among replicates and possibly tests for outliers.

Certain factors intrinsic to natural sediment samples may confound the relationship between the concentrations of sediment contaminants and toxicity. The objective of sediment toxicity testing is to evaluate the response of the test species to target chemicals contained in the sediment sample. It is preferable that the species not be responsive to other sediment characteristics such as grain size or organic carbon content. If such responses occur, toxicity may be incorrectly attributed to target chemicals. Changes in the following factors can restrict the application of a particular test or have a confounding effect on test results:

- Sediment grain size
- Organic carbon content
- Oxidation-reduction conditions
- pH
- Alkalinity
- Temperature
- Turbidity
- Water hardness
- Ultraviolet light intensity
- Mold or pathogens.

Information on potential interferences in sediment toxicity tests is provided in Appendix D of the Technical Resource Document.

SAMPLING ISSUES

The collection of representative sediment samples is essential for ensuring that the results of the subsequent toxicity tests are indicative of the true conditions in the field. A representative sample is one that is collected in a relatively undisturbed state from the intended field location; one that is collected using an appropriate collection device; and one for which proper handling, preservation, and documentation procedures have been observed after collection. A deficiency in any one of the above elements can affect the integrity of the sample and thereby influence the results of the toxicity testing so that they are not indicative of the true field conditions. Each of these elements is described below.

Sample Location

Sediment samples should be collected as close to their intended locations as required to satisfy the study objectives. This usually means that accurate positioning methods should be used both to locate the station initially and to allow the station to be revisited, if necessary, for subsequent sample collection.

Sample Collection

Sediment samples should be collected using appropriate collection devices that ensure that the sediment is collected with minimal disturbance, that an adequate penetration depth is achieved, and that the sample is retrieved in a relatively undisturbed state. When the results for different samples will be compared with each other (e.g., along spatial gradients, during different time periods), it is advisable to use the same sampling device to collect all of the samples so that biases that may occur from the use of different sample collection devices can be avoided.

Sediment samples should be collected in a relatively undisturbed state. The most common means of disturbing sediments are by excessive bow wake in front of the sample collection device immediately before the device contacts the sediment and by leakage of overlying water from the sample collection device as it is retrieved. In both cases, fine-grained surface organic material can be lost from the sample, thus biasing the grain-size characteristics of the sample toward the coarse mineral fraction.

Sample Handling

Sediment samples should be subsampled and homogenized in a controlled and noncontaminating manner. To avoid contaminating sediments, all utensils should be constructed of stainless steel and should be chemically cleaned between different samples. Sediments should be removed from the sampling device in an unbiased manner, especially if the characteristics of the sediments are heterogeneous. In general, all of the sediment collected from a station that will be evaluated for toxicity, chemical concentrations, and sediment conventional variables should be pooled and homogenized prior to being distributed to sample jars. This process ensures that the various kinds of analytical results will be related as closely as possible. Homogenization is considered complete when the sediments are visually uniform with respect to texture and color.

Sediments that will be analyzed for unstable chemicals such as volatile organic compounds and acid volatile sulfides should not be homogenized prior to distribution because the resulting sample disturbance could alter those chemicals. Therefore, sediments that are suspected to contain unstable chemicals should be transferred directly from the sampling device to the sample jar, leaving minimal or no headspace. To provide representative sediments for unstable chemicals, it is best to take several random subsamples from various parts of each sediment sample.

Chemicals in interstitial water samples are likely to be modified during the collection and preparation process. Guidance on sample collection procedures for interstitial water samples is contained in Burton (1992).

Sample Preservation

Sediment samples should be preserved in a manner that maintains their integrity during storage prior to laboratory analysis and should be analyzed within the specified maximum holding times. Proper sample preservation is essential for minimizing potential changes in the toxicity of the sediments during storage. Typically, sediments should be held unfrozen at 4°C for toxicity tests that rely on exposure to whole sediments. The maximum allowable holding time prior to testing for those sediments is generally specified as 2 weeks. However, sediment characteristics change during storage, even under controlled conditions. Therefore, it is preferable to conduct toxicity testing as soon as possible after field collection. For toxicity tests that rely on exposure to sediment extracts, sediments can sometimes be stored frozen if the test method allows.

Sample Documentation

All field collection procedures should be properly documented to verify that appropriate methods were used and that the security of samples was maintained at all times. Proper documentation generally involves the use of a field logbook to record pertinent information for each station and sediment sample and the use of chain-of-custody forms to document the transfer of samples among different parties.

DATA ANALYSIS ISSUES

Toxicity data should be analyzed using methods that are appropriate for the kinds of data available. To ensure that the data are appropriate for the planned analytical methods, it is essential that those methods be identified when the toxicity study is being designed. The study design specifications can then be tailored to provide data that are appropriate for the planned data analysis methods.

In monitoring programs and cleanup assessments, hypotheses regarding the toxicity of sediments at a specific site are usually tested using statistical methods to provide an objective analysis of the data. Statistical analysis allows quantification of the uncertainty associated with test results and typically ensures that several investigators would reach the same conclusions if each one analyzed the data separately. Statistical analyses are especially important for determining whether the results of a site-specific toxicity test differ significantly from the reference sediment results. For example, a statistical approach might be used to evaluate the following null hypothesis: *There is no significant ($P>0.05$) difference between the site and a reference area in sediment toxicity as measured by the amphipod mortality test.* Rejection of the null hypothesis based on statistical comparison of the sediment toxicity test data from the site with data from the reference area generally leads to acceptance of the alternative hypothesis that the site sediments are toxic (at least as measured by a laboratory toxicity test). However, a regulatory program may require further analyses to assess the implications of the laboratory test results. A specific probability level ($P\leq0.05$ in the example above) is associated with the statistical test to quantify the level of confidence in the result if the null hypothesis is rejected. If the null hypothesis is not rejected, acceptance of the alternative hypothesis of "significant toxicity" may be supported by a further evaluation technique known as statistical power analysis that determines the probability of detecting a specified level of toxicity.

When designing a study for which the data will be analyzed statistically, there are two major considerations that should be addressed. One consideration is whether to use parametric or nonparametric statistical methods. The parametric tests assume a normal frequency distribution for the data, whereas the nonparametric tests make no assumptions about the form of the data distribution. Typically, it is desirable to use parametric methods because they generally are more powerful than nonparametric methods. However, it is important to evaluate the assumptions of the selected statistical test for each data set. If one or more parametric assumptions are not met, the data can be transformed and the assumptions can then be evaluated for the transformed data. If the transformed data satisfy the assumptions, they can be evaluated using parametric methods. Otherwise, nonparametric methods should be used to evaluate untransformed data.

A second consideration is the specific kind of statistical test that will be used to analyze the data. The kind of test is usually determined by the study objectives. If the objective is to compare the toxicity results between a potential problem area and a reference area, analysis of variance can be used to conduct the evaluation. If the objective is to evaluate whether a gradient of toxicity exists with distance from a potential problem area, an analysis of variance or a correlation analysis can be used. In many cases, the kinds of statistical procedures that are used to analyze toxicity test results will be specified in a permit. Other details such as sample comparisons, statistical confidence levels, and other interpretive guidelines may also be specified. For an evaluation of permit specifications or design of testing programs refer to Gad and Weil (1986).

GLOSSARY

Acute toxicity — The ability of a chemical to cause a toxic response in organisms immediately or shortly after exposure to the chemical.

Adverse effect — An impairment of biological functions or description of ecological processes that results in unfavorable changes in an ecological system.

Amphipod — A small shrimp-like member of one subgroup of the large group of animals called Crustacea, which includes crayfish, lobsters, shrimps, and crabs.

Aquatic — Living or growing in water.

Benthic — Pertaining to, or associated with, the bottom of a body of water.

Biomass — The total weight of live organisms in a sampled population.

Biotic group — A group of related organisms with generally similar body structure and function.

Chronic toxicity — The ability of a chemical to produce a toxic response when an organism is exposed over a long period of time, generally corresponding to a substantial part of the organism's life cycle.

Concentration — The amount of a chemical expressed relative to amount of environmental medium (e.g., μg/L [micrograms of chemical per liter of water] or μg/g [micrograms of chemical per gram of sediment]).

Control sediment — A sediment essentially free of chemicals and compatible with the biological needs of the test organisms such that it has no discernable influence on the response being measured in the test. Control sediment may be the sediment from which the test organisms are collected or a laboratory sediment, provided the organisms meet control standards. Test procedures are conducted with the control sediment in the same way as the reference sediment and test material. The purpose of the control sediment is to confirm the biological acceptability of the test conditions and to help verify the health of the organisms during the test. Excessive mortality in the control sediment indicates a problem with the test conditions or organisms and can invalidate the results of the corresponding test.

Ecosystem — An ecological community, together with its physical habitat, considered as a unit.

Embryo	A plant or animal in the very early stages of development following fertilization of the egg.
Elutriate	A liquid solution used for toxicity testing, which is prepared by adding water to the sediment, shaking, and centrifuging to separate the solids.
Endpoint	The biological or ecological unit or variable being measured or assessed. The number of organisms dead at the end of an exposure is a lethal endpoint.
Epibenthic	Inhabiting the sediment surface, or closely associated with the sediment surface, rather than dwelling buried within the sediments.
Estuarine	Surface water containing greater than 0.5 parts per thousand (ppt) salinity and less than 28 ppt salinity.
Exposure	Contact between an organism and a chemical in the environment.
Fresh water	Surface water containing less than or equal to 0.5 ppt salinity.
Foundation species	A species that provides important physical habitat for other species in a biological community (e.g., marsh grass).
Hardness	A measure of the calcium and magnesium concentrations in water.
In situ	In the natural or original position (occurring in nature, and not in the laboratory).
Infaunal	Refers to animals living in the sediments, including such forms as worms and clams.
Interference	Physical elements or chemical compounds that cause bias in the results of a toxicity test.
Keystone species	A species that controls the species composition and relative abundances of species in a community by its predatory (or grazing) effects (e.g., by grazing on kelp, purple urchins prevent the establishment of kelp beds and maintain open rocky subtidal communities).
Interstitial water	Water that fills the spaces between sediment particles. Often referred to as "pore water."
Larval	Relating to the juvenile form of certain invertebrate animals that must undergo metamorphosis before assuming adult characteristics.

Lethal	Causing death; mortality (or survival) is the endpoint for lethal toxicity tests.
Life stage	A developmental stage of an organism (e.g., egg, larva, embryo, juvenile, adult).
Macroinvertebrate	An invertebrate (without a backbone) organism visible to the naked eye (e.g., >1.0 mm). Often refers to animals such as insects, worms, clams, and snails.
Marine	Surface water containing 28 ppt salinity or greater.
Medium (plural: media)	The substance in which a chemical may exist. Air, sediment, and water are all media.
Midge	A group of true flies (similar to mosquitos) that have aquatic larvae and non-biting adults. They are one of the most abundant groups of aquatic insects.
Monitoring	Periodic testing of water and sediment quality or of biota to verify continued compliance with the requirements of a discharge permit or other authorization.
Nektonic	Refers to the nekton, the group of active swimmers that are capable of strong, independent movement in the water. Examples include many juvenile and adult fishes and large invertebrates (e.g., squid).
Organism	An individual plant or animal.
Ovigerous	Refers to females bearing eggs.
Planktonic	Refers to the plankton, the group of small plants and animals that are weak swimmers and tend to drift with the current.
Population	A group of individuals of the same species interacting within a given habitat.
Precision	The ability to replicate a value; the degree to which observations or measurements of the same property, usually obtained under similar conditions, conform to themselves. Usually expressed as standard deviation, variance, or range.
Quality assurance and quality control	A system of procedures, checks, audits, and corrective actions to ensure that all research design and performance, environmental monitoring and sampling, and other technical and reporting activities are of the highest achievable quality.

Reference sediment	A sediment, substantially free of chemicals, that is as similar as practicable to the grain size of the test material and the sediment at the disposal site and that reflects the conditions that would exist in the vicinity of the site had no anthropogenic activity ever taken place but had all other influences on sediment condition taken place.
Reference area	An area that has similar characteristics to a site being evaluated but that is unaffected by chemicals of potential concern. The reference area is compared to the site to assess the effects of chemicals of potential concern.
Route	The mechanism of contact between an organism and a toxic chemical (e.g., ingestion or dermal contact).
Site-specific	Of or relating to a particular area or location.
Sediments	Material, such as sand, silt, or clay, suspended in or settled on the bottom of a water body.
Sublethal	Causing an endpoint other than death; growth is a sublethal endpoint in toxicity tests.
Terrestrial	Living or growing on land.
Toxicity test	A test in which organisms are exposed to chemicals in a test medium (e.g., waste, sediment, soil) to determine the effects of exposure.
Trophic	Relating to food or feeding relationships. Trophic levels consist of producers (plants), herbivores or primary consumers, carnivores or secondary consumers, and top carnivores or tertiary consumers.

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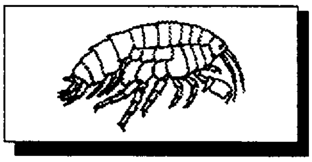
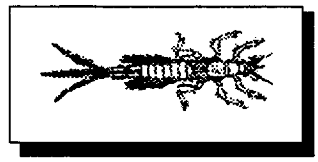
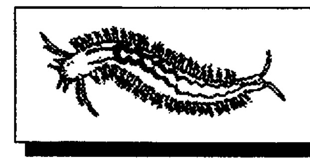
Technical Resource Document:

Evaluation of Sediment Toxicity Tests for Biomonitoring Programs







ABSTRACT

Sediment toxicity test methods are available for marine, estuarine, and freshwater sediments and organisms. The methods can be used for a variety of purposes: for example, assessment of existing environmental conditions, monitoring changes with time, or for NPDES permit compliance. Use of inappropriate test methods or species for a given purpose can impact the toxicity results and their interpretation. This Technical Resource document has been prepared as a detailed resource for environmental support staff for petroleum industry facilities (refineries, marketing terminals, and production locations) in their selection of sediment toxicity test methods and species which are reliable, scientifically valid, and appropriate for the habitat. Test methods are evaluated for their reliability, ecological relevance, exposure relevance, availability, interferences, and ability to discriminate toxicants. The tests are categorized by habitat type (marine, estuarine, or freshwater) and test endpoint (lethal or sublethal). Species' descriptions, test protocols, and documentation of method evaluations are contained in appendices. A companion report (User's Guide) has been prepared to provide an introduction to sediment toxicity test methods and to summarize their use.

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EXECUTIVE SUMMARY

Federal and state agencies are developing and implementing water quality-based approaches to regulate permitted wastewater discharges. As part of this overall approach, sediment toxicity testing may be required as part of discharge monitoring programs. Toxicity tests of sediments provide an integrated measure of sediment contamination that eliminates much of the uncertainty associated with theoretical predictions of sediment toxicity at a specific site. The value of the biological effects-based approach can be diminished, however, by using inappropriate test methods or species or by not considering the many physical and chemical factors that can affect biological testing. To date, the selection of a sediment toxicity test(s) for a specific site has not been a straightforward process. The selection process is complicated by the constant introduction of new test species and methods. There have been few objective comparisons among alternative tests and there is a general lack of recommendations for the most appropriate sediment test procedures for a given situation.

OBJECTIVES AND APPROACH

The purpose of this document is to provide a resource tool that will enable environmental staff at petroleum facilities to readily select sediment toxicity tests and test methods that are reliable, scientifically valid, and appropriate for a specific site.

The test methods and species identified during the information review were classified into six groups based on the following physical and biological test characteristics:

- Habitat type (marine, estuarine, or fresh water)
- Test endpoint (lethal or sublethal).

Each toxicity test within each of the six groups is summarized in a classification table that also contains information on the organisms, including the broad taxonomic group, common name, scientific name, and life stage of the species used in the test; exposure type (whole sediment, interstitial water, water elutriate, or solvent extract); the exposure duration; and the primary literature references. In some cases, tests differed only by the species, life stage, or exposure period used. Even so, the tests were maintained as separate entities for the evaluation because minor differences in organisms or exposure duration may significantly influence test sensitivity.

The tests in the initial screening were evaluated according to various technical criteria. An overall technical rating was based on the following evaluation criteria:

- Reliability
- Ecological relevance
- Exposure relevance
- Availability
- Interferences
- Chemical discrimination.

RECOMMENDED TESTS

This report contains a series of evaluation tables that describe the group of relevant toxicity tests for each habitat (marine, estuarine, or freshwater) and for each endpoint type (lethal and sublethal). In most cases, the highest ranking tests were the ones based on the exposure of infaunal organisms to whole sediments. These kinds of tests were ranked highly because they rely on exposure conditions that closely mimic field conditions. Many of the test species are available by field collection during most of the year. The highly ranked marine infaunal tests predominantly used amphipods as test organisms, whereas most of the highly ranked freshwater infaunal tests predominantly used insects (mayfly nymphs and midge larvae) as test organisms.

In many cases, the lowest ranking toxicity tests were those that rely on the exposure of planktonic organisms to whole sediments. Those tests have a relatively low exposure relevance because the test organisms are rarely exposed to sediments in the field. In addition, the tests are sensitive to potential interferences from sediments in the test chambers.

In some cases, a highly ranked lethal test was also a highly ranked sublethal test. The inclusion of a test in both categories is a particularly desirable characteristic when both lethal and sublethal endpoints will be evaluated in a biomonitoring program. By using the same species to evaluate both endpoints, potential interpretive problems related to interspecific differences in factors such as sensitivity to toxicity can be avoided.

USER'S GUIDE

A companion document, *User's Guide: Evaluation of Sediment Toxicity Tests for Biomonitoring Programs* (PTI, 1994) has been prepared to provide an introduction to sediment toxicity testing and to describe how to use this resource manual for those readers who are unfamiliar with sediment toxicity testing.

INTRODUCTION

The U.S. Environmental Protection Agency (EPA) has established a water-quality based approach to control potentially toxic substances in wastewater effluents that are discharged to inland and coastal waters (U.S. EPA 1991c). Toxicity testing of effluents and ambient water or sediments in the vicinity of wastewater discharges is a key element in predicting the potential biological effects of discharges and in monitoring existing impacts. For example, requirements for toxicity testing of whole effluents are being incorporated into many wastewater discharge permits under the National Pollutant Discharge Elimination System (NPDES). At the same time, development of sediment toxicity tests that could be used to monitor discharge sites is proceeding at a relatively rapid pace at the federal and state levels (U.S. EPA 1992).

Toxicity tests of sediments provide an integrated measure of sediment contamination that eliminates much of the uncertainty associated with theoretical predictions of sediment toxicity at a specific site. The value of the biological effects-based approach can be diminished, however, by using inappropriate test methods or species, or by not considering the many physical and chemical factors that can affect biological testing. To date, the selection of a sediment toxicity test for a specific site has not been a straightforward process.

OBJECTIVES

The purpose of this resource document is to provide information that will enable environmental staff at petroleum facilities to select sediment toxicity tests and test methods that are reliable, scientifically valid, and appropriate for a specific site. This document also summarizes the technical requirements and appropriate conditions for use of sediment toxicity test methods anticipated for future NPDES permit compliance. This document was developed specifically for use by petroleum industry operations (refineries, marketing terminals, and production facilities) that have effluent discharges to surface waters. However, this Technical Resource Document and the User's Guide contain information that is applicable to other industries and could be used by any wastewater discharger.

USE OF DOCUMENT

The first step when using the decision framework and information in this document is a **review of available information** on the ecological characteristics of the discharge site. Available data on site-specific chemicals and physical properties of the sediments can be useful in selecting test species that are sensitive to the chemicals of concern at the site, but that are minimally affected by the natural properties of the sediment (e.g., grain size, organic carbon, ammonia). An equally important step in the selection of sediment toxicity tests is a **thorough understanding of the applicable regulatory requirements** that are driving the testing program. Regulatory programs frequently include explicit requirements that immediately limit the field of potential toxicity tests. These confining factors can include specifications for lethal or sublethal tests, exposure duration, seasons for testing, single species vs. a battery of species for testing, and data quality objectives.

This report contains a series of evaluation tables that describe the group of relevant toxicity tests for each habitat system (marine, estuarine, or fresh water) and for each test type (lethal and sublethal). These tables summarize the key information used in selecting toxicity tests. **Users enter the evaluation tables for a given system and test type** (e.g., estuarine/lethal). In each evaluation table, candidate tests are ranked from best overall tests to least appropriate tests. In most cases, the higher ranked tests may have very similar overall scores. Therefore, **the user should select the most appropriate high-ranked test** for a particular program based on a consideration of site-specific factors and regulatory considerations. The selected test should match the site-specific requirements for the biotic group and geographic zone.

The candidate toxicity test(s) that were tentatively selected in the previous step should be compared with the regulatory requirements identified. If the test (or tests) meets these requirements, then the selection process is complete and the actual test (or tests) can be performed. If the selected toxicity test (or tests) does not meet the requirements of the applicable regulatory program, then users have the option of entering into negotiations with the regulatory agencies. The information provided in the evaluation tables and the appendices will be useful in developing the arguments for and against particular tests. Detailed guidance on test selection is provided in a companion document, *User's Guide: Evaluation of Sediment Toxicity Tests for Biomonitoring Programs* (PTI, 1994).

USER'S GUIDE AND RELATED DOCUMENTS

A companion document, *User's Guide: Evaluation of Sediment Toxicity Tests for Biomonitoring Programs* (PTI, 1994) has been prepared to provide an introduction to sediment toxicity testing and to present how to use this resource manual for those readers who are unfamiliar with sediment toxicity testing. The User's Guide contains descriptions of habitat type, sediment test systems, and biological endpoints. Site-specific concerns are identified to aid in test selection. Lastly, brief summaries of sampling and data analysis issues are presented.

The Society of Environmental Toxicology and Chemistry has recently released a guidance document on use of sediment toxicity tests (Hill *et al.* 1993). Other useful review documents include Dillon and Gibson (1990), Burton (1991), and U.S. EPA (1992).

REPORT ORGANIZATION

The next section presents the approach used to compile and evaluate information on sediment toxicity test methods. The following sections classify available test methods using technical criteria. Technical evaluation results and guidance on application of toxicity tests in the context of NPDES programs are then provided in a sequence of tables for each system and toxicity test type (e.g., freshwater/sublethal). The highest ranked tests are shown first. Supporting summary materials are in appendices. Appendix A classifies the available sediment toxicity tests that were considered in this evaluation. Appendix B presents profiles of the species commonly used in the tests that ranked highest in the evaluation. Appendix C provides an overview of the key test methods. Appendix D is a summary table of the rationale for the evaluation results, organized by evaluation criteria. Key technical terms used herein are defined in the *Glossary*.

TEST SCREENING APPROACH

Toxicity test information, the classification of tests, the selection of tests for evaluation, and the development and application of evaluation criteria are described below.

INFORMATION REVIEW

The sources consulted to identify available test methods and species included Current Contents (a compilation of information contained in agricultural, biological, and environmental science journals); a bibliography of the Ecological Risk Assessment Library at PTI Environmental Services; and recent reviews of test methods, including Giesy and Hoke (1990), Burton (1991, 1992a), Burton *et al.* (1992), Lamberson *et al.* (1992), Bennett and Cubbage (1992), and the proceedings of the EPA workshop on *Tiered Testing Issues for Freshwater and Marine Sediments* (U.S. EPA 1992). Major documents discussing test methods and test strategies (e.g., U.S. EPA 1991b; U.S. EPA and U.S. COE 1991, 1993; Environment Canada 1990a–e, 1992a–f) were also reviewed to identify candidate tests. Finally, information from colleagues and agencies that are conducting ongoing investigations of test methods was collected and used to identify relatively new tests that show promise for testing of contaminated sediments.

CLASSIFICATION OF TESTS

The toxicity test methods and species identified during the information review were classified into six groups based on a series of physical and biological test characteristics. The objective of this step was to identify each group of test methods that have common characteristics and that, therefore, should be considered as possible alternatives for use at all facilities with similar receiving waters. The characteristics used to classify available test methods and species into the six groups are:

- Habitat type (marine, estuarine, or freshwater)
- Toxicity test endpoint (lethal or sublethal).

Habitat type is a key physical factor in narrowing the list of appropriate test species for use at a facility. For example, few marine sediment toxicity tests will be applicable at a facility that discharges to fresh water (one exception is the Microtox® test, which can be adjusted for different salinity regimes). For purposes of this study, habitat categories were defined as follows:

- Marine ($\geq$28 ppt)
- Estuarine ($>$0.5 and $<$28 ppt)
- Freshwater ($\leq$0.5 ppt)

Because the division between habitat categories is an artificial distinction, use of a particular habitat designation for a test in this report should not necessarily preclude the application of a test to sediments in other habitats. For example, some tests that are classified as marine tests may be applied to high salinity estuarine sediments, and in some cases, adjusting the salinity of a sediment sample to allow the use of a particular test may be appropriate. The test endpoint is a key biological factor in comparing and evaluating performance data among toxicity tests and satisfying regulatory requirements. Each toxicity test is summarized in a classification table that also contains information on the organisms (see *Classification of Available Test Methods* section and Appendix A).

SELECTION OF TESTS FOR EVALUATION

All of the individual toxicity tests identified in this review are listed in the classification tables (Appendix A). To facilitate detailed consideration of the most promising tests, tests were selected for further evaluation based on the following criteria:

- Tests recommended in guidance documents issued recently by national agencies in the United States and Canada (e.g., Dillon and Gibson 1990; U.S. EPA and U.S. COE 1991, 1993; U.S. EPA 1991b, 1992; Environment Canada 1990a–e, 1992a–f)

- Tests recommended in recent evaluations of sediment toxicity tests (e.g., Dillon and Gibson 1990; Giesy and Hoke 1990; Bennett and Cubbage 1992)
- Tests that are under development in several laboratories and show promise for application to NPDES sediment testing for petroleum refineries (e.g., relatively new tests included in EPA research and development; U.S. EPA 1992).

Tests that were not evaluated (Appendix A, Attachment 1) included relatively old ones (>10 years old) that are generally not used now, very new ones that need substantial method development, and tests with inadequate specification of the method to classify them.

EVALUATION CRITERIA AND APPROACH

Criteria to evaluate sediment toxicity tests include repeatability, precision, dose-responsiveness, discrimination among chemicals or sites, sensitivity, ecological relevance, appropriateness of the sample phase (e.g., particulate, extract), exposure period, relationship to field effects, availability, ease of use, and stage of method development (Pastorok and Becker 1989; Giesy and Hoke 1990; Burton 1991). An overall technical rating was based on the following technical evaluation criteria:

- Reliability
- Ecological relevance
- Exposure relevance
- Availability
- Interferences
- Chemical discrimination.

Each test was rated according to each criterion using a scoring system of 0 to 4, with 4 being the most favorable score. The overall technical rating was derived by summing the scores for the individual criteria. The scores for "interferences" and "chemical discrimination" were weighted by a factor of 0.5 to reduce their influence on the overall rating. Less information was available for these two criteria and, therefore, confidence in their scores was lower than those for other criteria. Because the cost of a given sediment toxicity test varies greatly with its status for regulatory use and with the laboratory conducting the test, cost was not considered in the overall rating. Details of the scoring system used to rank the toxicity tests are presented in Appendix D.

Factors considered in evaluating the tests relative to the technical criteria listed above and relative to the regulatory status of tests are described in more detail in the following sections.

Reliability

The reliability of a test method is high when the endpoint can be measured accurately, the results are repeatable, and the negative control results generally meet quality assurance criteria. High reliability ratings were assigned to tests that have been subjected to an interlaboratory comparison and found to have an acceptable level of variability within and among laboratories (e.g., ≤50 percent coefficient of variation). However, most sediment toxicity tests have not undergone interlaboratory studies. A test method was assigned low reliability if determination of the endpoint involves a high degree of subjectivity, if the response is easily biased by laboratory artifacts (e.g., organism stress due to confinement), or if repeated tests yield highly variable results. For example, many lethal tests involve a relatively straightforward determination of whether a fish or larger invertebrate is dead or alive. Such situations have minimal potential for error or bias on the part of the observer. Alternatively, some sublethal tests have a relatively subjective endpoint such as "abnormality" of a microscopic larvae. In such cases, the potential for individual bias is much greater. Tests were also assigned a low rating for reliability if the negative controls are prone to failures (e.g., high susceptibility of test organisms to laboratory stress).

Ecological Relevance

The ecological relevance of a test method is high if the results of a test method are directly applicable to indigenous species under field conditions. A test method has low ecological relevance if there is little or no basis for associating the method to the field conditions or if the method results display no concordance with the responses of indigenous organisms. Field validation studies provide the best evidence for judging ecological relevance, but most sediment toxicity tests have not undergone field validation tests. In lieu of information on field validation, any test that uses a species that is commercially harvested (e.g., oysters) or a species that provides habitat structure for other species (e.g., marsh grass) was assigned a high rating for ecological relevance.

Exposure Relevance

The exposure relevance of a test method is high when the pathway of exposure used in a test is analogous to exposure under field conditions (e.g., whole sediment exposure for chemicals that may be taken up by several routes such as pore water and ingestion of sediment particles). Other test procedures involve exposure of water column organisms to extracts of sediments or sediment interstitial water, which is not necessarily a good analog of field conditions. Exposure relevance is, therefore, low when test conditions mimic only some of the exposure pathways applicable to field conditions. The general ecological niche of the test organism was also considered in assessing exposure relevance. Thus, exposure of infaunal test organisms to whole sediment would have a higher exposure relevance than exposure of planktonic organisms to any of the test media.

Availability

The availability of a test method is high when test organisms can be easily obtained, the method is standardized and well documented, and commercial laboratories routinely perform the test. Test organisms that are easily cultured or are available from the field throughout all seasons in a broad geographic area were assigned high ratings for this criterion. Test methods that are constrained by a limited supply of organisms (by season or geography) or a lack of experienced laboratories were assigned low scores for availability. A low rating was assigned to test methods that are still in the early stages of documentation and standardization; these methods will be difficult or costly to develop and use as quickly as routine methods.

Interferences

The interference criterion is a measure of the extent of confounding physical or chemical factors. The objective of sediment toxicity testing is to evaluate the response of the test species to target chemicals contained in the sediment sample. It is preferable that the species not be responsive to other sediment characteristics such as particle size or organic content. If such responses occur, toxicity may be incorrectly attributed to target chemicals. Changes in the following factors can restrict the application of a particular test or have a confounding effect on test results:

- Sediment grain size
- Organic carbon content
- Oxidation-reduction conditions
- pH
- Alkalinity
- Temperature
- Turbidity
- Water hardness
- Ultraviolet light intensity
- Mold or pathogens.

A high rating for interference was assigned to test methods that have a low susceptibility to interferences. A low rating for interference was assigned to test methods that exhibit a large number of interferences or a narrow range of applicable environmental conditions.

Chemical Discrimination

The chemical discrimination of a test method is high if the test results are dose-responsive over at least a moderate range of chemical concentrations. Test methods that exhibit high chemical discrimination are expected to be useful in defining gradients of sediment toxicity in the environment. Test methods that are either insensitive or always highly-sensitive to chemical contamination were assigned a low rating for chemical discrimination.

Regulatory Status

The rating for regulatory status was based on information from national and regional EPA offices and whether a test was recommended in guidance documents for potential use in NPDES programs, cleanup assessments, baseline monitoring, or dredged material testing. The guidance documents considered as the basis for rating regulatory status included the method documents issued by the Canadian government (Environment Canada 1990a–e, 1992a–f), the dredged material testing documents issued by United States government agencies (U.S. EPA and U.S. COE 1991, 1993), and a major research and development planning document issued by EPA (U.S. EPA 1992). If a test was included in 3–4 of these document categories, it was assigned a rating of "high" for regulatory status. If a test was included in 1–2 of these document categories, it was assigned a rating of "medium." Toxicity tests that were not included in these documents and were not known to be recommended for use in current regulatory programs were assigned a rating of "low."

CLASSIFICATION OF AVAILABLE TEST METHODS

The available toxicity test methods were classified by type of habitat (marine, estuarine, and fresh water) to which each method applies and the general endpoint type (lethal or sublethal) specified for each test. This classification scheme resulted in the following six major categories of tests:

- Marine lethal
- Marine sublethal
- Estuarine lethal
- Estuarine sublethal
- Freshwater lethal
- Freshwater sublethal.

Within each category, tests are distinguished by three key characteristics: 1) the exposure medium, 2) characteristics of the test organisms, and 3) the test duration.

Using the classification criteria and key characteristics described above, 336 unique tests were identified (Appendix A). Each test was assigned a number to allow users of this document to track a given test through the various evaluation tables. In many cases, several of the specified tests were actually variations of a single test method and were assigned the same test number. For example, if the test species is tolerant of both marine and estuarine conditions and if both lethal and sublethal endpoints are possible (e.g., the amphipod test using *Ampelisca abdita*), then the test can be classified four different ways.

CLASSIFICATION CRITERIA

Use of general habitat type and endpoint type to classify sediment toxicity tests provides a broad classification scheme. This approach should facilitate review of evaluation results and selection of tests at specific sites.

Habitat Type

The primary characteristic that distinguishes marine, estuarine, and freshwater habitat types is water salinity. Salinity strongly influences the distributions of most of the test organisms. In some cases, test organisms are tolerant of both marine and estuarine conditions or both estuarine and freshwater conditions. However, few test organisms tolerate both marine and freshwater conditions.

Endpoint Type

The two major types of endpoints for most toxicity tests are lethal and sublethal. The lethal endpoint is based on the percentage of test organisms that die during the exposure period. The use of mortality as a test endpoint has several advantages. Mortality represents an unambiguous adverse effect (i.e., death). In addition, the ecological significance of the endpoint is relatively certain. If the test organisms cannot survive in association with the test sediments, it is likely that significant alterations of aquatic assemblages would be found in the environment. Although mortality may appear to not be an environmentally protective endpoint, tests that use a sensitive species may be protective of less-sensitive species.

Sublethal endpoints represent a wide variety of organism responses other than mortality. Some of the most commonly measured sublethal endpoints include:

- Reduced growth
- Reproductive effects
- Developmental abnormality
- Histopathological abnormalities.

The reliability of any sublethal endpoint test depends on the use of experienced laboratory personnel.

Reduced growth is generally measured in juvenile organisms (i.e., when growth is generally expected to be rapid) and can be estimated directly by measuring the size of organisms (e.g., length, biomass) prior to and following exposure to test media. Growth can also be represented as an instantaneous measurement called "scope for growth," which is based on physiological variables such as feeding rate, absorption efficiency, respiration rate, and excretion. Growth itself does not require extensive expertise to measure, but scope for growth does. Both measures of growth are quantitative and objective. However, the ecological relevance of reductions in organism growth is uncertain or varies considerably among test species. If organisms can maintain their normal level of fecundity, then no reductions in population levels may be experienced. A reduction in size of adult organisms may cause organisms to be less desirable to some predators and more desirable to others, thereby influencing trophic relationships. Growth reductions may also enhance the risk of being preyed on because organisms cannot reach a size large enough to escape predation.

Reproductive effects are frequently measured in adult female organisms as number of eggs per individual, percentage of ovigerous individuals, and time to sexual maturity. These measurements can be made in a relatively quantitative and objective manner. Reproductive effects may be more ecologically relevant than growth because they imply that the local supply of recruits to adult populations may be reduced. However, adult populations could be sustained by recruitment of pelagic larvae from surrounding areas or immigration by adult organisms.

Developmental abnormalities are generally evaluated in the early life stages (e.g., embryos, larvae) of the test organisms. Bivalve molluscs and echinoderms are two groups of organisms that are frequently used to evaluate developmental abnormalities. Because the determination of abnormal development can be somewhat subjective, the abnormality endpoint can be somewhat ambiguous. Much of the potential subjectivity of the abnormality endpoint can be avoided by standardizing and clearly defining abnormalities. The ecological relevance of the abnormality endpoint is uncertain. Although the presence of abnormalities in early life stages suggests that recruitment to adult assemblages may be curtailed, it is possible that those assemblages could be maintained by a limited level of recruitment or by immigration of adult organisms (for mobile species). Abnormality endpoints are typically selected to represent conditions that limit the successful growth or reproduction of individuals. Because larvae represent a sensitive life stage and because abnormalities may be expected to occur prior to the onset of mortality, the abnormality endpoint can generally be considered environmentally protective.

Histopathological abnormalities include measures of degeneration, necrosis, and other abnormalities in cells and tissues. The determination of these disorders requires a highly trained pathologist and frequently is subjective. The ecological relevance of histopathological disorders are uncertain because they may or may not influence an organism's life functions. For example, a malignant tumor will lead to negative consequences. With other kinds of abnormalities, the affected individual may experience no negative consequences and may eventually recover.

KEY CHARACTERISTICS

Exposure Medium

The kind of exposure medium was used to classify the various toxicity tests because each kind of exposure medium has favorable and unfavorable characteristics that can profoundly influence the toxicity test results. The four kinds of exposure media considered are whole sediments, sediment elutriates, interstitial water, and sediment extracts (Table 1). The first two media are commonly used in tests, whereas use of the latter two media is infrequent.

The use of whole sediments is probably the most realistic exposure scenario because it mimics the manner in which most organisms are exposed to chemicals in the environment. Ankley *et al.* (1991) and Green *et al.* (1993) showed that toxicity may be detected in pore water or interstitial water tests

TABLE 1. SEDIMENT PHASES USED IN TOXICITY TESTS

Phase	Strengths	Weaknesses	Routine Uses
Whole sediment	Use with all sediment types Relative realism high Holistic (whole) vs. reductionist toxicity approach (water, interstitial water, elutriate, extract) Determine sediment quality criteria Use site or reconstituted water to isolate sediment toxicity	Some physical/chemical/microbiological alteration from field collection Dose-response methods tentative Testing more difficult with some species and some sediments Few standard methods Indigenous biota may occur in sample	Rapid screen Chronic studies Initial surveys Sediment criteria
Sediment elutriate (water extractable)	Use with all sediment types Readily available fraction Mimics anoxic toxic environmental process Large variety of available assay endpoints Exposure methods relatively standardized Determine dose response	Ecosystem realism uncertain: Only one oxidizing condition used; only one solid:water ratio; exposure for extended period of one-phase condition that never occurs *in situ* or never occurs in equilibrium *in situ* Extract conditions vary with investigator Filtration affects response, sometimes used	Rapid screen Endpoints not possible with whole sediments Dredging evaluation
Interstitial water	Relevant direct route of uptake for most species Large variety of available assay endpoints Exposure methods relatively standardized Determine dose response Determine sediment quality criteria	Cannot collect interstitial water from some sediments Limited volumes can be collected efficiently Optimal collection method unknown, constituents altered by all methods Exposure phase altered chemically <u>and</u> physically when isolated from whole sediment Relevance for some organisms uncertain	Rapid screen Endpoints not possible with whole sediments Initial surveys Sediment criteria
Sediment extract (organic solutes)	Use with all sediment types Sequentially extract different degrees of bioavailable fractions Variety of available assay endpoints Determine dose response	Ecosystem realism uncertain: Bioavailability unknown, chemical alternation Organic solvent extracts may interact with chemicals in sediment to produce interference (toxicity)	Rapid screen Unique endpoints, so component of test battery

Source: Adapted from Burton (1991).

when whole sediments are nontoxic. The degree to which the sampling of interstitial waters or the eluviation process modifies the toxicity of the sample is often unknown. Harkey *et al.* (1994) concluded that the bioavailability of nonionic hydrophobic compounds cannot be accurately predicted by exposures to elutriates or interstitial water.

For most whole sediment tests, the sediments are carefully placed in the exposure chamber with minimal disturbance; the chamber is then filled with test water. The water is obtained from a source known to be uncontaminated, or in the case of seawater, is generated using commercial "artificial seawater" salts. After the whole sediments and overlying test water have equilibrated, the test organisms are introduced and the test is initiated. At the end of testing, the test organisms are sorted from the sediment and the test endpoint results are determined. Generally, infaunal test organisms are expected to have the highest potential for exposure to chemicals because they live within the sediments. By contrast, planktonic and nektonic test organisms are expected to have a relatively low exposure potential because they spend all or much of their time in the water column above the sediments. Epifaunal organisms are expected to have an intermediate exposure potential because they reside primarily at the sediment-water interface.

Sediment elutriates are prepared by mixing sediments and test water for a prescribed period of time and then removing the sediments by methods such as filtration, centrifugation, and decanting after a settling period. The test organisms are then introduced to the test water in the absence of sediments. Elutriates are useful for representing the exposure to chemicals that can occur after sediments have been resuspended into the water column or after they have passed through the water column as part of dredged material disposal operations. The use of an elutriate as an exposure medium is probably most realistic for planktonic and nektonic test organisms because those organisms would generally remain in the water column after the contaminated sediments have settled to the bottom. The use of an elutriate for testing infaunal organisms is unrealistic because those organisms would be in direct contact with the contaminated sediments after they settle.

Interstitial water as an exposure medium is prepared by removing it from sediments by methods such as filtration and centrifugation. The test organisms are then introduced to the interstitial water in the absence of sediments. The use of interstitial water as an exposure medium is representative of one exposure route that may be experienced by infaunal organisms. The use of this medium is not representative of realistic exposure scenarios for epifaunal, planktonic, and nektonic organisms.

Sediment extracts are prepared by mixing sediments with an organic solvent that is capable of removing specific kinds of chemicals from the sediments. After the extraction process is completed, the sediments are removed by methods such as filtration, centrifugation, and decanting after a settling period. The test organisms are then either introduced to the extractant or the extracted chemicals are first exchanged with a less toxic carrier medium and then the test organisms are introduced to the carrier medium. In either case, sediments are absent from the exposure medium. The use of an extractant to prepare an exposure medium is generally considered an unrealistic exposure scenario because it does not mimic events that occur in the environment. However, the use of an extractant may be useful for providing a worst-case evaluation of sediment toxicity because it may release more chemicals from the sediments than would be released under normal conditions. A potential problem with the use of extract as exposure medium is the possibility that the extractant or solvent used as a carrier for the extract is potentially toxic to the test organisms and thereby capable of interfering with the test results.

Characteristics of Test Organisms

The key characteristics of the test organisms evaluated are biotic group, species, and life stage. Whether the test organisms are infaunal, epifaunal, planktonic, or nektonic affects the representativeness of the exposure scenarios. For example, Ankley *et al.* (1991) concluded that upper-water-column species such as fathead minnows (*Pimephales promelas*) and cladocerans (*Ceriodaphnia dubia*) are inappropriate organisms for use in whole sediment tests focused on evaluating *in situ* toxicity to benthic species. Because different species and different life stages often exhibit different levels of sensitivity to toxicity, those two variables can also influence the test results. For example, crustaceans are generally considered more sensitive to toxic chemicals than are polychaetes in marine and estuarine sediment tests. Insects are considered more sensitive than oligochaetes in freshwater sediment tests. In addition, early life stages (e.g., embryos, larvae) are typically more sensitive than later life stages (e.g., juveniles, adults).

Test Duration

Test duration is an important distinguishing characteristic for the various toxicity tests because it influences the likelihood that toxic effects will occur. Tests with relatively short exposure periods that do not cover a substantial portion of the life cycle of the test organisms are referred to as acute tests, and tests with longer exposure periods that cover a substantial portion of the life cycle of the test organisms are called chronic tests. Because of their shorter exposure period, acute tests are typically less sensitive for detecting toxicity than chronic tests. However, if the test species for an acute test is particularly sensitive or the test species for a chronic test is particularly insensitive, the acute test may be more sensitive than the chronic test.

EVALUATION FOR TEST SELECTION

Several general patterns were evident from the technical evaluation of sediment toxicity tests. In most cases, the highest ranking tests were the ones based on the exposure of infaunal organisms to whole sediments because: 1) they rely on exposure conditions that closely mimic field conditions; 2) most of the test species are available by field collection during most of the year; and 3) many of the tests have well-developed methods. Most of the highly ranked marine and estuarine infaunal tests were based on the use of amphipods as test organisms, whereas most of the highly ranked freshwater infaunal tests were based on the use of insects (mayflies and midges) as test organisms. These species groups are ecologically important, especially as key prey items for various fishes. In many cases, the lowest ranking toxicity tests were those that rely on the exposure of planktonic organisms to whole sediments. Those tests have a relatively low exposure relevance because the test organisms are rarely exposed to sediments in the field. In addition, the tests are sensitive to potential interferences from sediments in the test chambers. Many planktonic organisms are filter-feeders, and resuspended sediments may interfere with feeding mechanisms. Physical contact with whole sediments may also decrease the efficiency of locomotion and feeding in planktonic organisms.

In some cases, a highly ranked lethal test was also a highly ranked sublethal test. The inclusion of a test in both categories is a particularly desirable characteristic when both lethal and sublethal endpoints will be evaluated during the same monitoring program. By using the same species to evaluate both endpoints, potential interpretive problems based on interspecific differences in factors, such as sensitivity to toxicity, can be avoided.

The detailed results of the evaluation of sediment toxicity tests are presented in Tables 2 through 7. Within each table, the tests are ordered from the most desirable to the least desirable based on the previously discussed technical criteria (see *Test Screening Approach* for method used to derive ratings). Tables 2 through 7 also show the regulatory status for each toxicity test. The rating for regulatory status was determined primarily on the inclusion of a test in guidance documents for use of sediment toxicity testing in regulatory programs (Environment Canada 1990a–e, 1992a–f; U.S. EPA and U.S. COE 1991, 1993; U.S. EPA 1992). A rating of "high" for regulatory status indicates that a test was included in 3–4 of these document categories. A rating of "medium" indicates that a test was included in 1–2 of these document categories. Toxicity tests that were not included in these documents and were not known to be required for use in current regulatory programs were assigned a rating of "low."

In the remainder of this section, the highest ranking tests in each evaluation table are identified and discussed. The reasons for their high rankings are also provided. The discussion is limited to the top 5–10 tests that, in most cases, have been used extensively. Because tests based on a variety of amphipod species are primarily included among the top ranked marine lethal and estuarine lethal test categories, a greater number of tests is discussed for these categories than for others. Thus, more than one species group is discussed for each test category.

MARINE LETHAL TESTS

Seven of the eight highest ranking marine lethal tests were based on the exposure of juvenile or adult infaunal amphipods to whole sediments for a 10-day exposure period. These seven tests ranked highest largely because of: 1) a high degree of exposure relevance, 2) year-round availability of the test species by field collection (six of the seven species) or culture (*Grandidierella japonica*), and 3) well-developed methods by either the American Society for Testing and Materials (ASTM) or Environment Canada with available laboratories.

The tests based on *A. abdita* and *Rhepoxynius abronius* are the only ones with a high regulatory

TABLE 2. EVALUATION OF SEDIMENT TOXICITY TESTS—MARINE LETHAL

Test No.	Exposure Media	Common Name	Life Stage	Test Species	Tech. Rating	Reli-ability	Ecol. Rel.	Exp. Rel.	Avail-ability	Inter-ferences	Chem. Discrim.	Reg. Status	Comments
001	S	Amphipod	Juveniles or adult females	*Ampelisca abdita*	16.5	3	3	4	3	3	4	High	B,C,E
008	S	Amphipod	Adults	*Rhepoxynius abronius*	16.5	4	2	4	3	3	4	High	C,E
003	S	Amphipod	Juveniles or young adults	*Eohaustorius washingtonianus*	16.5	4	2	4	3	3	4	Med.	E
002	S	Amphipod	Juveniles or young adults	*Amphiporeia virginiana*	15.5	4	2	4	3	3	2	Med.	E
006	S	Amphipod	Juveniles or young adults	*Leptocheirus pinguis*	14.5	3	2	4	3	3	2	Med.	
005	S	Amphipod	Immature	*Grandidierella japonica*	14.5	3	2	4	3	3	2	Low	A,B
004	S	Amphipod	Juveniles or young adults	*Foxiphalus xiximeus*	13.5	4	2	4	1	3	2	Med.	E
020	S	Littleneck clam	Juveniles	*Protothaca staminea*	13.5	3	3	4	2	3	0	Low	
017	EL	Blue mussel	Embryos	*Mytilus edulis*	13	2	4	0	3	4	4	Med.	B
033	S	Shrimp	Juveniles	*Pandalus* sp.	13	3	3	3	1	4	2	Low	
029	S	Cancer crab	Juveniles	*Cancer* sp.	13	3	3	3	1	4	2	Low	
027	S	Blue crab	Juveniles	*Callinectes sapidus*	13	3	3	3	1	4	2	Low	
034	S	Shrimp	Post-larvae	*Penaeus* sp.	13	3	3	3	1	4	2	Low	
071	S	Polychaete	Juveniles	*Neanthes* sp.	12.5	3	2	4	1	3	2	Med.	B
021	S	Japanese clam	Juveniles	*Tapes japonica*	12.5	3	3	4	1	3	0	Low	
016	S	Blue mussel	Embryos	*Mytilus edulis*	12	2	4	1	3	0	4	Med.	B,C
011	EL	Pacific oyster	Embryos	*Crassostrea gigas*	12	2	4	0	2	4	4	Med.	B,D
009	EL	Eastern oyster	Embryos	*Crassostrea virginica*	12	2	4	0	2	4	4	Med.	B
055	EL	Sheepshead minnow	1-14 days old	*Cyprinodon variegatus*	12	4	2	0	4	3	1	Med.	B,E
037	S	Ridge-back prawn	Juveniles	*Sicyonia ingentis*	12	3	2	3	1	4	2	Low	A
030	S	Sand shrimp	Juveniles	*Crangon* sp.	12	3	2	3	1	4	2	Low	
043	S	White sea urchin	8- to 22-mm diameter	*Lytechinus pictus*	12	3	2	3	1	4	2	Low	
015	S	Clam	Juveniles	*Mulinia lateralis*	12	3	2	4	1	4	0	Low	
039	S	Sand dollar	Juveniles	*Dendraster excentricus*	12	3	2	3	1	4	2	Low	
023	S	Yoldia clam	Juveniles	*Yoldia limatula*	11.5	3	2	4	1	3	0	Low	
032	EL	Shrimp	Embryo-larval	*Pandalus* sp.	11.5	3	3	1	1	3	4	Low	
046	S	Purple sea urchin	Embryos	*Strongylocentrotus purpuratus*	11	2	4	1	2	0	4	Med.	C
045	EL	Purple sea urchin	Embryos (<1 hr old)	*Strongylocentrotus purpuratus*	11	2	4	0	1	4	4	Med.	
010	S	Pacific oyster	Embryos	*Crassostrea gigas*	11	2	4	1	2	0	4	Low	B,C,D
014	EL	Quahog clam	Embryos	*Mercenaria mercenaria*	11	2	3	0	2	4	4	Low	B

TABLE 2. (cont.)

Test No.	Exposure Media	Common Name	Life Stage	Test Species	Tech. Rating	Reli-ability	Ecol. Rel.	Exp. Rel.	Avail-ability	Inter-ferences	Chem. Discrim.	Reg. Status	Comments
026	EL	Blue crab	Embryo-larval	*Callinectes sapidus*	11	3	3	0	1	4	4	Low	
028	EL	Cancer crab	Embryo-larval	*Cancer* sp.	11	3	3	0	1	4	4	Low	
035	EL	Shrimp	Post-larvae	*Penaeus* sp.	10.5	3	3	1	1	3	2	Low	
031	EL	Grass shrimp	Post-hatch	*Palaemonetes* sp.	10.5	3	2	1	1	3	4	Low	
056	EL	3-spine stickleback	Juveniles	*Gasterosteus aculeatus*	10	3	2	0	3	4	0	Med.	B
057	S	Surf smelt	Larvae	*Hypomesus pretiosus*	10	3	3	1	1	2	2	Low	
058	S	Surf smelt	Larvae	*Hypomesus pretiosus*	10	3	3	1	1	2	2	Low	
059	EL	Pinfish	Embryo-larval	*Lagodon rhomboides*	10	3	3	0	1	4	2	Low	
060	EL	Spot	Embryo-larval	*Leiostomus xanthurus*	10	3	3	0	1	4	2	Low	
061	EL	Grunion	Embryo-larval	*Leuresthes tenuis*	10	3	3	0	1	4	2	Low	A
065	EL	Mysid	1–5 days old	*Mysidopsis* sp.	9.5	3	2	0	1	3	4	Med.	
063	EL	Mysid	1–5 days old	*Holmesimysis* sp.	9.5	3	2	0	1	3	4	Med.	
018	EL	Oyster	Embryo-larval	*Ostrea* sp.	9	2	2	0	1	4	4	Med.	
041	EL	Sand dollar	Embryos	*Dendraster excentricus*	9	2	2	0	1	4	4	Med.	
054	EL	Shiner perch	Embryo-larval	*Cymatogaster aggregata*	9	3	2	0	1	4	2	Med.	
062	S	Mysid	1–5 days old	*Holmesimysis* sp.	9	3	2	1	1	0	4	Low	
064	S	Mysid	1–5 days old	*Mysidopsis* sp.	9	3	2	1	1	0	4	Low	
044	EL	White sea urchin	Embryos (<1 hr old)	*Lytechinus pictus*	9	3	2	0	1	4	4	Low	
049	EL	Sea urchin	Embryos (<1 hr old)	*Strongylocentrotus* sp.	9	2	2	0	1	4	4	Low	
053	EL	Dolphinfish	Embryo-larval	*Coryphaena hippurus*	9	3	3	0	0	4	2	Low	
048	S	Green sea urchin	Embryos	*Strongylocentrotus droebachiensis*	8	2	2	1	2	0	2	Med.	C
040	S	Sand dollar	Embryos	*Dendraster excentricus*	8	2	2	1	1	0	4	Med.	C
038	S	Atlantic urchin	Embryos	*Arbacia punctulata*	8	2	2	1	1	0	4	Low	C
051	EL	Speckled sanddab	Juveniles	*Citharichthys stigmaeus*	7.5	3	2	0	1	3	0	Low	

Note: EL - elutriate
S - sediment

A - geographically restricted or alien to United States
B - widely distributed and/or cultured
C - potential sediment interferences
D - field validated to benthos
E - interlaboratory comparisons available

TABLE 3. EVALUATION OF SEDIMENT TOXICITY TESTS—MARINE SUBLETHAL

Test No.	Exposure Media	Common Name	Life Stage	Test Species	Tech. Rating	Reli-ability	Ecol. Rel.	Exp. Rel.	Avail-ability	Inter-ferences	Chem. Discrim.	Reg. Status	Comments
001	S	Amphipod	Juveniles or adult females	*Ampelisca abdita*	14.5	1	3	4	3	3	4	Low	B,C
121	S	Polychaete	Juveniles	*Neanthes* sp.	14.5	3	2	4	3	3	2	Low	B,E
109	EL	Purple sea urchin	Gametes	*Strongylocentrotus purpuratus*	14	3	4	0	3	4	4	Med.	E
008	S	Amphipod	Adults	*Rhepoxynius abronius*	13.5	1	2	4	3	3	4	Low	C
005	S	Amphipod	Immature	*Grandidierella japonica*	13.5	1	2	4	3	3	4	Low	B
017	EL	Blue mussel	Embryos	*Mytilus edulis*	13	2	4	0	3	4	4	Low	B,E
111	EL	Purple sea urchin	Gametes	*Strongylocentrotus purpuratus*	13	3	4	0	2	4	4	Low	E
047	EL	Purple sea urchin	Gametes	*Strongylocentrotus purpuratus*	12	3	4	0	1	4	4	Med.	
102	EL	Sand dollar	Gametes	*Dendraster excentricus*	12	3	2	0	3	4	4	Med.	E
100	EL	Atlantic urchin	Gametes	*Arbacia punctulata*	12	3	2	0	3	4	4	Med.	E
115	EL	Green sea urchin	Gametes	*Strongylocentrotus droebachiensis*	12	3	2	0	3	4	4	Med.	E
015	S	Clam	Juveniles	*Mulinia lateralis*	12	2	2	3	1	4	4	Low	
107	EL	White sea urchin	Gametes	*Lytechinus pictus*	12	3	2	0	3	4	4	Med.	A,E
112	EL	Purple sea urchin	Gametes	*Strongylocentrotus purpuratus*	12	2	4	0	2	4	4	Low	
011	EL	Pacific oyster	Embryos	*Crassostrea gigas*	12	2	4	0	2	4	4	Low	B,D,E
081	EL	Microtox	Cells	*Photobacterium phosphoreum*	12	2	1	2	4	3	3	Low	B,D
016	S	Blue mussel	Embryos	*Mytilus edulis*	11	1	4	1	3	0	4	Med.	B,C
009	EL	Eastern oyster	Embryos	*Crassostrea virginica*	11	1	4	0	2	4	4	Low	B
080	INT	Microtox	Cells	*Photobacterium phosphoreum*	11	2	1	1	4	3	3	Low	B,D
010	S	Pacific oyster	Embryos	*Crassostrea gigas*	10	1	4	1	2	0	4	Med.	B,C,D
046	S	Purple sea urchin	Embryos	*Strongylocentrotus purpuratus*	10	1	4	1	2	0	4	Med.	C
042	EL	Sand dollar	Gametes	*Dendraster excentricus*	10	3	2	0	1	4	4	Med.	E
043	S	White sea urchin	8- to 22-mm diameter	*Lytechinus pictus*	10	2	2	3	1	4	0	Low	
039	S	Sand dollar	Juveniles	*Dendraster excentricus*	10	2	2	3	1	4	0	Low	
014	EL	Quahog clam	Embryos	*Mercenaria mercenaria*	10	1	3	0	2	4	4	Low	B
098	EL	Atlantic urchin	Gametes	*Arbacia punctulata*	10	3	2	0	1	4	4	Low	E
097	INT	Atlantic urchin	Gametes	*Arbacia punctulata*	9	2	2	0	1	4	4	Low	
048	S	Green sea urchin	Embryos	*Strongylocentrotus droebachiensis*	8	1	2	1	2	0	4	Med.	C
058	S	Surf smelt	Larvae	*Hypomesus pretiosus*	8	2	3	1	1	0	2	Low	
038	S	Atlantic urchin	Gametes	*Arbacia punctulata*	8	2	2	1	1	0	4	Low	C

TABLE 3. (cont.)

Test No.	Exposure Media	Common Name	Life Stage	Test Species	Tech. Rating	Reli-ability	Ecol. Rel.	Exp. Rel.	Avail-ability	Inter-ferences	Chem. Discrim.	Reg. Status	Comments
083	S	Microtox®	Cells	*Photobacterium phosphoreum*	7	2	1	0	2	1	3	Med.	B
084	EX	Mod. Ames/HPTLC	Cells	*Salmonella typhimurium*	7	2	1	0	2	1	3	Low	B
040	S	Sand dollar	Embryos	*Dendraster excentricus*	6	1	2	1	1	0	2	Med.	C

Note: EL - elutriate
EX - extract
INT - interstitial water
S - sediment

A - geographically restricted or alien to United States
B - widely distributed and/or cultured
C - potential sediment interferences
D - field validated to benthos
E - interlaboratory comparisons available

TABLE 4. EVALUATION OF SEDIMENT TOXICITY TESTS—ESTUARINE LETHAL

Test No.	Exposure Media	Common Name	Life Stage	Test Species	Tech. Rating	Reli-ability	Ecol. Rel.	Exp. Rel.	Avail-ability	Inter-ferences	Chem. Discrim.	Reg. Status	Comments
001	S	Amphipod	Juveniles or adult females	*Ampelisca abdita*	16.5	3	3	4	3	3	4	High	B,C,E
129	S	Amphipod	Large juveniles and adults	*Eohaustorius estuarius*	16.5	4	2	4	3	3	4	High	C
003	S	Amphipod	Juveniles or young adults	*Eohaustorius washingtonianus*	16.5	4	2	4	3	3	4	Med.	E
133	S	Amphipod	Juveniles	*Leptocheirus plumulosus*	16	4	2	4	3	2	4	Low	B,E
135	S	Amphipod	Mixed sexes	*Leptocheirus plumulosus*	15.5	3	2	4	3	3	4	High	B
126	S	Amphipod	Juveniles or young adults	*Corophium volutator*	15.5	4	2	3	3	3	4	Med.	E
002	S	Amphipod	Juveniles or young adults	*Amphiporeia virginiana*	15.5	4	2	4	3	3	2	Med.	E
128	S	Amphipod	Adults	*Corophium volutator*	15.5	4	2	3	3	3	4	Low	E
134	S	Amphipod	Juveniles	*Leptocheirus plumulosus*	15.5	3	2	4	3	3	4	Low	B
131	S	Amphipod	7-14 days old	*Hyalella azteca*	14	3	2	3	2	4	4	High	B
020	S	Littleneck clam	Juveniles	*Protothaca staminea*	13.5	3	3	4	2	3	0	Low	
017	EL	Blue mussel	Embryos	*Mytilus edulis*	13	2	4	0	3	4	4	Med.	B
027	S	Blue crab	Juveniles	*Callinectes sapidus*	13	3	3	3	1	4	2	Low	
151	S	Grass shrimp	Post-hatch (1–4 days)	*Palaemonetes* sp.	13	3	2	3	1	4	4	Low	
016	S	Blue mussel	Embryos	*Mytilus edulis*	12	2	4	1	3	0	4	Med.	B,C
009	EL	Eastern oyster	Embryos	*Crassostrea virginica*	12	2	4	0	2	4	4	Med.	B
011	EL	Pacific oyster	Embryos	*Crassostrea gigas*	12	2	4	0	2	4	4	Med.	B,D
030	S	Sand shrimp	Juveniles	*Crangon* sp.	12	3	2	3	1	4	2	Low	
153	EL	Sheepshead minnow	1–14 days old	*Cyprinodon variegatus*	11.5	3	2	0	4	4	1	Low	B
010	S	Pacific oyster	Embryos	*Crassostrea gigas*	11	2	4	1	2	0	4	Med.	B,C,D
157	EL	Silverside	9–14 days old	*Menidia* sp.	11	4	2	0	3	3	1	Med.	B,E
158	EL	Silverside	1–14 days old	*Menidia* sp.	11	4	2	0	3	3	1	Med.	B,E
014	EL	Quahog clam	Embryos	*Mercenaria mercenaria*	11	2	3	0	2	4	4	Low	B
026	EL	Blue crab	Embryo-larval	*Callinectes sapidus*	11	3	3	0	1	4	4	Low	
065	EL	Mysid	1–5 days old	*Mysidopsis* sp.	10.5	4	2	0	1	3	4	Med.	E
162	EL	Mysid	1–5 days old	*Neomysis* sp.	10.5	3	2	0	2	3	4	Med.	
161	S	Mysid	1–5 days old	*Neomysis* sp.	10	3	2	1	2	0	4	Low	
056	EL	3-spine stickleback	Juveniles	*Gasterosteus aculeatus*	10	3	2	0	3	4	0	Med.	B
064	S	Mysid	1–5 days old	*Mysidopsis* sp.	9	3	2	1	1	0	4	Low	
156	EL	Grunion	9–14 days old	*Leuresthes tenuis*	9	3	3	0	1	3	1	Low	A
165	INT	Polychaete	Females (1–2 days)	*Dinophilus gyrociliatus*	8.5	3	1	1	1	3	2	Low	

Footnotes on following page.

TABLE 4. (cont.)

Note: EL - elutriate
INT - interstitial water
S - sediment

A - geographically restricted or alien to United States
B - widely distributed and/or cultured
C - potential sediment interferences
D - field validated to benthos
E - interlaboratory comparisons available

TABLE 5. EVALUATION OF SEDIMENT TOXICITY TESTS—ESTUARINE SUBLETHAL

Test No.	Exposure Media	Common Name	Life Stage	Test Species	Tech. Rating	Reli-ability	Ecol. Rel.	Exp. Rel.	Avail-ability	Inter-ferences	Chem. Discrim.	Reg. Status	Comments
001	S	Amphipod	Juveniles or adult females	*Ampelisca abdita*	16.5	3	3	4	3	3	4	Low	B,C
134	S	Amphipod	Juveniles	*Leptocheirus plumulosus*	14.5	2	2	4	3	3	4	Low	B
135	S	Amphipod	Mixed sexes	*Leptocheirus plumulosus*	14.5	2	2	4	3	3	4	Low	B
129	S	Amphipod	Large juveniles and adults	*Eohaustorius estuarius*	14.5	2	2	4	3	3	4	Low	C
128	S	Amphipod	Adults	*Corophium volutator*	14.5	3	2	3	3	3	4	Low	E
133	S	Amphipod	Juveniles	*Leptocheirus plumulosus*	14	2	2	4	3	2	4	Low	B
017	EL	Blue mussel	Embryos	*Mytilus edulis*	13	2	4	0	3	4	4	Low	B,E
011	EL	Pacific oyster	Embryos	*Crassostrea gigas*	12	2	4	0	2	4	4	Low	B,D,E
010	S	Pacific oyster	Embryos	*Crassostrea gigas*	11	2	4	1	2	0	4	Med.	B,C,D
016	S	Blue mussel	Embryos	*Mytilus edulis*	11	1	4	1	3	0	4	Med.	B,C
014	EL	Quahog clam	Embryos	*Mercenaria mercenaria*	11	1	4	0	2	4	4	Low	B
009	EL	Eastern oyster	Embryos	*Crassostrea virginica*	11	1	4	0	2	4	4	Low	B
187	EL	Sheepshead minnow	Adults	*Cyprinodon variegatus*	11	1	2	0	4	4	4	Low	B
176	EX	Microtox	Cells	*Photobacterium phosphoreum*	10	2	2	0	4	1	3	Low	B,D
191	INT	Polychaete	Females	*Dinophilus gyrociliatus*	8.5	2	1	1	1	3	4	Low	
175	S	Microtox	Cells	*Photobacterium phosphoreum*	6.5	2	1	0	2	0	3	Low	B,C

Note: EL - elutriate
EX - extract
INT - interstitial water
S - sediment

B - widely distributed and/or cultured
C - potential sediment interferences
D - field validated to benthos
E - interlaboratory comparisons available

TABLE 6. EVALUATION OF SEDIMENT TOXICITY TESTS—FRESHWATER LETHAL

Test No.	Exposure Media	Common Name	Life Stage	Test Species	Tech. Rating	Reli-ability	Ecol. Rel.	Exp. Rel.	Avail-ability	Inter-ferences	Chem. Discrim.	Reg. Status	Comments
241	S	Mayfly	Nymphs	*Hexagenia limbata*	16.5	3	3	4	3	3	4	Low	B,D
239	S	Mayfly	Young nymphs	*Hexagenia limbata*	16.5	3	3	4	3	3	4	Low	B,D
238	S	Midge	2nd instar larvae	*Chironomus tentans*	16	3	2	4	4	2	4	High	B,D
237	S	Midge	Neonates or larvae	*Chironomus riparius*	16	3	2	4	4	2	4	Med.	B
200	S	Amphipod	Juveniles	*Hyalella azteca*	15.5	3	2	3	4	3	4	High	B
242	S	Mayfly	Nymphs	*Hexagenia limbata*	15.5	3	2	4	3	3	4	Low	B,D
209	INT	Water flea	Neonates	*Daphnia magna*	15	4	3	0	4	4	4	Low to Med.	B,D,E
256	S	Marsh grass	Seedlings	*Echinochloa crusgalli*	15	3	4	4	1	4	2	Low	
250	S	Oligochaete	Large worms	*Lumbriculus variegatus*	14.5	3	2	4	3	3	2	Low	B
249	S	Oligochaete	Mixed-age	*Lumbriculus variegatus*	14.5	3	2	4	3	3	2	Low	B
212	EL	Water flea	Neonates (<24 hrs)	*Daphnia magna*	14	4	2	0	4	4	4	Med.	B,D,E
240	INT	Mayfly	Nymphs	*Hexagenia limbata*	14	3	3	2	3	2	4	Low	B,D
205	EL	Water flea	Neonates	*Ceriodaphnia dubia*	13.5	4	2	0	4	3	4	Med.	B,E
213	EL	Water flea	Neonates (<24 hrs)	*Daphnia pulex*	13.5	4	2	0	4	3	4	Med.	B,E
208	EL	Water flea	Neonates	*Daphnia magna*	13.5	4	2	0	4	3	4	Med.	B,D,E
214	EL	Water flea	Neonates (<24 hrs)	*Daphnia pulex*	13	3	2	0	4	4	4	Med.	B
202	S	Paper pondshell clam	Juveniles	*Anodonta imbecillis*	13	3	2	4	2	3	1	Low	
252	S	Oligochaete	Mixed age; similar size	*Pristina leidyi*	13	3	2	3	2	4	2	Low	B
255	S	Oligochaete	Mixed age	*Tubifex tubifex*	13	3	2	3	2	4	2	Low	B,D
222	EL	Bluegill	Approx. 4 days	*Lepomis macrochirus*	13	4	3	0	4	4	0	Low	B,E
226	EL	Fathead minnow	Larvae (<24 hrs old)	*Pimephales promelas*	12.5	4	2	0	4	4	1	Med.	B,E
234	EL	Rainbow trout	Fry or fingerlings	*Oncorhynchus mykiss*	12.5	3	3	0	4	4	1	Med.	B
235	S	Rainbow trout	Egg-sac stage	*Oncorhynchus mykiss*	12.5	3	3	1	3	1	4	Low	B
199	INT	Amphipod	Juveniles	*Hyalella azteca*	12.5	3	2	1	3	3	4	Low	B
220	EL	Channel catfish	Approx. 4 days	*Ictalurus punctatus*	12	3	3	0	4	4	0	Low	B
229	EL	Fathead minnow	Approximately 4 days	*Pimephales promelas*	11	3	2	0	4	4	0	Med.	B
207	S	Water flea	Neonates	*Ceriodaphnia dubia*	11	3	2	1	3	0	4	Low	B,C
211	S	Water flea	Neonates	*Daphnia magna*	11	3	2	1	3	0	4	Low	B,C,D
251	EL	Oligochaete	Similar size	*Pristina leidyi*	10.5	3	2	1	2	3	2	Low	B
232	EX	Fathead minnow	Embryo-larval	*Pimephales promelas*	10.5	3	2	0	4	2	1	Low	B

TABLE 6. (cont.)

Test No.	Exposure Media	Common Name	Life Stage	Test Species	Tech. Rating	Reli-ability	Ecol. Rel.	Exp. Rel.	Avail-ability	Inter-ferences	Chem. Discrim.	Reg. Status	Comments
233	S	Fathead minnow	Juveniles	*Pimephales promelas*	10	3	2	1	3	2	0	Low	B
244	EL	Nematode	L1 stage juveniles	*Panagrellus redivivus*	9	3	1	1	2	2	2	Low	
195	EX	African clawed frog	Embryos	*Xenopus laevis*	9	3	0	0	3	2	4	Low	A,B

Note: EL - elutriate
EX - extract
INT - interstitial water
S - sediment

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B - widely distributed and/or cultured
C - potential sediment interferences
D - field validated to benthos
E - interlaboratory comparisons available

TABLE 7. EVALUATION OF SEDIMENT TOXICITY TESTS—FRESHWATER SUBLETHAL

Test No.	Exposure Media	Common Name	Life Stage	Test Species	Tech. Rating	Reli-ability	Ecol. Rel.	Exp. Rel.	Avail-ability	Inter-ferences	Chem. Discrim.	Reg. Status	Comments
238	S	Midge	2nd instar larvae	*Chironomus tentans*	15	2	2	4	4	2	4	Low	B,D
237	S	Midge	Neonates or larvae	*Chironomus riparius*	15	2	2	4	4	2	4	Low	B
200	S	Amphipod	Juveniles	*Hyalella azteca*	14.5	2	2	3	4	3	4	Low to Med.	B
242	S	Mayfly	Nymphs	*Hexagenia limbata*	14.5	2	2	4	3	3	4	Low	B,D
333	S	Aquatic vascular plant	Apical shoots	*Hydrilla verticillata*	14	2	3	4	2	4	2	Low	
256	S	Marsh grass	Seedlings	*Echinochloa crusgalli*	14	2	3	4	1	4	4	Low	
269	S	Amphipod	Juveniles (<1 week)	*Hyalella azteca*	14	2	2	3	3	4	4	Low	B
285	EL	Water flea	Neonates	*Ceriodaphnia dubia*	13.5	4	2	0	4	3	4	Med.	B,E
330	S	Oligochaete	Adults	*Tubifex tubifex*	13	2	2	3	2	4	4	Low	B,D
304	INT	Midge	2nd instar larvae	*Chironomus tentans*	12.5	2	3	2	3	1	4	Low to Med.	B,D
250	S	Oligochaete	Large worms	*Lumbriculus variegatus*	12.5	2	0	4	3	3	4	Low	B
323	S	Oligochaete	Juveniles and adults	*Lumbriculus variegatus*	12.5	2	2	4	1	3	4	Low	B
324	S	Oligochaete	15 mm in length	*Lumbriculus variegatus*	12.5	2	2	4	1	3	4	Low	B
226	EL	Fathead minnow	Larvae (<24 hrs old)	*Pimephales promelas*	11.5	3	2	0	4	4	1	Med.	B,E
260	EL	Green algae	Cells	*Selenastrum capricornutum*	11.5	2	2	0	4	4	3	Med.	B
272	INT	Microtox	Cells	*Photobacterium phosphoreum*	11.5	2	2	0	4	4	3	Low to Med.	B,D
235	S	Rainbow trout	Egg-sac stage	*Oncorhynchus mykiss*	11.5	2	3	1	3	1	4	Low	B
261	INT	Green algae	Cells	*Selenastrum capricornutum*	11.5	2	2	0	4	4	3	Low	B
252	S	Oligochaete	Mixed age; similar size	*Pristina leidyi*	11	1	2	3	2	4	2	Low	B
176	EX	Microtox®	Cells	*Photobacterium phosphoreum*	10.5	2	2	0	4	2	3	Low	B,D
211	S	Water flea	Juvenile (5 days)	*Daphnia magna*	10	2	2	1	3	0	4	Low	B,C,D
232	EX	Fathead minnow	Embryo-larval	*Pimephales promelas*	9.5	2	2	0	4	2	1	Low to Med.	B
233	S	Fathead minnow	Juveniles	*Pimephales promelas*	9.5	2	2	1	3	2	1	Low	B
263	EL	Algae	Cells	*Chlorella vulgaris*	9.5	2	2	0	2	4	3	Low	B
302	S	Rainbow trout	Gonad cell (RTG-2)	*Oncorhynchus mykiss*	9	2	3	1	1	0	4	Low	B,C
244	EL	Nematode	L1 stage juveniles	*Panagrellus redivivus*	9	2	2	1	2	2	2	Low	
262	S	Algae	Cells	*Chlorella vulgaris*	8.5	2	2	1	2	0	3	Low	B,C
195	EX	African clawed frog	Embryos	*Xenopus laevis*	8	2	0	0	3	2	4	Low	A,B
278	S	Mutatox	Cells	*Vibrio fischeri*	7.5	2	1	1	2	0	3	Low	B,C
273	S	Microtox®	Cells	*Photobacterium phosphoreum*	7.5	2	1	1	2	0	3	Low	B,C

TABLE 7. (cont.)

Test No.	Exposure Media	Common Name	Life Stage	Test Species	Tech. Rating	Reli-ability	Ecol. Rel.	Exp. Rel.	Avail-ability	Inter-ferences	Chem. Discrim.	Reg. Status	Comments
274	S	Microtox®	Cells	*Photobacterium phosphoreum*	7.5	2	1	1	2	0	3	Low	B,C
270	EX	Aquatic bacterium	Cells	*Aeromonas hydrophila*	7.5	2	1	0	2	2	3	Low	B
320	EX	Nematode	L2 stage juveniles	*Panagrellus redivivus*	7	2	2	1	1	0	2	Low	
279	EL	Mutatox	Cells	*Vibrio fischeri*	6.5	2	1	0	2	0	3	Low	B
308	EL	Microbial enzyme	N/A	*Alkaline phosphatase* (APA)	6.5	2	1	1	1	0	3	Low	B
084	EX	Mod. Ames/HPTLC	Cells	*Salmonella typhimurium*	6.5	2	1	0	2	0	3	Low	B

Note: EL - elutriate
EX - extract
INT - interstitial water
S - sediment

A - geographically restricted or alien to United States
B - widely distributed and/or cultured
C - potential sediment interferences
D - field validated to benthos
E - interlaboratory comparisons available

status. *R. abronius* is known to sometimes be sensitive to interferences from fine-grained sediments; in nature, this species is restricted to medium- to coarse-grained sediment habitats. Effects of sediment grain-size composition on *A. abdita* and *Eohaustorius washingtonianus* have also been demonstrated (DeWitt *et al.* 1989; U.S. EPA 1992). By contrast, *Amphiporeia virginiana* is not influenced markedly by sediment grain size, and *G. japonica* is found in a wide variety of sediment types in the field. *Foxiphalus xiximeus* inhabits fine to medium sands. Information on the sediment grain-size preference of *Leptocheirus pinguis* was not available. Although *G. japonica* can be cultured, the test has a low regulatory status and the test species has a limited geographical distribution (i.e., San Francisco Bay to Southern California). Although *A. abdita* can not be consistently maintained in culture (Redmond *et al.* 1994), this species has the broadest geographic distribution (i.e., East Coast, Gulf of Mexico, and San Francisco Bay) among the amphipod test species. This species also has the highest ecological relevance because it is considered a foundation species (i.e., its tube mats provide habitat for other species).

The juvenile clam mortality test based on a 10-day exposure of the infaunal species *Protothaca staminea* to whole sediment ranked among the top eight marine lethal tests (Table 2). This test ranked high because of the relatively high scores for reliability, ecological relevance, and exposure relevance. The test is easy to perform, and at least some laboratories have experience with this test or similar tests. A relatively developed method is available, but a standardized method has not been issued by ASTM or EPA. The species is widely distributed throughout the West Coast of North America and is harvested commercially and recreationally. The only serious drawback of this test is the potentially low sensitivity of juvenile bivalves to chemicals in sediments. However, little information on the actual performance of this test is available.

MARINE SUBLETHAL TESTS

Four of the five highest ranking marine sublethal tests are based on the exposure of juvenile or adult infaunal organisms to whole sediments. Interestingly, all four infaunal tests have a low regulatory status. These four infaunal tests ranked highest largely because of: 1) a high degree of exposure relevance, 2) year-round availability of the test species by field collection (*A. abdita* and *R. abronius*) or culture (*G. japonica* and *Neanthes* sp.), and 3) well developed methods by either ASTM or Environment Canada with available laboratories. Three of the four infaunal tests are based on amphipods (i.e., *A. abdita, G. japonica*, and *R. abronius*) and were among the highest ranking marine lethal tests. The primary sublethal endpoint for all three of these tests is reburial, which, as a behavioral endpoint, scored low for reliability because of the potential variability. The major attributes of the three amphipod tests are discussed in *Marine Lethal Tests*. The fourth infaunal test is a long-term test based on the polychaete *Neanthes* sp. (Test 121) and does not include a lethal endpoint (see Table 2, Test 071 for a 10-day lethality test with *Neanthes* sp.). The sublethal endpoint for Test 121 is growth, which is generally easier to interpret than reburial. The exposure period for the three amphipod tests is 10 days, whereas the exposure period for the polychaete sublethal test is 20–28 days.

Although the echinoderm gamete test on elutriate sediment (Test 109) based on the purple sea urchin, *Strongylocentrotus purpuratus*, is not an infaunal test, it was ranked high, largely because of: 1) a sensitive life stage (i.e., gametes) and a reliable endpoint (i.e., fertilization), 2) a high degree of ecological relevance, and 3) well developed methods by Environment Canada. The purple sea urchin is considered an important species because its grazing activity can influence the distribution of kelp beds. The major drawback to this echinoderm test is the low exposure relevance of the elutriate exposure scenario. In the field, it is unlikely that planktonic gametes are commonly exposed to conditions similar to a sediment elutriate.

Although the bivalve development abnormality test (Test 017) based on exposure of the blue mussel, *Mytilus edulis*, to elutriates (or potentially to interstitial water) is not an infaunal test, it was ranked high, largely because it is based on a sensitive life stage (i.e., embryos) of a species having a relatively high ecological relevance and high overall availability. In addition, the test is not affected by physical interferences because sediment is not included in the test chambers. The test has been evaluated in an intra- and interlaboratory variability study using complex effluents and reference toxicants (Pastorok *et al.* 1994). The test species has a wide distribution (i.e., East and West coasts), is commercially and recreationally harvested, and is considered a foundation species because it provides habitat for other species. The test species can also be cultured and embryos are available for at least half the year. The major drawback to the bivalve test is the low exposure relevance of the elutriate exposure scenario. In the field, it is unlikely that planktonic embryos are commonly exposed to conditions similar to a sediment elutriate. For the corresponding lethal test (Table 2, Test 017), highly variable results may be obtained when imprecise pipetting procedures are used to count initial and final larval densities. This problem affects all lethal tests with bivalve embryos, but not the corresponding sublethal tests.

ESTUARINE LETHAL TESTS

The 10 highest ranking estuarine lethal tests were based on the exposure of juvenile or adult infaunal amphipods to whole sediments for a 10-day exposure period. These tests ranked highest largely because of: 1) a high degree of exposure relevance, 2) good availability of the test species by field collection (8 of the 10 species) or culture (*Leptocheirus plumulosus* and *Hyalella azteca*), and 3) well developed methods by ASTM or Environment Canada with available laboratories. Two of these tests (i.e., those based on *A. abdita* and *E. washingtonianus*) were also included among the highest ranking marine lethal tests. Three of the 10 tests use *L. plumulosus* as the test species, and two tests use *Corophium volutator* as the test species. There are well-developed methods by ASTM and Environment Canada for selected tests based on *L. plumulosus* and *C. volutator*, and both test species have relatively wide tolerances for sediment grain size. The tests based on *A. abdita* and *Eohaustorius estuarius* and one of the tests based on *L. plumulosus* have a high regulatory status. One of the tests based on each of the species *L. plumulosus* and *C. volutator* has a low regulatory status.

The bivalve test based on exposure of the littleneck clam, *P. staminea*, to whole sediments was the eleventh ranked test. Refer to the previous section entitled *Marine Lethal Tests* for a discussion of this test.

ESTUARINE SUBLETHAL TESTS

The six highest ranking estuarine sublethal tests are based on the exposure of juvenile or adult infaunal amphipods to whole sediments. The primary sublethal endpoint for the tests based on *A. abdita*, *E. estuarius*, and *C. volutator* is reburial. By contrast, the primary sublethal endpoints for the tests based on *L. plumulosus* are fertility, reproduction, and growth, all of which are generally easier to interpret than reburial. All six tests were also included in the nine highest ranking estuarine lethal tests, although the exposure period is increased from 10 days to 28–30 days for the three sublethal tests that use *L. plumulosus* as the test species. These six test ranked highly as sublethal tests for the same reasons discussed previously for the estuarine lethal tests. All six tests have a low regulatory status.

FRESHWATER LETHAL TESTS

Five of the six highest ranking freshwater lethal tests were based on the exposure of infaunal insects (i.e., the mayfly *Hexagenia limbata* and the midges *Chironomus riparius* and *Chironomus tentans*) to whole sediments. The fifth ranked test was based on the exposure of an epifaunal amphipod, *H. azteca*, to whole sediments. The exposure periods varied from 7 to 30 days. These tests ranked highest largely because of their high degree of exposure relevance and their relatively high availability. All of the test species are available by field collection during most of the year, and *C. riparius*, *C. tentans*, and *H. azteca* can be cultured.

The tests based on *C. riparius, C. tentans*, and *H. azteca* have well-developed methods by ASTM. The tests based on *C. tentans* and *H. azteca* have a high regulatory status, whereas all three tests based on *H. limbata* have a low regulatory status. All of the test species are distributed throughout North America, except *C. tentans*, which is limited to the mid-continental areas of the United States. *C. riparius* and *H. azteca* are tolerant of a wide range of sediment grain sizes. For *H. limbata*, large individuals often exhibit cannibalistic behavior.

FRESHWATER SUBLETHAL TESTS

Four of the six highest ranking freshwater sublethal tests were also included as the highest ranking freshwater lethal tests. They included the tests based on the amphipod *H. azteca*, the mayfly *H. limbata*, and the midges *C. riparius* and *C. tentans*. The sublethal endpoints for all four tests is growth. All of the tests have a low regulatory status, except the one based on *H. azteca*, which has a low to medium regulatory status. The four tests ranked high as sublethal tests for the same reasons discussed previously for the freshwater lethal tests.

The other two highest ranking freshwater sublethal tests were based on plants (*Hydrilla verticillata* and *Echinochloa crusgalli*) that are exposed to whole sediments for 14 days. These tests ranked high primarily because of their high degrees of exposure and ecological relevance and their relatively low susceptibility to interferences. The high ecological relevance of the two tests is based on the importance of the plants in providing habitat for other organisms. The major drawback of these two tests is their relatively low availability because of the small number of laboratories that have experience with them. Both tests have a low regulatory status. The sublethal endpoints for the test based on *H. verticillata* is growth, whereas the sublethal endpoint for the test based on *E. crusgalli* is seed germination.

APPLICATION OF SEDIMENT TOXICITY TESTS

In this section, a brief overview is presented of the major kinds of considerations that should be addressed when using sediment toxicity tests to evaluate the potential effects of chemicals discharged to aquatic environments. Although many of these considerations may be specified as part of NPDES permits, knowledge of some of the most important considerations may be useful for addressing items that are not specified in permits or for requesting changes in the permit specifications to enhance the scientific basis or the cost-effectiveness of the toxicity testing program. The major kinds of considerations include the following:

- Study design specifications
- Sample collection, storage, and handling methods
- Laboratory methods
- Data analysis.

Study design specifications are addressed below. Sampling, laboratory, and data analysis methods are addressed in the User's Guide (PTI 1994). For more detailed discussions of sediment collection and handling procedures, laboratory techniques, and quality assurance considerations, the user is referred to information in specific methods. ASTM (1989, 1990, 1991a,b) and 40 CFR Part 792 contain discussions that may be applicable for a variety of sediment testing procedures.

STUDY DESIGN SPECIFICATIONS

The study design specifications lay the groundwork for the success or failure of a toxicity testing program. If a study is not designed properly, the best field collection methods, laboratory methods, and data analysis techniques may not overcome a poor design to provide a reliable assessment of sediment toxicity. Perhaps the most important study design consideration is a clearly defined statement of the objectives of the study. Objectives should be the ultimate guidelines for all subsequent decisions related to a toxicity evaluation. If the objectives are too general or are stated poorly, they will provide little guidance and the usefulness of the results of the toxicity evaluation may be jeopardized.

It is essential that the study objectives be sufficiently detailed and clear to adequately guide the evaluation. For example, the selection of species for toxicity testing is influenced by study objectives. If the study objective is to assess potential effects of contaminated sediments on the ecosystem, then a variety of species and organism level endpoints could be selected as surrogates for ecosystem level effects. If the real objective of a study is to assess potential sediment toxicity to a commercially harvested clam species at a site, use of a bivalve larval development test would be more appropriate than a test with a worm species or a fish species. Thus, failure to define the specific species or ecological components to be protected could reduce the value of the results.

Additional major study design specifications that are guided by the study objectives include the general assessment approach, the kind of toxicity tests to use, the method of assessing adverse effects, and the collection of ancillary information (e.g., sediment grain size, sediment chemistry).

GENERAL ASSESSMENT APPROACH

A key consideration when developing the general approach for a toxicity evaluation is whether to conduct the evaluation in a tiered manner. Using a tiered approach, a sensitive screening evaluation precedes one or more detailed, definitive evaluations. The definitive evaluations are conducted only at stations where the screening evaluation indicates that sediments are toxic. For example, a rapid and relatively inexpensive toxicity test (e.g., Microtox®) could be used to select those stations at which additional toxicity tests or other kinds of information (e.g., sediment chemical concentrations, evaluations of benthic macroinvertebrate assemblages) should be evaluated. The primary benefits of a tiered approach are that it focuses the evaluation effort on a subset of stations and thereby reduces the cost of the overall evaluation. Within each tier, the data analysis methods should be specified based on the study objectives and the planned uses of the data (for further discussion, see *Data Analysis Issues* in the User's Guide [PTI 1994]).

The general assessment strategy should also address the primary uses of the data, which in turn will influence the data analysis approach. For example, assessment of the potential effects of a point discharge may involve collecting data from a geographic distribution of stations along an expected gradient of suspected contamination and effects. The data would be analyzed graphically or statistically to evaluate gradual trends or gradients in toxicity in relation to chemical concentrations in sediments. In contrast, an assessment of a heterogeneous study area may need to rely on comparison of the results from each station with those from a reference site.

SELECTION OF TOXICITY TESTS

To provide an accurate evaluation of sediment toxicity, it is essential that the appropriate toxicity tests are selected for a specific discharge site. Some key considerations for selecting a test include the test species; the life stage tested; the test endpoints; the exposure period; and the reliability, ecological relevance, exposure relevance, and availability of the test. All of these considerations are addressed in other sections of this report.

EFFECTS ASSESSMENT

The method of effects assessment influences both the number and locations of sampling stations. Two of the most common methods are the reference area approach and the gradient approach. For the reference area approach, effects are evaluated by comparing the toxicity results found in a potential problem area with those from a reference area. Usually, these comparisons are made statistically and an effect is identified when the results for the potential problem area are significantly different ($P \leq 0.05$) from the reference results. The reference area is defined as an area that is as similar as possible to the potential problem area, except that it is not influenced by the chemicals of interest. In streams, reference areas are typically located upstream from the discharge area. The application of the reference area approach is sometimes limited by difficulty in finding a suitable reference area, especially with regard to sediment characteristics. In coastal environments, many discharge sites are located in embayments with relatively fine-grained sediments (i.e., silts and fine sands). In such cases, it is especially important that the reference area also have a similar sediment grain size. For some sediment tests, there is a potential for reaching an erroneous conclusion concerning significant toxicity if a coarser sediment is used for reference tests. For example, the optimal habitat of the test species may be coarse sediments and any suboptimal survival, growth, or reproduction that was observed in the test sediments relative to the reference could be related to habitat preference. Use of formulated reference sediments in toxicity testing programs can provide a standard material for comparison with study site sediments, while avoiding some of the problems of using naturally collected reference sediments (Suedel and Rodgers 1994).

For the gradient approach to effects assessment, three or more stations are located at increasing distances from a discharge point. These stations are usually located downstream or downcurrent. Effects are evaluated by determining whether a decrease in toxicity occurs with increasing distance from a potential problem area. These comparisons can be made graphically or statistically using correlation analysis. Effects are identified when an obvious relationship or a statistically significant ($P \leq 0.05$) correlation is found between toxicity and distance from the potential problem area. The application of the gradient approach can be limited by other chemicals or conventional variables (e.g., sediment grain size, sediment organic carbon content) that vary along the spatial gradient and thereby confound the toxicity results.

ANCILLARY INFORMATION

During most toxicity evaluations, it is useful to collect various kinds of ancillary information that may assist in the interpretation of the toxicity results, in the selection of reference areas, and in the evaluation of environmental gradients. Three major kinds of ancillary information are supplemental sediment variables, sediment chemical concentrations, and *in situ* biological effects.

Supplemental Sediment Variables

Because the results of toxicity tests can be affected by supplemental variables, it is advisable to measure the major variables that are known to influence each kind of test. For example, an important supplemental variable for most tests that rely on exposure to whole sediments is the grain size of the sediments. Other important supplemental variables that should be considered include sediment organic content (measured as percent organic carbon) and the concentrations of ammonia and sulfides in interstitial water.

Most organisms living in or on sediments have specific preferences and/or requirements concerning the physical/chemical characteristics of those sediments. The characteristics of sediments can independently affect the response of test organisms, thus confounding the interpretation of test results. For example, water column organisms (e.g., plankton, fishes) may not encounter whole sediments but still may be adversely affected by high concentrations of suspended sediments. Although it is widely acknowledged that physical/chemical characteristics of sediments may directly affect responses, there has been little experimental work conducted in this area. For most species, there is no information available. In tests of naturally occurring reference sediments and in experimental manipulations of grain size composition, DeWitt *et al.* (1988, 1989) found that the amphipod *R. abronius* was apparently sensitive to fine-grained material or some correlated factor. Recently, Suedel and Rodgers (in press) showed that survival of the freshwater midge *C. tentans* was reduced in formulated reference sediments with less than 0.76–0.96 percent organic matter. These authors demonstrated that this species was tolerant of a wide range of sediment particle sizes (0–100 percent sand, 0–100 percent silt, and 0–60 percent clay). *H. azteca* was tolerant of a similarly wide range of particle sizes and various levels of organic matter (0.12–7.8 percent). PTI (1991) tested 21 naturally occurring reference sediments that varied greatly in percent sand/silt fraction (3.2–96 percent) and total organic carbon (0.2–2.6 percent) using *R. abronius* mortality, *Neanthes* sp. biomass, Microtox® bacteria luminescence, *Crassostrea gigas* larval abnormality, and *Dendraster excentricus* embryo abnormality tests. Only the latter two tests showed a correlation between the response and sediment parameters (i.e., sediment grain size). Even in these cases, embryo abnormality was less than 11 percent after exposure to the reference sediments.

The effects of sediment particle size on survival of the amphipod *R. abronius* have been evaluated in detail. DeWitt *et al.* (1988, 1989) concluded that elevated mortality in the *R. abronius* test can result solely from exposure of the organisms to sediments that have a high percentage of fine-grained material (i.e., silt/clay). Elevated mortality found in reference area sediments without apparent chemical contamination was attributed to physical characteristics of the sediments or some other natural factor that correlated with the physical characteristics. The distribution of observations compiled by DeWitt *et al.* (1988, 1989) suggests that adverse effects of fine-grained sediments on *R. abronius* are found only in some reference area sediments and that high survival in fine-grained sediments may be observed for some reference sites. In an independent study, PTI (1991) evaluated the relationship between amphipod (*R. abronius*) mortality and percent fine-grained material in sediments by analyzing samples from 21 stations in reference areas of Puget Sound. For this data set, amphipod survival was not significantly related to percent fine-grained material in sediment.

Although the response of *R. abronius* to fine-grained particles is not consistent, the measured toxicity in some cases probably reflects the natural preference of this species for sandy substrates. Other amphipods may have broader sediment preferences and may be better test species in extremely fine-grained sediments (e.g., >90 percent silt/clay). For example, recent studies of the East Coast amphipod *L. plumulosus* indicate that its survival is not adversely affected by sediments with silt/clay fractions of more than 90 percent and that this species commonly occurs in both muddy and sandy habitats (Schlekat *et al.* 1992). Similarly, the West Coast estuarine amphipod *E. estuarius* generally shows high survival in fine-grained sediments (DeWitt *et al.* 1989).

In summary, there is little definitive information on the effects of potentially confounding variables on sediment toxicity test results. However, it is important that such factors be considered in the selection of candidate tests. The user should consult the available information in Appendix D.

Sediment Chemical Concentrations

The evaluation of sediment chemical concentrations in conjunction with sediment toxicity is often useful for identifying the chemicals that may be causing any observed toxicity and for determining whether sediment conventional variables or other confounding factors may have influenced the results of toxicity results. The influence of confounding factors is suspected when significant toxicity results are found in the absence of elevated sediment chemical concentrations. In designing a sediment chemistry evaluation, it is important to define the chemicals of potential concern based on knowledge of sources, the level of accuracy and sensitivity (i.e., detection limits) required, and the quality assurance and quality control procedures for analytical laboratories. Some analytical requirements, such as detection limits, may be defined in guidance from a relevant regulatory agency. The level of chemical analysis should be matched to that of the toxicity test design. For example, a screening-level chemistry method would be appropriate for a reconnaissance survey of a large area to focus further investigations on specific high priority areas. Complete characterization of sediment chemicals in an area might require specialized analytical methods and considerable documentation of results, which would only be appropriate when detailed toxicity evaluations are conducted. PTI (1992) provides detailed guidance on designing chemical analyses and evaluating results. Because the costs of sediment chemical evaluations can be high, these evaluations are used most efficiently as part of a tiered assessment strategy in which they are conducted only at selected stations.

In Situ Biological Effects

The evaluation of *in situ* biological effects in conjunction with toxicity testing provides real world verification of the toxicity results. Because there are many uncertainties associated with extrapolating the results of laboratory evaluations to the field setting, the actual measurement of biological effects in the field is a useful method of addressing those uncertainties and confirming the presence or absence of toxicity. The group of organisms that is used most frequently for field assessments of sediment toxicity is benthic macroinvertebrates because many of these organisms are relatively stationary and therefore highly susceptible to chemical exposure from fixed sources. Because the costs of evaluations of *in situ* biological effects can be high, these evaluations are used most efficiently as part of a tiered assessment strategy in which they are conducted only at selected stations.

Diaz (1992) and La Point and Fairchild (1992) discuss approaches for using benthic community structure analyses in assessing contaminated sediments. La Point and Fairchild (1992) emphasize assessing benthic macroinvertebrate community structure because of the limitations in using fishes or periphyton. These authors and Diaz (1992) provide information on the advantages and limitations of various measures of community structure, including species composition, diversity, evenness, multivariate resemblance indices (i.e., indicators of similarity in faunal composition and abundance between sites), biomass, and indicator species abundances. Each study should use several measures of community structure because no one measure is a reliable indicator of contaminant effects. Caution is needed to avoid confounding effects of natural factors that may obscure the potential effects of contaminants or lead to false positives. For example, comparisons of benthic community measures between potentially affected sites and reference sites should be stratified by season and by habitat features (e.g., sediment grain size and organic carbon).

Chapman *et al.* (1987) illustrate the use of macroinvertebrate community measures in combination with sediment toxicity tests and sediment chemistry in an integrated approach termed the sediment quality triad. Synoptic collection of all three types of data is preferred to minimize uncertainty in the combined results (Chapman *et al.* 1992). Although aliquots for analysis of sediment chemistry and toxicity can be taken from the same sample, separate samples are needed for analysis of benthic macroinvertebrates. Despite its relatively high cost and the high level of expertise needed for interpretation of the data, the triad approach represents one of the most scientifically defensible and relevant approaches to assessment of sediment contamination. Chapman *et al.* (1992) provide further details on the application of the triad approach.

CONCLUSIONS

Reliable sediment toxicity tests are currently available for testing of marine, estuarine, and freshwater sediments. Many of the well standardized tests are documented in methods or guidance manuals developed by ASTM, EPA, and Environment Canada. However, fewer than 25 percent of the sediment toxicity tests in each category (e.g., marine lethal or estuarine sublethal) have undergone intra- and inter-laboratory variability studies. Few comparative studies of multiple tests have been conducted. Also, most sediment toxicity tests have not undergone field validation tests in which the toxicity results are correlated with chemical concentrations in sediments and effects on indigenous benthic fauna.

In this study, the criteria used to evaluate sediment toxicity tests were reliability, ecological relevance, exposure relevance, availability, interferences, and chemical discrimination. Because little information was available on the latter two categories for most tests, their influence on the overall technical rating scores was moderated by use of a weighting factor.

Most of the highly ranked marine and estuarine infaunal tests were based on the use of amphipods as test organisms, whereas most of the highly ranked freshwater infaunal tests were based on the use of insects (mayfly nymphs and midge larvae) as test organisms. These species groups are ecologically important, especially as key prey items for various fishes. In most cases, the highest ranking tests were the ones based on the exposure of infaunal organisms to whole sediments because: 1) exposure conditions closely mimic field conditions; 2) most of the test species are available by field collection during most of the year; and 3) many of the tests have well-developed methods.

Many of the lowest ranking toxicity tests involve exposure of planktonic organisms to whole sediments. The exposure relevance of these tests is relatively low because the test species are rarely exposed to sediments in the field and they may be sensitive to interference of suspended sediments with feeding mechanisms.

The species included in the highest ranking marine and estuarine tests for lethality include the following amphipods: *A. abdita*, *R. abronius*, *G. japonica*, *E. washingtonianus*, *E. estuarius*, *A. virginiana*, *F. xiximeus*, *C. volutator*, *L. pinguis*, and *L. plumulosus*. Reproductive endpoints are also well developed for the *L. plumulosus* test. Although behavioral endpoints (e.g., reburial at exposure termination) are used in many of these amphipod tests, the behavioral endpoints have generally not been field validated. The tests based on *A. abdita* and *R. abronius* are the only ones with a high regulatory status.

Taxonomic groups other than amphipods also ranked high among the marine and estuarine sublethal tests, including the polychaete (*Neanthes* sp.) growth test based on a 20–28 day exposure to whole sediments, the purple sea urchin (*S. purpuratus*) fertilization test of sediment elutriates, and the bivalve (*M. edulis*) larval abnormality test of sediment elutriates. Although these elutriate tests have a lower exposure relevance than the whole sediment tests, they use sensitive life stages of ecologically important species, are widely available, and have well developed methods. Although these elutriate tests are generally reliable, their variability can be high and the negative controls fail quality assurance limits more frequently than those in the tests involving juveniles and adults of these or other species.

The highest ranking freshwater tests for lethal and sublethal endpoints were based on the exposure of infaunal insects (i.e., nymphs of the mayfly *H. limbata* and larvae of the midges *C. riparius* and *C. tentans*) and an epifaunal amphipod (*H. azteca*) to whole sediments. Only the *H. azteca* and *C. tentans* lethal tests have high regulatory status. Whole sediment tests with vascular plants (*H. verticillata* and *E. crusgalli*) were among the top six ranked tests in the freshwater lethal category. These tests ranked high primarily because of their high degrees of exposure and ecological relevance and their relatively low susceptibility to interferences. The high ecological relevance of the two tests is based on the importance of the plants in providing habitat for other organisms. The major drawback of these two tests is their infrequent use in regulatory programs.

There is a relative paucity of information on interferences and chemical discrimination for sediment toxicity tests. Further research in these areas and more comparative studies of toxicity tests with corresponding data on the bioavailability of sediment chemicals are needed.

GLOSSARY

Acute toxicity — The ability of a chemical to cause a toxic response in organisms immediately or shortly after exposure to the chemical.

Adverse effect — An impairment of biological functions or description of ecological processes that results in unfavorable changes in an ecological system.

Amphipod — A small shrimp-like member of one subgroup of the large group of animals called Crustacea, which includes crayfish, lobsters, shrimps, and crabs.

Aquatic — Living or growing in water.

Benthic — Pertaining to, or associated with, the bottom of a body of water.

Biomass — The total weight of live organisms in a sampled population.

Biotic group — A group of related organisms with generally similar body structure and function.

Chronic toxicity — The ability of a chemical to produce a toxic response when an organism is exposed over a long period of time, generally corresponding to a substantial part of the organism's life cycle.

Concentration — The amount of a chemical expressed relative to amount of environmental medium (e.g., μg/L [micrograms of chemical per liter of water] or μg/g [micrograms of chemical per gram of sediment]).

Control sediment — A sediment essentially free of chemicals and compatible with the biological needs of the test organisms such that it has no discernable influence on the response being measured in the test. Control sediment may be the sediment from which the test organisms are collected or a laboratory sediment, provided the organisms meet control standards. Test procedures are conducted with the control sediment in the same way as the reference sediment and test material. The purpose of the control sediment is to confirm the biological acceptability of the test conditions and to help verify the health of the organisms during the test. Excessive mortality in the control sediment indicates a problem with the test conditions or organisms and can invalidate the results of the corresponding test.

Ecosystem	An ecological community, together with its physical habitat, considered as a unit.
Embryo	A plant or animal in the very early stages of development following fertilization of the egg.
Elutriate	A liquid solution used for toxicity testing, which is prepared by adding water to the sediment, shaking, and centrifuging to separate the solids.
Endpoint	The biological or ecological unit or variable being measured or assessed. The number of organisms dead at the end of an exposure is a lethal endpoint.
Epibenthic	Inhabiting the sediment surface, or closely associated with the sediment surface, rather than dwelling buried within the sediments.
Estuarine	Surface water containing greater than 0.5 parts per thousand (ppt) salinity and less than 28 ppt salinity.
Exposure	Contact between an organism and a chemical in the environment.
Fresh water	Surface water containing less than or equal to 0.5 ppt salinity.
Foundation species	A species that provides important physical habitat for other species in a biological community (e.g., marsh grass).
Hardness	A measure of the calcium and magnesium concentrations in water.
In situ	In the natural or original position (occurring in nature, and not in the laboratory).
Infaunal	Refers to animals living in the sediments, including such forms as worms and clams.
Interference	Physical elements or chemical compounds that cause bias in the results of a toxicity test.
Keystone species	A species that controls the species composition and relative abundances of species in a community by its predatory (or grazing) effects (e.g., by grazing on kelp, purple urchins prevent the establishment of kelp beds and maintain open rocky subtidal communities).
Interstitial water	Water that fills the spaces between sediment particles. Often referred to as "pore water."

Larval	Relating to the juvenile form of certain invertebrate animals that must undergo metamorphosis before assuming adult characteristics.
Lethal	Causing death; mortality (or survival) is the endpoint for lethal toxicity tests.
Life stage	A developmental stage of an organism (e.g., egg, larva, embryo, juvenile, adult).
Macroinvertebrate	An invertebrate (without a backbone) organism visible to the naked eye (e.g., >1.0 mm). Often refers to animals such as insects, worms, clams, and snails.
Marine	Surface water containing 28 ppt salinity or greater.
Medium (plural: media)	The substance in which a chemical may exist. Air, sediment, and water are all media.
Midge	A group of true flies (similar to mosquitos) that have aquatic larvae and non-biting adults. They are one of the most abundant groups of aquatic insects.
Monitoring	Periodic testing of water and sediment quality or of biota to verify continued compliance with the requirements of a discharge permit or other authorization.
Nektonic	Refers to the nekton, the group of active swimmers that are capable of strong, independent movement in the water. Examples include many juvenile and adult fishes and large invertebrates (e.g., squid).
Organism	An individual plant or animal.
Ovigerous	Refers to females bearing eggs.
Planktonic	Refers to the plankton, the group of small plants and animals that are weak swimmers and tend to drift with the current.
Population	A group of individuals of the same species interacting within a given habitat.
Precision	The ability to replicate a value; the degree to which observations or measurements of the same property, usually obtained under similar conditions, conform to themselves. Usually expressed as standard deviation, variance, or range.
Quality assurance and quality control	A system of procedures, checks, audits, and corrective actions to ensure that all research design and performance, environmental

monitoring and sampling, and other technical and reporting activities are of the highest achievable quality.

Reference sediment A sediment, substantially free of chemicals, that is as similar as practicable to the grain size of the test material and the sediment at the disposal site and that reflects the conditions that would exist in the vicinity of the site had no industrial activity ever taken place but had all other influences on sediment condition taken place.

Reference area An area that has similar characteristics to a site being evaluated but that is unaffected by chemicals of potential concern. The reference area is compared to the site to assess the effects of chemicals of potential concern.

Route The mechanism of contact between an organism and a toxic chemical (e.g., ingestion or dermal contact).

Site-specific Of or relating to a particular area or location.

Sediments Material, such as sand, silt, or clay, suspended in or settled on the bottom of a water body.

Sublethal Causing an endpoint other than death; growth is a sublethal endpoint in toxicity tests.

Terrestrial Living or growing on land.

Toxicity test A test in which organisms are exposed to chemicals in a test medium (e.g., waste, sediment, soil) to determine the effects of exposure.

Trophic Relating to food or feeding relationships. Trophic levels consist of producers (plants), herbivores or primary consumers, carnivores or secondary consumers, and top carnivores or tertiary consumers.

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APPENDIX A

Classification of Sediment Toxicity Tests

CLASSIFICATION OF SEDIMENT TOXICITY TESTS

The tables contained in this appendix list the sediment toxicity tests considered in the detailed evaluation (Table A-1) and those that were eliminated early in an initial screening (Attachment 1) (see *Test Screening Approach*, *Selection of Tests for Evaluation* in the main text for the criteria for selecting tests for the detailed evaluation). The major characteristics of the tests that were used to classify them included:

- Habitat type
  - Marine
  - Estuarine
  - Freshwater
- Endpoint type
  - Lethal (i.e., mortality)
  - Sublethal (e.g., growth, reproduction, abnormality)

Within each category, the tests were distinguished by the following key characteristics:

- Exposure medium
  - Sediment
  - Sediment elutriate (water extractable)
  - Interstitial water
  - Sediment extract (solvent extractable)
- Characteristics of the test organism
  - Biotic group (e.g., infauna, planktonic)
  - Species
  - Life stage
- Test duration.

These characteristics are discussed in the main text (see *Classification of Available Test Methods*).

TABLE A-1. CLASSIFICATON OF SEDIMENT TOXICITY TESTS – MARINE LETHAL

Test No.	Exposure Media	Biotic Group	Life Stage	Test Species	Duration	Reference
001	S	Amphipod	Juv or adult females	Ampelisca abdita	10 days	ASTM (1990) (E1367–90); U.S. EPA (1991b); U.S. EPA and U.S. COE (1991, 1993); Dewitt et al. (1992)
002	S	Amphipod	Juv or young adults	Amphiporeia virginiana	10 days	Environment Canada (1992a)
003	S	Amphipod	Juv or young adults	Eohaustorius washingtonianus	10 days	Environment Canada (1992a)
004	S	Amphipod	Juv or young adults	Foxiphalus xiximeus	10 days	Environment Canada (1992a), based on ASTM (1990) (E1367–90); Swartz et al. (1985)
005	S	Amphipod	Immature	Grandidierella japonica	10 days	ASTM (1990) (E1367–90); Reish and Lemay (1988); U.S. EPA and U.S. COE (1993)
006	S	Amphipod	Juv or young adults	Leptocheirus pinguis	10 days	Environment Canada (1992a), based on ASTM (1990) (E1367–90); Swartz et al. (1985)
008	S	Amphipod	Adults	Rhepoxynius abronius	10 days	ASTM (1990) (E1367–90); U.S. EPA (1991b); U.S. EPA and U.S. COE (1991, 1993); Environment Canada (1992a)
009	EL	Bivalve	Embryos	Crassostrea virginica	48 hrs	ASTM (1989) (E724–89); U.S. EPA and U.S. COE (1991, 1993)
010	S	Bivalve	Embryos	Crassostrea gigas	48–60 hrs	U.S. EPA (1991b)
011	EL	Bivalve	Embryos	Crassostrea gigas	48 hrs	ASTM (1989) (E724–89); U.S. EPA and U.S. COE (1993)
014	EL	Bivalve	Embryos	Mercenaria mercenaria	48 hrs	ASTM (1989) (E724–89)
015	S	Bivalve	Juveniles	Mulinia lateralis	7 days	Burgess et al. (1992)
016	S	Bivalve	Embryos	Mytilus edulis	48–60 hrs	U.S. EPA (1991b)
017	EL	Bivalve	Embryos	Mytilus edulis	48 hrs	ASTM (1989) (E724–89); Chapman and Morgan (1983); Long et al. (1990); U.S. EPA and U.S. COE (1991, 1993)
018	EL	Bivalve	Embryo–larval	Ostrea sp.	48 hrs	U.S. EPA and U.S. COE (1991)
020	S	Bivalve	Juveniles	Protothaca staminea	10 days	Swartz et al. (1979); U.S. EPA and U.S. COE (1991)
021	S	Bivalve	Juveniles	Tapes japonica	10 days	U.S. EPA and U.S. COE (1991)
023	S	Bivalve	Juveniles	Yoldia limatula	10 days	U.S. EPA and U.S. COE (1991)
026	EL	Crustacean	Embryo–larval	Callinectes sapidus	48 hrs	U.S. EPA and U.S. COE (1991)
027	S	Crustacean	Juveniles	Callinectes sapidus	10 days	U.S. EPA and U.S. COE (1991)
028	EL	Crustacean	Embryo–larval	Cancer sp.	48 hrs	U.S. EPA and U.S. COE (1991)
029	S	Crustacean	Juveniles	Cancer sp.	10 days	U.S. EPA and U.S. COE (1991)
030	S	Crustacean	Juveniles	Crangon sp.	10 days	U.S. EPA and U.S. COE (1991)
031	EL	Crustacean	Post–hatch	Palaemonetes sp.	96 hrs	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
032	EL	Crustacean	Embryo–larval	Pandalus sp.	48 hrs	U.S. EPA and U.S. COE (1991)
033	S	Crustacean	Juveniles	Pandalus sp.	10 days	U.S. EPA and U.S. COE (1991)
034	S	Crustacean	Post–larvae	Penaeus sp.	10 days	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
035	EL	Crustacean	Post–larvae	Penaeus sp.	96 hrs	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
037	S	Crustacean	Juveniles	Sicyonia ingentis	10 days	U.S. EPA and U.S. COE (1991)
038	S	Echinoderm	Embryos	Arbacia punctulata	72–96 hrs	Lamberson et al. (1992), following Dinnel and Stober (1985); U.S. EPA (1991b)
039	S	Echinoderm	Juveniles	Dendraster excentricus	28 days	Casillas et al. (1992a)
040	S	Echinoderm	Embryos	Dendraster excentricus	48–96 hrs	U.S. EPA (1991b)
041	EL	Echinoderm	Embryos	Dendraster excentricus	48 hrs	U.S. EPA and U.S. COE (1993)
043	S	Echinoderm	8–22 mm diameter	Lytechinus pictus	60 days	Thompson et al. (1989)

TABLE A-1. (cont.)

Test No.	Exposure Media	Biotic Group	Life Stage	Test Species	Duration	Reference
044	EL	Echinoderm	Embryos (< 1 hr old)	Lytechinus pictus	48-96 hrs	Reish and Lemay (1988); U.S. EPA and U.S. COE (1993)
045	EL	Echinoderm	Embryos (< 1 hr old)	Strongylocentrotus purpuratus	48 hrs	U.S. EPA and U.S. COE (1993); Dinnel et al. (1982)
046	S	Echinoderm	Embryos	Strongylocentrotus purpuratus	48-96 hrs	U.S. EPA (1991b)
048	S	Echinoderm	Embryos	Strongylocentrotus droebachiensis	48-96 hrs	U.S. EPA (1991b)
049	EL	Echinoderm	Embryos (< 1 hr old)	Strongylocentrotus sp.	48 hrs	U.S. EPA and U.S. COE (1993)
051	EL	Fish	Juveniles	Citharichthys stigmaeus	96 hrs	Reish and Lemay (1988); U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
053	EL	Fish	Embryo-larval	Coryphaena hippurus	48 hrs	U.S. EPA and U.S. COE (1991)
054	EL	Fish	Embryo-larval	Cymatogaster aggregata	48 hrs	U.S. EPA and U.S. COE (1991)
055	EL	Fish	1-14 days old	Cyprinodon variegatus	96 hrs	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
056	EL	Fish	Juveniles	Gasterosteus aculeatus	96 hrs	Environment Canada (1990c)
057	S	Fish	Larvae	Hypomesus pretiosus	10 days	Chapman et al. (1985)
058	S	Fish	Larvae	Hypomesus pretiosus	96 hrs	Casillas et al. (1992b)
059	EL	Fish	Embryo-larval	Lagodon rhomboides	48 hrs	U.S. EPA and U.S. COE (1991)
060	EL	Fish	Embryo-larval	Leiostomus xanthurus	48 hrs	U.S. EPA and U.S. COE (1991)
061	EL	Fish	Embryo-larval	Leuresthes tenuis	48 hrs	Reish and Lemay (1988); U.S. EPA and U.S. COE (1991)
062	S	Mysid	1-5 days old	Holmesimysis sp.	10 days	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
063	EL	Mysid	1-5 days old	Holmesimysis sp.	96 hrs	Reish and Lemay (1988); U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
064	S	Mysid	1-5 days old	Mysidopsis sp.	10 days	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
065	EL	Mysid	1-5 days old	Mysidopsis sp.	96 hrs	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
071	S	Polychaete	Juveniles	Neanthes sp.	10 days	Tay et al. (1992); U.S. EPA and U.S. COE (1991, 1993); Dillon et al. (1993)

Notes: EL - elutriate
EX - extract
INT - interstitial water
S - sediment

TABLE A-2. CLASSIFICATION OF SEDIMENT TOXICITY TESTS – MARINE SUBLETHAL

Test No.	Exposure Media	Biotic Group	Life Stage	Test Species	Duration	Reference
001	S	Amphipod	Juv or adult females	Ampelisca abdita	10 days	ASTM (1990) (E1367-90)
005	S	Amphipod	Immature	Grandidierella japonica	10 days	ASTM (1990) (E1367-90)
008	S	Amphipod	Adults	Rhepoxynius abronius	10 days	ASTM (1990) (E1367-90)
042	EL	Echinoderm	Gametes	Dendraster excentricus	80 min	U.S. EPA and U.S. COE (1993)
047	EL	Echinoderm	Gametes	Strongylocentrotus purpuratus	80 min	U.S. EPA and U.S. COE (1993); Dinnel et al. (1982)
080	INT	Bacterium	Cells	Photobacterium phosphoreum	15 min	Tay et al. (1992)
081	EL	Bacterium	Cells	Photobacterium phosphoreum	15 min	Microbics (1992)
083	S	Bacterium	Cells	Photobacterium phosphoreum	5-60 min	Environment Canada (1992f)
084	EX	Bacterium	Cells	Salmonella typhimurium	N/S	Jarvis and Reilly (1992)
009	EL	Bivalve	Embryos	Crassostrea virginica	48 hrs	ASTM (1989) (E724-89)
010	S	Bivalve	Embryos	Crassostrea gigas	48-60 hrs	U.S. EPA (1991b)
011	EL	Bivalve	Embryos	Crassostrea gigas	48 hrs	ASTM (1989) (E724-89)
014	EL	Bivalve	Embryos	Mercenaria mercenaria	48 hrs	ASTM (1989) (E724-89)
015	S	Bivalve	Juveniles	Mulina lateralis	7 days	Burgess et al. (1992)
016	S	Bivalve	Embryos	Mytilus edulis	48-60 hrs	U.S. EPA (1991b)
017	EL	Bivalve	Embryos	Mytilus edulis	48 hrs	ASTM (1989) (E724-89)
097	INT	Echinoderm	Gametes	Arbacia punctulata	1.3 hrs	Burgess et al. (1993)
098	EL	Echinoderm	Gametes	Arbacia punctulata	1.3 hrs	Burgess et al. (1993)
038	S	Echinoderm	Gametes	Arbacia punctulata	72-96 hrs	Lamberson et al. (1992), following Dinnel and Stober (1985), U.S. EPA (1991b)
100	EL	Echinoderm	Gametes	Arbacia punctulata	20, 40, 60 min	Environment Canada (1992b)
101	S	Echinoderm	Gametes	Arbacia punctulata	20, 40, 60 min	Environment Canada (1992b)
040	S	Echinoderm	Embryos	Dendraster excentricus	48-96 hrs	U.S. EPA (1991b)
039	S	Echinoderm	Juveniles	Dendraster excentricus	28 days	Casillas et al. (1992a)
102	EL	Echinoderm	Gametes	Dendraster excentricus	20, 40, 60 min	Environment Canada (1992b)
103	S	Echinoderm	Gametes	Dendraster excentricus	20, 40, 60 min	Environment Canada (1992b)
043	S	Echinoderm	8-22 mm diameter	Lytechinus pictus	60 days	Thompson et al. (1989)
107	EL	Echinoderm	Gametes	Lytechinus pictus	20, 40, 60 min	Environment Canada (1992b)
108	S	Echinoderm	Gametes	Lytechinus pictus	20, 40, 60 min	Environment Canada (1992b)
111	EL	Echinoderm	Gametes	Strongylocentrotus purpuratus	80 min	Long et al. (1990)
112	EL	Echinoderm	Gametes	Strongylocentrotus purpuratus	48 hrs	Long et al. (1990)
046	S	Echinoderm	Embryos	Strongylocentrotus purpuratus	48-96 hrs	U.S. EPA (1991b)
109	EL	Echinoderm	Gametes	Strongylocentrotus purpuratus	20, 40, 60 min	Environment Canada (1992b)
110	S	Echinoderm	Gametes	Strongylocentrotus purpuratus	20, 40, 60 min	Environment Canada (1992b)
048	S	Echinoderm	Embryos	Strongylocentrotus droebachiensis	48-96 hrs	U.S. EPA (1991b)
115	EL	Echinoderm	Gametes	Strongylocentrotus droebachiensis	20, 40, 60 min	Environment Canada (1992b)
116	S	Echinoderm	Gametes	Strongylocentrotus droebachiensis	20, 40, 60 min	Environment Canada (1992b)
058	S	Fish	Larvae	Hypomesus pretiosus	96 hours	Casillas et al. (1992b)
121	S	Polychaete	Juveniles	Neanthes sp.	20-28 days	Dillon et al. (1993); U.S. EPA (1990); Johns et al. (1992)

Notes

EL – elutriate
EX – extract
INT – interstitial water
S – sediment

N/A – not applicable
N/S – not specified

EC – Environment Canada (1990, 1992)
GB – U.S. EPA and U.S. COE (1991)
IN – U.S. EPA and U.S. COE (1993)
TT – U.S. EPA (1992)

TABLE A-3. CLASSIFICATION OF SEDIMENT TOXICITY TESTS – ESTUARINE LETHAL

Test No.	Exposure Media	Biotic Group	Life Stage	Test Species	Duration	Reference
001	S	Amphipod	Juv or adult females	Ampelisca abdita	10 days	ASTM (1990) (E1367-90); U.S. EPA (1991b); U.S. EPA and U.S. COE (1991, 1993)
002	S	Amphipod	Juv or young adults	Amphiporeia virginiana	10 days	Environment Canada (1992a)
126	S	Amphipod	Juv or young adults	Corophium volutator	10 days	Environment Canada (1992a), based on ASTM (1990) (E1367-90); Swartz et al. (1985)
128	S	Amphipod	Adults	Corophium volutator	10 days	Tay et al. (1992), based on Swartz et al. (1985); U.S. EPA and U.S. COE (1993); ASTM (1990) (E1367-90)
129	S	Amphipod	Large juv and adults	Eohaustorius estuarius	10 days	ASTM (1990) (E1367-90); U.S. EPA and U.S. COE (1993); Environment Canada (1992a)
003	S	Amphipod	Juv or young adults	Eohaustorius washingtonianus	10 days	Environment Canada (1992a)
131	S	Amphipod	7-14 days old	Hyalella azteca	10 days	ASTM (1991b) (E1383-90); U.S. EPA and U.S. COE (1993)
133	S	Amphipod	Juveniles	Leptocheirus plumulosus	10 days	Dewitt et al. (1992)
134	S	Amphipod	Juveniles	Leptocheirus plumulosus	10 days	McGee et al. (1993)
135	S	Amphipod	Mixed sexes	Leptocheirus plumulosus	10 days	Schlekat et al. (1992); U.S. EPA and U.S. COE (1993)
009	EL	Bivalve	Embryos	Crassostrea virginica	48 hrs	ASTM (1989) (E724-89); U.S. EPA and U.S. COE (1991, 1993)
010	S	Bivalve	Embryos	Crassostrea gigas	48-60 hrs	U.S. EPA (1991b)
011	EL	Bivalve	Embryos	Crassostrea gigas	48 hrs	ASTM (1989) (E724-89); U.S. EPA and U.S. COE (1993)
014	EL	Bivalve	Embryos	Mercenaria mercenaria	48 hrs	ASTM (1989) (E724-89)
016	S	Bivalve	Embryos	Mytilus edulis	48-60 hrs	U.S. EPA (1991b)
017	EL	Bivalve	Embryos	Mytilus edulis	48 hrs	ASTM (1989) (E724-89); U.S. EPA and U.S. COE (1991, 1993)
020	S	Bivalve	Juveniles	Protothaca staminea	10 days	Swartz et al. (1979); U.S. EPA and U.S. COE (1991)
026	EL	Crustacean	Embryo-larval	Callinectes sapidus	48 hrs	U.S. EPA and U.S. COE (1991)
027	S	Crustacean	Juveniles	Callinectes sapidus	10 days	U.S. EPA and U.S. COE (1991)
030	S	Crustacean	Juveniles	Crangon sp.	10 days	U.S. EPA and U.S. COE (1991)
151	S	Crustacean	Post-hatch (1-4 days)	Palaemonetes sp.	10 days	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
153	EL	Fish	1-14 days old	Cyprinodon variegatus	96 hrs	U.S. EPA and U.S. COE (1993); U.S. EPA (1991a)
056	EL	Fish	Juveniles	Gasterosteus aculeatus	96 hrs	Environment Canada (1990c)
156	EL	Fish	9-14 days old	Leuresthes tenuis	96 hrs	U.S. EPA and U.S. COE (1993); U.S. EPA (1991a)
157	EL	Fish	9-14 days old	Menidia sp.	96 hrs	U.S. EPA and U.S. COE (1993); U.S. EPA (1991a)
158	EL	Fish	1-14 days old	Menidia sp.	96 hrs	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
064	S	Mysid	1-5 days old	Mysidopsis sp.	10 days	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
065	EL	Mysid	1-5 days old	Mysidopsis sp.	96 hrs	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
161	S	Mysid	1-5 days old	Neomysis sp.	10 days	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
162	EL	Mysid	1-5 days old	Neomysis sp.	96 hrs	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
165	INT	Polychaete	Females (1-2 days)	Dinophilus gyrociliatus	7 days	Long et al. (1990); Carr et al. (1989)

Notes: EL – elutriate
EX – extract
INT – interstitial water
S – sediment

TABLE A–4. CLASSIFICATION OF SEDIMENT TOXICITY TESTS – ESTUARINE SUBLETHAL

Test No.	Exposure Media	Biotic Group	Life Stage	Test Species	Duration	Reference
001	S	Amphipod	Juv or adult females	Ampelisca abdita	10 days	ASTM (1990) (E1367–90)
128	S	Amphipod	Adults	Corophium volutator	10 days	Tay et al. (1992)
129	S	Amphipod	Large juv and adults	Eohaustorius estuarius	10 days	ASTM (1990) (E1367–90)
133	S	Amphipod	Juveniles	Leptocheirus plumulosus	28 days	Dewitt et al. (1992)
134	S	Amphipod	Juveniles	Leptocheirus plumulosus	30 days	McGee et al. (1993)
135	S	Amphipod	Mixed sexes	Leptocheirus plumulosus	28 days	Schlekat et al. (1992)
175	S	Bacterium	Cells	Photobacterium phosphoreum	30 min	Brouwer et al. (1990)
176	EX	Bacterium	Cells	Photobacterium phosphoreum	15 min	Jacobs et al. (1992); Microbics (1992); U.S. EPA (1991b)
009	EL	Bivalve	Embryos	Crassostrea virginica	48 hrs	ASTM (1989) (E724–89)
010	S	Bivalve	Embryos	Crassostrea gigas	48–60 hrs	U.S. EPA (1991b)
011	EL	Bivalve	Embryos	Crassostrea gigas	48 hrs	ASTM (1989) (E724–89)
014	EL	Bivalve	Embryos	Mercenaria mercenaria	48 hrs	ASTM (1989) (E724–89)
016	S	Bivalve	Embryos	Mytilus edulis	48–60 hrs	U.S. EPA (1991b)
017	EL	Bivalve	Embryos	Mytilus edulis	48 hrs	ASTM (1989) (E724–89)
187	EL	Fish	Adults	Cyprinodon variegatus	96 hrs	Alden et al. (1988)
191	INT	Polychaete	Females (1–2 days)	Dinophilus gyrociliatus	7 days	Carr et al. (1989); Long et al. (1990)

Notes: EL – elutriate
EX – extract
INT – interstitial water
S – sediment

TABLE A-5. CLASSIFICATION OF SEDIMENT TOXICITY TESTS – FRESHWATER LETHAL

Test No.	Exposure Media	Biotic Group	Life Stage	Test Species	Duration	Reference
195	EX	Amphibian	Embryos	Xenopus laevis	96 hours	Dawson et al. (1988)
199	INT	Amphipod	Juveniles	Hyalella azteca	96 hrs	Ankley et al. (1991)
200	S	Amphipod	Juveniles	Hyalella azteca	10–14 days	ASTM (1991b) (E1383–90); U.S. EPA and U.S. COE (1993)
202	S	Bivalve	Juveniles	Anodonta imbecillis	10 days	U.S. EPA and U.S. COE (1993)
205	EL	Cladoceran	Neonates	Ceriodaphnia dubia	48–96hrs	Ankley et al. (1991); U.S. EPA and U.S. COE (1993); U.S. EPA (1991a); Environment Canada (1992e)
207	S	Cladoceran	Neonates	Ceriodaphnia dubia	48 hrs	Sassoon–Brickson and Burton (1991); Stemmer et al. (1990b)
208	EL	Cladoceran	Neonates	Daphnia magna	48–96 hrs	Nebeker et al. (1984); U.S. EPA and U.S. COE (1993); U.S. EPA (1991a)
209	INT	Cladoceran	Neonates	Daphnia magna	48 hrs	Giesy et al. (1988, 1990)
211	S	Cladoceran	Neonates	Daphnia magna	48 hrs	Nebeker et al. (1984); Stemmer et al. (1990a,b)
212	EL	Cladoceran	Neonates (< 24 hrs)	Daphnia magna	48 hrs	Environment Canada (1990a,d)
213	EL	Cladoceran	Neonates (< 24 hrs)	Daphnia pulex	96 hrs	U.S. EPA and U.S. COE (1993); U.S. EPA (1991a)
214	EL	Cladoceran	Neonates (< 24 hrs)	Daphnia pulex	48 hrs	Environment Canada (1990a)
220	EL	Fish	Approx. 4 days	Ictalurus punctatus	96 hrs	U.S. EPA and U.S. COE (1993); U.S. EPA (1991a)
222	EL	Fish	Approx. 4 days	Lepomis macrochirus	96 hrs	U.S. EPA and U.S. COE (1993); U.S. EPA (1991a)
226	EL	Fish	Larvae (<24 hrs old)	Pimephales promelas	7 days	Environment Canada (1992d)
229	EL	Fish	Approx. 4 days	Pimephales promelas	96 hrs	U.S. EPA and U.S. COE (1993); U.S. EPA (1991a)
232	EX	Fish	Embryo–larval	Pimephales promelas	6 days	Dawson et al. (1988)
233	S	Fish	Juveniles	Pimephales promelas	21 days	Krantzberg and Boyd (1992)
234	EL	Fish	Fry or fingerlings	Oncorhynchus mykiss	96 hrs	Environment Canada (1990b,e); U.S. EPA and U.S. COE (1993); U.S. EPA (1991a)
235	S	Fish	Egg–sac stage	Oncorhynchus mykiss	21 days	Birge et al. (1984); Krantzberg (1989); Krantzberg and Boyd (1992)
237	S	Insect	Neonates or larvae	Chironomus riparius	10–30 days	ASTM (1991b) (E1383–90); U.S. EPA and U.S. COE (1993); Ingersoll and Nelson (1990); Nelson et al. (1990)
238	S	Insect	2nd instar larvae	Chironomus tentans	10–14 days	ASTM (1991b) (E1383–90); U.S. EPA and U.S. COE (1993); Ingersoll and Nelson (1990); Nelson et al. (1990)
239	S	Insect	Young nymphs	Hexagenia limbata	10 days	Malueg et al. (1984a,b); Nebeker et al. (1984); U.S. EPA and U.S. COE (1993)
240	INT	Insect	Nymphs	Hexagenia limbata	168 hrs	Giesy et al. (1990)
241	S	Insect	Nymphs	Hexagenia limbata	168 hrs	Giesy et al. (1990)
242	S	Insect	Nymphs	Hexagenia limbata	21 days	Krantzberg and Boyd (1992)
244	EL	Nematode	L1 stage juveniles	Panagrellus redivivus	96 hrs	Burnett et al. (no date); Samoiloff et al. (1980)
249	S	Oligochaete	Mixed–age	Lumbriculus variegatus	10 days	U.S. EPA and U.S. COE (1993); Ankley et al. (1992); Bailey and Lui (1980)
250	S	Oligochaete	Large worms	Lumbriculus variegatus	14 days	Dermott and Munawar (1992)
251	EL	Oligochaete	Similar size	Pristina leidyi	48 hrs	Smith et al. (1991)
252	S	Oligochaete	Mixed age; similar size	Pristina leidyi	10–18 days	Smith et al. (1991); U.S. EPA and U.S. COE (1993)
255	S	Oligochaete	Mixed age	Tubifex tubifex	10 days	U.S. EPA and U.S. COE (1993); Reynoldson et al. (1991)
256	S	Plant	Seedlings	Echinochloa crusgalli	2 weeks	Walsh et al. (1991)

Notes: EL – elutriate
EX – extract
INT – interstitial water
S – sediment

TABLE A-6. CLASSIFICATION OF SEDIMENT TOXICITY TESTS – FRESHWATER SUBLETHAL

Test No.	Exposure Media	Biotic Group	Life Stage	Test Species	Duration	Reference
260	EL	Algae	Cells	Selenastrum capricornutum	48 hrs	Environment Canada (1992c)
261	INT	Algae	Cells	Selenastrum capricornutum	48 hrs	Burton et al. (1989)
262	S	Algae	Cells	Chlorella vulgaris	4–96 hrs	Munawar and Munawar (1987)
263	EL	Algae	Cells	*Chlorella vulgaris*	4–96 hrs	Munawar and Munawar (1987)
195	EX	Amphibian	Embryos	Xenopus laevis	96 hrs	Dawson et al. (1988)
200	S	Amphipod	Juveniles	Hyalella azteca	10–30 days	ASTM (1991b) (E1383–90)
269	S	Amphipod	Juveniles	Hyalella azteca	8–10 wks	Borgmann et al. (1989)
270	EX	Bacterium	Cells	Aeromonas hydrophila	20 hrs	Dutka and Kwan (1981); Flemming and Trevors (1989)
176	EX	Bacterium	Cells	Photobacterium phosphoreum	15 min	Jacobs et al. (1992)
272	*INT*	*Bacterium*	*Cells*	Photobacterium phosphoreum	30 min	Hoke et al. (1992)
273	S	Bacterium	Cells	Photobacterium phosphoreum	20 min	Microbics (1991)
274	S	Bacterium	Cells	Photobacterium phosphoreum	30 min	Brouwer et al. (1990)
084	EX	Bacterium	Cells	Salmonella typhimurium	N/S	Jarvis and Reilly (1992)
278	S	Bacterium	Cells	Vibrio fischeri	16–24 hrs	Microbics (1993)
279	EL	Bacterium	Cells	Vibrio fischeri	16–24 hrs	Microbics (1993)
285	EL	Cladoceran	Neonates	Ceriodaphnia dubia	7 ± 1 days	Environment Canada (1992e)
211	S	Cladoceran	Juvenile	Daphnia magna	7 days	Nebeker et al. (1984); see Ceriodaphnia dubia (Environment Canda 1992e)
233	S	Fish	Juveniles	Pimephales promelas	21 days	Krantzberg and Boyd (1992)
226	EL	Fish	Larvae (<24 hrs old)	Pimephales promelas	7 days	Environment Canada (1992d)
232	EX	Fish	Embryo–larval	Pimephales promelas	6 days	Dawson et al. (1988)
235	S	Fish	Egg–sac stage	Oncorhynchus mykiss	21 days	Birge et al. (1984); Krantzberg (1989); Krantzberg and Boyd (1992)
302	S	Fish	Gonad cell (RTG–2)	Oncorhynchus mykiss	48 hrs	Chapman et al. (1985); U.S. EPA (1991b)
237	S	Insect	Neonates or larvae	Chironomus riparius	10–30 days	ASTM (1991b) (E1383–90)
304	INT	Insect	2nd instar larvae	Chironomus tentans	10 days	Giesy et al. (1990)
238	S	Insect	2nd instar larvae	Chironomus tentans	10–25 days	ASTM (1991b) (E1383–90); Giesy et al. (1990); Wentsel et al. (1977a)
242	S	Insect	Nymphs	Hexagenia limbata	21 days	Krantzberg and Boyd (1992)
308	EL	Microbe	N/A	Alkaline phosphatase (APA)	40–130 min	Burton et al. (1989)
320	EX	Nematode	L2 stage juveniles	Panagrellus redivivus	96 hrs	Samoiloff et al. (1983)
244	EL	Nematode	L1 stage juveniles	Panagrellus redivivus	96 hrs	Burnett et al. (no date); Samoiloff et al. (1980)
323	S	Oligochaete	Juveniles and adults	Lumbriculus variegatus	10–28 days	Phipps et al. (1993)
324	S	Oligochaete	15 mm in length	Lumbriculus variegatus	14 days	Dermott and Munawar (1992)
250	S	Oligochaete	Large worms	Lumbriculus variegatus	24,48 hrs	Dermott and Munawar (1992)
252	S	Oligochaete	Mixed age; similar size	Pristina leidyi	18 days	Smith et al. (1991)
330	S	Oligochaete	Adults	Tubifex tubifex	28 days	Reynoldson et al. (1991)
256	S	Plant	Seedlings	Echinochloa crusgalli	2 weeks	Walsh et al. (1991)
333	S	Plant	Apical shoots	Hydrilla verticillata	14 days	Klaine and Hinman (no date)

Notes: EL – elutriate
EX – extract
INT – interstitial water
S – sediment

ATTACHMENT 1. SEDIMENT TOXICITY TESTS DELETED FROM FURTHER REVIEW

Test No.	Site	Exposure Media	Biotic Group	Common Name	Life Stage	Recom.	Test Species	Duration	References
007	M/L a,e	S	Amphipod	Amphipod	N/S		Paraphoxus epistomus	10 days	Swartz et al. (1979)
012	M/L a,e	S	Bivalve	Clam	N/S		Macoma inquinata	10 days	Swartz et al. (1979)
013	M/L d,e	S	Bivalve	Bent-nose clam	N/S		Macoma nasuta	96 hrs	Reish and Lemay (1988)
019	M/L d,e	S	Bivalve	Littleneck clam	N/S		Protothaca staminea	96 hrs	Reish and Lemay (1988)
022	M/L e	S	Bivalve	Bivalve	N/S		Tellina liliana	10 days	Roper and Hickey (1992)
024	M/L c,e	EL	Copepod	Copepod	N/S	GB	Acartia sp.	48 hrs	U.S. EPA and U.S. COE (1991)
025	M/L b	S	Copepod	Copepod	Adults		Microarthridion littorale	96 hrs	DiPinto et al. (1992)
036	M/L e	S	Crustacean	Ridge-back prawn	N/S		Sicyonia *ingentis*	96 hrs	Reish and Lemay (1988)
050	M/L e	EL	Fish	Topsmelt	N/S		Atherinopes affinis		Reish and Lemay (1988)
052	M/L e	S	Fish	Arrow goby	N/S		Clevelanda ios	96 hrs	Reish and Lemay (1988)
066	M/L e	S	Polychaete	Polychaete	N/S		Abarenicola sp.	10 days	U.S. EPA and U.S. COE (1991)
067	M/L e	S	Polychaete	Polychaete	N/S		Arenicola sp.	10 days	U.S. EPA and U.S. COE (1991)
068	M/L e	S	Polychaete	Polychaete	N/S		Ctenodrilus serratus	N/S	Reish and Lemay (1988)
069	M/L e	S	Polychaete	Polychaete	N/S		Glycera sp.	10 days	U.S. EPA and U.S. COE (1991)
070	M/L a,e	S	Polychaete	Polychaete	N/S		Glycinde picta	10 days	Swartz et al. (1979)
072	M/L e	S	Polychaete	Polychaete	N/S		Nephthys sp.	10 days	U.S. EPA and U.S. COE (1991)
073	M/L e	S	Polychaete	Polychaete	N/S	GB	Nereis sp.	10 days	U.S. EPA and U.S. COE (1991)
074	M/L e	S	Polychaete	Polychaete	N/S		Ophrotrocha diadema	N/S	Reish and Lemay (1988)
077	M/S	S	Amphipod	Amphipod	<48 hr old juvs.		Grandidierella japonica	28 days	Nipper et al. (1989)
078	M/S b	S	Amphipod	Amphipod	Adults		Rhepoxynius abronius	30 days	Swartz et al. (in prep.)
082	M/S	EL	Bacterium	Microtox	Cells	EC	Photobacterium phosphoreum	5 and 15 min	Environment Canada (1992f)
092	M/S c	EL	Cells	Single cell gel	Eukaryotic cells		Eukaryotic cells	N/S	Jarvis and Reilly (1992)
093	M/S c	S	Cells	Single cell gel	Eukaryotic cells		Eukaryotic cells	N/S	Jarvis and Reilly (1992)
094	M/S	S	Copepod	Copepod	Adults		Amphiascus tenuiremis	21 days	Strawbridge et al. (1992)
095	M/S b	S	Copepod	Copepod	Adults		Microarthridion littorale	12 days	DiPinto et al. (1992)
096	M/S b	S	Echinoderm	Brittlestar	N/S		Amphiodia urtica	N/S	Bay and Greestein (1991)
117	M/S a	S	Fish	Surf smelt	Larvae		Hypomesus pretiosus	10 days	Chapman et al. (1985)
119	M/S a	EX	Fish	Rainbow trout	Gonad cell (RTG-2)		Oncorhynchus mykiss	48 hrs	Chapman et al. (1985); U.S. EPA (1991b)
120	M/S f	S	Polychaete	Polychaete	Juveniles		Armandia brevis	20 days	Casillas et al. (1991)
124	E/L c,e	S	Amphipod	Amphipod	N/S		Corophium insidiosum	10 days	Reish and Lemay (1988); U.S. EPA and U.S. COE (1991)
125	E/L	S	*Amphipod*	*Amphipod*	*Mature, 5-8 mm*		*Corophium sp.*	10 days	Reish and Lemay (1988); Swartz et al. (1990); U.S. EPA and U.S. COE (1991, 1993)
132	E/L b,c,e	S	Amphipod	Amphipod	N/S		Lepidactylus dytiscus	10, 20 days	Alden and Deaver (1991)
139	E/L a,e	S	Bivalve	Clam	N/S		Macoma inquinata	10 days	Swartz et al. (1979)
140	E/L e	S	Bivalve	Bent-nose clam	N/S		Macoma nasuta	96 hrs	Reish and Lemay (1988)
144	E/L e	S	Bivalve	Littleneck clam	N/S		Protothaca staminea	96 hrs	Reish and Lemay (1988)
146	E/L b	S	Copepod	Copepod	F1-2 offspring		Namopus palustris	7 days	Chandler and Scott (1991)
147	E/L b	S	Copepod	Copepod	F1-2 offspring		Pseudobradya pulchella	7 days	Chandler and Scott (1991)
154	E/L b	S	Fish	Sheepshead minnow	Embryos		Cyprinodon variegatus	7 days	Adolphson et al. (1991)
163	E/L a	S	Polychaete	Polychaete	Larvae		Capitella capitata	35 days	Chapman and Fink (1984)
164	E/L a	EL	Polychaete	Polychaete	Larvae		Capitella capitata	50 days	Chapman and Fink (1984)
166	E/L b	EL	Polychaete	Polychaete	Juveniles		Streblospio benedicti	96 hrs	Ehret et al. (1991)
167	E/L b	S	Polychaete	Polychaete	Juveniles		Streblospio benedicti	10 days	Ehret et al. (1991)

ATTACHMENT 1. (cont.)

Test No.	Site	Exposure Media	Biotic Group	Common Name	Life Stage	Recom.	Test Species	Duration	References
171	E/S b,e	S	Amphipod	Amphipod	N/S		Lepidactylus dytiscus	10, 20 days	Alden and Deaver (1991)
183	E/S b	S	Copepod	Copepod	F1–2 offspring		Namopus palustris	7 days	Chandler and Scott (1991)
184	E/S b	S	Copepod	Copepod	F1–2 offspring		Pseudobradya pulchella	7 days	Chandler and Scott (1991)
185	E/S b	EL	Crustacean	Grass shrimp	Small shrimp		Palaemonetes pugio	96 hrs	Alden et al. (1988)
186	E/S b	S	Fish	Sheepshead minnow	Embryos		Cyprinodon variegatus	7 days	Adolphson et al. (1991)
188	E/S b	EX	Fish	Rainbow trout	Gonad cell (RTG–2)		Salmo gairdneri	48 hrs	Chapman et al. (1985); U.S. EPA (1991b)
189	E/S a	S	Polychaete	Polychaete	Larvae		Capitella capitata	35 days	Chapman and Fink (1984)
190	E/S a	EL	Polychaete	Polychaete	Larvae		Capitella capitata	50 days	Chapman and Fink (1984)
192	E/S b	S	Polychaete	Polychaete	Larvae		Streblospio benedicti	7 days	Chandler and Scott (1991)
193	F/L a	S	Amphibian	Narrow–mouthed toad	Embryo–larval		Gastrophyrne carolinensis	7–8 days	Birge et al. (1984)
194	F/L a	S	Amphibian	Leopard frog	Embryo–larval		Rana pipiens	6–7 days	Birge et al. (1984); Francis et al. (1984)
196	F/L b,e	S	Amphipod	Amphipod	N/S		Diporeia sp. (Pontoporeia hoyi)	3–31 days	Gossiaux et al. (1992); Landrum et al. (1989, 1991)
197	F/L a	S	Amphipod	Amphipod	Juveniles		Gammarus lacustris	10 days	Nebeker et al. (1984)
198	F/L f	S	Amphipod	Amphipod	Juveniles		Hyalella azteca	96 hrs	Ankley et al. (1991)
201	F/L f	EL	Amphipod	Amphipod	Juveniles		Hyalella azteca	96 hrs	Ankley et al. (1991)
203	F/L b	S	Bivalve	Clam	Larvae		Corbicula	96 hrs	Phelps (1990)
204	F/L a,c,e	INT	Cladoceran	Water flea	N/S		Ceriodaphnia affinis	48 hrs	Adams et al. (1986)
206	F/L f	INT	Cladoceran	Water flea	Neonate	G (7 d)	Ceriodaphnia dubia	48 hrs	Ankley et al. (1991)
210	F/L a	S	Cladoceran	Water flea	Early–mid instars		Daphnia magna	96 hrs	Prater and Anderson (1977)
215	F/L a	EX	Fish	Goldfish	Embryos		Carassius auratus	6–8 days	Birge et al. (1985); Peddicord and McFarland (1978); U.S. EPA (1981)
216	F/L a	S	Fish	Goldfish	Embryo–larval		Carassius auratus	6–7 days	Birge et al. (1984); Francis et al. (1984)
217	F/L a	EX	Fish	Carp	Embryos		Cyprinus carpio	6–8 days	Birge et al. (1985); Peddicord and McFarland (1978); U.S. EPA (1981)
218	F/L b	S	Fish	Mosquitofish	Early life stages		Gambusia affinis	N/S	Blegger and Ross (1989)
219	F/L a	EX	Fish	Channel catfish	Embryos		Ictalurus punctatus	6–8 days	Birge et al. (1985); Peddicord and McFarland (1978); U.S. EPA (1981)
221	F/L a	EX	Fish	Bluegill	Embryos		Lepomis macrochirus	6–8 days	Birge et al. (1985); Peddicord and McFarland (1978); U.S. EPA (1981)
223	F/L a	EX	Fish	Largemouth bass	Embryos		Micropterus salmoides	6–8 days	Birge et al. (1985); Peddicord and McFarland (1978); U.S. EPA (1981)
224	F/L a	S	Fish	Largemouth bass	Embryo–larval		Micropterus salmoides	6–7 days	Birge et al. (1984); Francis et al. (1984)
225	F/L b	S	Fish	Medaka	Embryo–larval		Oryzias latipes	N/S	Cooper et al. (1993)
227	F/L f	EL	Fish	Fathead minnow	Larvae (<24hrs old)		Pimephales promelas	96 hrs	Ankley et al. (1991)
228	F/L f	S	Fish	Fathead minnow	Larvae (<24hrs old)		Pimephales promelas	96 hrs	Ankley et al. (1991)
230	F/L f	INT	Fish	Fathead minnow	Larvae (<24hrs old)	G (7 d)	Pimephales promelas	96 hrs	Ankley et al. (1991)
231	F/L a	S	Fish	Fathead minnow	Small fish		Pimephales promelas	96 hrs	Prater and Anderson (1977)
236	F/L a	S	Insect	Midge	4th instar larvae		Chironomus decorus	72 hrs	Kosalwat and Knight (1987)
243	F/L a	S	Isopod	Isopod	Early–mid instars		Asellus communis	96 hrs	Prater and Anderson (1977)
245	F/L c,e	S	Oligochaete	Oligochaete	N/S		Limnodrilus hoffmeisteri	96 hrs	Keilty et al. (1988)
246	F/L f	EL	Oligochaete	Oligochaete	Mixed–age		Lumbriculus variegatus	96 hrs	Ankley et al. (1991)
247	F/L f	INT	Oligochaete	Oligochaete	Mixed–age		Lumbriculus variegatus	96 hrs	Ankley et al. (1991)
248	F/L f	S	Oligochaete	Oligochaete	Mixed–age		Lumbriculus variegatus	96 hrs	Ankley et al. (1991)
253	F/L c,e	S	Oligochaete	Oligochaete	N/S		Stylodrilus heringianus	96 hrs	Keilty et al. (1988)
254	F/L c,e	S	Oligochaete	Oligochaete	N/S		Stylodrilus heringianus	42 days	Keilty and Landrum (1990)
257	F/L b,e	EL	Plant	Duckweed	N/S		Lemna minor	N/S	Burton et al. (1990)
258	F/L b,e	S	Plant	Duckweed	N/S		Lemna minor	N/S	Burton et al. (1990)

ATTACHMENT 1. (cont.)

Test No.	Site	Exposure Media	Biotic Group	Common Name	Life Stage	Recom.	Test Species	Duration	References
259	F/S a	EL	Algae	Green algae	Cells		Selenastrum capricornutum	4hr, 4–8 days	Blaise et al. (1986)
265	F/S b	S	Amphibian	Leopard frog	Embryo–larval		Rana pipiens	7–8 days	Birge et al. (1984)
266	F/S b	S	Amphibian	Narrow–mouthed toad	Embryo–larval		Gastrophyrne carolinensis	7–8 days	Birge et al. (1984)
267	F/S e	S	Amphipod	Amphipod	N/S		Diporeia sp. (Pontoporeia hoyi)	3–31 days	Gosslaux et al. (1992); Landrum et al. (1989, 1991)
275	F/S a	EX	Bacterium	Bacterium	Cells		Pseudomonas flourescens	20 hrs	Dutka and Kwan (1981); Trevors et al. (1981)
277	F/S a	EX	Bacterium	Aquatic bacterium	Cells		Spirillum volutans	120 min	Coleman and Qureshi (1985); Dutka and Bitton (1986); Dutka et al. (1988)
280	F/S b	S	Bivalve	Mussel	8 d old juveniles		Anodonta imbecilis	90 days	Schweinforth and Wade (1990)
281	F/S b,e	N/S	Bivalve	Fingernail clam	N/S		Musculium transversum	N/S	Dillon and Ross (1992)
282	F/S c	S	Cells	Single cell gell	Cells		Eukaryotic cells	N/S	Jarvis and Reilly (1992)
283	F/S c	EL	Cells	Single cell gell	Cells		Eukaryotic cells	N/S	Jarvis and Reilly (1992)
284	F/S a,e	S	Cladoceran	Water flea	N/S		Ceriodaphnia affinis	7 days	Adams et al. (1986)
287	F/S a	EX	Fish	Goldfish	Embryos		Carassius auratus	6–8 days	Birge et al. (1985); Peddicord and McFarland (1978); U.S. EPA (1981)
288	F/S a	S	Fish	Goldfish	Embryo–larval		Carassius auratus	7–8 days	Birge et al. (1984)
289	F/S a	EX	Fish	Carp	Embryos		Cyprinus carpio	6–8 days	Birge et al. (1985); Peddicord and McFarland (1978); U.S. EPA (1981)
290	F/S b	S	Fish	Mosquitofish	Early life stages		Gambusia affinis	N/S	Biegger and Ross (1989)
291	F/S a	EX	Fish	Channel catfish	Embryos		Ictalurus punctatus	6–8 days	Birge et al. (1985); Peddicord and McFarland (1978); U.S. EPA (1981)
292	F/S a	EX	Fish	Bluegill	Embryos		Lepomis macrochirus	6–8 days	Birge et al. (1985); Peddicord and McFarland (1978); U.S. EPA (1981)
293	F/S a	EX	Fish	Largemouth bass	Embryos		Micropterus salmoides	6–8 days	Birge et al. (1985); Peddicord and McFarland (1978); U.S. EPA (1981)
294	F/S a	S	Fish	Largemouth bass	Embryo–larval		Micropterus salmoides	7–8 days	Birge et al. (1984)
295	F/S b	S	Fish	Medaka	Embryo–larval		Oryzias latipes	N/S	Cooper et al. (1993)
296	F/S b,e	EX	Fish	Medaka	N/S		Oryzias latipes	N/S	Fabacher et al. (1991)
300	F/S f	S	Fish	Fathead minnow	Embryos		Pimephales promelas	6 days	Westerman (1988)
306	F/S a	S	Insect	Midge	3rd instar larvae		Chironomus tentans	5 days	Wentsel et al. (1977b)
309	F/S b	S	Microbes	Microbial enzyme	N/A		Alkaline phosphatase (APA)	40–130 min	Burton (1984); Burton et al. (1989); Stemmer et al. (1990b)
310	F/S b	S	Microbes	Microbial enzyme	N/A		Amylase	40–130 min	Burton (1988)
311	F/S b	S	Microbes	Microbial enzyme	N/A		Arlysulfatase	40–130 min	Burton (1988)
312	F/S b	EL	Microbes	Microbial enzyme	N/A		b–galactosidase (GAL)	40–130 min	Burton (1984); Burton et al. (1989)
313	F/S b	S	Microbes	Microbial enzyme	N/A		b–galactosidase (GAL)	40–130 min	Burton (1984); Burton et al. (1989)
314	F/S b	S	Microbes	Microbial enzyme	N/A		b–glucosidase (GLU)	40–130 min	Burton (1984); Burton et al. (1989)
315	F/S b	EL	Microbes	Microbial enzyme	N/A		b–glucosidase (GLU)	40–130 min	Burton (1984); Burton et al. (1989)
316	F/S b	S	Microbes	Microbial enzyme	N/A		C–14 prot. hydrolysate turnover	40–130 min	Burton (1988)
317	F/S b	EL	Microbes	Microbial enzyme	N/A		Dehydrogenase (DHA)	40–130 min	Burton (1984); Burton et al. (1989)
318	F/S b	S	Microbes	Microbial enzyme	N/A		Dehydrogenase (DHA)	40–130 min	Burton (1984); Burton et al. (1989)
319	F/S b	S	Microbes	Microbial enzyme	N/A		Protease	40–130 min	Burton (1988)
322	F/S b,c,e	S	Oligochaete	Oligochaete	N/S		Limnodrilus hoffmeisteri	96 hrs	Keilty et al. (1988)
327	F/S b,c,e	S	Oligochaete	Oligochaete	N/S		Stylaria lacustris	N/S	Kennedy et al. (1988)

ATTACHMENT 1. (cont.)

Test No.	Site		Exposure Media	Biotic Group	Common Name	Life Stage	Recom.	Test Species	Duration	References
328	F/S	b,c,e	S	Oligochaete	Oligochaete	N/S		Stylodrilus heringianus	96 hrs	Kelity et al. (1988)
329	F/S	b,c,e	S	Oligochaete	Oligochaete	N/S		Stylodrilus heringianus	42 days	Kelity and Landrum (1990)
331	F/S	b,c,e	N/S	Ostracod	Ostracods	N/S		Ostracoda	N/S	Bennett and Cubbage (1992)
334	F/S	b,e	S	Plant	Duckweed	N/S		Lemna minor	N/S	Burton et al. (1990)
335	F/S	b,e	EL	Plant	Duckweed	N/S		Lemna minor	N/S	Burton et al. (1990)

EC = Environment Canada; GB = "Greenbook" U.S. EPA and U.S. COE (1991); IN = Inland Testing Manual U.S. EPA and U.S. COE (1993); TT = Tiered Testing Proceedings (1992)

Site:
M/L = Marine Lethal
M/S = Marine Sublethal
E/L = Estuarine Lethal
E/S = Estuarine Sublethal
F/L = Freshwater Lethal
F/S = Freshwater Sublethal

Type:
EL = Elutriate
EX = Extract
INT = Interstital Water
S = Sediment

Screening remarks:
a Very old, little current use.
b Very new, under development, little promise, or too little information too evaluate.
c Inadequate protocol specifications.
d Exposure duration inadequate.
e Life stage of organism not specified.
f Definitive protocol not available or not specified.

N/S = Not Specified
N/A = Not Apllicable

APPENDIX B

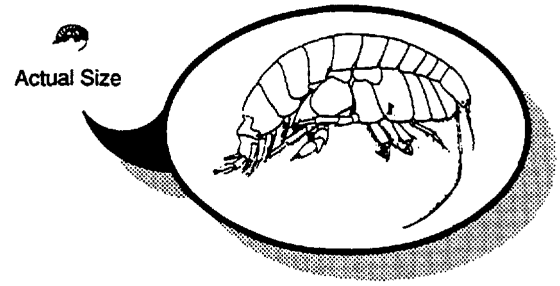
Species Summaries

SPECIES SUMMARIES

The life history and ecology of species used in various sediment toxicity tests are described in this appendix. The species were selected to provide representatives of various taxonomic groups and various habitats. Many of the species are used in the top ranked tests or in commonly used tests. The following characteristics are described for each species: major taxonomic group, life stages, geographic distribution, habitat, environmental tolerances, food, and sensitivity to contaminants. When information on a particular topic is not provided, it was not available in the references reviewed. Key references on the ecology and toxicity test protocols are also given for each species.

Ampelisca abdita

Used in Test No. 001



Ampelisca abdita is a tube-dwelling amphipod, which is a type of crustacean. Crustaceans represent a large group of invertebrates that include shrimp and crabs. Amphipods are one of the most abundant groups of crustaceans in the benthic environment. *A. abdita* ranges from south-central Florida to central Maine and is also found along the eastern Gulf of Mexico and in San Francisco Bay. Because amphipods are the main source of benthic prey for many fish, birds, and larger invertebrates and an important element in the diet of some marine mammals (e.g., grey whales), they are considered to be ecologically important organisms. Amphipods are characteristically absent from benthic communities that are heavily impacted by pollution. This species primarily inhabits protected areas, from the low intertidal zone to depths of 60 m. It is generally found in sediments ranging from mud and silt without shell to fine sand and is often abundant in sediments with high organic content. *A. abdita* also can be found in larger size sediments. It can live in areas that have a wide range of temperature (−2 to 27°C) and salinity (10 ppt to fully marine). These amphipods are scavengers and feed using a constructed tube to create a feeding current that draws in food particles from th3e overlying water and from the sediment surface. *A. abdita* is known to ingest algal material, sediment, and organic detritus. *A. abdita* may reproduce continuously throughout the year in warmer waters, but it only reproduces twice a year in colder regions. Full moon and spring tides have been correlated with intense breeding activity. After 2 weeks in a brood pouch, juveniles require 40–80 days to become mature adults. Only females (immature and mature) are recommended for toxicity testing. Desirable characteristics of *A. abdita* for toxicity testing include wide geographic distribution, ease of collecting, and sensitivity of growth and survival to a variety of contaminants.

SOURCES

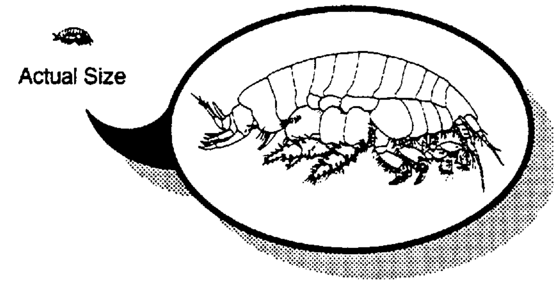
ASTM. 1990. Standard guide for conducting 10-day static sediment toxicity tests with marine and estuarine amphipods. Annual Book of ASTM Standards, Vol. 11.04. E1367-90. American Society for Testing and Materials, Philadelphia, PA.

Bousfield, E.L. 1973. Shallow-water Gammaridean Amphipoda of New England. Cornell University Press, Ithaca, NY.

Mills, E.L. 1967. The biology of an Amphipod Crustacean sibling species pair. J. Fish. Res. Board Can. 24:305-355.

Rhepoxynius abronius

Used in Test No. 008



Rhepoxynius abronius is a free-burrowing amphipod, which is a type of crustacean. Crustaceans represent a large group of invertebrates that include shrimp and crabs. Amphipods are one of the most abundant groups of crustaceans in the benthic environment. *R. abronius* has a wide geographical distribution along the West Coast, extending from Puget Sound, Washington, to southern California. Because amphipods are the main source of benthic prey for many fish, birds, and larger invertebrates and an important element in the diet of some marine mammals (e.g., grey whales), they are considered to be ecologically important organisms. Amphipods are characteristically absent from benthic communities that are heavily impacted by pollution. *R. abronius* lives in the upper 2–6 cm of sediments. It prefers to live in unvegetated, coarse-grained environments (i.e., fine sand to silty sand). *R. abronius* lives in sediments where the annual temperature ranges from 8 to 16°C and the annual salinity ranges from 22 to 33 ppt. *R. abronius* is very sensitive to salinities below 15 ppt. This amphipod consumes very small animals living among the sediment particles and plant matter (algae and detritus) that is 0.1–1 mm in size. Mature male and female amphipods, which are used for toxicity testing, are 3–5 mm in length. Use of *R. abronius* as a toxicity test organism should be limited to sediment samples collected from subtidal and lower intertidal regions (to 274 m) and higher-salinity portions of estuaries. *R. abronius* is moderately sensitive to sediment-associated contaminants.

SOURCES

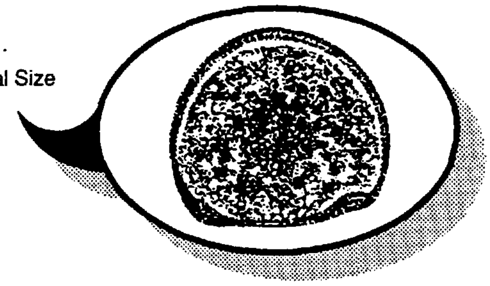
ASTM. 1990. Standard guide for conducting 10-day static sediment toxicity tests with marine and estuarine amphipods. Annual Book of ASTM Standards, Vol. 11.04. E1367–90. American Society for Testing and Materials, Philadelphia, PA.

Barnard, J.L., and C.M. Barnard. 1982. The genus *Rhepoxynius* (Crustacea: Amphipoda: Phoxocephalidae) in American Seas. Smithsonian Contributions for Zoology. No. 357. Smithsonian Institution Press, Washington, DC.

Kemp, P.F., F.A. Cole, and R.C. Swartz. 1985. Life history and production of the Phoxocephalid amphipod *Rhepoxynius abronius*. J. Crustacean Biol. 5:449–464.

Crassostrea gigas

Used in Tests No. 010 and 011



Crassostrea gigas (the Japanese or Pacific oyster) is a filter-feeding oyster and a member of the class Bivalvia (phylum Mollusca), which also includes clams, scallops, and mussels. *C. gigas* was introduced to the West Coast of North America from Japan in the early 1900s and is now well established in many places. This oyster is an important economic crop and is cultured and harvested for human consumption. It is a very large oyster, enclosed within a shell made up of two lateral halves. Individuals normally begin life as males, but they can change to females and then back to males several times during the course of their life cycle. These sex changes are associated with temperature changes and food supply. In general, *C. gigas* will spawn when the temperature is between 25 and 30°C. Females can produce and discharge 500,000 eggs in a season. Embryos and young larvae live in the open water and can passively drift for up to 2–3 weeks. Once oyster larvae settle to the bottom, they become firmly attached to mother shells (cultch) and are then called spat. *C. gigas* is epibenthic, inhabiting marine and estuarine bottoms in the intertidal and subtidal zones. This species is adapted to a wide range of substrates, salinities, temperatures, and dissolved oxygen concentrations. Like other oysters, *C. gigas* is a filter-feeder, collecting and consuming materials suspended in the water (e.g., bacteria, protozoa, small single-cell plants, larvae, detritus). Starfish, crabs, flatworms, oyster drillers, and some species of fish are predators of *C. gigas*. The embryos and larvae of *C. gigas* have been shown to be sensitive to sediment-associated contaminants. Mortality and abnormal growth in the early life stages of *C. gigas* serve as effective measurement endpoints in toxicity tests.

SOURCES

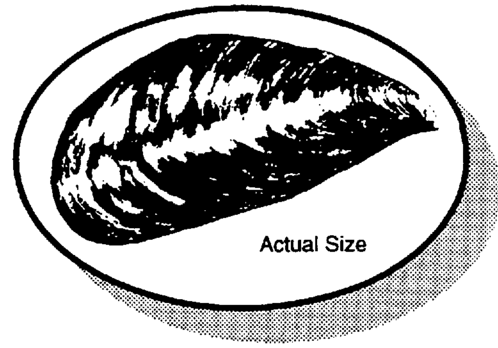
ASTM. 1989. Standard practice for conducting static acute toxicity tests with larvae of four species of bivalve. pp. 256–272. In: Annual Book of ASTM Standards, Vol. 11.04. E724-80. American Society for Testing and Materials, Philadelphia, PA.

Cheney, D.P., and T.F. Mumford, Jr. 1986. Shellfish and seaweed harvests of Puget Sound. pp. 11–36. University of Washington Press, Seattle, WA.

Kozloff, E.N. 1973. Seashore life of Puget Sound, the Strait of Georgia, and the San Juan archipelago. University of Washington Press, Seattle, WA.

Mytilus edulis

Used in Tests No. 016 and 017



Mytilus edulis (the blue or bay mussel) is a filter-feeding mussel and a member of the class Bivalvia (phylum Mollusca), which also includes clams, scallops, and oysters. *M. edulis* is a marine/estuarine species that is widely distributed around the world in northern temperate regions. This mussel is an important economic crop and is cultured and harvested for human consumption. It is enclosed within a wedge-shaped shell made up of two lateral halves. Full-grown mussels rarely exceed 6 cm in length. *M. edulis* exhibits separate sexes, and spawning of the eggs and sperm usually occurs in spring to early summer. The fertilized eggs live in the open water and develop into free-swimming planktonic larvae in about 48 hours. When larvae develop to where they can both swim and crawl (pedivelger stage), they will begin to settle out on a stable substrate (e.g., wharf pilings). Larvae can delay settlement for up to 10 weeks while they search for a suitable solid surface to settle on. Upon settling, larvae attach themselves to the substrate using a series of byssal threads. These tough, fibrous threads also enable young mussels to move and adjust their position to a more favorable environment. They live in clusters and attain sexual maturity in 1 year. *M. edulis* prefers to inhabit calmer waters and is abundant on wharf pilings, floats, docks, and rocks. This species is found occasionally on outer coasts. This mussel is most common below mid-tide zones, but can be found from high-tide zones to depths of 36 m. *M. edulis* can tolerate temperatures between 1.7–27°C, and salinities between 5 and 37 ppt. It is also able to withstand low oxygen concentrations for short periods. *M. edulis* feeds by separating small particles of organic matter from the current of water that passes through its gills. Birds, crabs, starfish, and humans are the principle predators of this mussel. This species has been shown to be sensitive to sediment-associated contaminants.

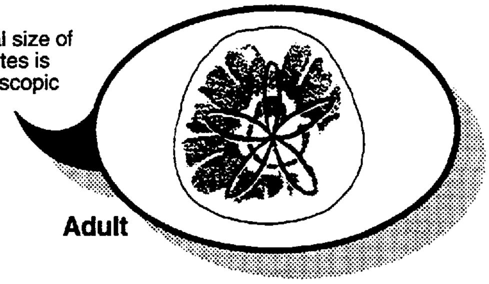
SOURCES

Cheney, D.P., and T.F. Mumford, Jr. 1986. Shellfish and seaweed harvests of Puget Sound. pp. 11-36. University of Washington Press, Seattle, WA.

Kozloff, E.N. 1973. Seashore life of Puget Sound, the Strait of Georgia, and the San Juan archipelago. University of Washington Press, Seattle, WA.

Dendraster excentricus

Used in Tests No. 039, 040, 041, 042, 102, and 103



Dendraster excentricus (sand dollar) is a member of the phylum Echinodermata, which also includes starfish and sea urchins. This echinoderm is found on the West Coast of North America from Baja California to Alaska. Sand dollars are preyed on by starfish. *D. excentricus* lives along quiet water beaches with flat sandy bottoms extending from the low-tide mark to depths of 100 m. The sand dollar grows to be about 8 cm in diameter. Gametes, embryos, and juveniles (3–7 mm in diameter) are used for toxicity tests. Sand dollars have separate sexes, and release sperm and eggs into the surrounding water, relying on their chance union for reproduction. Embryos develop into free-swimming larvae, which eventually settle out onto suitable substrate and grow into adults. Young *D. excentricus* will selectively ingest the heaviest sand grains to gain weight, thus enabling them to secure themselves to a shifting substrate. As young *D. excentricus* grow, the sand grains disappear from their gut. This echinoderm is almost completely enclosed in a skeleton of calcarious plates. The sand dollar generally remains partially exposed above the sediment surface and orients itself nearly vertically and perpendicular to the current. *D. excentricus* is somewhat mobile, plowing or pushing itself through the sand. *D. excentricus* feeds on fine particles of organic matter (e.g., diatoms) removed from the sand or water by tube feet (cilia) that carry the food to its mouth. Early life stages of *D. excentricus* have been shown to be sensitive to sediment-associated contaminants.

SOURCES

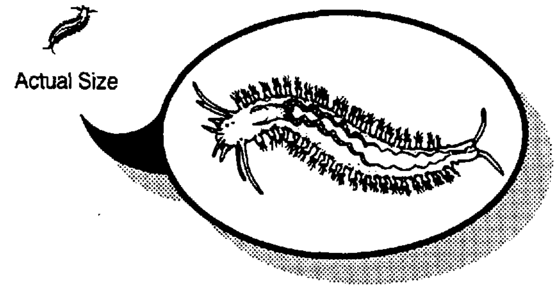
Casillas, E., D. Weber, C. Haley, and S. Sol. 1992. Comparison of growth and mortality in juvenile sand dollars (*Dendraster excentricus*) as indicators of contaminated marine sediments. Environ. Toxicol. Chem. 11:559-569.

Kozloff, E.N. 1973. Seashore life of Puget Sound, the Strait of Georgia, and the San Juan archipelago. University of Washington Press, Seattle, WA.

Ricketts, E.F., J. Calvin, and J.W. Hedgepeth. 1968. Between Pacific tides. pp. 278–280. Stanford University Press, Stanford, CA.

Neanthes sp.

Used in Tests No. 071 and 121



Neanthes sp. is a deposit-feeding polychaetous annelid (worm) inhabiting marine and high salinity environments throughout the world. This species belongs to the family Nereidae, which is a dominant taxon of intertidal and subtidal habitats. Because *Neanthes* sp. is an important element in the diet of many fish, they are considered to be ecologically important organisms. *Neanthes* sp. is a relatively sedentary species that has been shown to tolerate a wide range of sediment grain sizes. Interstitial and overlying water below 20 ppt salinity may adversely affect the growth and survival of *Neanthes* sp. This polychaete constructs and lives in tubes made of organic material within the sediment or on firm surfaces. Reproduction is sexual. Females deposit eggs within the worm tubes and then die in 2-3 days. The surviving male then cares for the eggs. After the eggs hatch, larvae feed on yolk sacs within the tubes for approximately 3 weeks. Shortly thereafter, the larvae emerge from the parental tube, construct independent tubes, and begin feeding on algae and detritus. This worm is easily cultured in laboratories. Under laboratory conditions at 20°C, *Neanthes* sp. takes 3–4 months to complete its life cycle. Juvenile worms weighing 0.5–1.0 mg dry weight (i.e., 2-3 weeks after emergence) are recommended for sediment toxicity tests. Desirable characteristics of *Neanthes* sp. include wide geographic distribution, ease of culturing, and sensitivity of growth and survival to a variety of contaminants.

SOURCES

Johns, D.M., R.A. Pastorok, and T.C. Ginn. 1992. A sublethal sediment toxicity test using juvenile *Neanthes* sp. (Polychaeta: Nereidae). pp. 280–293. In: Aquatic Toxicology and Risk Assessment. Fourteenth Volume. ASTM STP 1124. M.A. Mayes and M.G. Barron (eds). American Society for Testing and Materials, Philadelphia, PA.

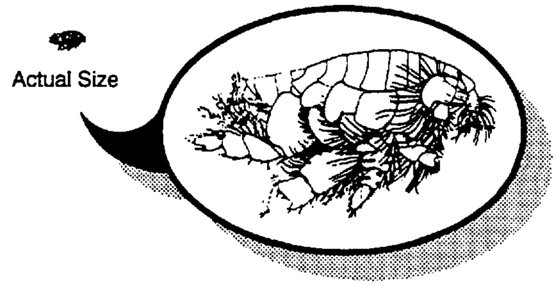
Reish, D.J. 1980. Use of polychaetous annelids as test organisms for marine bioassay experiments. pp. 140–154. In: Aquatic Invertebrate Bioassays. ASTM STP 715. A.L. Buikema, Jr., and J. Cairns, Jr. (eds). American Society for Testing Materials. Philadelphia, PA.

Reish, D.J. 1985. The use of the polychaetous annelid *Neanthes arenaceodentata* as a laboratory experiment animal. Tethys. 11:335–341.

U.S. EPA. 1990. Protocol for juvenile *Neanthes* sediment bioassay. Prepared by PTI Environmental Services, Bellevue, WA. EPA 910/9-90-011. U.S. Environmental Protection Agency, Puget Sound Estuary Program, Seattle, WA.

Eohaustorius estuarius

Used in Test No. 129



Eohaustorius estuarius is an estuarine amphipod, which is a type of crustacean. Crustaceans represent a large group of invertebrates that include shrimp and crabs. Amphipods are one of the most abundant groups of crustaceans in the benthic environment. *E. estuarius* has a wide geographic distribution along the West, East, and Gulf coasts of North America. Because amphipods are the main source of benthic prey for many fish, birds, and larger invertebrates, they are considered to be ecologically important organisms. Amphipods are characteristically absent from benthic communities that are heavily impacted by pollution. *E. estuarius* appears to exhibit an annual life cycle. Mature male and females of the species, which are used for toxicity tests, are 3–5 mm in length. *E. estuarius* is a free-burrowing deposit feeder. This amphipod is generally found in the upper 10 cm of intertidal, estuarine sands, +0.15 to +0.61 m above mean low water. It occurs in maximum densities near the mouths of streams and rivers. *E. estuarius* tolerates temperature ranges between 0°C and 21°C, salinity ranges between near 0 to 35 ppt, and a broad range of sediment grain sizes. *E. estuarius* has been shown to be sensitive to sediment-associated contaminants. Desirable characteristics of *E. estuarius* for toxicity testing include almost constant contact with sediment particles and interstitial water, broad tolerance of natural environmental conditions, wide geographic distribution, and ease of collecting.

SOURCES

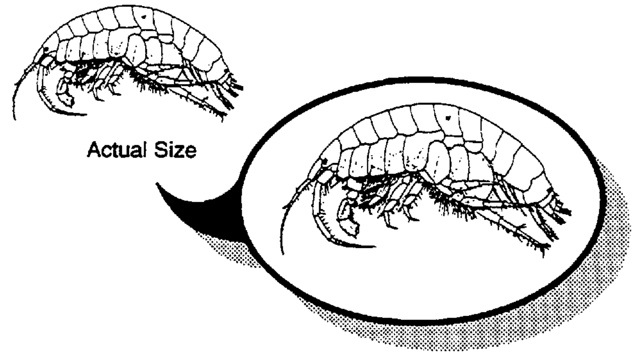
ASTM. 1990. Standard guide for conducting 10-day static sediment toxicity tests with marine and estuarine amphipods. Annual Book of ASTM Standards, Vol. 11.04. E1367–90. American Society for Testing and Materials, Philadelphia, PA.

Sameoto, D.D. 1969. Comparative ecology, life histories, and behavior of intertidal sand-burrowing amphipods (Crustacea: Haustoriidae) at Cape Cod. J. Fish. Res. Board Can. 26:361–388.

Sattery, P.N. 1985. Life histories of infaunal amphipods from subtidal sands of Monterey Bay, California. J. Crust. Biol. 5:635–649.

Leptocheirus plumulosus

Used in Tests No. 133, 134, and 135



Leptocheirus plumulosus is a deposit-feeding amphipod, which is a type of crustacean. Crustaceans represent a large group of invertebrates that include shrimp and crabs. Amphipods are one of the most abundant groups of crustaceans in the benthic environment. *L. plumulosus* is found along the East Coast of the United States from Massachusetts to northern Florida. Because amphipods are the main source of benthic prey for many fish, birds, and larger invertebrates, they are considered to be ecologically important organisms. Amphipods are characteristically absent from benthic communities that are heavily impacted by pollution. *L. plumulosus* is an annual species and produces up to two broods per year. The timing of reproduction is variable, but reproductive peaks usually occur in early to mid-spring and in the fall. *L. plumulosus* lives in bottom sediments that range from fine sand to very fine mud. L. plumulosus constructs U-shaped burrows within the sediment, where it feeds on organic detritus from ingested particulates. *L. plumulosus* tolerates a wide range of salinity (1.5–32 ppt) and sediment grain size (96.5 percent silt/clay particles to 98.1 percent sand particles). Juvenile organisms are recommended for sublethal toxicity tests. Adults of mixed sexes, ranging in size from 4 to 8 mm, are recommended for lethal toxicity tests. Adults have been shown to be sensitive to sediment-associated contaminants. Other desirable characteristics of *L. plumulosus* for toxicity testing are ease of collecting and sensitivity of sublethal endpoints (e.g., growth inhibition) in toxicity tests.

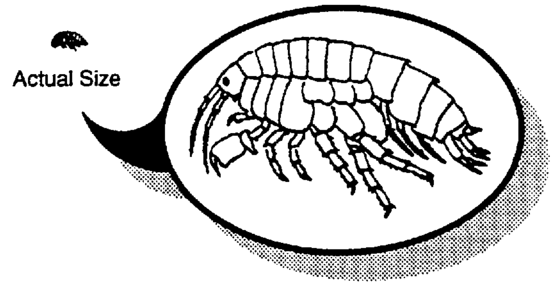
SOURCES

McGee, B.L., C.E. Schlekat, and E. Reinharz. 1993. Assessing the sublethal levels of sediment contamination using the estuarine amphipod *Leptocheirus plumulosus*. Environ. Toxicol. Chem. 12:577–587.

Schlekat, C.E., B.L. McGee, and E. Reinharz. 1992. Testing sediment toxicity in Chesapeake Bay with the amphipod *Leptocheirus plumulosus*: an evaluation. Environ. Toxicol. Chem. 11:225–236.

Hyalella azteca

Used in Tests No. 199, 200, 131, and 269



Hyalella azteca is a freshwater amphipod, which is a type of crustacean. Crustaceans represent a large group of invertebrates that include shrimp and crabs. Fish are the chief predators of *H. azteca*, but birds, predatory aquatic insects, and amphibians probably consume appreciable quantities as well. *H. azteca* can be found in permanent lakes, ponds, and streams throughout North America. This amphipod is usually lives in shallow waters. Amphipods are characteristically absent from benthic communities that are heavily impacted by pollution. The life cycle of *H. azteca* can be completed in 27 days or more. Adult *H. azteca* males are larger than adult females. *H. azteca* exhibits a wide tolerance of sediment grain size (from > 90 percent silt/clay particles to 100 percent sand particles showed no adverse effects). Reproduction is optimal at temperatures between 26° and 28°C. This amphipod will burrow into the sediment surface and is an epibenthic detritivore (general scavenger) capable of digesting bacteria and algae from ingested sediments (<65 μm). An environmental requirement for *H. azteca* appears to be an abundance of dissolved oxygen. This species has been shown to be a sensitive indicator of sediment-associated contaminants. Desirable characteristics of *H. azteca* for toxicity testing include a short generation time and ease of collecting or culturing.

SOURCES

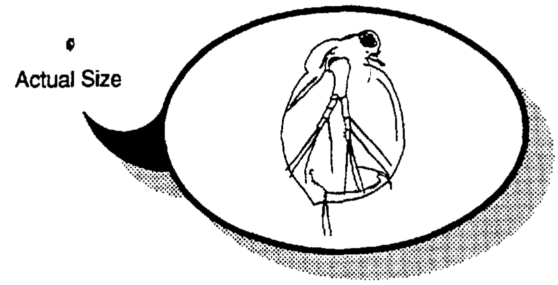
ASTM. 1991. Standard guide for conducting sediment toxicity tests with freshwater invertebrates. Annual Book of ASTM Standards. E1383-90. American Society for Testing and Materials, Philadelphia, PA. 20 pp.

Pennak, R.W. 1978. Freshwater invertebrates of the United States. Second Edition. pp. 451–463. John Wiley & Sons, Inc. New York, NY.

Strong, D.R. 1972. Life history variation among populations of an amphipod (*Hyalella azteca*). Ecology 53:1103–1111.

Ceriodaphnia dubia

Used in Tests No. 205, 207, and 285



Ceriodaphnia dubia is a planktonic, filter-feeding microcrustacean, commonly referred to as a water flea. *C. dubia* is widely distributed in freshwater locations throughout temperate North America and is most abundant in lakes and ponds, occasionally occurring in quiescent sections of streams and rivers. It represents an ecologically important component of food webs. This species forms a significant portion of the diet of many fish and other crustaceans. Adult females range in size up to 0.9 mm. *C. dubia* reproduces rapidly after birth and exhibits a short life cycle. Reproduction is parthenogenetic (without fertilization) and primarily produces females. Sexual reproduction can be stimulated by environmental stress, which produces "resting eggs" that can withstand conditions such as drying and freezing, thus facilitating the survival and transport of *C. dubia*. The species is adapted to a wide range of temperatures, but cannot survive low dissolved oxygen concentrations (i.e., <5 mg/L). *C. dubia* is planktonic, freely swimming among the weeds and coarse detritus. It feeds on small floating plants (phytoplankton), bacteria, and suspended detritus. *C. dubia* is easily cultured and has been shown to be sensitive to sediment-associated contaminants. Toxicity tests are started with *C. dubia* neonates that are less than 24 hours old. To control for initial (neonate) age in reproductive toxicity tests, only cultured individuals of the species should be used in the tests.

SOURCES

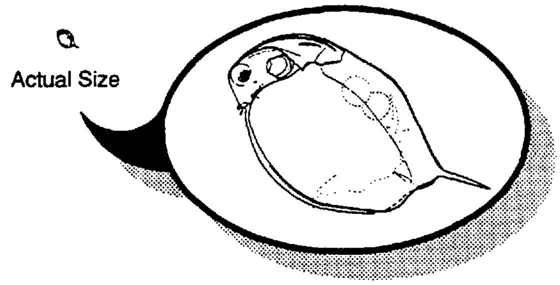
Brandlova, J., Z. Brandl, and C.H. Fernando. 1972. The Cladocera of Ontario with remarks on some species and distribution. Can. J. Zoo. 50:1373–1403.

Environment Canada. 1992. Biological test method: test of reproduction and survival using the Cladoceran *Ceriodaphnia dubia*. Report EPS 1/RM/21. Environmental Publications, Conservation, and Protection, Environment Canada, Ottawa, Ontario, Canada.

Pennak, R.W. 1978. Freshwater invertebrates of the United States. Second Edition. John Wiley & Sons, Inc., New York, NY.

Daphnia magna

Used in Tests No. 208, 209, 211, and 212



Daphnia magna is a microcrustacean, commonly referred to as a water flea. *D. magna* is a freshwater organism, with a wide distribution based on suitable habitat. This water flea inhabits small lakes, ponds, and temporary pools of clean, weedy water. Adult females are larger than adult males, with females ranging in size up to 5 mm and males ranging to greater than 2 mm. Reproduction is parthenogenetic (without fertilization) and primarily produces females. *D. magna* are capable of producing a brood of up to 65 individuals, every other day. Sexual reproduction can be stimulated by environmental stress, which produces "resting eggs" that can withstand conditions such as drying and freezing, thus facilitating the survival and transport of *D. magna*. *D. magna* has been shown to survive for periods of 28 and 108 days at 8°C and 28°C, respectively. *D. magna* is primarily free-swimming and normally is not found on the sediment bottom. It lives chiefly among weeds and course detritus. *D. magna* is a filter feeder, ingesting suspended or settled fine particles down to 0.5 μm. Their diet primarily consists of algae and protozoa, but it will consume any edible item (e.g., detritus, bacteria). Both young and adult fish are predators of *D. magna*, as are salamanders and aquatic insects. The species is easily collected in the field or can be cultured. *D. magna* has been shown to be relatively sensitive to many sediment-associated contaminants. To control for initial (neonate) age in reproductive toxicity tests, only cultured individuals of the species should be used in these tests.

SOURCES

ASTM. 1984. Standard practice for conducting acute toxicity tests on wastewaters with Daphnia. pp. 64–76. In: Annual Book of ASTM Standards. D4229-84. American Society for Testing and Materials, Philadelphia, PA.

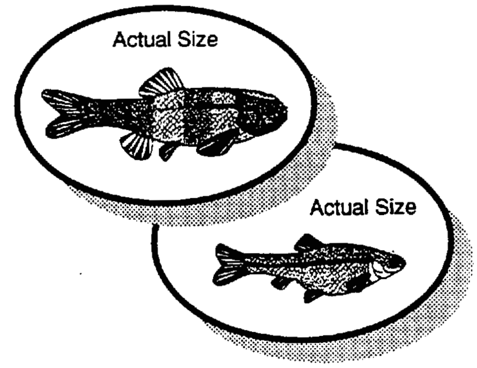
Brandlova, J., Z. Brandl, and C.H. Fernando. 1972. The Cladocera of Ontario with remarks on some species and distribution. Can. J. Zoo. 50:1373–1403.

Brooks, J.L. 1959. Cladocera. pp. 587-656. In: Freshwater Biology. Second Edition. Edmondson, W.T. (ed). John Wiley & Sons, Inc. New York, NY.

Pennak, R.W. 1978. Freshwater invertebrates of the United States. Second Edition. John Wiley & Sons, Inc., New York, NY.

Pimephales promelas

Used in Tests No. 226, 229, 232, and 233



Pimephales promelas (flathead minnow) is a small freshwater fish of the minnow family that is distributed throughout most of central North America. It ranges from New Brunswick to Alberta, Canada; south to Louisiana; and into Chihuahua, Mexico. This minnow is often found in muddy ditches, ponds, lakes, and warm muddy brooks. Fish and fish-eating birds are predators of this species. *P. promelas* primarily spawns in the spring when water temperatures reach 16°C, but spawning may continue into August. Females deposit eggs on the underside of logs, branches, and large rocks. Eggs hatch on the average in 4.5–6 days. Newly hatched young are approximately 5 mm in length. In warmer regions, *P. promelas* can attain adult sizes (50–70 mm in length) and will complete spawning by July. *P. promelas* is a relatively short-lived species, rarely living beyond 2 years. In cooler waters, at least 2 years are probably required to reach adult size. Males grow more rapidly and attain larger sizes than females. There is a high rate of post-spawning mortality. *P. promelas* can withstand extremely low oxygen concentrations and tolerates salinities greater than 10 ppt. While algae is the principle food supply for this minnow, it also consumes organic detritus, bottom mud, aquatic insect larvae (mosquitoes), and zooplankton. *P. promelas* is suitable for culturing and has been used as a food source for pond-reared gamefish in the United States. Desirable characteristics of *P. promelas* for sediment toxicity tests include wide distribution, small size, highly prolific, prolonged spawning period, association with sediment, and importance in aquatic food webs.

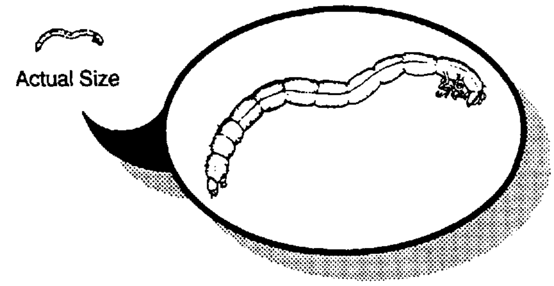
SOURCES

Eddy, S., and J.C. Underhill. 1974. Northern fishes. Third Edition. pp. 248–249. University of Minnesota, Minneapolis, MN.

Scott, W.B., and E.J. Crossman. 1975. Freshwater fishes of Canada. pp. 480–483. Information Canada, Ottawa, Canada.

Chironomus riparius

Used in Test No. 237



Chironomus riparius is a fairly large freshwater midge belonging to the Diptera order of insects (flies, gnats, or mosquitos), which are distributed worldwide. This midge is an important dietary component of young and adult fish and dabbling ducks. The larvae of *C. riparius* inhabit muddy bottomed, shallow water sites and are frequently found in eutrophic lakes, ponds, and streams. The larval stages have been observed in gravel, limestone, marl, plants, and silt environments. *C. riparius* exhibits three distinct life stages: an aquatic burrowing larval stage, a pupal stage, and an adult (fly) stage. Larvae and pupae live in fresh water where they emerge as adults in 15–21 days (under optimal conditions at 20°C); adults are terrestrial. The larval stages tolerate a wide range of environmental conditions (e.g., sediment grain size ranging from > 90 percent silt/clay particles to 100 percent sand particles, temperatures between 0 and 33°C, pH between 5 and 9, and dissolved oxygen concentrations as low as 1 mg/L). *C. riparius* larvae burrow into the sediments to construct a case and consume a variety of food materials. *C. riparius* has been shown to be sensitive to many sediment-associated contaminants. Desirable characteristics of *C. riparius* for toxicity testing include a fairly large size that facilitates handling and observation, short generation time, ease of culture, and direct contact with sediments. Successful toxicity tests have been performed with larvae that are less than 24 hours old and with larvae that are 3 days old.

SOURCES

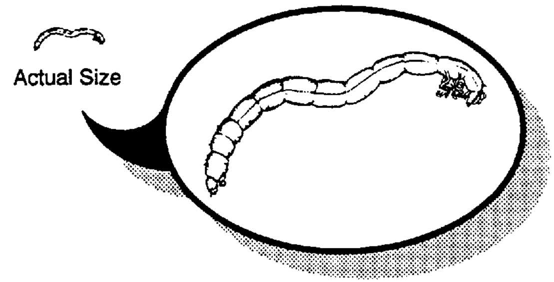
ASTM. 1991. Standard guide for conducting sediment toxicity tests with freshwater invertebrates. Annual Book of ASTM Standards. E1383-90. American Society for Testing and Materials, Philadelphia, PA.

Oliver, D.R. 1971. Life history of the Chironomidae. Annual Review of Entomology 16:211–230.

Rasmussen, J.B. 1984. The life-history, distribution and production of *Chironomus riparius* and *Glyptotendipes paripes* in a prairie pond. Hydrobiologia 119:65–72.

Chironomus tentans

Used in Tests No. 238 and 304



Chironomus tentans is a fairly large freshwater midge belonging to the Diptera order of insects (flies, gnats, or mosqüitos), which are distributed worldwide. *C. tentans* is often found in the mid-continental areas of North America. *C. tentans* larvae form an important dietary component of young and adult fish and dabbling ducks. *C. tentans* exhibits three distinct life stages: an aquatic larval stage, a pupal stage, and an adult stage. Larvae and pupae live in fresh water; adults are terrestrial. Eggs hatch in about 2-3 days after deposition in water. Larvae will develop and emerge as adults in 24–28 days (under optimal conditions at 20°C). Second to fourth larval instars range in size from approximately 0.5 to 2.5 cm, respectively. The larval stages tolerate a wide range of sediment grain-size particles (<0.15–2.0 mm), but they occur most frequently in fine sediment and detritus. The vast majority (>95 percent) of *C. tentans* larvae are located in the upper 10 cm of substrate, where the larvae burrow in the sediments to build a case. Larval stages tolerate a wide variety of environmental conditions (e.g., temperatures between 0 and 35°C, pH between 7 and 10, and dissolved oxygen concentrations as low as 1 mg/L). The larval stages consume primarily algae and organic detritus. This midge has been shown to be sensitive to many sediment-associated contaminants. Desirable characteristics of *C. tentans* for toxicity tests include fairly large size, which facilitates handling and observation; short generation time; ease of culture; and direct contact with sediments. Successful toxicity tests have been performed with second instar larvae, which are 10–14 days old.

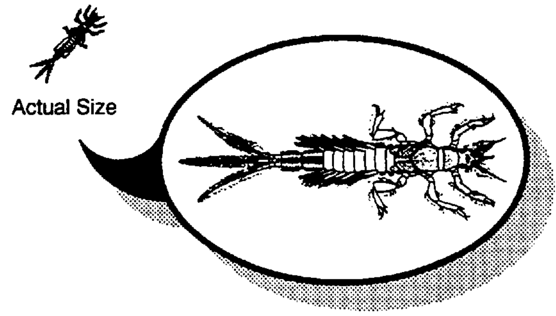
SOURCES

ASTM. 1991. Standard guide for conducting sediment toxicity tests with freshwater invertebrates. Annual Book of ASTM Standards. E1383-90. American Society for Testing and Materials, Philadelphia, PA.

Oliver, D.R. 1971. Life history of the Chironomidae. Annual Review of Entomology. 16:211–230.

Hexagenia limbata

Used in Tests No. 239, 240, 241, and 242



Hexagenia limbata (burrowing mayfly) belongs to the class Insecta and order Ephemeroptera. This mayfly is widely distributed throughout North America and occurs in association with freshwater ecosystems (e.g. lakes, ponds, streams). Nymphs are often abundant and a typical source of food for trout. In lakes and ponds, *H. limbata* is believed to be an important dietary component of other fish species. Immature *H. limbata* (nymphs) are aquatic. The first nymphal stage of *H. limbata* is usually less than 1 mm in length. In colder environments, the nymphal life stages can last for up to 2 years. The adults are terrestrial and are short-lived (relative to nymphal stages). *H. limbata* nymphs are most characteristically found in shallow waters, but they can be found to depths of 15 m. Nymphs may occur in virtually all types of fresh water where there is suitable substrate and an abundance of oxygen (dissolved oxygen concentrations of < 5 ppm can be lethal). *H. limbata* nymphs are burrowers that plow through soft-bottom sediments. Because they do not construct their burrows from particulates or mucus, they require fine-textured, high organic content sediment so that their burrow will not collapse. Burrows are usually found in the top 5 cm of sediments and contain two openings. *H. limbata* maintain a constant current of water through their burrows, but will frequently leave their burrows to feed on surface-sediment debris. *H. limbata* are chiefly opportunistic feeders, ingesting sediment and digesting the organic component (e.g., phytoplankton and detritus). They will, however, also feed on living animals and have exhibited cannibalism in laboratory toxicity tests. Survival, growth, and emergence of *H. limbata* nymphs have been shown to be sensitive to sediment-associated contaminants.

SOURCES

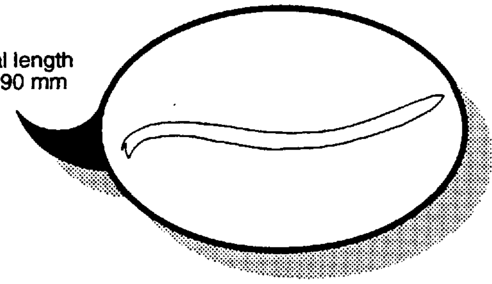
Fremling, C.R., and W.L. Mauck. 1980. Methods for using nymphs of burrowing mayflies (Ephemeroptera, Hexagenia) as toxicity test organisms. pp. 81-97. In: Aquatic Invertebrate Bioassays. A.L. Buikema, Jr., and J. Cairns, Jr. (eds). ASTM STP 715. American Society for Testing and Materials, Philadelphia, PA.

Pennak, R.W. 1978. Freshwater invertebrates of the United States. Second Edition. pp. 350-387. John Wiley & Sons, Inc., New York, NY.

Schloesser, D.W. 1988. Zonation of mayfly nymphs and caddisfly larvae in the St. Mary's River. J. Great Lakes Res. 14:227-233.

Lumbriculus variegatus

Used in Tests No. 249, 250, 323, and 324



Lumbriculus variegatus is a segmented worm and a member of the phylum Annelida and class Oligochaeta (aquatic earthworms). *L. variegatus* is restricted to the temperate and cold temperate zones of North America, Europe, Asia, and Africa. This species inhabits freshwater ecosysems and is commonly found in reservoirs, lakes, rivers, ponds, and marshes. *L. variegatus* is an epibenthic/benthic species that normally lives in silty to sandy sediments under water at depths of 2–60 m. The species ranges in size from 40 to 90 mm in length and from 1.0 to 1.5 mm in diameter. Although the potential for sexual reproduction exists, individuals containing sexual organs are rare. Reproduction is accomplished primarily by architomy, whereby new individuals are "budded" off parents. The parent will subsequently replace this lost portion with eight new segments of its own. In 10–14 days (at 20°C), populations of cultured worms can double their number through this budding process. *L. variegatus* usually tunnels within the aerobic zone of sediments, keeping its anterior portion buried and its posterior portion exposed to the overlying water for respiration. *L. variegatus* feeds by ingesting substrate materials and digesting the organic components (e.g., filamentous algae, diatoms, plant and animal detritus). This worm tolerates a wide range of natural environmental conditions (e.g., sediment grain size, temperature, dissolved oxygen content, pH). Cultured worms, which are recommended for toxicity tests, have been shown to be sensitive to some sediment-associated contaminants.

SOURCES

Bailey, H.C., and D.H.W. Liu. 1980. *Lumbriculus variegatus*, a benthic Oligochaete as a bioassay organism. pp. 205–215. In: Aquatic Toxicology. Eaton, J.C., P.R. Parrish, and A.C. Hendricks (eds). ASTM STP 707. American Society for Testing and Materials, Philadelphia, PA.

Cook, D.G. 1969. Observations on the life history and ecology of some Lumbriculidae (Annelida, Oligochaeta). Hydrobiologia 34:561–574.

Pennak, R.W. 1978. Freshwater invertebrates of the United States. Second Edition. pp. 275–290. John Wiley & Sons, Inc., New York, NY.

Phipps, G.L., G.T. Ankley, D.A. Benoit, and V.R. Mattson. 1993. Use of the aquatic Oligochaete *Lumbriculus variegatus* for assessing the toxicity and bioaccumulation of sediment associated contaminants. Environ. Toxicol. Chem. 12:269–279.

APPENDIX C

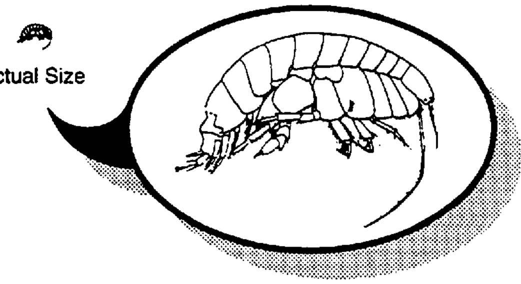
Overview of Test Protocols

OVERVIEW OF TEST PROTOCOLS

Summaries of sediment toxicity test protocols are provided in this appendix. This information is intended to familiarize the reader with the major characteristics of the protocols and is not intended to be a guide to actually performing the toxicity tests. Information is presented on endpoints, test duration, controlled environmental conditions, test solution, sources of organisms, feeding regime, sample holding, test acceptability criteria, and other protocol characteristics. The species included in this appendix are the same as those described in Appendix B. These species were selected to provide representatives of various taxonomic groups and various habitats. Many of the tests described here are among the top ranked tests or are commonly used tests.

Ampelisca abdita

Test No. 001

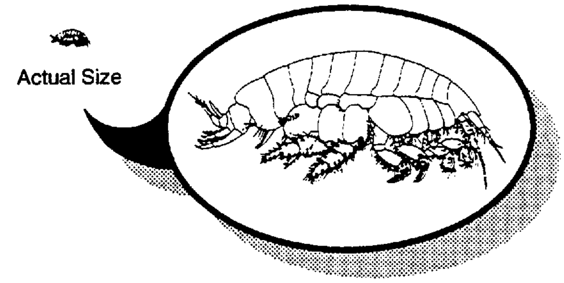


Test type:	Static nonrenewal
Test endpoint(s):	Survival and growth
Test duration:	10 days
Test water temperature:	20 ± 3°C
Test water salinity:	28–35 ppt
Photoperiod:	Continuous light
Test solution volume:	Not specified
Sediment volume and depth:	4 cm
Renewal of test solutions:	None
Test organisms:	Small juveniles of similar size
Source of test organisms:	3–4 days before test is initiated, collect approx. ⅓ more amphipods than are required for the bioassay; obtain amphipods from clean sediment by benthic grabs (e.g., van Veen, Smith-McIntyre) or small, short-haul (10 m) dredge
Number of organisms per test chamber:	20–30
Number of replicate chambers per sediment type:	5
Feeding regime:	Diatom culture in excess (0.5–1.0 L of algae/gal)
Test chamber cleaning:	Before and after
Test solution aeration:	Gentle aeration with glass tipped pipette placed above sediment surface
Positive control:	ASTM (1990) does not recommend a positive control; however, cadmium chloride is often used
Sample holding requirements:	<2 weeks in dark at 4 ± 3°C, avoid freezing and drying
Test acceptability criterion:	≥ 90 percent survival in controls

Source: ASTM. 1990. Standard guide for conducting 10-day static sediment toxicity tests with marine and estuarine amphipods. Annual Book of ASTM Standards, Vol. 11.04. E1367–90. American Society for Testing and Materials, Philadelphia, PA. 23 pp.

Rhepoxynius abronius

Test No. 008

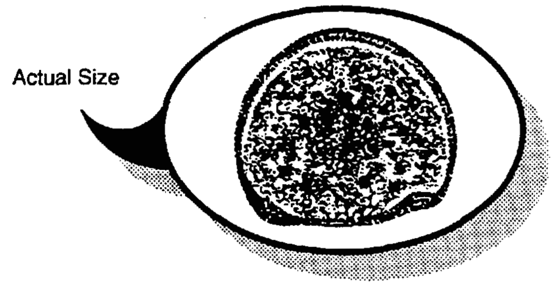


Test type:	Static nonrenewal
Test endpoint(s):	Survival and reburial
Test duration:	10 days
Test water temperature:	15 ± 3°C
Test water salinity:	28 ppt
Photoperiod:	Continuous light
Test solution volume:	Volume to 700 mL
Sediment volume and depth:	157 cm^2 and 2 cm (minimum depth)
Renewal of test solutions:	None
Test organisms:	Mature 3–5 mm; mixed sexes
Source of test organisms:	3–4 days before the test is initiated, collect approx. ⅓ more amphipods than are required for the bioassay; obtain amphipods from clean sediment by benthic grabs (e.g., van Veen, Smith-McIntyre) or small, short-haul (10 m) dredge
Number of organisms per test chamber:	20
Number of replicate chambers per sediment type:	5
Feeding regime:	None
Test chamber cleaning:	Before and after
Test solution aeration:	Trickle-flow (<100 bubbles/minute)
Positive control:	ASTM (1990) does not recommend a specific chemical for the positive control; however, cadmium chloride is often used
Sample holding requirements:	<2 weeks at 4 ± 3°C
Test acceptability criterion:	≥90 percent survival in controls

Source: ASTM. 1990. Standard guide for conducting 10-day static sediment toxicity tests with marine and estuarine amphipods. Annual Book of ASTM Standards, Vol. 11.04. E1367-90. American Society for Testing and Materials, Philadelphia, PA.

Crassostrea gigas

Test No. 011

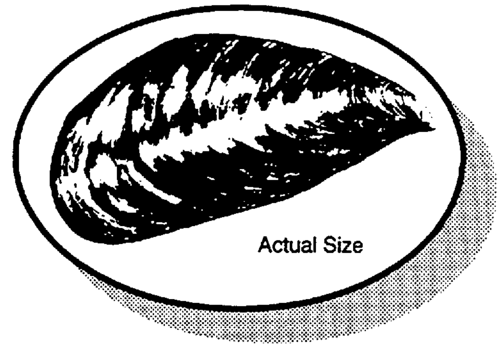


Test type:	Static nonrenewal
Test endpoint(s):	Survival and percent abnormal larvae
Test duration:	48 hours
Test water temperature:	20°C
Test water salinity:	20–35 ppt
Photoperiod:	Not specified
Test solution volume:	Variable
Sediment volume and depth:	Not applicable (use sediment extracts at various dilutions)
Renewal of test solutions:	None
Test organisms:	<1-hour old post-spawning embryos
Source of test organisms:	Collect fertilized eggs <1 hour post-spawning from laboratory cultures of adult bivalves, conditioned and stimulated to spawn when test organisms are needed
Number of organisms per test chamber:	Constant density between chambers, 15–30 embryos/mL (15/mL for routine tests)
Number of replicate chambers per sediment type:	5 concentrations of each sediment, conforming to a geometric progression, and replicated 2–3 times (3 preferably)
Feeding regime:	None
Test chamber cleaning:	Before and after
Test solution aeration:	None
Positive control:	Not specified
Sample holding requirements:	Not specified
Test acceptability criterion:	±70 percent survival and <10 percent abnormalities in controls

Source: ASTM. 1989. Standard practice for conducting static acute toxicity tests with larvae of four species of bivalve molluscs. ASTM E724-89. Annual Book of ASTM Standards, Vol. 11.04. American Society for Testing and Materials, Philadelphia, PA. pp. 256–272.

Mytilus edulis

Test No. 016

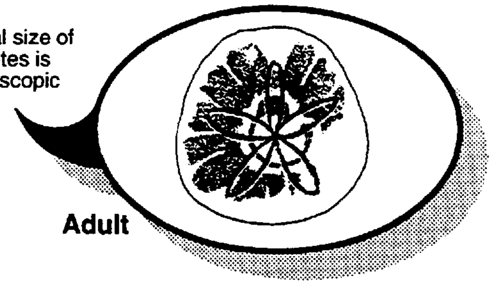


Test type:	Static nonrenewal
Test endpoint(s):	Survival and percent abnormal larvae
Test duration:	48–60 hours
Test water temperature:	16 ± 1°C
Test water salinity:	28 ± 1 ppt
Photoperiod:	14:10 (L:D)
Test solution volume:	1 L
Sediment volume and depth:	20 g
Renewal of test solutions:	None
Test organisms:	Embryos <2 hours old post-fertilization
Source of test organisms:	Collect fertilized eggs <1 hour after spawning from laboratory cultures of adult bivalves, conditioned and stimulated to spawn when test organisms needed
Number of organisms per test chamber:	Approx. 20,000–40,000; establish a concentration density of 20–40 embryos/mL in each test chamber
Number of replicate chambers per sediment type:	5
Feeding regime:	None
Test chamber cleaning:	Before and after
Test solution aeration:	Gentle aeration if dissolved oxygen concentration falls below 60 percent of saturation
Positive control:	Cadmium chloride or sodium dodecyl sulfate at 5 logarithmic concentrations and a control
Sample holding requirements:	<2 weeks at 4°C in the dark
Test acceptability criterion:	≥70 percent survival and <10 percent abnormalities in controls

Source: U.S. EPA. 1991. Recommended guidelines for conducting laboratory bioassays on Puget Sound sediments. Recommended Protocols for Measuring Environmental Variables in Puget Sound. U.S. Environmental Protection Agency Region 10, Office of Puget Sound, Puget Sound Estuary Program, Seattle, WA.

Dendraster excentricus

Test No. 103

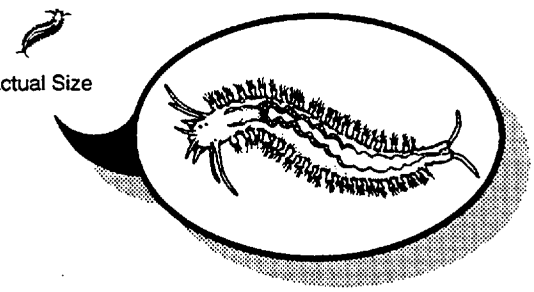


Test type:	Static nonrenewal
Test endpoint(s):	Fertilization
Test duration:	20, 40, and 60 minutes
Test water temperature:	15 ± 1°C
Test water salinity:	28–34 ppt
Photoperiod:	Not applicable
Test solution volume:	10 mL
Sediment volume and depth:	Not applicable
Renewal of test solutions:	None
Test organisms:	Gametes
Source of test organisms:	From commercial harvesters or field collected adults; conditioned and spawned in laboratory environment
Number of organisms per test chamber:	0.1 mL sperm and 0.1 mL eggs per test replicate
Number of replicate chambers per sediment type:	3
Feeding regime:	None
Test chamber cleaning:	Before and after
Test solution aeration:	Gentle aeration if dissolved oxygen falls below 40 percent of saturation
Positive control:	Reagent grade copper chloride or copper sulfate
Sample holding requirements:	≤14 days, in dark at 4°C
Test acceptability criterion:	≥50 percent fertilization in controls

Source: Environment Canada. 1992. Biological test method: fertilization assay using echinoids (sea urchins and sand dollars). Report EPS 1/RM/27. Environment Canada, Conservation and Protection, Environmental Protection, Ottawa, Canada.

Neanthes sp.

Test No. 121



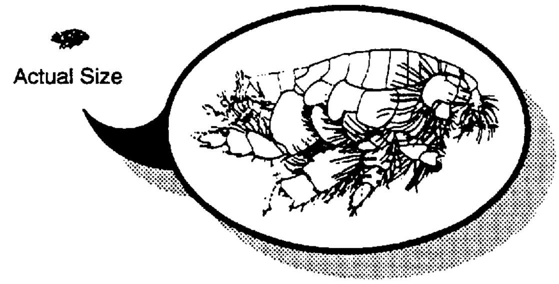
Test type:	Static renewal
Test endpoint(s):	Survival and growth
Test duration:	10 or 20 days
Test water temperature:	20 ± 1°C
Test water salinity:	28 ± 2 ppt
Photoperiod:	Continuous light
Test solution volume:	750 mL
Sediment volume and depth:	250 mL minimum and 2 cm
Renewal of test solutions:	Every third day, replace ⅓ of chamber solution with fresh seawater
Test organisms:	Juvenile worms 0.5–1.0 mg dry weight (i.e., 2–3 weeks post-emergence)
Source of test organisms:	From laboratory cultures
Number of organisms per test chamber:	5
Number of replicate chambers per sediment type:	5
Feeding regime:	40 mg (i.e., 8 mg/individual) of TetraMarin every other day
Test chamber cleaning:	Before and after
Test solution aeration:	150–300 mL/minute from glass pipette suspended 3–4 mm below water surface
Positive control:	Reagent-grade cadmium chloride, 96-hr LC_{50} exposure on 10 juveniles/chamber, no feeding
Sample holding requirements:	<2 weeks in the dark at 4°C
Test acceptability criterion:	Not specified

Source: Johns, D.M., R.A. Pastorok, and T.C. Ginn. 1992. A sublethal sediment toxicity test using juvenile *Neanthes* sp. (Polychaeta: Nereidae). pp. 280–293. In: Aquatic Toxicology and Risk Assessment. Fourteenth Volume. ASTM STP 1124. M.A. Mayes and M.G. Barron (eds). American Society for Testing and Materials, Philadelphia, PA.

U.S. EPA. 1990. Protocol for juvenile *Neanthes* sediment bioassay. Prepared by PTI Environmental Services, Bellevue, WA. EPA 910/9-90-011. U.S. Environmental Protection Agency, Puget Sound Estuary Program, Seattle, WA.

Eohaustorius estuarius

Test No. 129

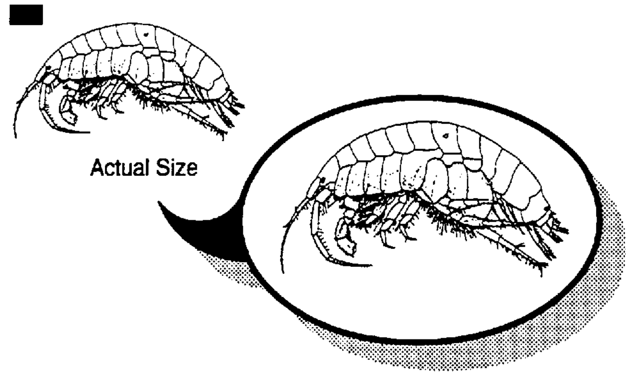


Test type:	Static nonrenewal
Test endpoint(s):	Survival and reburial
Test duration:	10 days
Test water temperature:	15 ± 3°C
Test water salinity:	2–28 ppt
Photoperiod:	Continuous light
Test solution volume:	950 mL
Sediment volume and depth:	200 mL and 2 cm
Renewal of test solutions:	None
Test organisms:	Large immatures and adults, 3–5 mm in length; mixed sexes
Source of test organisms:	3–4 days before test is initiated, collect approx. ⅓ more amphipods than are required for the bioassay; collect amphipods with shovel from clean sediment at low tide
Number of organisms per test chamber:	20
Number of replicate chambers per sediment type:	5
Feeding regime:	None
Test chamber cleaning:	Before and after
Test solution aeration:	1 mL glass pipette placed at a depth >2 cm from sediment surface
Positive control:	ASTM (1990) does not recommend a positive control; however, cadmium chloride is often used
Sample holding requirements:	<2 weeks in dark at 4 ± 3°C, avoid freezing or drying
Test acceptability criterion:	≥90 percent survival in controls

Source: ASTM. 1990. Standard guide for conducting 10-day static sediment toxicity tests with marine and estuarine amphipods. Annual Book of ASTM Standards, Vol. 1.04. E1367-90. American Society for Testing and Materials, Philadelphia, PA. 23 pp.

Leptocheirus plumulosus

Test No. 134



Test type:	Static renewal
Test endpoint(s):	Survival, growth, development, and reproduction
Test duration:	≤10 days (short-term), ≤30 days (long-term)
Test water temperature:	20°C
Test water salinity:	6 ppt
Photoperiod:	16:8 (L:D)
Test solution volume:	700 mL
Sediment volume and depth:	2 cm
Renewal of test solutions:	⅓ of overlying water replaced 2 times per week
Test organisms:	Juveniles, 1–2 weeks old and 1–2 mm in length
Source of test organisms:	From benthic grab in subtidal areas, followed by transport and conditioning in laboratory for up to 4 days before testing
Number of organisms per test chamber:	20
Number of replicate chambers per sediment type:	4
Feeding regime:	6 mg of TetraMin plus Tetra (1:1, w:w), 3 times per week per test chamber between days 0–10; 12 mg as above after day 10
Test chamber cleaning:	Before and after
Test solution aeration:	2 bubbles per second from a 1 mL pipette
Positive control:	Cadmium chloride
Sample holding requirements:	4°C
Test acceptability criterion:	Not specified

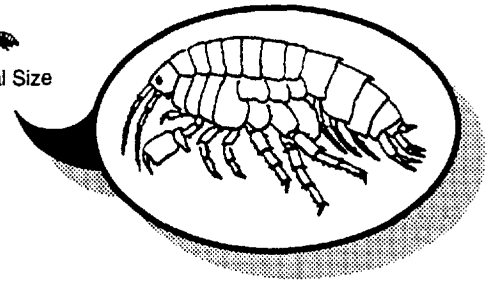
Source: McGee, B.L., C.E. Schlekat, and E. Reinharz. 1993. Assessing the sublethal levels of sediment contamination using the estuarine amphipod *Leptocheirus plumulosus*. Environ. Toxicol. Chem. 12:577–587.

Schlekat, C.E., B.L. McGee, and E. Reinharz. 1992. Testing sediment toxicity in Chesapeake Bay with the amphipod *Leptocheirus plumulosus*: an evaluation. Environ. Toxicol. Chem. 11:225–236.

Hyalella azteca

Test No. 200

Actual Size

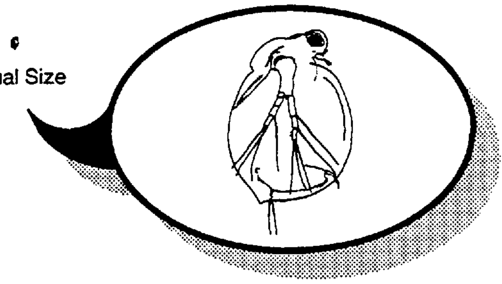


Test type:	Static nonrenewal or flow-through
Test endpoint(s):	Survival, growth, and reproductive capacity
Test duration:	≤10 days (short-term), >10–30 days (long-term)
Test water temperature:	20–25°C
Test water salinity:	Freshwater
Photoperiod:	16:8 (L:D)
Test solution volume:	800 mL in 1 L, or 15 cm depth in 20 L
Sediment volume and depth:	200 mL and 2 cm in 1 L, or 2-3 cm in 20 L
Renewal of test solutions:	None
Test organisms:	Juveniles 2–3 mm in length
Source of test organisms:	Obtain juveniles from a culture of wild, laboratory, or commercial brood stock
Number of organisms per test chamber:	20 in 1 L, or 100 in 20 L
Number of replicate chambers per sediment type:	4 using 1 L chambers, or 2+ using 20 L chambers
Feeding regime:	Static: 14 mg rabbit pellets, 3 times weekly, for 20 organisms in 1 L; 200 mg rabbit pellets, twice weekly, for 100 organisms in 20 L chamber Flow-through: 20 mg rabbit pellets, once prior to test, then 3 times weekly for duration of test
Test chamber cleaning:	Before and after
Test solution aeration:	Gentle aeration to overlying water
Positive control:	Not specified
Sample holding requirements:	<2 weeks at 4 ± 2°C, no freezing or drying
Test acceptability criterion:	≥80 percent survival in controls

Source: ASTM. 1991. Standard guide for conducting sediment toxicity tests with freshwater invertebrates. ASTM E1383-90. American Society for Testing and Materials, Philadelphia, PA. 20 pp.

Ceriodaphnia dubia

Test No. 285



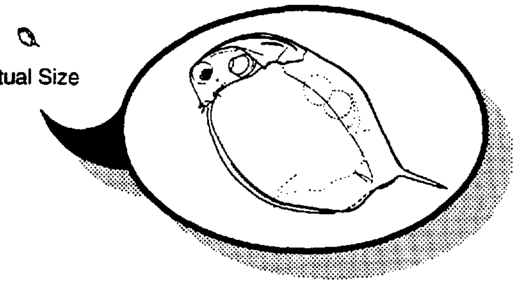
Test type:	Three brood, static renewal, elutriate test
Test endpoint(s):	Mortality of first generation daphnids and reduction in reproduction of survivors
Test duration:	7 ± 1 days (when 60 percent of controls have 3 broods)
Test water temperature:	25 ± 1°C
Test water salinity:	≤10 ppt
Photoperiod:	16:8 (L:D)
Test solution volume:	≥15 mL
Sediment volume and depth:	Not applicable (use sediment elutriates)
Renewal of test solutions:	At least once daily
Test organisms:	Neonate larvae <24 hours old (best if all organisms within 4 hours of same age and <12 hours old)
Source of test organisms:	At least 3 weeks prior to tests, initiate laboratory culture of organisms obtained from a biological supply house or government laboratory; obtain test organisms from the third or subsequent broods of a single organism <14 days old
Number of organisms per test chamber:	1
Number of replicate chambers per sediment type:	≥10
Feeding regime:	Daily, 0.1 mL YCT and 0.1 mL algal suspension
Test chamber cleaning:	Before and after
Test solution aeration:	None, except in special instances
Positive control:	Sodium chloride, phenol, or zinc sulfate
Sample holding requirements:	<10 days in the dark at 4 ± 2°C (no freezing); start test <72 hours after preparation of elutriate
Test acceptability criterion:	>80 percent survival in controls, or average >15 live young per surviving adult in controls

Source: Environment Canada. 1992. Biological test method: test of reproduction and survival using the Cladoceran *Ceriodaphnia dubia*. Report EPS 1/Rm/21. Environment Canada, Conservation and Protection, Environmental Protection, Ottawa, Ontario. 72 pp.

Daphnia magna

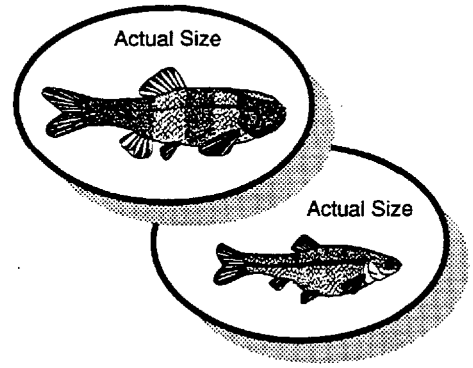
Test No. 211

Actual Size



Test type:	Static nonrenewal
Test endpoint(s):	Survival and biomass
Test duration:	48 hours
Test water temperature:	20°C
Test water salinity:	Freshwater
Photoperiod:	16:8 (L:D)
Test solution volume:	200 mL
Sediment volume and depth:	Not applicable (use sediment elutriates)
Renewal of test solutions:	None
Test organisms:	Juveniles <24 hours old
Source of test organisms:	Laboratory cultures
Number of organisms per test chamber:	10
Number of replicate chambers per sediment type:	3
Feeding regime:	None
Test chamber cleaning:	None
Test solution aeration:	Gentle aeration with glass tipped pipette placed 3 cm below water surface
Positive control:	Not specified
Sample holding requirements:	<2 weeks at 4°C
Test acceptability criterion:	Not specified

Source: Nebeker, A.V., M.A. Cairns, J.H. Gakstatter, K.W. Malueg, G.S. Schuytema, and D.F. Krawczyk. 1984. Biological methods for determining toxicity of contaminated freshwater sediments to invertebrates. Environ. Toxicol. Chem. 3:617–630.



Pimephales promelas

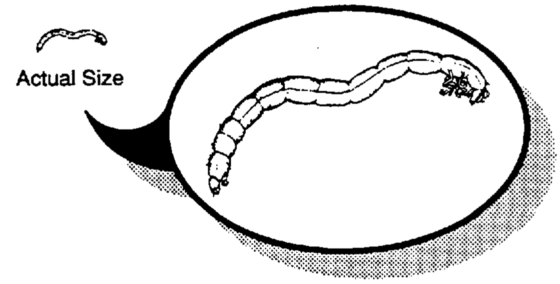
Test No. 232

Test type:	Static renewal
Test endpoint(s):	Survival and growth
Test duration:	6 days
Test water temperature:	22–24°C
Test water salinity:	Freshwater
Photoperiod:	Not specified
Test solution volume:	10 mL
Sediment volume and depth:	Not applicable (use sediment extracts from 50 g of sediment in 1 L of modified FETAX solution)
Renewal of test solutions:	Complete renewal every 24 hours
Test organisms:	Normally developing embryos of the gastrula stage
Source of test organisms:	Laboratory culture
Number of organisms per test chamber:	10–20
Number of replicate chambers per sediment type:	2
Feeding regime:	None
Test chamber cleaning:	Before and after
Test solution aeration:	None
Positive control:	Zinc sulfate
Sample holding requirements:	Not specified
Test acceptability criterion:	Not specified

Source: Dawson, D.A., E.F. Stebler, S.L. Burks, and J.A. Bantle. 1988. Evaluation of the developmental toxicity of metal-contaminated sediments using short-term fathead minnow and frog embryo-larval assays. Environ. Toxicol. Chem. 7:27–34.

Chironomus riparius

Test No. 237

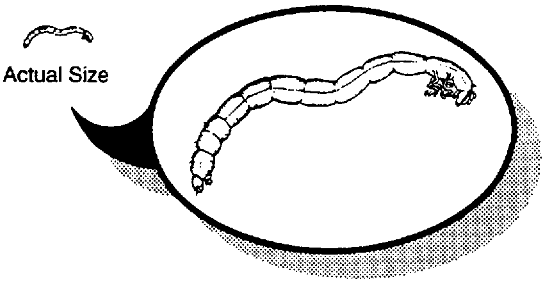


Test type:	Static nonrenewal or flow-through
Test endpoint(s):	Survival, emergence, and growth
Test duration:	10–30 days
Test water temperature:	20–22°C ± 3°C
Test water salinity:	Freshwater
Photoperiod:	16:8 (L:D)
Test solution volume:	800 mL in 1 L (static or flow-through), or 11 L in 13 L (static)
Sediment volume and depth:	200 mL in 1 L (static or flow-through), or 2 L in 13 L (static)
Renewal of test solutions:	None (static)
Test organisms:	First instar larvae <24 hours old, or 3-day-old larvae
Source of test organisms:	3–6 days prior to test collect at least 3 freshly laid egg masses from laboratory cultures, then collect larvae when eggs hatch in approx. 3 days
Number of organisms per test chamber:	50 in 1 L, or 130 in 13 L
Number of replicate chambers per sediment type:	Not specified
Feeding regime:	Various combinations of ground cereal leaves, green algae, and dog treats for static and flow-through tests using 1 L, or 200 mg of fish food flakes every other day in 13 L chamber
Test chamber cleaning:	Before and after
Test solution aeration:	Gentle aeration in static tests
Positive control:	Not specified
Sample holding requirements:	<2 weeks at 4 ± 2°C, avoid freezing
Test acceptability criterion:	≥70 percent survival in controls

Source: ASTM. 1991. Standard guide for conducting sediment toxicity tests with freshwater invertebrates. ASTM E1383-90. American Society for Testing and Materials, Philadelphia, PA. 20 pp.

Chironomus tentans

Test No. 238

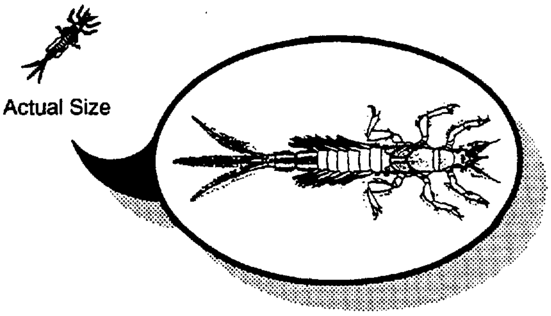


Test type:	Static nonrenewal or flow-through
Test endpoint(s):	Survival, emergence, and growth
Test duration:	10–14 days
Test water temperature:	20-23°C ± 3°C
Test water salinity:	Freshwater
Photoperiod:	16:8 (L:D)
Test solution volume:	800 mL in 1 L or 1.5 L in 2 L (static); 2 L in 3 L (static or flow-through)
Sediment volume and depth:	200 mL in 1 L or 2 cm in 2 L (static); 100 g in 3L (static or flow-through)
Renewal of test solutions:	None (static)
Test organisms:	Second instar juveniles approx. 0.5 cm in length (10–14 days old)
Source of test organisms:	12–16 days before test, collect at least 3 freshly laid egg masses from laboratory cultures, then collect larvae 10–14 days after egg hatch
Number of organisms per test chamber:	15 using 1 L, 20 using 2 L, or 25 using 3 L
Number of replicate chambers per sediment type:	3 using 1 L, or 2 using 3 L
Feeding regime:	50 mg of fish food flakes per day using 3 L chamber containing 25 larvae
Test chamber cleaning:	Before and after
Test solution aeration:	Overnight prior to addition of test organisms provide gentle aeration with glass tipped pipette placed 3 cm below water surface
Positive control:	Not specified
Sample holding requirements:	<2 weeks at 4 ± 2°C, avoid freezing
Test acceptability criterion:	≥70 percent survival in controls

Source: ASTM. 1991. Standard guide for conducting sediment toxicity tests with freshwater invertebrates. ASTM E1383-90. American Society for Testing and Materials, Philadelphia, PA. 20 pp.

Hexagenia limbata

Test No. 240

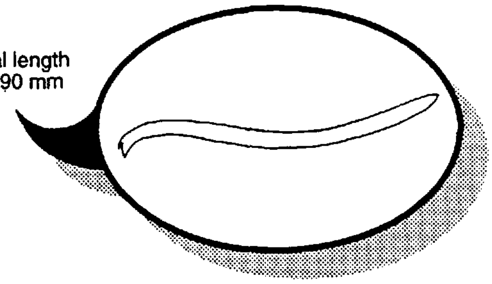


Test type:	Static nonrenewal
Test endpoint(s):	Survival
Test duration:	7 days
Test water temperature:	17°C
Test water salinity:	Freshwater
Photoperiod:	16:8 (L:D)
Test solution volume:	50 mL (containing interstitial water from sediment)
Sediment volume and depth:	Not applicable
Renewal of test solutions:	None
Test organisms:	Nymphs 150 days old post-hatch
Source of test organisms:	Collect and culture fertilized eggs from gravid females sampled in the field
Number of organisms per test chamber:	1
Number of replicate chambers per sediment type:	10
Feeding regime:	Not specified
Test chamber cleaning:	Wash in detergent and acetone, then bake in drying oven for 24 hours
Test solution aeration:	Gentle aeration with glass tipped pipette placed below water surface
Positive control:	Not specified
Sample holding requirements:	<30 days at 4°C
Test acceptability criterion:	Not specified

Source: Giesy, J.P., C.J. Rosiu, R.L. Graney, and M.G. Henry. 1990. Benthic invertebrate bioassays with toxic sediment and pore water. Environ. Toxicol. Chem. 9:233-248.

Lumbriculus variegatus

Test No. 323



Test type:	Flow-through
Test endpoint(s):	Survival, reproduction, and dry weight
Test duration:	10–28 days
Test water temperature:	20°C
Test water salinity:	Freshwater
Photoperiod:	16:8 (L:D)
Test solution volume:	100–150 mL
Sediment volume and depth:	100 mL
Renewal of test solutions:	4–10 water volume additions daily to overlying water
Test organisms:	Animals of equal size
Source of test organisms:	From cultures of brood stock available from several laboratories
Number of organisms per test chamber:	10
Number of replicate chambers per sediment type:	8
Feeding regime:	20 mg of salmon starter per 100 mL of sediment on days 0, 3, 6, and 9 of a 10-day test
Test chamber cleaning:	Before and after
Test solution aeration:	Aerate if necessary to maintain dissolved oxygen >60 percent of saturation
Positive control:	None
Sample holding requirements:	Avoid freezing and drying
Test acceptability criterion:	Not specified

Source: Phipps, G.L., G.T. Ankley, D.A. Benoit, and V.R. Mattson. 1993. Use of the aquatic Oligochaete *Lumbriculus variegatus* for assessing the toxicity of sediment-associated contaminants. Environ. Toxicol. Chem. 12:269–279.

APPENDIX D

Detailed Evaluation Tables

DETAILED EVALUATION TABLES

An overall technical rating for each toxicity test was based on the following major evaluation criteria:

- Reliability
- Ecological relevance
- Exposure relevance
- Availability
- Interferences
- Chemical discrimination.

General factors considered in rating the tests are discussed in the main text of this report (see *Evaluation Criteria and Approach*). Each major evaluation criterion was broken down into subcriteria, and rank scores were assigned based on consideration of test characteristics. The subcriteria are as follows:

- **Reliability**—The determination of reliability was based on the bias, precision, and endpoint of the proposed toxicity test
- **Ecological Relevance**—Field validation and species importance were used to determine the ecological relevance of a toxicity test
- **Exposure Relevance**—Exposure relevance was determined by comparing the test medium (e.g., sediment, elutriate) with the habitat group of the test organism (e.g., epifauna, infauna, plankton)
- **Availability**—Seasonality, geographic coverage, culturability, and protocol development were used to determine the overall availability of an organism for testing
- **Interferences**—Several interferences were targeted as important: nitrates, sulfides, grain size, food (i.e., mold problems), and the overall contamination potential
- **Chemical Discrimination**—Overall sensitivity of a test protocol or test organism was studied to determine its suitability.

Relative ranks were assigned to sediment toxicity tests for the major evaluation criteria according to the scoring systems shown in Tables D-1 through D-8. Ranking of reliability considered scores for endpoints (Tables D-7 and D-8) and whether an interlaboratory comparison had been conducted (Table D-1). Scoring of the ecological relevance of a test was based mainly on the importance of the test species to human and ecological communities (Table D-2). Exposure relevance scores were assigned according to the realism of the exposure medium and the ecological niche of the test species (Table D-3). Whole sediment exposures were considered most relevant, interstitial water and elutriate exposures were considered moderately relevant, and organic solvent extracts were considered least relevant. The order of exposure relevance of test species was infauna (most relevant), epibiota (moderately relevant), and plankton or nekton (least relevant), based on the degree of contact that an organism would have with sediments. Availability scores were based on consideration of temporal availability of species and life stage, species geographic distribution, the availability of laboratories to perform the test, and the degree of protocol development (Table D-4).

The potential for interferences in a sediment toxicity test was based on consideration of the tolerance of organisms for a wide range of sediment grain-size distributions, the presence or absence of sediment in the test chamber, use of solvent extracts, effects of turbidity, and potential for problems with mold, anoxia, or ammonia (Table D-5). Epibiota were considered to have a potentially wider tolerance range for sediment particle size than infauna because the latter live directly in the sediments, while the former live on the sediment surface. The presence of sediment in a chamber was assumed to be potentially stressful to plankton and nekton, while its absence was assumed to be potentially stressful to infauna and possibly stressful to epibiota. Organic solvent extracts are potentially toxic, and interaction between ambient chemicals in the sediment and the exposure to the extract is possible but rarely accounted for in tests that use such extracts. Sediment in the test chamber may be resuspended, which was assumed to represent a potential interference with filter-feeders and with any test that used light emission as an endpoint (e.g., Microtox®). Tests that require feeding of organisms were scored lower than those that do not because of the potential problems of determining the proper diet and the potential for mold problems in the test chamber.

Finally, the ability of a test to discriminate among toxic and nontoxic sediments was based mainly on consideration of the relative sensitivity of tests (Table D-6). Little direct information is available on discriminatory ability.

The evaluation of selected sediment toxicity based on technical and environmental criteria tests is provided in Tables D-9 through D-14.

TABLE D-1. OVERALL RELIABILITY OF SEDIMENT TOXICITY TESTS

Endpoint Characteristics[a]		Interlaboratory Comparison[b]	Reliability Score[c]
Objectivity	Accuracy		
H	H	Yes	4
		No	3
H	M or L	Yes	3
		No	2
M or L	H	Yes	3
		No	2
M	M	Yes	3
		No	2
M	L	Yes	2
		No	1
L	M	Yes	2
		No	1
L	L	Yes	1
		No	0

[a] See Table D-7 for scoring system for endpoints.

H - high
M - medium
L - low.

[b] Entry in this column indicates whether an interlaboratory comparison has been conducted for the sediment toxicity test of interest. Interlaboratory comparisons based on wastewater effluents or reference toxicants were considered relevant only for sediment tests using interstitial water or extracts as the exposure medium.

[c] 4 - most reliable
0 - least reliable.

TABLE D-2. ECOLOGICAL RELEVANCE OF SEDIMENT TOXICITY TEST SPECIES

Species Characteristics	Example	Relevance Score[a]
Commercial or recreational harvest species and a foundation or keystone species[b]	Pacific oyster Purple urchin	4
Commercial or recreational harvest species or a foundation or keystone species	Blue crab Marsh grass	3
Other fish; other invertebrates; major microalgae	Fathead minnow Sand dollar Green algae	2
Bacteria; minor microalgae	Microtox® bacteria	1
Alien species used as surrogate in toxicity testing	African clawed frog	0

[a] 4 - most relevant
0 - least relevant.

[b] A foundation species is a species that provides important physical habitat for other species in a biological community (e.g., marsh grass). A keystone species is a species that controls the species composition and relative abundances of species in a community by its predatory (or grazing) effects (e.g., by grazing on kelp, purple urchins prevent the establishment of kelp beds and maintain open rocky subtidal communities).

TABLE D-3. EXPOSURE RELEVANCE OF SEDIMENT TOXICITY TESTS

	Ecological Niche		
Exposure Medium	Infauna	Epifauna	Plankton/Nekton
Sediment	4	3	1
Interstitial Water	2	1	0
Elutriate (water)	1	1	0
Extract (organic solvent)	0	0	0

Note: Scores assigned to test methods based on ecological relevance of exposure medium and ecological niche of test organism:

4 - most relevant
0 - least relevant.

TABLE D-4. AVAILABILITY OF SEDIMENT TOXICITY TESTS

Species Availability	Laboratory Availability	Protocol Development	Availability Score[a]
Wide distribution; cultured	Many	Standardized protocol	4
Wide distribution; not cultured but available most of the year	Many	Standardized protocol	3
Narrow distribution or not cultured and available only for one or two seasons	Somewhat limited	Protocol relatively developed	2
Very narrow distribution or not cultured and available for a limited portion of the year	Limited	Protocol without details	1
Very narrow distribution and not cultured and available for a very limited portion of the year	Very limited	Protocol without details	0

Note: Mixed combinations of toxicity test attributes (i.e., species availability, laboratory availability, and protocol development) were considered in addition to the combination of examples shown in the table.

[a] 4 - most available
0 - least available.

TABLE D-5. POTENTIAL FOR INTERFERENCES WITH SEDIMENT TOXICITY TESTS

				Plankton	
Media	Epibiota	Infauna	Nekton	Embryo/Larva	Juvenile/Adult
Whole sediment	4	3	2	0	1
Interstitial water	3	2	4	4	4
Elutriate (water)	3	2	4	4	4
Extract (organic solvent)	1	0	2	2	2

Note: 4 - least potential for interferences with a test's assessment of sediment toxicity
0 - highest potential for interferences.

Scores were decreased by one unit for tests that required feeding of the organisms because of the potential for formation of molds and the sorption of some chemicals to organic matter.

TABLE D-6. DISCRIMINATORY ABILITY OF SEDIMENT TOXICITY TESTS

Species and Life Stage Characteristics	Example	Discrimination Score[a]
Sensitive species[b] and sensitive life stage[c] expected to provide good discrimination among toxic and non-toxic sediments	Amphipods Cladocerans Bivalve larvae	4
Overly sensitive test/species that may have less discriminatory power, but response has been calibrated to other tests/species	Microtox® bacteria	3
Insensitive species or insensitive life stage; moderate sensitivity, but not as well known as other tests	Oligochaetes	2
Insensitive species or insensitive life stage, with little or no information on relative sensitivity of test	Rainbow trout fry	1
Insensitive species[d] and insensitive life stage[e]	Juvenile clam Adult fathead minnow	0

Note: Relative sensitivity varies greatly among species within a biotic group, and the resulting generalized scoring scheme used here should not be solely relied upon for selecting tests. Actual test results in comparative studies should be examined for tests of interest.

[a] 4 - most discriminatory ability
0 - least discriminatory ability.

Sensitivity of sublethal tests was scored separately from lethal tests and included consideration of endpoints.

[b] Sensitive species include crustaceans, insects, echinoderms, molluscs (larvae or earlier life stage), and amphibians.

[c] Sensitive life stages include gametes, embryos, and larvae (or neonates).

[d] Insensitive species include polychaetes, fish, molluscs (adults, juveniles), oligochaetes, nematodes, and plants.

[e] Insensitive life stages include juveniles and adults.

TABLE D-7. RELIABILITY OF ENDPOINTS

Endpoint	Objectivity[a]	Accuracy[b]
Molecules/Cells		
Mutation	H	M
Anaphase aberration	M	M
Enzyme production	H	L
Organisms		
Metabolism		
Luminescence	H	L
Carbon-14 (productivity)	H	L
Chlorophyll ratios	H	L
Respiration	M	L
Behavior		
Avoidance	H	L
Reburial	H	L
Growth/development		
Embryo abnormality	L	M
Emergence/molting	H	M
Biomass	H	M
Structure/morphology	M	M
Reproduction		
Fecundity	H	M
Gonad biomass	H	M
Egg fertilization	H	M
Egg sac stage	M	M
Survival	H	H
Populations and Communities		
Intrinsic rate of increase	M	L
Abundance	H	L
Ecosystems	--[c]	--

Note: H - high
M - medium
L - low

[a] Ability to determine endpoint; repeatability.

[b] Lack of bias and laboratory artifacts.

[c] -- - ecosystem (e.g., microcosm) tests for sediment are not developed.

TABLE D-8. ECOLOGICAL RELEVANCE OF ENDPOINTS

Endpoint	Ecological Relevance
Molecules/Cells	
Mutation	L
Anaphase aberration	L
Enzyme production	L
Organisms	
Metabolism	
Luminescence	L
Carbon-14 (productivity)	M–H
Chlorophyll ratios	M–H
Respiration	M
Behavior	
Avoidance	M
Reburial	M
Growth/development	
Embryo abnormality	H
Emergence/molting	H
Biomass	H
Structure/morphology	M–H
Reproduction	
Fecundity	H
Gonad biomass	M–H
Egg fertilization	H
Egg sac stage	H
Survival	H
Populations and Communities	
Intrinsic rate of increase	H
Abundance	H
Ecosystems	--[a]

Note: H - high
M - medium
L - low.

[a] -- - ecosystem tests (e.g., microcosms) for sediment toxicity are not developed.

TABLE D-9. BASIS FOR SEDIMENT TOXICITY TEST EVALUATION – MARINE LETHAL

Test No.	Site	Exposure Media	Biotic Group	Common Name	Life Stage	Recommended	Test Species
001	M/L	S	Amphipod	Amphipod	Juv or adult females	GB,TT,IN	Ampelisca abdita
008	M/L	S	Amphipod	Amphipod	Adults (3–5 mm)	GB,TT,IN, EC	Rhepoxynius abronius
003	M/L	S	Amphipod	Amphipod	Juv or young adults	EC	Eohaustorius washingtonianus
002	M/L	S	Amphipod	Amphipod	Juv or young adults	EC	Amphiporela virginiana
006	M/L	S	Amphipod	Amphipod	Juv or young adults (4–10 mm)	EC	Leptocheirus pinguis
005	M/L	S	Amphipod	Amphipod	Immature (3–6mm); no females with embryos		Grandidierella japonica
004	M/L	S	Amphipod	Amphipod	Juv or young adults (3–6 mm)	EC	Foxiphalus xiximeus
020	M/L	S	Bivalve	Littleneck clam	Juveniles		Protothaca staminea
017	M/L	EL	Bivalve	Blue mussel	Embryos	GB,IN	Mytilus edulis
033	M/L	S	Crustacean	Shrimp	Juveniles		Pandalus sp.
029	M/L	S	Crustacean	Cancer crab	Juveniles		Cancer sp.
027	M/L	S	Crustacean	Blue crab	Juveniles		Callinectes sapidus
034	M/L	S	Crustacean	Shrimp	Post-larvae (8–10 days)		Penaeus sp.
071	M/L	S	Polychaete	Polychaete	Juveniles	GB	Neanthes sp.
021	M/L	S	Bivalve	Japanese clam	Juveniles		Tapes japonica
016	M/L	S	Bivalve	Blue mussel	Embryos		Mytilus edulis
011	M/L	EL	Bivalve	Pacific oyster	Embryos	IN	Crassostrea gigas
009	M/L	EL	Bivalve	Eastern oyster	Embryos	GB,IN	Crassostrea virginica
055	M/L	EL	Fish	Sheepshead minnow	1–14 days old	IN	Cyprinodon variegatus
037	M/L	S	Crustacean	Ridge-back prawn	Juveniles		Sicyonia ingentis
030	M/L	S	Crustacean	Sand shrimp	Juveniles		Crangon sp.
043	M/L	S	Echinoderm	White sea urchin	8–22 mm diameter		Lytechinus pictus
015	M/L	S	Bivalve	Clam	Juveniles		Mulinia lateralis
039	M/L	S	Echinoderm	Sand dollar	Juveniles		Dendraster excentricus
023	M/L	S	Bivalve	Yoldia clam	Juveniles		Yoldia limatula
032	M/L	EL	Crustacean	Shrimp	Embryo-larval		Pandalus sp.
046	M/L	S	Echinoderm	Purple sea urchin	Embryos		Strongylocentrotus purpuratus
045	M/L	EL	Echinoderm	Purple sea urchin	Embryos (< 1 hr old)	IN	Strongylocentrotus purpuratus
010	M/L	S	Bivalve	Pacific oyster	Embryos		Crassostrea gigas
014	M/L	EL	Bivalve	Quahog clam	Embryos		Mercenaria mercenaria
026	M/L	EL	Crustacean	Blue crab	Embryo-larval		Callinectes sapidus
028	M/L	EL	Crustacean	Cancer crab	Embryo-larval		Cancer sp.
035	M/L	EL	Crustacean	Shrimp	Post-larvae (8–10 days)		Penaeus sp.
031	M/L	EL	Crustacean	Grass shrimp	Post-hatch (1–4 days)		Palaemonetes sp.
056	M/L	EL	Fish	3-Spine stickleback	Juveniles (0.1–3.0 g)	EC	Gasterosteus aculeatus

TABLE D-9. (cont.)

Test No.	Site	Exposure Media	Biotic Group	Common Name	Life Stage	Recommended	Test Species
057	M/L	S	Fish	Surf smelt	Larvae		Hypomesus pretiosus
058	M/L	S	Fish	Surf smelt	Larvae		Hypomesus pretiosus
059	M/L	EL	Fish	Pinfish	Embryo-larval		Lagodon rhomboides
060	M/L	EL	Fish	Spot	Embryo-larval		*Leiostomus* xanthurus
061	M/L	EL	Fish	Grunion	Embryo-larval		Leuresthes tenuis
065	M/L	EL	Mysid	Mysid	1-5 days old	GB	Mysidopsis sp.
063	M/L	EL	Mysid	Mysid	*1-5 days old*	GB	Holmesimysis sp.
018	M/L	EL	Bivalve	Oyster	Embryo-larval	GB	Ostrea sp.
041	M/L	EL	Echinoderm	Sand dollar	Embryos	IN	Dendraster excentricus
054	M/L	EL	Fish	Shiner perch	Embryo-larval	GB	Cymatogaster aggregata
062	M/L	S	Mysid	Mysid	1-5 days old		Holmesimysis sp.
064	M/L	S	Mysid	Mysid	1-5 days old		Mysidopsis sp.
044	M/L	EL	Echinoderm	White sea urchin	Embryos (< 1 hr old)		*Lytechinus pictus*
049	M/L	EL	Echinoderm	Sea urchin	Embryos (< 1 hr old)		Strongylocentrotus sp.
053	M/L	EL	Fish	Dolphinfish	Embryo-larval		Coryphaena hippurus
048	M/L	S	*Echinoderm*	*Green sea urchin*	*Embryos*		Strongylocentrotus droebachiensis
040	M/L	S	Echinoderm	Sand dollar	Embryos		Dendraster excentricus
038	M/L	S	Echinoderm	Atlantic urchin	Embryos		Arbacia punctulata
051	M/L	EL	Fish	Speckled sanddab	Juveniles (< 8 cm)		Citharichthys stigmaeus

TABLE D-9. (cont.)

Test No.	Test Species	Salinity (ppt)	Duration	Media	Habitat Group	Ecorating	Notes
001	Ampelisca abdita	28-35	10 days	S	I	4	Particle-feeding, tube dweller
008	Rhepoxynius abronius	28	10 days	S	I	4	Free-burrowing, sand dweller
003	Eohaustorius washingtonianus	12-33	10 days	S	I	4	Free-burrowing, sand dweller
002	Amphiporeia virginiana	31	10 days	S	I	4	Lives in the sediment
006	Leptocheirus pinguis	>25	10 days	S	I	4	Contructs flimsy tubes of bottom sediment and debris; also a deposit feeder.
005	Grandidierella japonica	30-35	10 days	S	I	4	Tube dweller
004	Foxiphalus xiximeus	>25	10 days	S	I	4	
020	Protothaca staminea	23.5-32.7	10 days	S	I	4	
017	Mytilus edulis	18-32	48 hrs	EL	P	0	
033	Pandalus sp.		10 days	S	E	3	
029	Cancer sp.		10 days	S	E	3	
027	Callinectes sapidus		10 days	S	E	3	
034	Penaeus sp.	30-35 +/- 10%	10 days	S	E	3	
071	Neanthes sp.	28 +/- 1	10 days	S	I	4	
021	Tapes japonica		10 days	S	I	4	
016	Mytilus edulis	28	48-60 hrs	S	P	1	
011	Crassostrea gigas	18-32	48 hrs	EL	P	0	
009	Crassostrea virginica	18-32	48 hrs	EL	P	0	
055	Cyprinodon variegatus	5-32 +/- 10%	96 hrs	EL	N	0	
037	Sicyonia ingentis		10 days	S	E	3	
030	Crangon sp.		10 days	S	E	3	
043	Lytechinus pictus		60 days	S	E	3	
015	Mulinia lateralis		7 days	S	I	4	
039	Dendraster excentricus	Seawater	28 days	S	E	3	
023	Yoldia limatula		10 days	S	I	4	
032	Pandalus sp.		48 hrs	EL	E	1	
046	Strongylocentrotus purpuratus	28	48-96 hrs	S	P	1	
045	Strongylocentrotus purpuratus	30 +/-2	48 hrs	EL	P	0	
010	Crassostrea gigas	28	48-60 hrs	S	P	1	
014	Mercenaria mercenaria	18-32	48 hrs	EL	P	0	
026	Callinectes sapidus		48 hrs	EL	P	0	
028	Cancer sp.		48 hrs	EL	P	0	
035	Penaeus sp.	30-35 +/- 10%	96 hrs	EL	E	1	
031	Palaemonetes sp.	30-35 +/- 10%	96 hrs	EL	E	1	
056	Gasterosteus aculeatus	Seawater	96 hrs	EL	N	0	

TABLE D–9. (cont.)

Test No.	Test Species	Salinity (ppt)	Duration	Exposure Relevance: Media	Habitat Group	Ecorating	Notes
057	Hypomesus pretiosus	25	10 days	S	P	1	
058	Hypomesus pretiosus	Seawater	96 hrs	S	P	1	
059	Lagodon rhomboides		48 hrs	EL	P	0	
060	Leiostomus xanthurus		48 hrs	EL	P	0	
061	Leuresthes tenuis	20–32 +/– 10%	48 hrs	EL	P	0	
065	Mysidopsis sp.	25–30 +/– 10%	96 hrs	EL	P	0	
063	Holmesimysis sp.	25–30 +/– 10%	96 hrs	EL	P	0	
018	Ostrea sp.		48 hrs	EL	P	0	
041	Dendraster excentricus	30–32	48 hrs	EL	P	0	
054	Cymatogaster aggregata		48 hrs	EL	P	0	
062	Holmesimysis sp.	25–30 +/– 10%	10 days	S	P	1	
064	Mysidopsis sp.	25–30 +/– 10%	10 days	S	P	1	
044	Lytechinus pictus	30–32	48–96 hrs	EL	P	0	
049	Strongylocentrotus sp.		48 hrs	EL	P	0	
053	Coryphaena hippurus		48 hrs	EL	P	0	
048	Strongylocentrotus droebachiensis	28	48–96 hrs	S	P	1	
040	Dendraster excentricus	28	48–96 hrs	S	P	1	
038	Arbacia punctulata		72–96 hrs	S	P	1	
051	Citharichthys stigmaeus	30 +/– 2	96 hrs	EL	N	0	

TABLE D–9. (cont.)

Test No.	Test Species	Availability: Seasonality	Availability: Geographic Coverage	Availability: Cultured
001	Ampelisca abdita	Year–round (juveniles sometimes difficult to obtain)	East coast of North America; Gulf of Mexico; San Francisco Bay	No; field collected
008	Rhepoxynius abronius	Year–round	Puget Sound to southern California	No; field collected
003	Eohaustorius washingtonianus	Year–round	Southeastern Alaska to Oregon	No; field collected
002	Amphiporeia virginiana	Females with eggs present April–July	Eastern Nova Scotia to North Carolina	No; field collected
006	Leptocheirus pinguis	Not available January–March	East coast of North America	No; field collected
005	Grandidierella japonica	Cultured organisms available	San Fransisco Bay; southern California	Yes; also field collected
004	Foxiphalus xiximeus		Aleutian Islands to southern California	No; field collected
020	Protothaca staminea		Aleutian Islands to Baja California	
017	Mytilus edulis	6 months; cultured organisms available	East and west coasts of North America	Yes
033	Pandalus sp.		Alaska to California	
029	Cancer sp.		West coast of North America	
027	Callinectes sapidus		East coast of North America; Gulf of Mexico	
034	Penaeus sp.		Southeastern coast of North America; Gulf of Mexico	
071	Neanthes sp.	Year–round; cultured organisms available	West coast of North America	Yes
021	Tapes japonica		West coast of North America	
016	Mytilus edulis	6 months; cultured organisms available	East and west coasts of North America	Yes
011	Crassostrea gigas	6 months (lab conditioning possible)	West coast of North America	Yes
009	Crassostrea virginica	Limited	East and gulf coasts of North America	Yes
055	Cyprinodon variegatus	Cultured organisms available	East coast of North America	Yes
037	Sicyonia ingentis		West coast of North America (mainly California)	
030	Crangon sp.		East and west coast of North America	
043	Lytechinus pictus	Year–round	West coast of North America; southern California to South America	No; field collected
015	Mulinia lateralis		East coast of North America	No
039	Dendraster excentricus	6 months	West coast of North America	No; field collected
023	Yoldia limatula			
032	Pandalus sp.		Alaska to California	
046	Strongylocentrotus purpuratus	6 months	West coast of North America (Alaska to Mexico)	No; field collected
045	Strongylocentrotus purpuratus	6 months	West coast of North America (Alaska to Mexico)	No; field collected
010	Crassostrea gigas	6 months (lab conditioning possible)	West coast of North America	Yes
014	Mercenaria mercenaria	Limited	East coast of North America	Yes
026	Callinectes sapidus		East coast of North America; Gulf of Mexico	
028	Cancer sp.		West coast of North America	
035	Penaeus sp.		Southeastern coast of North America; Gulf of Mexico	
031	Palaemonetes sp.		Isolated occurences, species specific; east coast of North America; Gulf of Mexico	
056	Gasterosteus aculeatus	Cultured organisms available	West and east coasts of North America	Yes; also field collected

TABLE D-9. (cont.)

Test No.	Test Species	Availability: Seasonality	Availability: Geographic Coverage	Availability: Cultured
057	Hypomesus pretiosus	Limited	Southeastern Alaska to northern California	No; field collected
058	Hypomesus pretiosus	Limited	Southeastern Alaska to northern California	No; field collected
059	Lagodon rhomboides	Limited	Southeastern coast of North America	
060	Leiostomus xanthurus	Limited	Inshore waters from southern New England to Texas	
061	Leuresthes tenuis	Limited	Southern California	
065	Mysidopsis sp.	Species-specific	East coast of North America; Gulf of Mexico	
063	Holmesimysis sp.	Species-specific	Southeastern coast of North America; Gulf of Mexico	
018	Ostrea sp.	Limited	Alaska to Baja California	
041	Dendraster excentricus	6 months	West coast of North America	No; field collected
054	Cymatogaster aggregata		Southeast Alaska to southern Clalfornia	
062	Holmesimysis sp.	Species-specific	Southeastern coast of North America; Gulf of Mexico	
064	Mysidopsis sp.	Species-specific	East coast of North America; Gulf of Mexico	
044	Lytechinus pictus	6 months	West coast of North America; southern California to South America	No; field collected
049	Strongylocentrotus sp.		West coast of North America (Alaska to Mexico)	No; field collected
053	Coryphaena hippurus		Southeastern, extreme southwestern, and gulf coasts of North America	
048	Strongylocentrotus droebachiensis		West coast of North America (Alaska to Mexico)	No; field collected
040	Dendraster excentricus	6 months	West coast of North America	No; field collected
038	Arbacia punctulata	June - August	East coast of North America; Gulf of Mexico	No; field collected
051	Citharichthys stigmaeus		Southern Alaska to southern California	

TABLE D–9. (cont.)

Test No.	Test Species	Availability: Protocol Development	Availability: Overall Availability	Ecological Relevance: Field Validation	Ecological Relevance: Species Importance	Ecological Relevance: Overall EcoRelevance
001	Ampelisca abdita	ASTM (1990)	3		High; foundation species	3
008	Rhepoxynius abronius	ASTM (1990)	3			2
003	Eohaustorius washingtonianus	EC (1992a)	3		Imp. prey to shorebirds and fishes	2
002	Amphiporeia virginiana	EC (1992a)	3			2
006	Leptocheirus pinguis	EC (1992a)	3			2
005	Grandidierella japonica	ASTM (1990)	3			2
004	Foxiphalus xiximeus	EC (1992a)	1			2
020	Protothaca staminea	Yes	2		Harvested	3
017	Mytilus edulis	ASTM (1989)	3		High; foundation species; harvested	4
033	Pandalus sp.	Yes	1		Harvested	3
029	Cancer sp.	Yes	1		Harvested	3
027	Callinectes sapidus	Yes	1		Harvested	3
034	Penaeus sp.	Yes	1		Harvested	3
071	Neanthes sp.	Dillon et al. (1993)	1			2
021	Tapes japonica	Yes	1		Harvested	3
016	Mytilus edulis	U.S. EPA (1991b)	3		High; foundation species; harvested	4
011	Crassostrea gigas	ASTM (1989)	2	Field validated (calibrated) to benthos (Becker et al. 1990)	High; sensitive bivalve lifestage; harvested	4
009	Crassostrea virginica	ASTM (1989)	2		High; sensitive bivalve lifestage; harvested	4
055	Cyprinodon variegatus	Yes	4			2
037	Sicyonia ingentis	Yes	1			2
030	Crangon sp.	Yes	1			2
043	Lytechinus pictus		1			2
015	Mulinia lateralis		1			2
039	Dendraster excentricus	Dinnel and Stober (1985)	1			2
023	Yoldia limatula	Yes	1			2
032	Pandalus sp.	Yes	1		Harvested	3
046	Strongylocentrotus purpuratus	U.S. EPA (1991b)	2		High; keystone species; grazer on kelp	4
045	Strongylocentrotus purpuratus	Yes	1		High; keystone species; grazer on kelp	4
010	Crassostrea gigas	U.S. EPA (1991b)	2	Field validated (calibrated) to benthos (Becker et al. 1990)	High; sensitive bivalve lifestage; harvested	4
014	Mercenaria mercenaria	ASTM (1989)	2		Harvested	3
026	Callinectes sapidus	Yes	1		Harvested	3
028	Cancer sp.	Yes	1		Harvested	3
035	Penaeus sp.	Yes	1		Harvested	3
031	Palaemonetes sp.	Yes	1			2
056	Gasterosteus aculeatus	EC (1990c)	3			2

TABLE D–9. (cont.)

Test No.	Test Species	Availability: Protocol Development	Availability: Overall Availability	Ecological Relevance: Field Validation	Ecological Relevance: Species Importance	Ecological Relevance: Overall EcoRelevance
057	Hypomesus pretiosus		1			3
058	Hypomesus pretiosus		1			3
059	Lagodon rhomboides	Yes	1			3
060	Leiostomus xanthurus	Yes	1			3
061	Leuresthes tenuis	ASTM (1980)	1			3
065	Mysidopsis sp.	Yes	1			2
063	Holmesimysis sp.	ASTM (1980)	1			2
018	Ostrea sp.		1			2
041	Dendraster excentricus	Yes	1			2
054	Cymatogaster aggregata	Yes	1			2
062	Holmesimysis sp.	Yes	1			2
064	Mysidopsis sp.	Yes	1			2
044	Lytechinus pictus	Adapted	1			2
049	Strongylocentrotus sp.	Yes	1			2
053	Coryphaena hippurus	Yes	0			3
048	Strongylocentrotus droebachiensis	U.S. EPA (1991b)	2			2
040	Dendraster excentricus	U.S. EPA (1991b)	1			2
038	Arbacia punctulata		1			2
051	Citharichthys stigmaeus	ASTM (1990)	1			2

TABLE D–9. (cont.)

Test No.	Test Species	Reliability: Accuracy	Objectivity	Endpoint	Overall Reliability	Notes
001	Ampelisca abdita	H	H	Survival	3	Interlab (U.S. EPA 1992); problem with inexperienced lab
008	Rhepoxynius abronius	H	H	Survival	4	Interlab (Mearns et al. 1986)
003	Eohaustorius washingtonianus	H	H	Survival	4	Interlab (Paine and McPherson 1991a,b)
002	Amphiporeia virginiana	H	H	Survival	4	Interlab (Paine and McPherson 1991a)
006	Leptocheirus pinguis	H	H	Survival	3	
005	Grandidierella japonica	H	H	Survival	3	
004	Foxiphalus xiximeus	H	H	Survival	4	Interlab (Paine and McPherson 1991a,b)
020	Protothaca staminea	H	H	Survival	3	
017	Mytilus edulis	H	H	Survival	2	
033	Pandalus sp.	H	H	Survival	3	
029	Cancer sp.	H	H	Survival	3	
027	Callinectes sapidus	H	H	Survival	3	
034	Penaeus sp.	H	H	Survival	3	
071	Neanthes sp.	H	H	Survival	3	
021	Tapes japonica	H	H	Survival	3	
016	Mytilus edulis	H	H	Survival	2	
011	Crassostrea gigas	H	H	Survival	2	
009	Crassostrea virginica	H	H	Survival	2	
055	Cyprinodon variegatus	H	H	Survival	4	Inter/intra lab (U.S. EPA 1991c)
037	Sicyonia ingentis	H	H	Survival	3	
030	Crangon sp.	H	H	Survival	3	
043	Lytechinus pictus	H	H	Survival	3	
015	Mulinia lateralis	H	H	Survival	3	
039	Dendraster excentricus	H	H	Survival	3	
023	Yoldia limatula	H	H	Survival	3	
032	Pandalus sp.	H	H	Survival	3	
046	Strongylocentrotus purpuratus	H	H	Survival	2	May be highly variable response
045	Strongylocentrotus purpuratus	H	H	Survival	2	May be highly variable response
010	Crassostrea gigas	H	H	Survival	2	
014	Mercenaria mercenaria	H	H	Survival	2	
026	Callinectes sapidus	H	H	Survival	3	
028	Cancer sp.	H	H	Survival	3	
035	Penaeus sp.	H	H	Survival	3	
031	Palaemonetes sp.	H	H	Survival	3	
056	Gasterosteus aculeatus	H	H	Survival	3	

TABLE D–9. (cont.)

Test No.	Test Species	Reliability: Accuracy	Objectivity	Endpoint	Overall Reliability	Notes
057	Hypomesus pretiosus	H	H	Survival	3	
058	Hypomesus pretiosus	H	H	Survival	3	
059	Lagodon rhomboides	H	H	Survival	3	
060	Leiostomus xanthurus	H	H	Survival	3	
061	Leuresthes tenuis	H	H	Survival	3	
065	Mysidopsis sp.	H	H	Survival	3	
063	Holmesimysis sp.	H	H	Survival	3	
018	Ostrea sp.	H	H	Survival	2	
041	Dendraster excentricus	H	H	Survival	2	May be highly variable response
054	Cymatogaster aggregata	H	H	Survival	3	
062	Holmesimysis sp.	H	H	Survival	3	
064	Mysidopsis sp.	H	H	Survival	3	
044	Lytechinus pictus	H	H	Survival	2	May be highly variable response
049	Strongylocentrotus sp.	H	H	Survival	2	May be highly variable response
053	Coryphaena hippurus	H	H	Survival	3	
048	Strongylocentrotus droebachiensis	H	H	Survival	2	May be highly variable response
040	Dendraster excentricus	H	H	Survival	2	May be highly variable response
038	Arbacia punctulata	H	H	Survival	2	May be highly variable response
051	Citharichthys stigmaeus	H	H	Survival	3	

TABLE D–9. (cont.)

Test No.	Test Species	Interferences: Notes	Interferences: Overall Interferences
001	Ampelisca abdita	Inhabits fine sand–mud/silt; species tolerance range established; not fed	3
008	Rhepoxynius abronius	Prefers fine, sandy sediments; silts & clays may interfere; not fed	3
003	Eohaustorius washingtonianus	Insufficient data	3
002	Amphiporeia virginiana	Not influenced markedly by sediment particle size.	3
006	Leptocheirus pinguis		3
005	Grandidierella japonica	Lives in wide variety of sediment types; not fed	3
004	Foxiphalus xiximeus	Inhabits clean, medium to fine sand	3
020	Protothaca staminea		3
017	Mytilus edulis	Elutriate test (N/A); not fed	4
033	Pandalus sp.		4
029	Cancer sp.		4
027	Callinectes sapidus		4
034	Penaeus sp.	Not fed	4
071	Neanthes sp.		3
021	Tapes japonica		3
016	Mytilus edulis	Increased response in fine grained sediments (Chapman et al. 1987); not fed	0
011	Crassostrea gigas	Elutriate test (N/A): not fed	4
009	Crassostrea virginica	Elutriate test (N/A); not fed	4
055	Cyprinodon variegatus	Elutriate test (N/A); fed at 48h	3
037	Sicyonia ingentis		4
030	Crangon sp.		4
043	Lytechinus pictus		4
015	Mulinia lateralis		4
039	Dendraster excentricus	Not fed	4
023	Yoldia limatula		3
032	Pandalus sp.	Elutriate test (N/A)	3
046	Strongylocentrotus purpuratus	Not fed; potential effects for fine sediments (PTI 1991)	0
045	Strongylocentrotus purpuratus	Elutriate test (N/A); not fed	4
010	Crassostrea gigas	Not fed; potential effects for fine sediments (PTI 1991)	0
014	Mercenaria mercenaria	Elutriate test (N/A); not fed	4
026	Callinectes sapidus	Elutriate test (N/A)	4
028	Cancer sp.	Elutriate test (N/A)	4
035	Penaeus sp.	Elutriate test (N/A); not fed	3
031	Palaemonetes sp.	Elutriate test (N/A); not fed	3
056	Gasterosteus aculeatus	Elutriate test (N/A)	4

TABLE D-9. (cont.)

Test No.	Test Species	Interferences: Notes	Interferences: Overall Interferences
057	Hypomesus pretiosus		2
058	Hypomesus pretiosus		2
059	Lagodon rhomboides		4
060	Leiostomus xanthurus	Elutriate test (N/A)	4
061	Leuresthes tenuis	Elutriate test (N/A)	4
065	Mysidopsis sp.	Elutriate test (N/A); fed daily	3
063	Holmesimysis sp.	Elutriate test (N/A); fed	3
018	Ostrea sp.	Elutriate test (N/A)	4
041	Dendraster excentricus	Elutriate test (N/A); not fed	4
054	Cymatogaster aggregata	Elutriate test (N/A)	4
062	Holmesimysis sp.	Fed	0
064	Mysidopsis sp.	Fed daily	0
044	Lytechinus pictus	Elutriate test (N/A); not fed	4
049	Strongylocentrotus sp.	Elutriate test (N/A)	4
053	Coryphaena hippurus	Elutriate test (N/A)	4
048	Strongylocentrotus droebachiensis	Not fed	0
040	Dendraster excentricus	Not fed; potential effects for fine sediments (PTI 1991)	0
038	Arbacia punctulata	Potential effects for fine sediments (PTI 1991)	0
051	Citharichthys stigmaeus	Elutriate test (N/A); fed at 48h	3

TABLE D–9. (cont.)

Test No.	Chemical Discrimination Sensitivity
001	Sensitive to wide range of anthropogenic materials (e.g., PAH, PCB, metals).
008	Among the most sensitive of sediment toxicity test organisms.
003	E. washingtonianus is the same or slightly more sensitive than R. abronius; more difficult to remove from the test sediment than R. abronius.
002	Similar sensitivity to R. abronius.
006	
005	Apparent insensitivity to some organic chemicals compared to other amphipods (Swartz et al. 1994).
004	Similar sensitivity to R. abronius and E. estuarius; the range of tolerance to salinity and temperature extremes has not been studied.
020	
017	Highly sensitive response of M. edulis to toxic sediment was reported in Chapman et al. (1987) and Long et al. (1990).
033	
029	
027	
034	
071	
021	
016	Highly sensitive response of M. edulis to toxic sediment was reported in Chapman et al. (1987) and Long et al. (1990).
011	
009	
055	
037	
030	
043	Variable results in comparative bioassay tests (metal and organic compounds). Ranged from 1–3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
015	
039	Variable results in comparative bioassay tests (metal and organic compounds). Ranged from 1–3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
023	
032	
046	Variable results in comparative bioassay tests (metal and organic compounds). Ranged from 1–3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
045	Variable results in comparative bioassay tests (metal and organic compounds). Ranged from 1–3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
010	
014	
026	
028	
035	
031	
056	

TABLE D-9. (cont.)

Test No.	Chemical Discrimination: Sensitivity
057	
058	
059	
060	
061	
065	
063	
018	
041	Variable results in comparative bioassay tests (metal and organic compounds). Ranged from 1-3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
054	
062	
064	
044	Variable results in comparative bioassay tests (metal and organic compounds). Ranged from 1-3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
049	
053	
048	Variable results in comparative bioassay tests (metal and organic compounds). Ranged from 1-3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
040	Variable results in comparative bioassay tests (metal and organic compounds). Ranged from 1-3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
038	Variable results in comparative bioassay tests (metal and organic compounds). Ranged from 1-3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
051	

TABLE D–9. (cont.)

Test No.	Test Species	Chemical Discrimination Overall Sensitivity	Regulatory Status
001	Ampelisca abdita	4	High
008	Rhepoxynius abronius	4	High
003	Eohaustorius washingtonianus	4	Medium
002	Amphiporeia virginiana	2	Medium
006	Leptocheirus pinguis	2	Medium
005	Grandidierella japonica	2	Low
004	Foxiphalus xiximeus	2	Medium
020	Protothaca staminea	0	Low
017	Mytilus edulis	4	Medium
033	Pandalus sp.	2	Low
029	Cancer sp.	2	Low
027	Callinectes sapidus	2	Low
034	Penaeus sp.	2	Low
071	Neanthes sp.	2	Medium
021	Tapes japonica	0	Low
016	Mytilus edulis	4	Medium
011	Crassostrea gigas	4	Medium
009	Crassostrea virginica	4	Medium
055	Cyprinodon variegatus	1	Medium
037	Sicyonia ingentis	2	Low
030	Crangon sp.	2	Low
043	Lytechinus pictus	2	Low
015	Mulinia lateralis	0	Low
039	Dendraster excentricus	2	Low
023	Yoldia limatula	0	Low
032	Pandalus sp.	4	Low
046	Strongylocentrotus purpuratus	4	Medium
045	Strongylocentrotus purpuratus	4	Medium
010	Crassostrea gigas	4	Low
014	Mercenaria mercenaria	4	Low
026	Callinectes sapidus	4	Low
028	Cancer sp.	4	Low
035	Penaeus sp.	2	Low
031	Palaemonetes sp.	4	Low
056	Gasterosteus aculeatus	0	Medium

TABLE D–9. (cont.)

Test No.	Test Species	Chemical Discrimination Overall Sensitivity	Regulatory Status
057	Hypomesus pretiosus	2	Low
058	Hypomesus pretiosus	2	Low
059	Lagodon rhomboides	2	Low
060	Leiostomus xanthurus	2	Low
061	Leuresthes tenuis	2	Low
065	Mysidopsis sp.	4	Medium
063	Holmesimysis sp.	4	Medium
018	Ostrea sp.	4	Medium
041	Dendraster excentricus	4	Medium
054	Cymatogaster aggregata	2	Medium
062	Holmesimysis sp.	4	Low
064	Mysidopsis sp.	4	Low
044	Lytechinus pictus	4	Low
049	Strongylocentrotus sp.	4	Low
053	Coryphaena hippurus	2	Low
048	Strongylocentrotus droebachiensis	2	Medium
040	Dendraster excentricus	4	Medium
038	Arbacia punctulata	4	Low
051	Citharichthys stigmaeus	0	Low

TABLE D-9. (cont.)

Test No.	General Notes
001	Control survivability declines in coarse sand; euryhaline; amphipods are more sensitive to contaminated sediments than other major taxa (Swartz 1987).
008	Sensitive to salinities <25 g/kg (limits its use to test marine sediments); amphipods are more sensitive to contaminated sediments than other major taxa (Swartz 1987).
003	High level of availability, easy to handle, tolerant to a wide range of salinities; amphipods are more sensitive to contaminated sediments than other major taxa (Swartz 1987); most common haustoriid species.
002	Tolerant to wide range of salinities, however, reduced survival if salinity <20 ppt (Doe and Wade 1991).
006	Preliminary data indicate that R. abronius may be more sensitive.
005	Broad application because it's possible to conduct tests in sands, silts, or clays; amphipods are more sensitive to contaminated sediments than other major taxa (Swartz 1987).
004	High level of availability, easy to handle, tolerant to a wide range of salinities; sensitive to contaminated sediments (Swartz 1987); reburies/swims quickly could be difficult at test end.
020	
017	Sensitivity of M. edulis and R. abronius tests appears to be similar (Chapman et al. 1987; Long et al. 1990); abnormal development if >30 embryos/mL
033	
029	
027	
034	
071	
021	
016	Sensitivity of M. edulis and R. abronius tests appears to be similar (Chapman et al. 1987; Long et al. 1990); abnormal development if >30 embryos/mL
011	Not too difficult to isolate and obtain large numbers of embryos from individual male–female pairs.
009	Can tolerate DO of 0.5 mg/L; however, growth is reduced at DO <4.2 mg/L; abnormal development if >30 embryos/mL.
055	Widely used as estuarine toxicity and physiological test organism; has been proposed for use in dredged material assessments.
037	
030	
043	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
015	
039	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
023	
032	
046	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
045	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
010	Not too difficult to isolate and obtain large numbers of embryos from individual male–female pairs.
014	Abnormal development if >30 embryos/mL
026	
028	
035	
031	
056	Well documented life history; easily captured; species is euryhaline; size suitable for acute toxicity tests; recommended by EC and EPA.

TABLE D-9. (cont.)

Test No.	General Notes
057	
058	
059	
060	
061	
065	
063	
018	
041	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
054	
062	
064	
044	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
049	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
053	
048	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
040	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
038	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
051	Adapted in part from the Menidia sp. protocol in U.S. EPA (1991a).

TABLE D–9. (cont.)

Test No.	Test Species	References
001	Ampelisca abdita	ASTM (1990) (E1367–90); U.S. EPA (1991b); U.S. EPA and U.S. COE (1991, 1993); DeWitt et al. (1992)
008	Rhepoxynius abronius	ASTM (1990) (E1367–90); U.S. EPA (1991b); U.S. EPA and U.S. COE (1991, 1993); Environment Canada (1992a)
003	Eohaustorius washingtonianus	Environment Canada (1992a)
002	Amphiporeia virginiana	Environment Canada (1992a)
006	Leptocheirus pinguis	Environment Canada (1992a), based on ASTM (1990) (E1367–90); Swartz et al. (1985)
005	Grandidierella japonica	ASTM (1990) (E1367–90); Reish and Lemay (1988); U.S. EPA and U.S. COE (1993)
004	Foxiphalus xiximeus	Environment Canada (1992a), based on ASTM (1990) (E1367–90); Swartz et al. (1985)
020	Protothaca staminea	Swartz et al. (1979); U.S. EPA and U.S. COE (1991)
017	Mytilus edulis	ASTM (1989) (E724–89); Chapman and Morgan (1983); Long et al. (1990); U.S. EPA and U.S. COE (1991, 1993)
033	Pandalus sp.	U.S. EPA and U.S. COE (1991)
029	Cancer sp.	U.S. EPA and U.S. COE (1991)
027	Callinectes sapidus	U.S. EPA and U.S. COE (1991)
034	Penaeus sp.	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
071	Neanthes sp.	Tay et al. (1992); U.S. EPA and U.S. COE (1991, 1993); Dillon et al. (1993)
021	Tapes japonica	U.S. EPA and U.S. COE (1991)
016	Mytilus edulis	U.S. EPA (1991b)
011	Crassostrea gigas	ASTM (1989) (E724–89); U.S. EPA and U.S. COE (1993)
009	Crassostrea virginica	ASTM (1989) (E724–89); U.S. EPA and U.S. COE (1991, 1993)
055	Cyprinodon variegatus	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
037	Sicyonia ingentis	U.S. EPA and U.S. COE (1991)
030	Crangon sp.	U.S. EPA and U.S. COE (1991)
043	Lytechinus pictus	Thompson et al. (1989)
015	Mulinia lateralis	Burgess et al. (1992)
039	Dendraster excentricus	Casillas et al. (1992a)
023	Yoldia limatula	U.S. EPA and U.S. COE (1991)
032	Pandalus sp.	U.S. EPA and U.S. COE (1991)
046	Strongylocentrotus purpuratus	U.S. EPA (1991b)
045	Strongylocentrotus purpuratus	U.S. EPA and U.S. COE (1993); Dinnel et al. (1982)
010	Crassostrea gigas	U.S. EPA (1991b)
014	Mercenaria mercenaria	ASTM (1989) (E724–89)
026	Callinectes sapidus	U.S. EPA and U.S. COE (1991)
028	Cancer sp.	U.S. EPA and U.S. COE (1991)
035	Penaeus sp.	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
031	Palaemonetes sp.	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
056	Gasterosteus aculeatus	Environment Canada (1990c)

TABLE D-9. (cont.)

Test No.	Test Species	References
057	Hypomesus pretiosus	Chapman et al. (1985)
058	Hypomesus pretiosus	Casillas et al. (1992b)
059	Lagodon rhomboides	U.S. EPA and U.S. COE (1991)
060	Leiostomus xanthurus	U.S. EPA and U.S. COE (1991)
061	Leuresthes tenuis	Reish and Lemay (1988); U.S. EPA and U.S. COE (1991)
065	Mysidopsis sp.	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
063	Holmesimysis sp.	Reish and Lemay (1988); U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
018	Ostrea sp.	U.S. EPA and U.S. COE (1991)
041	Dendraster excentricus	U.S. EPA and U.S. COE (1993)
054	Cymatogaster aggregata	U.S. EPA and U.S. COE (1991)
062	Holmesimysis sp.	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
064	Mysidopsis sp.	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
044	Lytechinus pictus	Reish and Lemay (1988); U.S. EPA and U.S. COE (1993)
049	Strongylocentrotus sp.	U.S. EPA and U.S. COE (1993)
053	Coryphaena hippurus	U.S. EPA and U.S. COE (1991)
048	Strongylocentrotus droebachiensis	U.S. EPA (1991b)
040	Dendraster excentricus	U.S. EPA (1991b)
038	Arbacia punctulata	Lamberson et al. (1992), following Dinnel and Stober (1985); U.S. EPA (1991b)
051	Citharichthys stigmaeus	Reish and Lemay (1988); U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)

Notes: EL – elutriate
EX – extract
INT – interstitial water
S – sediment

N/A – not applicable
N/S – not specified

EC – Environment Canada (1990, 1992)
GB – U.S. EPA and U.S. COE (1991)
IN – U.S. EPA and U.S. COE (1993)
TT – U.S. EPA (1992)

TABLE D-10. BASIS FOR SEDIMENT TOXICITY TEST EVALUATION – MARINE SUBLETHAL

Test No.	Site	Exposure Media	Biotic Group	Common Name	Life Stage	Recommended	Test Species
001	M/S	S	Amphipod	Amphipod	Juv or adult females		Ampelisca abdita
121	M/S	S	Polychaete	Polychaete	Juveniles		Neanthes sp.
109	M/S	EL	Echinoderm	Purple sea urchin	Gametes	EC	Strongylocentrotus purpuratus
008	M/S	S	Amphipod	Amphipod	Adults (3–5 mm)		Rhepoxynius abronius
005	M/S	S	Amphipod	Amphipod	Immature (3–6mm); no females with embryos		Grandidierella japonica
017	M/S	EL	Bivalve	Blue mussel	Embryos		Mytilus edulis
111	M/S	EL	Echinoderm	Purple sea urchin	Gametes		Strongylocentrotus purpuratus
047	M/S	EL	Echinoderm	Purple sea urchin	Gametes	IN	Strongylocentrotus purpuratus
102	M/S	EL	Echinoderm	Sand dollar	Gametes	EC	Dendraster excentricus
100	M/S	EL	Echinoderm	Atlantic urchin	Gametes	EC	Arbacia punctulata
115	M/S	EL	Echinoderm	Green sea urchin	Gametes	EC	Strongylocentrotus droebachiensis
015	M/S	S	Bivalve	Clam	Juveniles		Mulina lateralis
107	M/S	EL	Echinoderm	White sea urchin	Gametes	EC	Lytechinus pictus
112	M/S	EL	Echinoderm	Purple sea urchin	Gametes		Strongylocentrotus purpuratus
011	M/S	EL	Bivalve	Pacific oyster	Embryos		Crassostrea gigas
081	M/S	EL	Bacterium	Microtox	Cells		Photobacterium phosphoreum
016	M/S	S	Bivalve	Blue mussel	Embryos		Mytilus edulis
009	M/S	EL	Bivalve	Eastern oyster	Embryos		Crassostrea virginica
080	M/S	INT	Bacterium	Microtox	Cells		Photobacterium phosphoreum
010	M/S	S	Bivalve	Pacific oyster	Embryos		Crassostrea gigas
046	M/S	S	Echinoderm	Purple sea urchin	Embryos		Strongylocentrotus purpuratus
042	M/S	EL	Echinoderm	Sand dollar	Gametes	IN	Dendraster excentricus
043	M/S	S	Echinoderm	White sea urchin	8–22 mm diameter		Lytechinus pictus
039	M/S	S	Echinoderm	Sand dollar	Juveniles		Dendraster excentricus
014	M/S	EL	Bivalve	Quahog clam	Embryos		Mercenaria mercenaria
098	M/S	EL	Echinoderm	Atlantic urchin	Gametes		Arbacia punctulata
097	M/S	INT	Echinoderm	Atlantic urchin	Gametes		Arbacia punctulata
048	M/S	S	Echinoderm	Green sea urchin	Embryos		Strongylocentrotus droebachiensis
058	M/S	S	Fish	Surf smelt	Larvae		Hypomesus pretiosus
038	M/S	S	Echinoderm	Atlantic urchin	Gametes		Arbacia punctulata
083	M/S	S	Bacterium	Microtox	Cells	EC	Photobacterium phosphoreum
084	M/S	EX	Bacterium	Mod. Ames/HPTLC	Cells		Salmonella typhimurium
040	M/S	S	Echinoderm	Sand dollar	Embryos		Dendraster excentricus

TABLE D-10. (cont.)

Test No.	Test Species	Salinity (ppt)	Duration	Exposure Relevance: Media	Habitat Group	Ecorating	Notes
001	Ampelisca abdita	28-35	10 days	S	I	4	Particle-feeding, tube dweller
121	Neanthes sp.	28 +/- 2	20-28 days	S	I	4	
109	Strongylocentrotus purpuratus	28-34	20, 40, 60 min	EL	P	0	
008	Rhepoxynius abronius	28	10 days	S	I	4	Free-burrowing, sand dweller
005	Grandidierella japonica	30-35	10 days	S	I	4	Tube dweller
017	Mytilus edulis	18-32	48 hrs	EL	P	0	
111	Strongylocentrotus purpuratus	32	80 min	EL	P	0	
047	Strongylocentrotus purpuratus	30-32	80 min	EL	P	0	
102	Dendraster excentricus	28-34	20, 40, 60 min	EL	P	0	
100	Arbacia punctulata	28-34	20, 40, 60 min	EL	P	0	
115	Strongylocentrotus droebachiensis	28-34	20, 40, 60 min	EL	P	0	
015	Mulina lateralis		7 days	S	E	3	
107	Lytechinus pictus	28-34	20, 40, 60 min	EL	P	0	
112	Strongylocentrotus purpuratus	32	48 hrs	EL	P	0	
011	Crassostrea gigas	18-32	48 hrs	EL	P	0	
081	Photobacterium phosphoreum		15 min	EL	P	2	
016	Mytilus edulis	28	48-60 hrs	S	P	1	
009	Crassostrea virginica	18-32	48 hrs	EL	P	0	
080	Photobacterium phosphoreum	31-34	15 min	INT	P	1	
010	Crassostrea gigas	28	48-60 hrs	S	P	1	
046	Strongylocentrotus purpuratus	28	48-96 hrs	S	P	1	
042	Dendraster excentricus	30 +/- 2	80 min	EL	P	0	
043	Lytechinus pictus		60 days	S	E	3	
039	Dendraster excentricus	Seawater	28 days	S	E	3	
014	Mercenaria mercenaria	18-32	48 hrs	EL	P	0	
098	Arbacia punctulata	28	1.3 hrs	EL	P	0	Modified elutriate
097	Arbacia punctulata	28	1.3 hrs	INT	P	0	
048	Strongylocentrotus droebachiensis	28	48-96 hrs	S	P	1	
058	Hypomesus pretiosus	Seawater	4 days	S	P	1	
038	Arbacia punctulata		72-96 hrs	S	P	1	
083	Photobacterium phosphoreum	>2	5 days 15 min	S	P	0	
084	Salmonella typhimurium		N/S	EX	N/A	0	
040	Dendraster excentricus	28	48-96 hrs	S	P	1	

TABLE D-10. (cont.)

Test No.	Test Species	Availability: Seasonality	Availability: Geographic Coverage	Availability: Cultured
001	Ampelisca abdita	Year-round (juveniles sometimes difficult to obtain)	East coast of North America; Gulf of Mexico; San Francisco Bay	No; field collected
121	Neanthes sp.	Year-round; cultured organisms available	West coast of North America	Yes
109	Strongylocentrotus purpuratus	6 months	West coast of North America (Alaska to Mexico)	No; field collected
008	Rhepoxynius abronius	Year-round	Puget Sound to southern California	No; field collected
005	Grandidierella japonica	Cultured organisms available	San Fransico Bay; southern California	Yes; also field collected
017	Mytilus edulis	6 months; cultured organisms available	West coast of North America	Yes
111	Strongylocentrotus purpuratus	6 months	West coast of North America (Alaska to Mexico)	No; field collected
047	Strongylocentrotus purpuratus	6 months	West coast of North America (Alaska to Mexico)	No; field collected
102	Dendraster excentricus	6 months	West coast of North America	No; field collected
100	Arbacia punctulata	June - August	East coast of North America; Gulf of Mexico	No; field collected
115	Strongylocentrotus droebachiensis	6 months	West coast of North America (Alaska to Mexico)	No; field collected
015	Mulina lateralis	Cultured organisms available	East and Gulf coasts of North America	Yes
107	Lytechinus pictus	6 months	West coast of North America; southern California to South America	No; field collected
112	Strongylocentrotus purpuratus	6 months	West coast of North America (Alaska to Mexico)	No; field collected
011	Crassostrea gigas	6 mo. (lab conditioning possible)	West coast of North America	Yes
081	Photobacterium phosphoreum	N/A; laboratory cultures	N/A; laboratory cultures	Yes
016	Mytilus edulis	6 months; cultured organisms available	West coast of North America	Yes
009	Crassostrea virginica	Limited	East and gulf coasts of North America	Yes
080	Photobacterium phosphoreum	N/A; laboratory cultures	N/A; laboratory cultures	Yes
010	Crassostrea gigas	6 mo. (lab conditioning possible)	West coast of North America	Yes
046	Strongylocentrotus purpuratus	6 months	West coast of North America (Alaska to Mexico)	No; field collected
042	Dendraster excentricus	6 months	West coast of North America	No; field collected
043	Lytechinus pictus	6 months	West coast of North America; southern California to South America	No; field collected
039	Dendraster excentricus	6 months	West coast of North America	No; field collected
014	Mercenaria mercenaria	Limited	East coast of North America	Yes
098	Arbacia punctulata	June - August	East coast of North America; Gulf of Mexico	No; field collected
097	Arbacia punctulata	June - August	East coast of North America; Gulf of Mexico	No; field collected
048	Strongylocentrotus droebachiensis		West coast of North America (Alaska to Mexico)	No; field collected
058	Hypomesus pretiosus		Southeastern Alaska to northern California	
038	Arbacia punctulata	June - August	East coast of North America; Gulf of Mexico	No; field collected
083	Photobacterium phosphoreum	N/A; laboratory cultures	N/A; laboratory cultures	Yes
084	Salmonella typhimurium	N/A; laboratory cultures	N/A; laboratory cultures	Yes
040	Dendraster excentricus	6 months	West coast of North America	No; field collected

TABLE D-10. (cont.)

		Availability		Ecological Relevance		
Test No.	Test Species	Protocol Development	Overall Availability	Field Validation	Species Importance	Overall EcoRelevance
001	Ampelisca abdita	ASTM (1990)	3		High; foundation species	3
121	Neanthes sp.	U.S. EPA (1990)	3			2
109	Strongylocentrotus purpuratus	EC (1992b)	3		High; keystone species; grazer on kelp	4
008	Rhepoxynius abronius	ASTM (1990)	3			2
005	Grandidierella japonica	ASTM (1990)	3			2
017	Mytilus edulis	ASTM (1989)	3		High; foundation species; harvested	4
111	Strongylocentrotus purpuratus		2		High; keystone species; grazer on kelp	4
047	Strongylocentrotus purpuratus	Yes	1		High; keystone species; grazer on kelp	4
102	Dendraster excentricus	EC (1992b)	3			2
100	Arbacia punctulata	EC (1992b)	3			2
115	Strongylocentrotus droebachiensis	EC (1992b)	3			2
015	Mulina lateralis	Yes	1			2
107	Lytechinus pictus	EC (1992b)	3			2
112	Strongylocentrotus purpuratus		2		High; keystone species; grazer on kelp	4
011	Crassostrea gigas	ASTM (1989)	2	Field validated (calibrated) to benthos (Becker et al. 1990)	High; sensitive bivalve lifestage; harvested	4
081	Photobacterium phosphoreum	Yes	4	Field validated (calibrated) to benthos (Gibsy et al. 1990; Rosiu et al. 1989; Williams et al. 1986).	Low	1
016	Mytilus edulis	U.S. EPA (1991b)	3		High; foundation species; harvested	4
009	Crassostrea virginica	ASTM (1989)	2		High; sensitive bivalve lifestage; harvested	4
080	Photobacterium phosphoreum		4	Field validated (calibrated) to benthos (Gibsy et al. 1990; Rosiu et al. 1989; Williams et al. 1986).	Low	1
010	Crassostrea gigas	U.S. EPA (1991b)	2	Field validated (calibrated) to benthos (Becker et al. 1990)	High; sensitive bivalve lifestage; harvested	4
046	Strongylocentrotus purpuratus	U.S. EPA (1991b)	2		High; keystone species; grazer on kelp	4
042	Dendraster excentricus	Yes	1			2
043	Lytechinus pictus		1			2
039	Dendraster excentricus		1			2
014	Merceraria mercenaria	ASTM (1989)	2		Harvested	3
098	Arbacia punctulata	Dinnel et al. (1983)	1			2
097	Arbacia punctulata	Dinnel et al. (1983)	1			2
048	Strongylocentrotus droebachiensis	U.S. EPA (1991b)	2			2
058	Hypomesus pretiosus		1			3
038	Arbacia punctulata	Dinnel and Stober (1985)	1			2
083	Photobacterium phosphoreum	EC (1992f)	2		Low	1
084	Salmonella typhimurium	Yes	2		Low	1
040	Dendraster excentricus	U.S. EPA (1991b)	1			2

TABLE D-10. (cont.)

Test No.	Test Species	Reliability: Accuracy	Reliability: Objectivity	Reliability: Endpoint	Reliability: Overall Reliability	Notes
001	Ampelisca abdita	L	H	Reburial (growth has also been tested/10 d)	1	Problem with inexperienced lab
121	Neanthes sp.	M	H	Growth	3	Inter/intra lab (Johns et al. 1991)
109	Strongylocentrotus purpuratus	M	H	Fertilization	3	Interlab (Anderson et al 1991)
008	Rhepoxynius abronius	L	H	Reburial	1	
005	Grandidierella japonica	L	H	Reburial	1	
017	Mytilus edulis	M	L	Abnormality	2	Inter/intra lab (Pastorok et al 1994)
111	Strongylocentrotus purpuratus	M	H	Fertilization	3	Inter/intra lab (Pastorok et al 1994)
047	Strongylocentrotus purpuratus	M	H	Fertilization	3	Inter/intra lab (Pastorok et al 1994)
102	Dendraster excentricus	M	H	Fertilization	3	Inter/intra lab (Anderson et al 1991; Pastorok et al 1994)
100	Arbacia punctulata	M	H	Fertilization	3	Interlab (Anderson et al 1991)
115	Strongylocentrotus droebachiensis	M	H	Fertilization	3	Interlab (Anderson et al 1991)
015	Mulina lateralis	M	H	Growth	2	
107	Lytechinus pictus	M	H	Fertilization	3	Interlab (Anderson et al 1991)
112	Strongylocentrotus purpuratus	M	H	Fertilization	2	
011	Crassostrea gigas	M	L	Abnormality	2	Inter/intra lab (Pastorok et al 1994)
081	Photobacterium phosphoreum	L	H	Luminescence	2	
016	Mytilus edulis	M	L	Abnormality	1	
009	Crassostrea virginica	M	L	Growth; abnormality	1	
080	Photobacterium phosphoreum	L	H	Luminescence	2	
010	Crassostrea gigas	M	L	Abnormality	1	
046	Strongylocentrotus purpuratus	M	L	Abnormality	1	
042	Dendraster excentricus	M	H	Fertilization	3	Inter/intra lab (Pastorok et al 1994)
043	Lytechinus pictus	M	H	Growth	2	
039	Dendraster excentricus	M	H	Growth	2	
014	Mercenaria mercenaria	M	L	Abnormality	1	
098	Arbacia punctulata	M	H	Fertilization	3	Interlab (U.S. EPA 1991c)
097	Arbacia punctulata	M	H	Fertilization	2	
048	Strongylocentrotus droebachiensis	M	L	Abnormality	1	
058	Hypomesus pretiosus	M	H	Growth	2	
038	Arbacia punctulata	M	H	Fertilization	2	
083	Photobacterium phosphoreum	L	H	Luminescence	2	
084	Salmonella typhimurium	M	H	Mutation	2	
040	Dendraster excentricus	M	L	Abnormality	1	

TABLE D-10. (cont.)

Test No.	Test Species	Interferences: Notes	Interferences: Overall Interferences
001	Ampelisca abdita	Inhabits fine sand-mud/silt; species tolerance range established; not fed (growth endpoint = fed)	3
121	Neanthes sp.		3
109	Strongylocentrotus purpuratus	Elutriate test (N/A); not fed	4
008	Rhepoxynius abronius	Prefers fine, sandy sediments; silts & clays may interfere; not fed	3
005	Grandidierella japonica	Lives in wide variety of sediment types; not fed	3
017	Mytilus edulis	Elutriate test (N/A); not fed	4
111	Strongylocentrotus purpuratus	Elutriate test (N/A)	4
047	Strongylocentrotus purpuratus	Elutriate test (N/A); not fed	4
102	Dendraster excentricus	Elutriate test (N/A); not fed	4
100	Arbacia punctulata	Elutriate test (N/A); not fed	4
115	Strongylocentrotus droebachiensis	Elutriate test (N/A); not fed	4
015	Mulina lateralis		4
107	Lytechinus pictus	Elutriate test (N/A); not fed	4
112	Strongylocentrotus purpuratus	Elutriate test (N/A)	4
011	Crassostrea gigas	Elutriate test (N/A); not fed	4
081	Photobacterium phosphoreum	Elutriate test (N/A);less sensitive to NH3 than Ceridiodaphia and Pimphales (Ankley et al. 1990); not fed	3
016	Mytilus edulis	Increased response in fine grained sediments (Chapman et al. 1987); not fed	0
009	Crassostrea virginica	Elutriate test (N/A); not fed	4
080	Photobacterium phosphoreum	Interstitial test (N/A); less sensitive to NH3 than Ceriodaphnia and Pimphales (Ankikey et al. 1990); not fed	3
010	Crassostrea gigas	Not fed	0
046	Strongylocentrotus purpuratus	Not fed	0
042	Dendraster excentricus	Elutriate test (N/A); not fed	4
043	Lytechinus pictus		4
039	Dendraster excentricus		4
014	Mercenaria mercenaria	Elutriate test (N/A); not fed	4
098	Arbacia punctulata	Elutriate test (N/A)	4
097	Arbacia punctulata	Interstitial test (N/A)	4
048	Strongylocentrotus droebachiensis	Not fed	0
058	Hypomesus pretiosus		0
038	Arbacia punctulata		0
083	Photobacterium phosphoreum	Less sensitive to NH3 than Ceriodaphnia and Pimephales (Ankley et al. 1990); not fed	1
084	Salmonella typhimurium	Extract test (N/A)	1
040	Dendraster excentricus	Not fed	0

TABLE D-10. (cont.)

Test No.	Chemical Discrimination: Sensitivity
001	Sensitive to wide range of anthropogenic materials (e.g., PAH, PCB, metals).
121	
109	Variable results obtained when comparing the results of several bioassay tests (metal and organic compounds). Ranged from 1-3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
008	Among the most sensitive of sediment toxicity test organisms.
005	Apparent insensitivity to some organic chemicals compared to other amphipods (Swartz et al. 1994).
017	Highly sensitive response of M. edulis to toxic sediment was reported in Chapman et al. (1987) and Long et al. (1990).
111	Variable results obtained when comparing the results of several bioassay tests (metal and organic compounds). Ranged from 1-3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
047	Variable results obtained in comparative bioassay tests (metal and organic compounds). Ranged from 1-3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
102	Variable results obtained when comparing the results of several bioassay tests (metal and organic compounds). Ranged from 1-3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
100	Variable results obtained when comparing the results of several bioassay tests (metal and organic compounds). Ranged from 1-3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
115	Variable results obtained when comparing the results of several bioassay tests (metal and organic compounds). Ranged from 1-3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
015	Sublethal growth endpoint increases sensitivity of test (Burgess and Morrison 1994).
107	Variable results obtained when comparing the results of several bioassay tests (metal and organic compounds). Ranged from 1-3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
112	Variable results obtained when comparing the results of several bioassay tests (metal and organic compounds). Ranged from 1-3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
011	
081	High sensitivity
016	Highly sensitive response of M. edulis to toxic sediment was reported in Chapman et al. (1987) and Long et al. (1990).
009	
080	High sensitivity
010	
046	Variable results obtained when comparing the results of several bioassay tests (metal and organic compounds). Ranged from 1-3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
042	Variable results in comparative bioassay tests (metal and organic compounds). Ranged from 1-3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
043	Variable results obtained when comparing the results of several bioassay tests (metal and organic compounds). Ranged from 1-3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
039	Variable results obtained when comparing the results of several bioassay tests (metal and organic compounds). Ranged from 1-3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
014	
098	Variable results obtained when comparing the results of several bioassay tests (metal and organic compounds). Ranged from 1-3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
097	Variable results obtained when comparing the results of several bioassay tests (metal and organic compounds). Ranged from 1-3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
048	Variable results obtained when comparing the results of several bioassay tests (metal and organic compounds). Ranged from 1-3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
058	
038	Variable results obtained when comparing the results of several bioassay tests (metal and organic compounds). Ranged from 1-3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).
083	High sensitivity
084	
040	Variable results obtained when comparing the results of several bioassay tests (metal and organic compounds). Ranged from 1-3 orders of magnitude more sensitive to 1 order of magnitude less sensitive (Nacci et al. 1986).

TABLE D-10. (cont.)

Test No.	Test Species	Chemical Discrimination: Overall Sensitivity	Regulatory Status
001	Ampelisca abdita	4	Low
121	Neanthes sp.	2	Low
109	Strongylocentrotus purpuratus	4	Medium
008	Rhepoxynius abronius	4	Low
005	Grandidierella japonica	4	Low
017	Mytilus edulis	4	Low
111	Strongylocentrotus purpuratus	4	Low
047	Strongylocentrotus purpuratus	4	Medium
102	Dendraster excentricus	4	Medium
100	Arbacia punctulata	4	Medium
115	Strongylocentrotus droebachiensis	4	Medium
015	Mulina lateralis	4	Low
107	Lytechinus pictus	4	Medium
112	Strongylocentrotus purpuratus	4	Low
011	Crassostrea gigas	4	Low
081	Photobacterium phosphoreum	3	Low
016	Mytilus edulis	4	Medium
009	Crassostrea virginica	4	Low
080	Photobacterium phosphoreum	3	Low
010	Crassostrea gigas	4	Medium
046	Strongylocentrotus purpuratus	4	Medium
042	Dendraster excentricus	4	Medium
043	Lytechinus pictus	0	Low
039	Dendraster excentricus	0	Low
014	Mercenaria mercenaria	4	Low
098	Arbacia punctulata	4	Low
097	Arbacia punctulata	4	Low
048	Strongylocentrotus droebachiensis	4	Medium
058	Hypomesus pretiosus	2	Low
038	Arbacia punctulata	4	Low
083	Photobacterium phosphoreum	3	Medium
084	Salmonella typhimurium	3	Low
040	Dendraster excentricus	2	Medium

TABLE D-10. (cont.)

Test No.	General Notes
001	Control survivability declines in coarse sand; euryhaline; feeding method causes exposure to overlying water (pore water enters tube); amphipods are more sensitive to contaminated sediments than other major taxa (Swartz 1987).
121	
109	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
008	Sensitive to salinities <25 g/kg (limits its use to test marine sediments); amphipods are more sensitive to contaminated sediments than other major taxa (Swartz 1987).
005	Broad application because it's possible to conduct tests in sands, silts, or clays; amphipods are more sensitive to contaminated sediments than other major taxa (Swartz 1987).
017	Sensitivity of M. edulis and R. abronius tests appears to be similar (Chapman et al. 1987; Long et al. 1990; Williams et al. 1986); abnormal development if >30 embryos/mL.
111	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
047	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
102	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
100	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
115	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
015	
107	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
112	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
011	Not too difficult to isolate and obtain large numbers of embryos from individual male-female pairs.
081	*Burton and Ingersoll (1994) reported that Microtox had equal sensitivity to Oncorhyncus mykiss, Pimephales promelas, Lepomis macrochirus, Cyprinidon variegatus*, and *Daphnia magna*.
016	Sensitivity of M. edulis and R. abronius tests appears to be similar (Chapman et al. 1987; Long et al. 1990; Williams et al. 1986); abnormal development if >30 embryos/mL.
009	Can tolerate DO of 0.5 mg/L; however, growth is reduced at DO <4.2 mg/L; abnormal development if >30 embryos/mL.
080	Burton and Ingersoll (1994) reported that Microtox had equal sensitivity to Oncorhyncus mykiss, Pimephales promelas, Lepomis macrochirus, Cyprinidon variegatus, and Daphnia magna.
010	Not too difficult to isolate and obtain large numbers of embryos from individual male-female pairs.
046	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
042	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
043	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
039	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
014	Abnormal development if >30 embryos/mL.
098	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
097	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
048	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
058	
038	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.
083	Burton and Ingersoll (1994) reported that Microtox had equal sensitivity to Oncorhyncus mykiss, Pimephales promelas, Lepomis macrochirus, Cyprinidon variegatus, and Daphnia magna.
084	
040	Echinoids have many similarities to chordate animals including basic pattern of embryonic development.

TABLE D-10. (cont.)

Test No.	Test Species	References
001	Ampelisca abdita	ASTM (1990) (E1367-90)
121	Neanthes sp.	Dillon et al. (1993); U.S. EPA (1990); Johns et al. (1992)
109	Strongylocentrotus purpuratus	Environment Canada (1992b)
008	Rhepoxynius abronius	ASTM (1990) (E1367-90)
005	Grandidierella japonica	ASTM (1990) (E1367-90)
017	Mytilus edulis	ASTM (1989) (E724-89)
111	Strongylocentrotus purpuratus	Long et al. (1990)
047	Strongylocentrotus purpuratus	U.S. EPA and U.S. COE (1993); Dinnel et al. (1982)
102	Dendraster excentricus	Environment Canada (1992b)
100	Arbacia punctulata	Environment Canada (1992b)
115	Strongylocentrotus droebachiensis	Environment Canada (1992b)
015	Mulina lateralis	Burgess et al. (1992)
107	Lytechinus pictus	Environment Canada (1992b)
112	Strongylocentrotus purpuratus	Long et al. (1990)
011	Crassostrea gigas	ASTM (1989) (E724-89)
081	Photobacterium phosphoreum	Microbics (1992)
016	Mytilus edulis	U.S. EPA (1991b)
009	Crassostrea virginica	ASTM (1989) (E724-89)
080	Photobacterium phosphoreum	Tay et al. (1992)
010	Crassostrea gigas	U.S. EPA (1991b)
046	Strongylocentrotus purpuratus	U.S. EPA (1991b)
042	Dendraster excentricus	U.S. EPA and U.S. COE (1993); Dinnel et al. (1982)
043	Lytechinus pictus	Thompson et al. (1989)
039	Dendraster excentricus	Casillas et al. (1992a)
014	Mercenaria mercenaria	ASTM (1989) (E724-89)
098	Arbacia punctulata	Burgess et al. (1993)
097	Arbacia punctulata	Burgess et al. (1993)
048	Strongylocentrotus droebachiensis	U.S. EPA (1991b)
058	Hypomesus pretiosus	Casillas et al. (1992b)
038	Arbacia punctulata	Lamberson et al. (1992), following Dinnel and Stober (1985), U.S. EPA (1991b)
083	Photobacterium phosphoreum	Environment Canada (1992f)
084	Salmonella typhimurium	Jarvis and Reilly (1992)
040	Dendraster excentricus	U.S. EPA (1991b)

Notes

EL – elutriate
EX – extract
INT – interstitial water
S – sediment

N/A – not applicable
N/S – not specified

EC – Environment Canada (1990, 1992)
GB – U.S. EPA and U.S. COE (1991)
IN – U.S. EPA and U.S. COE (1993)
TT – U.S. EPA (1992)

TABLE D-11. BASIS FOR SEDIMENT TOXICITY TEST EVALUATION – ESTUARINE LETHAL

Test No.	Site	Exposure Media	Biotic Group	Common Name	Life Stage	Recommended	Test Species
001	E/L	S	Amphipod	Amphipod	Juv or adult females	GB,TT,IN	Ampelisca abdita
129	E/L	S	Amphipod	Amphipod	Large juv and adults (3–5 mm)	GB,TT,IN, EC	Eohaustorius estuarius
003	E/L	S	Amphipod	Amphipod	Juv or young adults	EC	Eohaustorius washingtonianus
133	E/L	S	Amphipod	Amphipod	Juveniles (2–4 mm)		Leptocheirus plumulosus
135	E/L	S	Amphipod	Amphipod	Mixed sexes (4–8 mm)	TT,IN	Leptocheirus plumulosus
126	E/L	S	Amphipod	Amphipod	Juv or young adults (4–10 mm)	EC	Corophium volutator
002	E/L	S	Amphipod	Amphipod	Juv or young adults	EC	Amphiporeia virginiana
128	E/L	S	Amphipod	Amphipod	Adults		Corophium volutator
134	E/L	S	Amphipod	Amphipod	Juveniles		Leptocheirus plumulosus
131	E/L	S	Amphipod	Amphipod	7–14 days old	TT,IN	Hyalella azteca
020	E/L	S	Bivalve	Littleneck clam	Juveniles		Protothaca staminea
017	E/L	EL	Bivalve	Blue mussel	Embryos	GB,IN	Mytilus edulis
027	E/L	S	Crustacean	Blue crab	Juveniles		Callinectes sapidus
151	E/L	S	Crustacean	Grass shrimp	Post-hatch (1–4 days)		Palaemonetes sp.
016	E/L	S	Bivalve	Blue mussel	Embryos		Mytilus edulis
009	E/L	EL	Bivalve	Eastern oyster	Embryos	GB,IN	Crassostrea virginica
011	E/L	EL	Bivalve	Pacific oyster	Embryos	IN	Crassostrea gigas
030	E/L	S	Crustacean	Sand shrimp	Juveniles		Crangon sp.
153	E/L	EL	Fish	Sheepshead minnow	1–14 days old		Cyprinodon variegatus
010	E/L	S	Bivalve	Pacific oyster	Embryos		Crassostrea gigas
157	E/L	EL	Fish	Silverside	9–14 days old	IN	Menidia sp.
158	E/L	EL	Fish	Silverside	1–14 days old	GB,IN	Menidia sp.
014	E/L	EL	Bivalve	Quahog clam	Embryos		Mercenaria mercenaria
026	E/L	EL	Crustacean	Blue crab	Embryo-larval		Callinectes sapidus
065	E/L	EL	Mysid	Mysid	1–5 days old	GB	Mysidopsis sp.
162	E/L	EL	Mysid	Mysid	1–5 days old	GB	Neomysis sp.
161	E/L	S	Mysid	Mysid	1–5 days old		Neomysis sp.
056	E/L	EL	Fish	3-spine stickleback	Juveniles (0.1–3.0 g)	EC	Gasterosteus aculeatus
064	E/L	S	Mysid	Mysid	1–5 days old		Mysidopsis sp.
156	E/L	EL	Fish	Grunion	9–14 days old		Leuresthes tenuis
165	E/L	INT	Polychaete	Polychaete	Females (1–2 days)		Dinophilus gyrociliatus

TABLE D-11. (cont.)

Test No.	Test Species	Salinity (ppt)	Duration	Exposure Relevance: Media	Habitat Group	Ecorating	Notes
001	Ampelisca abdita	28-35	10 days	S	I	4	Particle-feeding, tube dweller
129	Eohaustorius estuarius	2-<28	10 days	S	I	4	
003	Eohaustorius washingtonianus	12-33	10 days	S	I	4	Free-burrowing, sand dweller
133	Leptocheirus plumulosus	5-25	10 days	S	I	4	
135	Leptocheirus plumulosus	2-30	10 days	S	I	4	
126	Corophium volutator	>2 to 28	10 days	S	I	3	
002	Amphiporeia virginiana	20-30	10 days	S	I	4	Lives in the sediment
128	Corophium volutator		10 days	S	I	3	
134	Leptocheirus plumulosus	6.9-7.2	10 days	S	I	4	
131	Hyalella azteca	0-15	10 days	S	E	3	
020	Protothaca staminea	23.5-32.7	10 days	S	I	4	
017	Mytilus edulis	18-32	48 hrs	EL	P	0	
027	Callinectes sapidus		10 days	S	E	3	
151	Palaemonetes sp.	2-<28	10 days	S	E	3	
016	Mytilus edulis	28	48-60 hrs	S	P	1	
009	Crassostrea virginica	18-32	48 hrs	EL	P	0	
011	Crassostrea gigas	18-32	48 hrs	EL	P	0	
030	Crangon sp.		10 days	S	E	3	
153	Cyprinodon variegatus	5-30 +/- 10%	96 hrs	EL	N	0	
010	Crassostrea gigas	28	48-60 hrs	S	P	1	
157	Menidia sp.	5-32 +/- 10%	96 hrs	EL	N	0	
158	Menidia sp.	5-32 +/- 10%	96 hrs	EL	N	0	
014	Mercenaria mercenaria	18-32	48 hrs	EL	P	0	
026	Callinectes sapidus		48 hrs	EL	P	0	
065	Mysidopsis sp.	25-30 +/- 10%	96 hrs	EL	P	0	
162	Neomysis sp.	20-30 +/- 10%	96 hrs	EL	P	0	
161	Neomysis sp.	20-30 +/- 10%	10 days	S	P	1	
056	Gasterosteus aculeatus	10-20	96 hrs	EL	N	0	
064	Mysidopsis sp.	25-30 +/- 10%	10 days	S	P	1	
156	Leuresthes tenuis	20-32 +/- 10%	96 hrs	EL	N	0	
165	Dinophilus gyrociliatus	25 +/- 1	7 days	INT	E	1	

TABLE D-11. (cont.)

Test No.	Test Species	Availability: Seasonality	Availability: Geographic Coverage	Availability: Cultured
001	Ampelisca abdita	Year-round (juveniles sometimes difficult to obtain)	East coast of North America; Gulf of Mexico; San Francisco Bay	No; field collected
129	Eohaustorius estuarius	Year-round	Central British Columbia to central California	No; field collected
003	Eohaustorius washingtonianus	Year-round	Southeastern Alaska to Oregon	No; field collected
133	Leptocheirus plumulosus	Year-round; cultured organisms available	East coast of North America	Yes; also field collected
135	Leptocheirus plumulosus	Year-round; cultured organisms available	East coast of North America	Yes; also field collected
126	Corophium volutator	Densities increase from June to September; low from January to May	Northeast coast of North America	
002	Amphiporeia virginiana	Females with eggs present April-July	Eastern Nova Scotia to North Carolina	No; field collected
128	Corophium volutator	Densities increase from June to September; low from January to May	Northeast coast of North America	
134	Leptocheirus plumulosus	Year-round; cultured organisms available	East coast of North America	Yes; also field collected
131	Hyalella azteca	Year-round	Widely distributed throughout North America	Yes; also field collected
020	Protothaca staminea		Aleutian Islands to Baja California	
017	Mytilus edulis	6 months; cultured organisms available	East and west coasts of North America	Yes
027	Callinectes sapidus		East coast of North America; Gulf of Mexico	
151	Palaemonetes sp.		Isolated occurances, species specific; east coast of North America; Gulf of Mexico	
016	Mytilus edulis	6 months; cultured organisms available	East and west coasts of North America	Yes
009	Crassostrea virginica	Limited	East and gulf coasts of North America	Yes
011	Crassostrea gigas	6 months (lab conditioning possible)	West coast of North America	Yes
030	Crangon sp.		East and west coasts of North America	
153	Cyprinodon variegatus	Cultured organisms available	East coast of North America	Yes
010	Crassostrea gigas	6 months (lab conditioning possible)	West coast of North America	Yes
157	Menidia sp.	Year-round	East coast of North America; Gulf of Mexico; Great Lakes	Yes
158	Menidia sp.	Year-round	East coast of North America; Gulf of Mexico; Great Lakes	Yes
014	Mercenaria mercenaria	Limited	East coast of North America	Yes
026	Callinectes sapidus		East coast of North America; Gulf of Mexico	
065	Mysidopsis sp.	Species-specific	East coast of North America; Gulf of Mexico	
162	Neomysis sp.		East coast of North America	
161	Neomysis sp.		East coast of North America	
056	Gasterosteus aculeatus	Cultured organisms available	West and east coasts of North America	Yes; also field collected
064	Mysidopsis sp.	Species-specific	East coast of North America; Gulf of Mexico	
156	Leuresthes tenuis	Limited	Southern California	
165	Dinophilus gyrociliatus			

TABLE D-11. (cont.)

Test No.	Availability: Protocol Development	Availability: Overall Availability	Ecological Relevance: Field Validation	Ecological Relevance: Species Importance	Ecological Relevance: Overall EcoRelevance
001	ASTM (1990)	3		High; foundation species	3
129	ASTM (1990)	3	Field validated (calibrated) to benthos (Swartz et al. 1994)	Free-burrowing, sand dweller; imp. prey to shorebirds and fishes	2
003	EC (1992a)	3		Free-burrowing, sand dweller; imp. prey to shorebirds and fishes	2
133	ASTM (1990)	3		Burrow-building	2
135	Modified	3		Burrow-building	2
126	EC (1992a)	3		Species forms a major food component of migrating seabirds	2
002	EC (1992a)	3			2
128		3		Species forms a major food component of migrating seabirds	2
134	Under development	3		Burrow-building	2
131	Yes	2		Detritovore that will burrow in the sediment surface; digests bacteria and algae from ingested sediment particles.	2
020		2		Harvested	3
017	ASTM (1989)	3		High; foundation species; harvested	4
027	Yes	1		Harvested	3
151	Yes	1		High	2
016	U.S. EPA (1991b)	3		High; foundation species; harvested	4
009	ASTM (1989)	2		High; sensitive bivalve lifestage; harvested	4
011	ASTM (1989)	2	Field validated (calibrated) to benthos (Becker et al. 1990)	High; sensitive bivalve lifestage; harvested	4
030	Yes	1			2
153	Yes	4			2
010	U.S. EPA (1991b)	2	Field validated (calibrated) to benthos (Becker et al. 1990)	High; sensitive bivalve lifestage; harvested	4
157	Yes	3			2
158	Yes	3			2
014	ASTM (1989)	2		Harvested	3
026	Yes	1		Harvested	3
065	Yes	1			2
162	Yes	2			2
161	Yes	2			2
056	EC (1990c)	3			2
064	Yes	1			2
156	Adapted	1			3
165		1			1

TABLE D-11. (cont.)

Test No.	Test Species	Reliability: Accuracy	Reliability: Objectivity	Reliability: Endpoint	Reliability: Overall Reliability	Notes
001	Ampelisca abdita	H	H	Survival	3	Interlab (U.S. EPA 1992); problem with inexperienced laboratory
129	Eohaustorius estuarius	H	H	Survival	4	
003	Eohaustorius washingtonianus	H	H	Survival	4	Interlab (Paine and McPherson 1991a,b)
133	Leptocheirus plumulosus	H	H	Survival	4	Interlab (DeWitt et al. 1992)
135	Leptocheirus plumulosus	H	H	Survival	3	
126	Corophium volutator	H	H	Survival	4	Interlab (Paine and McPherson 1991a)
002	Amphiporeia virginiana	H	H	Survival	4	Interlab (Paine and McPherson 1991a)
128	Corophium volutator	H	H	Survival	4	Interlab (Paine and McPherson 1991a)
134	Leptocheirus plumulosus	H	H	Survival	3	
131	Hyalella azteca	H	H	Survival	3	
020	Protothaca staminea	H	H	Survival	3	
017	Mytilus edulis	H	H	Survival	2	
027	Callinectes sapidus	H	H	Survival	3	
151	Palaemonetes sp.	H	H	Survival	3	
016	Mytilus edulis	H	H	Survival	2	
009	Crassostrea virginica	H	H	Survival	2	
011	Crassostrea gigas	H	H	Survival	2	
030	Crangon sp.	H	H	Survival	3	
153	Cyprinodon variegatus	H	H	Survival	3	
010	Crassostrea gigas	H	H	Survival	2	
157	Menidia sp.	H	H	Survival	4	Interlab (U.S. EPA 1991c)
158	Menidia sp.	H	H	Survival	4	Interlab (U.S. EPA 1991c)
014	Mercenaria mercenaria	H	H	Survival	2	
026	Callinectes sapidus	H	H	Survival	3	
065	Mysidopsis sp.	H	H	Survival	4	Interlab (U.S. EPA 1991c)
162	Neomysis sp.	H	H	Survival	3	
161	Neomysis sp.	H	H	Survival	3	
056	Gasterosteus aculeatus	H	H	Survival	3	
064	Mysidopsis sp.	H	H	Survival	3	
156	Leuresthes tenuis	H	H	Survival	3	
165	Dinophilus gyrociliatus	H	H	Survival	3	

TABLE D–11. (cont.)

Test No.	Test Species	Interferences: Notes	Overall Interferences
001	Ampelisca abdita	Inhabits fine sand–mud/silt; species tolerance range established; not fed	3
129	Eohaustorius estuarius	Prefers sand; not fed	3
003	Eohaustorius washingtonianus	Insufficient data	3
133	Leptocheirus plumulosus	Lives in medium to fine sand; wide tolerance (silt to sand) (McGee et al. 1993); fed	2
135	Leptocheirus plumulosus	Lives in medium to fine sand; wide tolerance (silt to sand) (McGee et al. 1993); not fed	3
126	Corophium volutator	Little effect of grain size or organic content on survival rates; 37% silt or clay, mud or muddy sand; not found in sulfide mud with excessive organic detritus	3
002	Amphiporeia virginiana	Not influenced markedly by sediment particle size.	3
128	Corophium volutator	Little effect of grain size or organic content on survival rates; 37% silt or clay, mud or muddy sand; not found in sulfide mud with excessive organic detritus	3
134	Leptocheirus plumulosus	Lives in medium to fine sand; wide tolerance (silt to sand) (McGee et al. 1993)	3
131	Hyalella azteca	Wide tolerance of sediment grain–size; >90% silt/clay to 100% sand did not reduce survival or growth.	4
020	Protothaca staminea		3
017	Mytilus edulis	Elutriate test (N/A); not fed	4
027	Callinectes sapidus		4
151	Palaemonetes sp.	Not fed	4
016	Mytilus edulis	Increased response in fine grained sediments (Chapman et al. 1987); not fed	0
009	Crassostrea virginica	Elutriate test (N/A); not fed	4
011	Crassostrea gigas	Elutriate test (N/A); not fed	4
030	Crangon sp.		4
153	Cyprinodon variegatus	Elutriate test (N/A)	4
010	Crassostrea gigas	Not fed; potential effects of fine sediments (PTI 1994)	0
157	Menidia sp.	Elutriate test (N/A); fed at 48h	3
158	Menidia sp.	Elutriate test (N/A); fed at 48h	3
014	Mercenaria mercenaria	Elutriate test (N/A); not fed	4
026	Callinectes sapidus	Elutriate test (N/A)	4
065	Mysidopsis sp.	Elutriate test (N/A); fed daily	3
162	Neomysis sp.	Elutriate test (N/A); fed daily	3
161	Neomysis sp.	Fed daily	0
056	Gasterosteus aculeatus	Elutriate test (N/A)	4
064	Mysidopsis sp.	Fed daily	0
156	Leuresthes tenuis	Elutriate test (N/A); fed at 48h	3
165	Dinophilus gyrociliatus	Interstitial test (N/A)	3

TABLE D-11. (cont.)

Test No.	Chemical Discrimination Sensitivity
001	Sensitive to wide range of anthropogenic materials (e.g., PAH, PCB, metals).
129	E. estuarius less sensitive to fines than R. abronius (DeWitt et al. 1989).
003	E. washingtonianus is the same or slightly more sensitive than Rhepoxynius; more difficult to remove from test sediment than R. abronius
133	
135	
126	High (>90%) survival rates have been noted for sediments with up to 102 g/kg total volatile residue (Paine and McPherson 1991b).
002	
128	High (>90%) survival rates have been noted for sediments with up to 102 g/kg total volatile residue (Paine and McPherson 1991b).
134	
131	
020	
017	Highly sensitive response of M. edulis to toxic sediment was reported in Chapman et al. (1987) and Long et al. (1990).
027	
151	
016	Highly sensitive response of M. edulis to toxic sediment was reported in Chapman et al. (1987) and Long et al. (1990).
009	
011	
030	
153	
010	
157	
158	
014	
026	
065	
162	
161	
056	
064	
156	
165	

TABLE D–11. (cont.)

Test No.	Test Species	Chemical Discrimination: Overall Sensitivity	Regulatory Status
001	Ampelisca abdita	4	High
129	Eohaustorius estuarius	4	High
003	Eohaustorius washingtonianus	4	Medium
133	Leptocheirus plumulosus	4	Low
135	Leptocheirus plumulosus	4	High
126	Corophium volutator	4	Medium
002	Amphiporeia virginiana	2	Medium
128	Corophium volutator	4	Low
134	Leptocheirus plumulosus	4	Low
131	Hyalella azteca	4	High
020	Protothaca staminea	0	Low
017	Mytilus edulis	4	Medium
027	Callinectes sapidus	2	Low
151	Palaemonetes sp.	4	Low
016	Mytilus edulis	4	Medium
009	Crassostrea virginica	4	Medium
011	Crassostrea gigas	4	Medium
030	Crangon sp.	2	Low
153	Cyprinodon variegatus	1	Low
010	Crassostrea gigas	4	Medium
157	Menidia sp.	1	Medium
158	Menidia sp.	1	Medium
014	Mercenaria mercenaria	4	Low
026	Callinectes sapidus	4	Low
065	Mysidopsis sp.	4	Medium
162	Neomysis sp.	4	Medium
161	Neomysis sp.	4	Low
056	Gasterosteus aculeatus	0	Medium
064	Mysidopsis sp.	4	Low
156	Leuresthes tenuis	1	Low
165	Dinophilus gyrociliatus	2	Low

TABLE D-11. (cont.)

Test No.	General Notes
001	Control survivability declines in coarse sand; euryhaline, amphipods are more sensitive to contaminated sediments than other major taxa (Swartz 1987).
129	High level of availability, easy to handle, tolerant to a wide range of salinities; amphipods are more sensitive to contaminated sediments than other major taxa (Swartz 1987).
003	High level of availability, easy to handle, tolerant to a wide range of salinities; amphipods are more sensitive to contaminated sediments than other major taxa (Swartz 1987); most common haustoriid species in British Columbia.
133	The concentration-sensitivity of L. plumulosus varied with the bioavailability of the toxicant.
135	The concentration-sensitivity of L. plumulosus varied with the bioavailability of the toxicant.
126	This species is generally less sensitive to contaminated sediments than R. abronius; animals selective deposit feeders, feeding on diatoms, microalgae, and bacteria associated with sediment particles; found over a wide range of salinities.
002	Tolerant to wide range of salinities, however, reduced survival if salinity <20 ppt (Doe and Wade 1991); similar sensitivity to R. abronius.
128	This species is generally less sensitive to contaminated sediments than R. abronius; animals selective deposit feeders, feeding on diatoms, microalgae, and bacteria associated with sediment particles; found over a wide range of salinities.
134	The concentration-sensitivity of L. plumulosus varied with the bioavailability of the toxicant.
131	Dominant role in many aquatic ecosystems; process organic matter (detritus) and are a primary food source for benthic feeding fish (Pennak 1978, 1989).
020	
017	
027	
151	
016	Sensitivity of M. edulis and R. abronius tests appears to be similar (Chapman et al. 1987; Long et al. 1990; Williams et al. 1986); abnormal development if >30 embryos/mL.
009	Can tolerate DO of 0.5 mg/L; however, growth is reduced at DO <4.2 mg/L; abnormal development if >30 embryos/mL.
011	Not too difficult to isolate and obtain large numbers of embryos from individual male-female pairs.
030	
153	Widely used as estuarine toxicity and physiological test organism; has been proposed for use in dredged material assessments.
010	Not too difficult to isolate and obtain large numbers of embryos from individual male-female pairs.
157	
158	
014	Abnormal development if >30 embryos/mL.
026	
065	
162	
161	
056	Well documented life history; easily captured; species is euryhaline; size suitable for acute toxicity tests; recommended by EC and EPA.
064	
156	
165	

TABLE D-11. (cont.)

Test No.	Test Species	References
001	Ampelisca abdita	ASTM (1990) (E1367-90); U.S. EPA (1991b); U.S. EPA and U.S. COE (1991, 1993)
129	Eohaustorius estuarius	ASTM (1990) (E1367-90); U.S. EPA and U.S. COE (1993); Environment Canada (1992a)
003	Eohaustorius washingtonianus	Environment Canada (1992a)
133	Leptocheirus plumulosus	DeWitt et al. (1992)
135	Leptocheirus plumulosus	Schlekat et al. (1992); U.S. EPA and U.S. COE (1993)
126	Corophium volutator	Environment Canada (1992a), based on ASTM (1990) (E1367-90); Swartz et al. (1985)
002	Amphiporeia virginiana	Environment Canada (1992a)
128	Corophium volutator	Tay et al. (1992), based on Swartz et al. (1985); U.S. EPA and U.S. COE (1993); ASTM (1990) (E1367-90)
134	Leptocheirus plumulosus	McGee et al. (1993)
131	Hyalella azteca	ASTM (1991b) (E1383-90); U.S. EPA and U.S. COE (1993)
020	Protothaca staminea	Swartz et al. (1979); U.S. EPA and U.S. COE (1991)
017	Mytilus edulis	ASTM (1989) (E724-89); U.S. EPA and U.S. COE (1991, 1993)
027	Callinectes sapidus	U.S. EPA and U.S. COE (1991)
151	Palaemonetes sp.	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
016	Mytilus edulis	U.S. EPA (1991b)
009	Crassostrea virginica	ASTM (1989) (E724-89); U.S. EPA and U.S. COE (1991, 1993)
011	Crassostrea gigas	ASTM (1989) (E724-89); U.S. EPA and U.S. COE (1993)
030	Crangon sp.	U.S. EPA and U.S. COE (1991)
153	Cyprinodon variegatus	U.S. EPA and U.S. COE (1993); U.S. EPA (1991a)
010	Crassostrea gigas	U.S. EPA (1991b)
157	Menidia sp.	U.S. EPA and U.S. COE (1993); U.S. EPA (1991a)
158	Menidia sp.	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
014	Mercenaria mercenaria	ASTM (1989) (E724-89)
026	Callinectes sapidus	U.S. EPA and U.S. COE (1991)
065	Mysidopsis sp.	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
162	Neomysis sp.	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
161	Neomysis sp.	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
056	Gasterosteus aculeatus	Environment Canada (1990c)
064	Mysidopsis sp.	U.S. EPA and U.S. COE (1991, 1993); U.S. EPA (1991a)
156	Leuresthes tenuis	U.S. EPA and U.S. COE (1993); U.S. EPA (1991a)
165	Dinophilus gyrociliatus	Long et al. (1990); Carr et al. (1989)

Notes

EL – elutriate
EX – extract
INT – interstitial water
S – sediment

N/A – not applicable
N/S – not specified

EC – Environment Canada (1990, 1992)
GB – U.S. EPA and U.S. COE (1991)
IN – U.S. EPA and U.S. COE (1993)
TT – U.S. EPA (1992)

TABLE D-12. BASIS FOR SEDIMENT TOXICITY TEST EVALUATION – ESTUARINE SUBLETHAL

Test No.	Site	Exposure Media	Biotic Group	Common Name	Life Stage	Recommended	Test Species
001	E/S	S	Amphipod	Amphipod	Juv or adult females		Ampelisca abdita
134	E/S	S	Amphipod	Amphipod	Juveniles		Leptocheirus plumulosus
135	E/S	S	Amphipod	Amphipod	Mixed sexes (4–8 mm)		Leptocheirus plumulosus
129	E/S	S	Amphipod	Amphipod	Large juv and adults (3–5 mm)		Eohaustorius estuarius
128	E/S	S	Amphipod	Amphipod	Adults		Corophium volutator
133	E/S	S	Amphipod	Amphipod	Juveniles (2–4 mm)		Leptocheirus plumulosus
017	E/S	EL	Bivalve	Blue mussel	Embryos		Mytilus edulis
011	E/S	EL	Bivalve	Pacific oyster	Embryos		Crassostrea gigas
010	E/S	S	Bivalve	Pacific oyster	Embryos		Crassostrea gigas
016	E/S	S	Bivalve	Blue mussel	Embryos		Mytilus edulis
014	E/S	EL	Bivalve	Quahog clam	Embryos		Mercenaria mercenaria
009	E/S	EL	Bivalve	Eastern oyster	Embryos		Crassostrea virginica
187	E/S	EL	Fish	Sheepshead minnow	Adults		Cyprinodon variegatus
176	E/S	EX	Bacterium	Microtox	Cells		Photobacterium phosphoreum
191	E/S	INT	Polychaete	Polychaete	Females (1–2 days)		Dinophilus gyrociliatus
175	E/S	S	Bacterium	Microtox	Cells		Photobacterium phosphoreum

TABLE D-12. (cont.)

Test No.	Test Species	Salinity (ppt)	Duration	Exposure Relevance: Media	Habitat Group	Ecorating	Notes
001	Ampelisca abdita	28-35	10 days	S	I	4	Particle-feeding, tube dweller
134	Leptocheirus plumulosus	6.3-8.0	30 days	S	I	4	
135	Leptocheirus plumulosus	2-30	28 days	S	I	4	
129	Eohaustorius estuarius	2-<28	10 days	S	I	4	
128	Corophium volutator		10 days	S	I	3	
133	Leptocheirus plumulosus	5-25	28 days	S	I	4	
017	Mytilus edulis	18-32	48 hrs	EL	P	0	
011	Crassostrea gigas	18-32	48 hrs	EL	P	0	
010	Crassostrea gigas	28	48-60 hrs	S	P	1	
016	Mytilus edulis	28	48-60 hrs	S	P	1	
014	Mercenaria mercenaria	18-32	48 hrs	EL	P	0	
009	Crassostrea virginica	18-32	48 hrs	EL	P	0	
187	Cyprinodon variegatus	30	96 hrs	EL	N	0	
176	Photobacterium phosphoreum	20	15 min	EX	P	0	
191	Dinophilus gyrociliatus	25 +/- 1	7 days	INT	E	1	
175	Photobacterium phosphoreum	20	30 min	S	P	0	

TABLE D-12. (cont.)

Test No.	Test Species	Availability: Seasonality	Availability: Geographic Coverage	Availability: Cultured
001	Ampelisca abdita	Year-round (juveniles sometimes difficult to obtain)	East coast of North America; Gulf of Mexico; San Francisco Bay	No; field collected
134	Leptocheirus plumulosus	Year-round; cultured organisms available	East coast of North America	Yes; also field collected
135	Leptocheirus plumulosus	Year-round; cultured organisms available	East coast of North America	Yes; also field collected
129	Eohaustorius estuarius	Year-round	Central British Columbia to central California	No; field collected
128	Corophium volutator	Densities increase from June to September; low from January to May	Northeast coast of North America	
133	Leptocheirus plumulosus	Year-round; cultured organisms available	East coast of North America	Yes; also field collected
017	Mytilus edulis	6 months; cultured organisms available	West coast of North America	Yes
011	Crassostrea gigas	6 months (lab conditioning possible)	West coast of North America	Yes
010	Crassostrea gigas	6 months (lab conditioning possible)	West coast of North America	Yes
016	Mytilus edulis	6 months; cultured organisms available	West coast of North America	Yes
014	Mercenaria mercenaria	Limited	East coast of North America	Yes
009	Crassostrea virginica	Limited	East and gulf coasts of North America	Yes
187	Cyprinodon variegatus	Cultured organisms available	East coast of North America	Yes; also field collected
176	Photobacterium phosphoreum	N/A; laboratory cultures	N/A; laboratory cultures	Yes
191	Dinophilus gyrociliatus			
175	Photobacterium phosphoreum	N/A; laboratory cultures	N/A; laboratory cultures	Yes

TABLE D–12. (cont.)

Test No.	Availability: Protocol Development	Availability: Overall Availability	Ecological Relevance: Field Validation	Ecological Relevance: Species Importance	Ecological Relevance: Overall EcoRelevance
001	ASTM (1990)	3		High; foundation species	3
134	Under development	3		Burrow–building	2
135	Modified	3		Burrow–building	2
129	ASTM (1990)	3	Field validated (calibrated) to benthos (Swartz et al. 1994)	Free–burrowing, sand dweller, Imp. prey to shorebirds and fishes	2
128		3		Species forms a major food component of migrating seabirds	2
133	ASTM (1990)	3		Burrow–building	2
017	ASTM (1989)	3		High; foundation species; harvested	4
011	ASTM (1989)	2	Field validated (calibrated) to benthos (Becker et al. 1990)	High; sensitive bivalve lifestage; harvested	3
010	U.S. EPA (1991b)	2	Field validated (calibrated) to benthos (Becker et al. 1990)	High; sensitive bivalve lifestage; harvested	4
016	U.S. EPA (1991b)	3		High; foundation species; harvested	4
014	ASTM (1989)	2		Harvested	4
009	ASTM (1989)	2		High; sensitive bivalve lifestage; harvested	4
187	Incomplete ref.	4			2
176	Modified	4	Field validated (calibrated) to benthos (Giesy et al. 1990; Rosiu et al. 1989; Williams et al. 1986).	Low	2
191	Yes	1			1
175	Yes	2		Low	1

TABLE D–12. (cont.)

Test No.	Test Species	Reliability: Accuracy	Reliability: Objectivity	Reliability: Endpoint	Reliability: Overall Reliability	Notes
001	Ampelisca abdita	L	H	Reburial and 10–day growth	2	Problem with inexperienced lab
134	Leptocheirus plumulosus	M	H	Growth and reproduction	2	
135	Leptocheirus plumulosus	M	H	Growth and reproduction	2	
129	Eohaustorius estuarius	L	H	Reburial	2	
128	Corophium volutator	L	H	Reburial	3	Interlab (Paine and McPherson 1991a)
133	Leptocheirus plumulosus	M	H	Growth and fertility	2	
017	Mytilus edulis	M	L	Abnormality	2	Inter/intra lab (Pastorok et al. 1994)
011	Crassostrea gigas	M	L	Abnormality	2	Inter/intra lab (Pastorok et al. 1994)
010	Crassostrea gigas	M	L	Abnormality	2	
016	Mytilus edulis	M	L	Abnormality	1	
014	Mercenaria mercenaria	M	L	Abnormality	1	
009	Crassostrea virginica	M	L	Growth; abnormality	1	
187	Cyprinodon variegatus	L	M	Respiration	1	
176	Photobacterium phosphoreum	L	H	Luminescence	2	
191	Dinophilus gyrociliatus	M	H	Fecundity	2	
175	Photobacterium phosphoreum	L	H	Luminescence	2	

TABLE D-12. (cont.)

Test No.	Test Species	Interferences: Notes	Interferences: Overall Interferences
001	Ampelisca abdita	Inhabits fine sand-mud/silt; Species tolerance range established: not fed	3
134	Leptocheirus plumulosus	Lives in medium to fine sand; wide tolerance (silt to sand) (McGee et al. 1993); not fed	3
135	Leptocheirus plumulosus	Lives in medium to fine sand; wide tolerance (silt to sand) (McGee et al. 1993)	3
129	Eohaustorius estuarius	Prefers sand; not fed	3
128	Corophium volutator	Little effect of grain size or organic content on survival rates; 37% silt or clay, mud or muddy sand; not found in sulfide mud with excessive organic detritus	3
133	*Leptocheirus plumulosus*	*Lives in medium to fine sand; wide tolerance (silt to sand) (McGee et al. 1993); fed*	2
017	Mytilus edulis	Elutriate test (N/A); not fed	4
011	Crassostrea gigas	Elutriate test (N/A); not fed	4
010	Crassostrea gigas	Not fed	0
016	Mytilus edulis	Increased response in fine grained sediments (Chapman et al. 1987); not fed	0
014	Mercenaria mercenaria	Elutriate test (N/A); not fed	4
009	Crassostrea virginica	Elutriate test (N/A); not fed	4
187	Cyprinodon variegatus	Elutriate test (N/A)	4
176	Photobacterium phosphoreum	Extract test (N/A); less sensitive to NH3 than Ceriodaphnia and Pimephales (Ankley et al. 1990); not fed	1
191	Dinophilus gyrociliatus	Interstitial test (N/A)	3
175	Photobacterium phosphoreum	Less sensitive to NH3 than Ceriodaphnia and Pimephales (Ankley et al. 1990); not fed	0

TABLE D-12. (cont.)

Test No.	Chemical Discrimination Sensitivity
001	Sensitive to wide range of anthropogenic materials (e.g., PAH, PCB, metals).
134	
135	
129	E. estuaris less sensitive to fines than R. abronius (DeWitt et al. 1989).
128	High (>90%) survival rates have been noted for sediments with up to 102 g/kg total volatile residue (Paine and McPherson 1991b).
133	
017	Highly sensitive response of M. edulis to toxic sediment was reported in Chapman et al. (1987) and Long et al. (1990).
011	
010	
016	Highly sensitive response of M. edulis to toxic sediment was reported in Chapman et al. (1987) and Long et al. (1990).
014	
009	
187	
176	High sensitivity
191	
175	High sensitivity; more sensitive to hydrophobic chemicals than elutriate Microtox.

TABLE D-12. (cont.)

Test No.	Test Species	Chemical Discrimination: Overall Sensitivity	Regulatory Status
001	Ampelisca abdita	4	Low
134	Leptocheirus plumulosus	4	Low
135	Leptocheirus plumulosus	4	Low
129	Eohaustorius estuarius	4	Low
128	Corophium volutator	4	Low
133	Leptocheirus plumulosus	4	Low
017	Mytilus edulis	4	Low
011	Crassostrea gigas	4	Low
010	Crassostrea gigas	4	Medium
016	Mytilus edulis	4	Medium
014	Mercenaria mercenaria	4	Low
009	Crassostrea virginica	4	Low
187	Cyprinodon variegatus	4	Low
176	Photobacterium phosphoreum	3	Low
191	Dinophilus gyrociliatus	4	Low
175	Photobacterium phosphoreum	3	Low

TABLE D-12. (cont.)

Test No.	General Notes
001	Control survivability declines in coarse sand; euryhaline; amphipods are more sensitive to contaminated sediments than other major taxa (Swartz 1987).
134	The concentration-sensitivity of L. plumulosus varied with the bioavailability of the toxicant.
135	The concentration-sensitivity of L. plumulosus varied with the bioavailability of the toxicant.
129	High level of availability, easy to handle, tolerant to a wide range of salinities; amphipods are more sensitive to contaminated sediments than other major taxa (Swartz 1987).
128	This species is generally less sensitive to contaminated sediments than R. abronius; animals selective deposit feeders, feeding on diatoms, microalgae, and bacteria associated with sediment particles; found over a wide range of salinities.
133	The concentration-sensitivity of L. plumulosus varied with the bioavailability of the toxicant.
017	Sensitivity of M. edulis and R. abronius tests appears to be similar (Chapman et al. 1987; Long et al. 1990; Williams et al. 1986); abnormal development if >30 embryos/mL.
011	Not too difficult to isolate and obtain large numbers of embryos from individual male-female pairs.
010	Not too difficult to isolate and obtain large numbers of embryos from individual male-female pairs.
016	Sensitivity of M. edulis and R. abronius tests appears to be similar (Chapman et al. 1987; Long et al. 1990; Williams et al. 1986); abnormal development if >30 embryos/mL.
014	Abnormal development if >30 embryos/mL
009	Can tolerate DO of 0.5 mg/L; however, growth is reduced at DO <4.2 mg/L; abnormal development if >30 embryos/mL
187	Widely used as estuarine toxicity and physiological test organism; has been proposed for use in dredged material assessments.
176	Burton and Ingersoll (1994) reported that Microtox had equal sensitivity to Oncorhynchus mykiss, Pimephales promelas, Lepomis macrochirus, Cyprinidon variegatus, and Daphnia magna.
191	
175	Burton and Ingersoll (1994) reported that Microtox had equal sensitivity to Oncorhynchus mykiss, Pimephales promelas, Lepomis macrochirus, Cyprinidon variegatus, and Daphnia magna.

TABLE D-12. (cont.)

Test No.	Test Species	References
001	Ampelisca abdita	ASTM (1990) (E1367-90)
134	Leptocheirus plumulosus	McGee et al. (1993)
135	Leptocheirus plumulosus	Schlekat et al. (1992)
129	Eohaustorius estuarius	ASTM (1990) (E1367-90)
128	Corophium volutator	Tay et al. (1992)
133	Leptocheirus plumulosus	DeWitt et al. (1992)
017	Mytilus edulis	ASTM (1989) (E724-89)
011	Crassostrea gigas	ASTM (1989) (E724-89)
010	Crassostrea gigas	U.S. EPA (1991b)
016	Mytilus edulis	U.S. EPA (1991b)
014	Mercenaria mercenaria	ASTM (1989) (E724-89)
009	Crassostrea virginica	ASTM (1989) (E724-89)
187	Cyprinodon variegatus	Alden et al. (1988)
176	Photobacterium phosphoreum	Jacobs et al. (1992); Microbics (1992); U.S. EPA (1991b)
191	Dinophilus gyrociliatus	Carr et al. (1989); Long et al. (1990)
175	Photobacterium phosphoreum	Brouwer et al. (1990)

Notes: EL – elutriate
EX – extract
INT – interstitial water
S – sediment

N/A – not applicable
N/S – not specified

EC – Environment Canada (1990, 1992)
GB – U.S. EPA and U.S. COE (1991)
IN – U.S. EPA and U.S. COE (1993)
TT – U.S. EPA (1992)

TABLE D-13. BASIS FOR SEDIMENT TOXICITY TEST EVALUATION – FRESHWATER LETHAL

Test No.	Site	Exposure Media	Biotic Group	Common Name	Life Stage	Recommended	Test Species
241	F/L	S	Insect	Mayfly	Nymphs		Hexagenia limbata
239	F/L	S	Insect	Mayfly	Young nymphs		Hexagenia limbata
238	F/L	S	Insect	Midge	2nd instar larvae	TT,IN, G	Chironomus tentans
237	F/L	S	Insect	Midge	Neonates or larvae	IN	Chironomus riparius
200	F/L	S	Amphipod	Amphipod	Juveniles (2-3 mm)	TT,IN	Hyalella azteca
242	F/L	S	Insect	Mayfly	Nymphs		Hexagenia limbata
209	F/L	INT	Cladoceran	Water flea	Neonates	IN	Daphnia magna
256	F/L	S	Plant	Marsh grass	Seedlings		Echinochloa crusgalli
250	F/L	S	Oligochaete	Oligochaete	Large worms (3-4 mg)		Lumbriculus variegatus
249	F/L	S	Oligochaete	Oligochaete	Mixed-age		Lumbriculus variegatus
212	F/L	EL	Cladoceran	Water flea	Neonates (< 24 hrs)	EC	Daphnia magna
240	F/L	INT	Insect	Mayfly	Nymphs		Hexagenia limbata
205	F/L	EL	Cladoceran	Water flea	Neonates	IN	Ceriodaphnia dubia
213	F/L	EL	Cladoceran	Water flea	Neonates (< 24 hrs)	IN	Daphnia pulex
208	F/L	EL	Cladoceran	Water flea	Neonates	IN	Daphnia magna
214	F/L	EL	Cladoceran	Water flea	Neonates (< 24 hrs)	EC	Daphnia pulex
202	F/L	S	Bivalve	Paper pondshell clam	Juveniles (8-10 days)		Anodonta imbecillis
252	F/L	S	Oligochaete	Oligochaete	Mixed age; similar size		Pristina leidyi
255	F/L	S	Oligochaete	Oligochaete	Mixed age		Tubifex tubifex
222	F/L	EL	Fish	Bluegill	Approx. 4 days		Lepomis macrochirus
226	F/L	EL	Fish	Fathead minnow	Larvae (<24 hrs old)	EC	Pimephales promelas
234	F/L	EL	Fish	Rainbow trout	Fry or fingerlings (15-30 days)	IN, EC	Oncorhynchus mykiss (Salmo gairdneri)
235	F/L	S	Fish	Rainbow trout	Egg-sac stage		Oncorhynchus mykiss (Salmo gairdneri)
199	F/L	INT	Amphipod	Amphipod	Juveniles		Hyalella azteca
220	F/L	EL	Fish	Channel catfish	Approx. 4 days		Ictalurus punctatus
229	F/L	EL	Fish	Fathead minnow	Approx. 4 days	IN	Pimephales promelas
207	F/L	S	Cladoceran	Water flea	Neonates		Ceriodaphnia dubia
211	F/L	S	Cladoceran	Water flea	Neonates		Daphnia magna
251	F/L	EL	Oligochaete	Oligochaete	Similar size		Pristina leidyi
232	F/L	EX	Fish	Fathead minnow	Embryo-larval		Pimephales promelas
233	F/L	S	Fish	Fathead minnow	Juveniles (3-4 months)		Pimephales promelas
244	F/L	EL	Nematode	Nematode	L1 stage juveniles		Panagrellus redivivus
195	F/L	EX	Amphibian	African clawed frog	Embryos		Xenopus laevis

TABLE D-13. (cont)

Test No.	Test Species	Salinity (ppt)	Duration	Media	Habitat Group	Ecorating	Notes
				Exposure Relevance			
241	Hexagenia limbata	N/A	168 hrs	S	I	4	
239	Hexagenia limbata	N/A	10 days	S	I	4	
238	Chironomus tentans	N/A	10-14 days	S	I	4	
237	Chironomus riparius	N/A	10-30 days	S	I	4	
200	Hyalella azteca	N/A	10-14 days	S	E	3	
242	Hexagenia limbata	N/A	21 days	S	I	4	
209	Daphnia magna	N/A	48 hrs	INT	P	0	
256	Echinochloa crusgalli	N/A	2 weeks	S	N/A	4	
250	Lumbriculus variegatus	N/A	14 days	S	I	4	
249	Lumbriculus variegatus	N/A	10 days	S	I	4	
212	Daphnia magna	N/A	48 hrs	EL	P	0	
240	Hexagenia limbata	N/A	168 hrs	INT	I	2	
205	Ceriodaphnia dubia	N/A	48-96hrs	EL	P	0	
213	Daphnia pulex	N/A	96 hrs	EL	P	0	
208	Daphnia magna	N/A	48-96 hrs	EL	P	0	
214	Daphnia pulex	N/A	48 hrs	EL	P	0	
202	Anodonta imbecillis	N/A	10 days	S	I	4	
252	Pristina leidyi	N/A	10-18 days	S	E	3	
255	Tubifex tubifex	N/A	10 days	S	E	3	
222	Lepomis macrochirus	N/A	96 hrs	EL	P	0	
226	Pimephales promelas	N/A	7 days	EL	P	0	
234	Oncorhynchus mykiss (Salmo gairdneri)	N/A	96 hrs	EL	N	0	
235	Oncorhynchus mykiss (Salmo gairdneri)	N/A	21 days	S	P	1	
199	Hyalella azteca	N/A	96 hrs	INT	E	1	
220	Ictalurus punctatus	N/A	96 hrs	EL	P	0	
229	Pimephales promelas	N/A	96 hrs	EL	P	0	
207	Ceriodaphnia dubia	N/A	48 hrs	S	P	1	
211	Daphnia magna	N/A	48 hrs	S	P	1	
251	Pristina leidyi	N/A	48 hrs	EL	E	1	
232	Pimephales promelas	N/A	6 days	EX	P	0	
233	Pimephales promelas	N/A	21 days	S	N	1	
244	Panagrellus redivivus	N/A	96 hrs	EL	I	1	
195	Xenopus laevis	N/A	4 days	EX	P	0	

TABLE D-13. (cont.)

Test No.	Test Species	Availability: Seasonality	Availability: Geographic Coverage	Availability: Cultured
241	Hexagenia limbata	Year-round	Widely distributed throughout North America	No; field collected
239	Hexagenia limbata	Year-round	Widely distributed throughout North America	No; field collected
238	Chironomus tentans	Year-round	Mid-continental areas of North America	Yes
237	Chironomus riparius	Year-round	Worldwide	Yes
200	Hyalella azteca	Year-round	Widely distributed throughout North America	Yes; also field collected
242	Hexagenia limbata	Year-round	Widely distributed throughout North America	No; field collected
209	Daphnia magna	Year-round	Widely distributed throughout North America	Yes
256	Echinochloa crusgalli			
250	Lumbriculus variegatus	Year-round	Widely distributed throughout North America	Yes
249	Lumbriculus variegatus	Year-round	Widely distributed throughout North America	Yes
212	Daphnia magna	Year-round	Widely distributed throughout North America	Yes
240	Hexagenia limbata	Year-round	Widely distributed throughout North America	No; field collected
205	Ceriodaphnia dubia	Year-round	Temperate zone worldwide; littoral areas of lakes, ponds, and marshes	Yes
213	Daphnia pulex	Cultured organisms available	Pond dweller; widely distributed throughout North America (not Florida)	Yes
208	Daphnia magna	Year-round	Widely distributed throughout North America	Yes
214	Daphnia pulex	Cultured organisms available	Pond dweller; widely distributed throughout North America (not Florida)	Yes
202	Anodonta imbecillis		Genus is widely distributed throughout North America	
252	Pristina leidyi	Cultured		Yes
255	Tubifex tubifex	Cultured organisms available	Wide-spread	Yes
222	Lepomis macrochirus	Cultured	Ontario to Georgia; northeastern Mexico to Texas; western North America	Yes
226	Pimephales promelas	Year-round	Canada and eastern United States (not native west of Rocky Mountains)	Yes
234	Oncorhynchus mykiss (Salmo gairdneri)	Year-round	Western North America	Yes
235	Oncorhynchus mykiss (Salmo gairdneri)	Year-round	Western North America	Yes
199	Hyalella azteca	Year-round	Widely distributed throughout North America	Yes; also field collected
220	Ictalurus punctatus		Widely distributed throughout North America	Yes (pond culture)
229	Pimephales promelas	Year-round	Canada and eastern United States (not native west of Rocky Mountains)	Yes
207	Ceriodaphnia dubia	Year-round	Temperate zone worldwide; littoral areas of lakes, ponds, and marshes	Yes
211	Daphnia magna	Year-round	Widely distributed throughout North America	Yes
251	Pristina leidyi	Cultured		Yes
232	Pimephales promelas	Year-round	Canada and eastern United States (not native west of Rocky Mountains)	Yes
233	Pimephales promelas	Year-round	Canada and eastern United States (not native west of Rocky Mountains)	Yes
244	Panagrellus redivivus			
195	Xenopus laevis	Cultured organisms available	Not native to North America (alien)	Yes

TABLE D-13. (cont)

Test No.	Test Species	Availability: Protocol Development	Availability: Overall Availability	Ecological Relevance: Field Validation
241	Hexagenia limbata		3	Field validated (calibrated) to benthos (Giesy et al. 1990), but 10-day test did not predict chronic effects in field (Malueg et al. 1984a, b).
239	Hexagenia limbata	Yes	3	Field validated (calibrated) to benthos (Giesy et al. 1990), but 10-day test did not predict chronic effects in field (Malueg et al. 1984a, b).
238	Chironomus tentans	ASTM (1991b)	4	Field validated (calibrated) to benthos (Giesy et al. 1988; 1990)
237	Chironomus riparius	ASTM (1991b)	4	
200	Hyalella azteca	ASTM (1991b)	4	
242	Hexagenia limbata	Krantzberg (1990)	3	Field validated (calibrated) to benthos (Giesy et al. 1990).
209	Daphnia magna		4	Field validated (calibrated) to benthos (Giesy et al. 1988; 1990)
256	Echinochloa crusgalli	Yes	1	
250	Lumbriculus variegatus	Yes	3	
249	Lumbriculus variegatus	Adapted	3	
212	Daphnia magna	EC (1990a, d)	4	Field validated (calibrated) to benthos (Giesy et al. 1988; 1990)
240	Hexagenia limbata		3	Field validated (calibrated) to benthos (Giesy et al. 1990).
205	Ceriodaphnia dubia	U.S. EPA (1991b)	4	
213	Daphnia pulex	Yes	4	
208	Daphnia magna	Yes	4	Field validated (calibrated) to benthos (Giesy et al. 1988; 1990)
214	Daphnia pulex	EC (1990 a)	4	
202	Anodonta imbecillis	Yes	2	
252	Pristina leidyi	Yes	2	
255	Tubifex tubifex	Yes	2	Field validated (calibrated) to benthos (Wiederholm and Dave 1989).
222	Lepomis macrochirus	Yes	4	
226	Pimephales promelas	EC (1992 d)	4	
234	Oncorhynchus mykiss (Salmo gairdneri)	EC (1990 b, e)	4	
235	Oncorhynchus mykiss (Salmo gairdneri)	Yes	3	
199	Hyalella azteca	Not specified	3	
220	Ictalurus punctatus	Yes	4	
229	Pimephales promelas	Yes	4	
207	Ceriodaphnia dubia	U.S. EPA (1985)	3	
211	Daphnia magna	Yes	3	Field validated (calibrated) to benthos (Giesy et al. 1988; 1990)
251	Pristina leidyi	Yes	2	
232	Pimephales promelas	Modified	4	
233	Pimephales promelas	Krantzberg (1990)	3	
244	Panagrellus redivivus		2	
195	Xenopus laevis	Modified	3	

TABLE D-13. (cont.)

Test No.	Ecological Relevance: Species Importance	Overall EcoRelevance
241		3
239		3
238	Imp. in the diet of young and adult fish and surface feeding ducks; high, imp. in benthic food webs and nutrient cycling	2
237	Imp. in the diet of young and adult fish and surface feeding ducks; high, imp. in benthic food webs and nutrient cycling	2
200	Detritovore that will burrow in the sediment surface; digests bacteria and algae from ingested sediment particles.	2
242		2
209	Frequently in contact with the sediment surface; exposed to both water soluble contamirants in the overlying water and particulate bound contamirants in the sediment surface.	3
256	High; foundation species	4
250		2
249		2
212	Frequently in contact with the sediment surface; exposed to both water soluble contamirants in the overlying water and particulate bound contamirants in the sediment surface.	2
240		3
205	Fine mesh, filter feeders; major group converting phytoplankton and bacteria into animal protein (Carpenter et al. 1985); significant portion of the diet of numerous fish species (including salmonids).	2
213	Frequently in contact with the sediment surface; exposed to both water soluble contamirants in the overlying water and particulate bound contamirants in the sediment surface.	2
208	Frequently in contact with the sediment surface; exposed to both water soluble contamirants in the overlying water and particulate bound contamirants in the sediment surface.	2
214	Frequently in contact with the sediment surface; exposed to both water soluble contamirants in the overlying water and particulate bound contamirants in the sediment surface.	2
202		2
252		2
255		2
222		3
226		2
234	Harvested; eggs and larvae develop in close association with the sediment.	3
235	Harvested; eggs and larvae develop in close association with the sediment.	3
199	Detritovore that will burrow in the sediment surface; digests bacteria and algae from ingested sediment particles.	2
220		3
229		2
207	Fine mesh, filter feeders; major group converting phytoplankton and bacteria into animal protein (Carpenter et al. 1985); significant portion of the diet of numerous fish species (including salmonids).	2
211	Frequently in contact with the sediment surface; exposed to both water soluble contamirants in the overlying water and particulate bound contamirants in the sediment surface.	2
251		2
232		2
233		2
244	Terrestrial	1
195	Low; alien to United States	0

TABLE D-13. (cont.)

Test No.	Test Species	Reliability: Accuracy	Objectivity	Endpoint	Overall Reliability	Notes
241	Hexagenia limbata	H	H	Survival	3	
239	Hexagenia limbata	H	H	Survival	3	
238	Chironomus tentans	H	H	Survival	3	
237	Chironomus riparius	H	H	Survival	3	
200	Hyalella azteca	H	H	Survival	3	
242	Hexagenia limbata	H	H	Survival	3	
209	Daphnia magna	H	H	Survival	4	Inter/intra lab (Buikema 1983; Grothe and Kimerle 1985)
256	Echinochloa crusgalli	H	H	Survival	3	
250	Lumbriculus variegatus	H	H	Survival	3	
249	Lumbriculus variegatus	H	H	Survival	3	
212	Daphnia magna	H	H	Survival	4	Inter/intra lab (Kovacs and Ferguson 1990)
240	Hexagenia limbata	H	H	Survival	3	
205	Ceriodaphnia dubia	H	H	Survival	4	Inter/intra lab (DeGraeve et al. 1989; EA 1984)
213	Daphnia pulex	H	H	Survival	4	Inter/intra lab (Dorn 1984)
208	Daphnia magna	H	H	Survival	4	Inter/intra lab (Buikema 1983; Grothe and Kimberle 1985)
214	Daphnia pulex	H	H	Survival	3	
202	Anodonta imbecillis	H	H	Survival	3	
252	Pristina leidyi	H	H	Survival	3	
255	Tubifex tubifex	H	H	Survival	3	
222	Lepomis macrochirus	H	H	Survival	4	Inter/intra lab (Dorn 1984)
226	Pimephales promelas	H	H	Survival	4	Interlab (API 1988; Anderson and Norberg-King 1991; Rue et al. 1988)
234	Oncorhynchus mykiss (Salmo gairdneri)	H	H	Survival	3	
235	Oncorhynchus mykiss (Salmo gairdneri)	H	H	Survival	3	
199	Hyalella azteca	H	H	Survival	3	
220	Ictalurus punctatus	H	H	Survival	3	
229	Pimephales promelas	H	H	Survival	3	
207	Ceriodaphnia dubia	H	H	Survival	3	
211	Daphnia magna	H	H	Survival	3	
251	Pristina leidyi	H	H	Survival	3	
232	Pimephales promelas	H	H	Survival	3	
233	Pimephales promelas	H	H	Survival	3	
244	Panagrellus redivivus	H	H	Survival	3	
195	Xenopus laevis	H	H	Survival	3	

TABLE D-13. (cont.)

Test No.	Test Species	Interferences: Notes	Interferences: Overall Interferences
241	Hexagenia limbata	Cannibalism in large individuals (Dillon and Gibson 1990)	3
239	Hexagenia limbata	Cannibalism in large individuals (Dillon and Gibson 1990)	3
238	Chironomus tentans	Inhabit fine sediment and <0.15 mm to 2.0 mm; not found where H_2S >0.3 mg\L; fed	2
237	Chironomus riparius	Wide tolerance to grain-size; >90% silt/clay to 100% sand; fed	2
200	Hyalella azteca	Wide tolerance of sediment grain-size; >90% silt/clay to 100% sand did not reduce survival or growth; fed	3
242	Hexagenia limbata	Cannibalism in large individuals (Dillon and Gibson 1990)	3
209	Daphnia magna	Interstitial test (N/A); more sensitive to NH_3 than to Microtox (Ankley et al. 1990); not fed	4
256	Echinochloa crusgalli		4
250	Lumbriculus variegatus		3
249	Lumbriculus variegatus	Not fed	3
212	Daphnia magna	Elutriate test (N/A); more sensitive to NH_3 than Microtox (Ankley et al. 1990); not fed	4
240	Hexagenia limbata	Interstitial test (N/A); cannibalism in large individuals (Dillon and Gibson 1990)	2
205	Ceriodaphnia dubia	Elutriate test (N/A); more sensitive to NH_3 than Microtox (Ankley et al. 1990); fed at -2h and 48h	3
213	Daphnia pulex	Elutriate test (N/A); fed at 48h	3
208	Daphnia magna	Elutriate test (N/A); more sensitive to NH_3 than Microtox (Ankley et al. 1990); fed at 48h (if test extends to 96h)	3
214	Daphnia pulex	Elutriate test (N/A); not fed	4
202	Anodonta imbecillis	Not fed	3
252	Pristina leidyi		4
255	Tubifex tubifex	Not fed	4
222	Lepomis macrochirus	Elutriate test (N/A); not fed	4
226	Pimephales promelas	Elutriate test (N/A); more sensitive to NH_3 than Microtox (Ankley et al. 1990)	4
234	Oncorhynchus mykiss (Salmo gairdneri)	Elutriate test (N/A)	4
235	Oncorhynchus mykiss (Salmo gairdneri)		1
199	Hyalella azteca	Interstitial test (N/A)	3
220	Ictalurus punctatus	Elutriate test (N/A); not fed	4
229	Pimephales promelas	Elutriate test (N/A); more sensitive to NH_3 than Microtox (Ankley et al. 1990)	4
207	Ceriodaphnia dubia	Sediment may interfere with feeding; more sensitive to NH_3 than Microtox (Ankley et al. 1990); fed at -2h and at 48h	0
211	Daphnia magna	Sediment may interfere with feeding; more sensitive to NH_3 than Microtox (Ankley et al. 1990); not fed	0
251	Pristina leidyi	Elutriate test (N/A)	3
232	Pimephales promelas	Extract test (N/A); more sensitive to NH_3 than Microtox (Ankley et al. 1990)	2
233	Pimephales promelas	More sensitive to NH_3 than Microtox (Ankley et al. 1990)	2
244	Panagrellus redivivus	Elutriate test (N/A)	2
195	Xenopus laevis	Extract test (N/A)	2

TABLE D-13. (cont.)

Test No.	Chemical Discrimination Sensitivity
241	
239	
238	Similar sensitivity as Microtox (15-min, pore water) test (Giesy et al. 1988); good discrimination (more than D. magna survival)
237	More sensitive than C. tentans sediment test
200	
242	
209	Sensitive species; less sensitive than Microtox (15-min, pore water test) and C. tentans (10-d, sediment test) (Giesy et al. 1988).
256	
250	Species similar in sensitivity to Daphnia spp. for copper but less sensitive to mixed chemicals in harbor muds (Nebeker et al. 1984).
249	Species similar in sensitivity to Daphnia spp. for copper but less sensitive to mixed chemicals in harbor muds (Nebeker et al. 1984).
212	Sensitive species; less sensitive than Microtox (15-min, pore water test) and C. tentans (10-d, sediment test) (Giesy et al. 1988).
240	More sensitive test than D. magna (48-h, pore water) and C. tentans (10-d, pore water) (Giesy et al. 1990).
205	In elutriate tests, C. dubia was less sensitive than H. azteca; and exhibited the same sensitivity as P. promelas and L. varigatus; more sensitive than D. magna and D. pulex; high sensitivity in general.
213	Sensitive species
208	Sensitive species; less sensitive than Microtox (15-min, pore water test) and C. tentans (10-d, sediment test) (Giesy et al. 1988).
214	Sensitive species
202	
252	
255	
222	
226	
234	
235	
199	Relatively sensitive species; in pore water tests, H. azteca was more sensitive than C. dubia, P. promelas, and L. variegatus.
220	
229	
207	More sensitive than L. variegatus; more sensitive than D. magna and D. pulex; high sensitivity in general.
211	Sensitive species; less sensitive than Microtox (15-min, pore water test) and C. tentans (10-d, sediment test) (Giesy et al. 1988).
251	
232	
233	
244	
195	Teratogenicity endpoint more sensitive than similar test with P. promelas (Dawson et al. 1988).

TABLE D–13. (cont.)

Test No.	Test Species	Particular Chemicals	Chemical Discrimination: Overall Sensitivity	Regulatory Status
241	Hexagenia limbata		4	Low
239	Hexagenia limbata		4	Low
238	Chironomus tentans		4	High
237	Chironomus riparius		4	Medium
200	Hyalella azteca		4	High
242	Hexagenia limbata		4	Low
209	Daphnia magna	Metals	4	Low to Medium
256	Echinochloa crusgalli		2	Low
250	Lumbriculus variegatus		2	Low
249	Lumbriculus variegatus		2	Low
212	Daphnia magna	Metals	4	Medium
240	Hexagenia limbata		4	Low
205	Ceriodaphnia dubia		4	Medium
213	Daphnia pulex		4	Medium
208	Daphnia magna	Metals	4	Medium
214	Daphnia pulex		4	Medium
202	Anodonta imbecillis		1	Low
252	Pristina leidyi		2	Low
255	Tubifex tubifex		2	Low
222	Lepomis macrochirus		0	Low
226	Pimephales promelas		1	Medium
234	Oncorhynchus mykiss (Salmo gairdneri)		1	Medium
235	Oncorhynchus mykiss (Salmo gairdneri)		4	Low
199	Hyalella azteca		4	Low
220	Ictalurus punctatus		0	Low
229	Pimephales promelas		0	Medium
207	Ceriodaphnia dubia		4	Low
211	Daphnia magna	Metals	4	Low
251	Pristina leidyi		2	Low
232	Pimephales promelas		1	Low
233	Pimephales promelas		0	Low
244	Panagrellus redivivus		2	Low
195	Xenopus laevis		4	Low

TABLE D-13. (cont.)

Test No.	General Notes
241	Important component of fish and waterfowl diets; provide critical link between changing organic detritus into a readily available food source (Burton and Ingersoll 1994).
239	Important component of fish and waterfowl diets; provide critical link between changing organic detritus into a readily available food source (Burton and Ingersoll 1994).
238	Sensitive to a wide range of contaminants; has short generation time, is easily cultured in the lab; 10-day exposure optimal.
237	Sensitive to a wide range of contaminants; has short generation time, is easily cultured in the lab; 10-day exposure optimal.
200	Dominant role in many aquatic ecosystems; process organic matter (detritus) and are a primary food source for benthic feeding fish (Pennak 1978, 1989).
242	Important component of fish and waterfowl diets; provide critical link between changing organic detritus into a readily available food source (Burton and Ingersoll 1994).
209	Test response matches well with other tests (Giesy et al. 1990); major grp. in many zooplankton communities (Burton and Ingersoll 1994); cladocerans one of the more sensitive groups of organisms used in tox. tests (Mayer and Ellersieck 1986)
256	Rooted aquatic plants can be used to evaluate the overlying water, interstitial water, and the sediment (Burton and Ingersoll 1994).
250	Oligochaetes are relatively insensitive to many classes of contaminants (Giesy and Hoke 1989); play a major role in the processing of organic material and as a food source for benthic feeding fish (Burton and Ingersoll 1994).
249	Oligochaetes are relatively insensitive to many classes of contaminants (Giesy and Hoke 1989); play a major role in the processing of organic material and as a food source for benthic feeding fish (Burton and Ingersoll 1994).
212	Major group in many zooplankton communities (Burton and Ingersoll 1994); cladocerans are one of the more sensitive groups of organisms used in toxicity testing (Mayer and Ellersieck 1986).
240	Lab growth response predicted by Daphnia (48-h, pore water) lethal and C. tentans (10-d, sed/pore water) growth test; imp. part of fish/waterfowl diets; provide imp. link btwn changing detritus into food (Burton and Ingersoll 1994).
205	Unlike other daphnid species, C. dubia can be cultured in hard or soft water; cannot survive dissolved oxygen levels <5 mg/L; organisms are delicate and should be handled carefully during testing.
213	Major group in many zooplankton communities (Burton and Ingersoll 1994); cladocerans are one of the more sensitive groups of organisms used in toxicity testing (Mayer and Ellersieck 1986).
208	Major group in many zooplankton communities (Burton and Ingersoll 1994); cladocerans are one of the more sensitive groups of organisms used in toxicity testing (Mayer and Ellersieck 1986).
214	Major group in many zooplankton communities (Burton and Ingersoll 1994); cladocerans are one of the more sensitive groups of organisms used in toxicity testing (Mayer and Ellersieck 1986).
202	
252	Oligochaetes are relatively insensitive to many classes of contaminants (Giesy and Hoke 1989); play a major role in the processing of organic material and as a food source for benthic feeding fish (Burton and Ingersoll 1994).
255	Oligochaetes traditionally have been regarded as relatively insensitive to many classes of contaminants (Giesy and Hoke 1989).
222	
226	Seven day test has shown an excellent correlation with ecological evaluations of polluted waters; toxic response between pore water and elutriate (relative to sediment test did not differ significantly; not cost effective for screening.
234	Toxicity tests have been done world-wide with this species; large database.
235	Toxicity tests have been done world-wide with this species; large database.
199	Dominant role in many aquatic ecosystems; process organic matter (detritus) and are a primary food source for benthic feeding fish (Pennak 1978, 1989).
220	
229	Toxic response between pore water and elutriate (relative to the bulk sediment test) did not differ significantly; not cost effective for screening.
207	Unlike other daphnid species, C. dubia can be cultured in hard or soft water; cannot survive dissolved oxygen levels <5 mg/L; organisms are delicate and should be handled carefully during testing.
211	Major group in many zooplankton communities (Burton and Ingersoll 1994); cladocerans are one of the more sensitive groups of organisms used in toxicity testing (Mayer and Ellersieck 1986).
251	Oligochaetes are relatively insensitive to many classes of contaminants (Giesy and Hoke 1989); play a major role in the processing of organic material and as a food source for benthic feeding fish (Burton and Ingersoll 1994).
232	Toxic response between pore water and elutriate (relative to the bulk sediment test) did not differ significantly; not cost effective for screening.
233	Fish feed on sediments during test (Burton 1991); not cost effective for screening.
244	
195	

TABLE D-13. (cont.)

Test No.	Test Species	References
241	Hexagenia limbata	Giesy et al. (1990)
239	Hexagenia limbata	Malueg et al. (1984a, b); Nebeker et al. (1984); U.S. EPA and U.S. COE (1993)
238	Chironomus tentans	ASTM (1991b) (E1383-90); U.S. EPA and U.S. COE (1993); Ingersoll and Nelson (1990); Nelson et al. (1990)
237	Chironomus riparius	ASTM (1991b) (E1383-90); U.S. EPA and U.S. COE (1993); Ingersoll and Nelson (1990); Nelson et al. (1990)
200	Hyalella azteca	ASTM (1991b) (E1383-90); U.S. EPA and U.S. COE (1993)
242	Hexagenia limbata	Krantzberg and Boyd (1992)
209	Daphnia magna	Giesy et al. (1988, 1990)
256	Echinochloa crusgalli	Walsh et al. (1991)
250	Lumbriculus variegatus	Dermott and Munawar (1992)
249	Lumbriculus variegatus	U.S. EPA and U.S. COE (1993); Ankley et al. (1992); Bailey and Lui (1980)
212	Daphnia magna	Environment Canada (1990a, d)
240	Hexagenia limbata	Giesy et al. (1990)
205	Ceriodaphnia dubia	Ankley et al. (1991); U.S. EPA and U.S. COE (1993); U.S. EPA (1991a); Environment Canada (1992e)
213	Daphnia pulex	U.S. EPA and U.S. COE (1993); U.S. EPA (1991a)
208	Daphnia magna	Nebeker et al. (1984); U.S. EPA and U.S. COE (1993); U.S. EPA (1991a)
214	Daphnia pulex	Environment Canada (1990a)
202	Anodonta imbecillis	U.S. EPA and U.S. COE (1993)
252	Pristina leidyi	Smith et al. (1991); U.S. EPA and U.S. COE (1993)
255	Tubifex tubifex	U.S. EPA and U.S. COE (1993); Reynoldson et al. (1991)
222	Lepomis macrochirus	U.S. EPA and U.S. COE (1993); U.S. EPA (1991a)
226	Pimephales promelas	Environment Canada (1992d)
234	Oncorhynchus mykiss (Salmo gairdneri)	Environment Canada (1990b, e); U.S. EPA and U.S. COE (1993); U.S. EPA (1991a)
235	Oncorhynchus mykiss (Salmo gairdneri)	Birge et al. (1984); Krantzberg (1989); Krantzberg and Boyd (1992)
199	Hyalella azteca	Ankley et al. (1991)
220	Ictalurus punctatus	U.S. EPA and U.S. COE (1993); U.S. EPA (1991a)
229	Pimephales promelas	U.S. EPA and U.S. COE (1993); U.S. EPA (1991a)
207	Ceriodaphnia dubia	Sassoon-Brickson and Burton (1991); Stemmer et al. (1990b)
211	Daphnia magna	Nebeker et al. (1984); Stemmer et al. (1990a, b)
251	Pristina leidyi	Smith et al. (1991)
232	Pimephales promelas	Dawson et al. (1988)
233	Pimephales promelas	Krantzberg and Boyd (1992)
244	Panagrellus redivivus	Burnett et al. (no date); Samoiloff et al. (1980)
195	Xenopus laevis	Dawson et al. (1988)

Notes: EL – elutriate
EX – extract
INT – interstitial water
S – sediment

N/A – not applicable
N/S – not specified

EC – Environment Canada (1990, 1992)
GB – U.S. EPA and U.S. COE (1991)
IN – U.S. EPA and U.S. COE (1993)
TT – U.S. EPA (1992)

TABLE D-14. BASIS FOR SEDIMENT TOXICITY TEST EVALUATION - FRESHWATER SUBLETHAL

Test No.	Site	Exposure Media	Biotic Group	Common Name	Life Stage	Recommended	Test Species
238	F/S	S	Insect	Midge	2nd instar larvae		Chironomus tentans
237	F/S	S	Insect	Midge	Neonates or larvae		Chironomus riparius
200	F/S	S	Amphipod	Amphipod	Juveniles (2-3 mm)	B	Hyalella azteca
242	F/S	S	Insect	Mayfly	Nymphs		Hexagenia limbata
333	F/S	S	Plant	Aq vascular plant	Apical shoots		Hydrilla verticillata
256	F/S	S	Plant	Marsh grass	Seedlings		Echinochloa crusgalli
269	F/S	S	Amphipod	Amphipod	Juveniles (<1 week)		Hyalella azteca
285	F/S	EL	Cladoceran	Water flea	Neonates	EC	Ceriodaphnia dubia
330	F/S	S	Oligochaete	Oligochaete	Adults		Tubifex tubifex
304	F/S	INT	*Insect*	*Midge*	*2nd instar larvae*	G	Chironomus tentans
250	F/S	S	Oligochaete	Oligochaete	Large worms (3-4 mg)		Lumbriculus variegatus
323	F/S	S	Oligochaete	Oligochaete	Juveniles and adults		Lumbriculus variegatus
324	F/S	S	Oligochaete	Oligochaete	15 mm in length		Lumbriculus variegatus
226	F/S	EL	Fish	Fathead minnow	Larvae (<24 hrs old)	EC	Pimephales promelas
260	F/S	EL	Algae	Green algae	Cells	EC	Selenastrum capricornutum
272	F/S	INT	Bacterium	Microtox	Cells	G	Photobacterium phosphoreum
235	F/S	S	Fish	Rainbow trout	Egg-sac stage		Oncorhynchus mykiss (Salmo gairdneri)
261	F/S	INT	Algae	Green algae	Cells		Selenastrum capricornutum
252	F/S	S	Oligochaete	Oligochaete	Mixed age; similar size		Pristina leidyi
176	F/S	EX	Bacterium	Microtox	Cells		Photobacterium phosphoreum
211	F/S	S	Cladoceran	Water flea	Juvenile (5 days)		Daphnia magna
232	F/S	EX	Fish	Fathead minnow	Embryo-larval	G (elutriate)	Pimephales promelas
233	F/S	S	Fish	Fathead minnow	Juveniles (3-4 months)		Pimephales promelas
263	F/S	EL	Algae	Algae	Cells		Chlorella vulgaris
302	F/S	S	Fish	Rainbow trout	Gonad cell (RTG-2)		Oncorhynchus mykiss (Salmo gairdneri)
244	F/S	EL	Nematode	Nematode	L1 stage juveniles		Panagrellus redivivus
262	F/S	S	Algae	Algae	Cells		Chlorella vulgaris
195	F/S	EX	Amphibian	African clawed frog	Embryos		Xenopus laevis
278	F/S	S	Bacterium	Mutatox	Cells		Vibrio fischeri
273	F/S	S	Bacterium	Microtox	Cells		Photobacterium phosphoreum
274	F/S	S	Bacterium	Microtox	Cells		Photobacterium phosphoreum
270	F/S	EX	Bacterium	Aquatic bacterium	Cells		Aeromonas hydrophila
320	F/S	EX	Nematode	Nematode	L2 stage juveniles		Panagrellus redivivus
279	F/S	EL	Bacterium	Mutatox	Cells		Vibrio fischeri
308	F/S	EL	Microbe	Microbial enzyme	N/A		Alkaline phosphatase (APA)
084	F/S	EX	Bacterium	Mod. Ames/HPTLC	Cells		Salmonella typhimurium

TABLE D-14. (cont.)

Test No.	Test Species	Salinity (ppt)	Duration	Media	Habitat Group	Ecorating	Notes
				Exposure Relevance			
238	Chironomus tentans	N/A	10-25 days	S	I	4	
237	Chironomus riparius	N/A	10-30 days	S	I	4	
200	Hyalella azteca	N/A	10-30 days	S	E	3	
242	Hexagenia limbata	N/A	21 days	S	I	4	
333	Hydrilla verticillata	N/A	14 days	S	N/A	4	
256	Echinochloa crusgalli	N/A	2 weeks	S	N/A	4	
269	Hyalella azteca	N/A	8-10 wks	S	E	3	
285	Ceriodaphnia dubia	N/A	7 ± 1 days	EL	P	0	
330	Tubifex tubifex	N/A	28 days	S	E	3	
304	Chironomus tentans	N/A	10 days	INT	I	2	
250	Lumbriculus variegatus	N/A	24,48 hrs	S	I	4	
323	Lumbriculus variegatus	N/A	10-28 days	S	I	4	
324	Lumbriculus variegatus	N/A	14 days	S	I	4	
226	Pimephales promelas	N/A	7 days	EL	P	0	
260	Selenastrum capricornutum	N/A	48 hrs	EL	P	0	
272	Photobacterium phosphoreum	N/A	30 min	INT	P	0	
235	Oncorhynchus mykiss (Salmo gairdneri)	N/A	21 days	S	P	1	
261	Selenastrum capricornutum	N/A	48 hrs	INT	P	0	
252	Pristina leidyi	N/A	18 days	S	E	3	
176	Photobacterium phosphoreum	N/A	15 min	EX	P	0	
211	Daphnia magna	N/A	7 days	S	P	1	
232	Pimephales promelas	N/A	6 days	EX	P	0	
233	Pimephales promelas	N/A	21 days	S	N	1	
263	Chlorella vulgaris	N/A	4-96 hrs	EL	P	0	
302	Oncorhynchus mykiss (Salmo gairdneri)	N/A	48 hrs	S	P	1	
244	Panagrellus redivivus	N/A	96 hrs	EL	I	1	
262	Chlorella vulgaris	N/A	4-96 hrs	S	P	1	
195	Xenopus laevis	N/A	4 days	EX	P	0	
278	Vibrio fischeri	N/A	16-24 hrs	S	N/A	1	
273	Photobacterium phosphoreum	N/A	20 min	S	P	1	
274	Photobacterium phosphoreum	N/A	30 min	S	P	1	
270	Aeromonas hydrophila	N/A	20 hrs	EX	P	0	
320	Panagrellus redivivus	N/A	96 hrs	EX	I	1	
279	Vibrio fischeri	N/A	16-24 hrs	EL	N/A	0	
308	Alkaline phosphatase (APA)	N/A	40-130 min	EL	N/A	1	
084	Salmonella typhimurium	N/A	N/S	EX	N/A	0	

TABLE D–14. (cont.)

Test No.	Test Species	Availability: Seasonality	Availability: Geographic Coverage	Availability: Cultured
238	Chironomus tentans	Year–round	Mid–continental areas of North America	Yes
237	Chironomus riparius	Year–round	Worldwide	Yes
200	Hyalella azteca	Year–round	Widely distributed throughout North America	Yes; also field collected
242	Hexagenia limbata	Year–round	Widely distributed throughout North America	No; field collected
333	Hydrilla verticillata			
256	Echinochloa crusgalli			
269	Hyalella azteca	Year–round	Widely distributed throughout North America	Yes; also field collected
285	Ceriodaphnia dubia	Year–round	Temperate zone worldwide; littoral areas of lakes, ponds, and marshes	Yes
330	Tubifex tubifex	Cultured organisms available	Wide–spread	Yes
304	Chironomus tentans	Year–round	Mid–continental areas of North America	Yes
250	Lumbriculus variegatus	Year–round	Widely distributed throughout North America	Yes
323	Lumbriculus variegatus	Year–round	Widely distributed throughout North America	Yes
324	Lumbriculus variegatus	Year–round	Widely distributed throughout North America	Yes
226	Pimephales promelas	Year–round	Canada and eastern United States (not native west of Rocky Mountains)	Yes
260	Selenastrum capricornutum	Cultured organisms available	Wide–spread	Yes
272	Photobacterium phosphoreum	N/A; laboratory cultures	N/A; laboratory cultures	Yes
235	Oncorhynchus mykiss (Salmo gairdneri)	Year–round	Western North America	Yes
261	Selenastrum capricornutum	Cultured organisms available	Wide–spread	Yes
252	Pristina leidyi	Cultured		Yes
176	Photobacterium phosphoreum	N/A; laboratory cultures	N/A; laboratory cultures	Yes
211	Daphnia magna	Year–round	Widely distributed throughout North America	Yes
232	Pimephales promelas	Year–round	Canada and eastern United States (not native west of Rocky Mountains)	Yes
233	Pimephales promelas	Year–round	Canada and eastern United States (not native west of Rocky Mountains)	Yes
263	Chlorella vulgaris	Cultured organisms available	Wide–spread	Yes
302	Oncorhynchus mykiss (Salmo gairdneri)	Year–round	Western North America	Yes
244	Panagrellus redivivus			
262	Chlorella vulgaris	Cultured organisms available	Wide–spread	Yes
195	Xenopus laevis	Cultured organisms available		Yes
278	Vibrio fischeri	Cultured organisms available		Yes
273	Photobacterium phosphoreum	N/A; laboratory cultures	N/A; laboratory cultures	Yes
274	Photobacterium phosphoreum	N/A; laboratory cultures	N/A; laboratory cultures	Yes
270	Aeromonas hydrophila			Yes
320	Panagrellus redivivus			
279	Vibrio fischeri	Cultured organisms available		Yes
308	Alkaline phosphatase (APA)			Yes
084	Salmonella typhimurium	N/A; laboratory cultures		Yes

TABLE D-14. (cont.)

Test No.	Test Species	Availability: Protocol Development	Availability: Overall Availability	Ecological Relevance: Field Validation
238	Chironomus tentans	ASTM (1991b)	4	Field validated (calibrated) to benthos (Giesy et al. 1988; 1990)
237	Chironomus riparius	ASTM (1991b)	4	
200	Hyalella azteca	ASTM (1991b)	4	
242	Hexagenia limbata	Krantzberg (1990)	3	Field validated (calibrated) to benthos (Giesy et al. 1990).
333	Hydrilla verticillata	Yes	2	
256	Echinochloa crusgalli	Yes	2	
269	Hyalella azteca		3	
285	Ceriodaphnia dubia	EC (1992e)	4	
330	Tubifex tubifex	Yes	2	Field validated (calibrated) to benthos (Wiederholm and Dave 1989).
304	Chironomus tentans		3	Field validated (calibrated) to benthos (Giesy et al. 1988; 1990)
250	Lumbriculus variegatus	Yes	3	
323	Lumbriculus variegatus	Phipps and Ankley (1990)	1	
324	Lumbriculus variegatus	Yes	1	
226	Pimephales promelas	EC (1992d)	4	
260	Selenastrum capricornutum	EC (1992c)	4	
272	Photobacterium phosphoreum	Yes	4	Field validated (calibrated) to benthos (Giesy et al. 1990; Rosiu et al. 1989; Williams et al. 1986).
235	Oncorhynchus mykiss (Salmo gairdneri)	Yes	3	
261	Selenastrum capricornutum	Yes	4	
252	Pristina leidyi	Yes	2	
176	Photobacterium phosphoreum	Modified	4	Field validated (calibrated) to benthos (Giesy et al. 1990; Rosiu et al. 1989; Williams et al. 1986).
211	Daphnia magna	Yes	3	Field validated (calibrated) to benthos (Giesy et al. 1988; 1990)
232	Pimephales promelas	Modified	4	
233	Pimephales promelas	Krantzberg (1990)	3	
263	Chlorella vulgaris		2	
302	Oncorhynchus mykiss (Salmo gairdneri)	U.S EPA (1991b)	1	
244	Panagrellus redivivus		2	
262	Chlorella vulgaris		2	
195	Xenopus laevis	Modified	3	
278	Vibrio fischeri	Yes	2	
273	Photobacterium phosphoreum	Yes	2	
274	Photobacterium phosphoreum	Yes	2	
270	Aeromonas hydrophila		2	
320	Panagrellus redivivus	Samoiloff et al. (1980)	1	
279	Vibrio fischeri	Yes	2	
308	Alkaline phosphatase (APA)		1	
084	Salmonella typhimurium	Yes	2	

TABLE D-14. (cont.)

Test No.	Ecological Relevance: Species Importance	Overall EcoRelevance
238	Imp. in the diet of young and adult fish and surface feeding ducks; high, imp. in benthic food webs and nutrient cycling	2
237	Imp. in the diet of young and adult fish and surface feeding ducks; high, imp. in benthic food webs and nutrient cycling	2
200	Detritovore that will burrow in the sediment surface; digests bacteria and algae from ingested sediment particles.	2
242		2
333	High; foundation species	3
256	High; foundation species	3
269	Detritovore that will burrow in the sediment surface; digests bacteria and algae from ingested sediment particles.	2
285	Fine mesh, filter feeders; major group converting phytoplankton and bacteria into animal protein (Carpenter et al. 1985); significant portion of the diet of numerous fish species (including salmonids).	2
330		2
304	Imp. in the diet of young and adult fish and surface feeding ducks; high, imp. in benthic food webs and nutrient cycling	3
250	Low (terrestrial; wetland species)	0
323		2
324		2
226		2
260		2
272	Low	2
235	Harvested; eggs and larvae develop in close association with the sediment.	3
261		2
252		2
176	Low	2
211	Frequently in contact with the sediment surface; exposed to both water soluble contaminants in the overlying water and particulate bound contaminants in the sediment surface.	2
232		2
233		2
263		2
302	Harvested; eggs and larvae develop in close association with the sediment.	3
244	Terrestrial	2
262		2
195	Low; non-native	0
278	Low	1
273	Low	1
274	Low	1
270	Low	1
320	Terrestrial	2
279	Low	1
308		1
084	Low	1

TABLE D–14. (cont.)

Test No.	Test Species	Accuracy	Objectivity	Endpoint	Overall Reliability	Notes
238	Chironomus tentans	M	H	Growth	2	
237	Chironomus riparius	M	H	Growth	2	
200	Hyalella azteca	M	H	Growth	2	
242	Hexagenia limbata	M	H	Growth	2	
333	Hydrilla verticillata	M	H	Growth	2	
256	Echinochloa crusgalli	M	H	Seed germination and growth	2	
269	Hyalella azteca	M	H	Growth	2	
285	Ceriodaphnia dubia	M	H	Reproduction (also mortality at end of first phase is measured)	4	Inter/intra lab (DeGraeve et al. 1989, 1992; U.S. EPA 1991c)
330	Tubifex tubifex	M	H	Reproduction	2	
304	Chironomus tentans	M	H	Growth	2	
250	Lumbriculus variegatus	M	H	Growth and reproduction	2	
323	Lumbriculus variegatus	M	H	Growth and reproduction	2	
324	Lumbriculus variegatus	M	H	Growth and reproduction	2	
226	Pimephales promelas	M	H	Growth	3	Inter/intra lab (API 1988; Anderson and Norberg–King 1991;
260	Selenastrum capricornutum	L	H	Growth inhibition	2	
272	Photobacterium phosphoreum	L	H	Luminescence	2	
235	Oncorhynchus mykiss (Salmo gairdneri)	M	H	Teratogenesis	2	
261	Selenastrum capricornutum	L	M	Cell growth or uptake of radio–labeled carbon	2	
252	Pristina leidyi	L	M	Population growth	1	
176	Photobacterium phosphoreum	L	H	Luminescence	2	
211	Daphnia magna	M	H	Biomass	2	
232	Pimephales promelas	M	H	Growth	2	
233	Pimephales promelas	M	H	Biomass	2	
263	Chlorella vulgaris	L	H	Carbon assimilation rates	2	
302	Oncorhynchus mykiss (Salmo gairdneri)	M	M	Anaphase aberration	2	
244	Panagrellus redivivus	M	H	Growth	2	
262	Chlorella vulgaris	L	H	Carbon assimilation rates	2	
195	Xenopus laevis	M	H	Growth	2	
278	Vibrio fischeri	M	H	Mutation	2	
273	Photobacterium phosphoreum	L	H	Luminescence	2	
274	Photobacterium phosphoreum	L	H	Luminescence	2	
270	Aeromonas hydrophila	L	H	Optical density	2	
320	Panagrellus redivivus	M	H	Growth	2	
279	Vibrio fischeri	M	H	Mutation	2	
308	Alkaline phosphatase (APA)	L	H	Production	2	
084	Salmonella typhimurium	M	H	Mutation	2	

TABLE D-14. (cont.)

Test No.	Test Species	Interferences: Notes	Interferences: Overall Interferences
238	Chironomus tentans	Inhabit fine sediment and <0.15 mm to 2.0 mm; not found where H2S >0.3 mg/L ; fed	2
237	Chironomus riparius	Wide tolerance to grain-size; >90% silt/clay to 100% sand; fed	2
200	Hyalella azteca	Wide tolerance of sediment grain-size; >90% silt/clay to 100% sand did not reduce survival or growth.; fed	3
242	Hexagenia limbata	Cannibalism in large individuals (Dillon and Gibson 1990)	3
333	Hydrilla verticillata		4
256	Echinochloa crusgalli		4
269	Hyalella azteca	Wide tolerance of sediment grain-size; >90% silt/clay to 100% sand did not reduce survival or growth.	4
285	Ceriodaphnia dubia	Elutriate test (N/A); more sensitive to NH3 than Microtox (Ankley et al 1990); fed daily	3
330	Tubifex tubifex		4
304	Chironomus tentans	Interstitial test (N/A); not found where H2S is >0.3 mg/L; fed	1
250	Lumbriculus variegatus		3
323	Lumbriculus variegatus		3
324	Lumbriculus variegatus		3
226	Pimephales promelas	Elutriate test (N/A); more sensitive to NH3 than Microtox (Ankley et al 1990)	4
260	Selenastrum capricornutum	Elutriate test (N/A)	4
272	Photobacterium phosphoreum	Interstitial test (N/A); less sensitive to NH3 than Ceriodaphnia and Pimaphales (Ankley et al. 1990); not fed	4
235	Oncorhynchus mykiss (Salmo gairdneri)	·	1
261	Selenastrum capricornutum	Interstial test (N/A)	4
252	Pristina leidyi		4
176	Photobacterium phosphoreum	Extract test (N/A); less sensitive to NH3 than Ceriodaphnia and Pimephales (Ankley et al. 1990); not fed	2
211	Daphnia magna	Sediment may interfere with feeding; more sensitive to NH3 than Microtrox (Ankley et al. 1990)	0
232	Pimephales promelas	More sensitive to NH3 than Microtox (Ankley et al. 1990)	2
233	Pimephales promelas	More sensitive to NH3 than Microtox (Ankley et al. 1990)	2
263	Chlorella vulgaris	Elutriate test (N/A)	4
302	Oncorhynchus mykiss (Salmo gairdneri)	Not fed; potential effects of sediment	0
244	Panagrellus redivivus	Elutriate test (N/A)	2
262	Chlorella vulgaris	Potential effects of sediment	0
195	Xenopus laevis	Extract test (N/A)	2
278	Vibrio fischeri	Potential effects of sediment	0
273	Photobacterium phosphoreum	Less sensitive to NH3 than Ceriodaphnia and Pimephales (Ankley et al. 1990); not fed; potential effects of sediment	0
274	Photobacterium phosphoreum	Less sensitive to NH3 than Ceriodaphnia and Pimephales (Ankley et al. 1990); not fed; potential effects of sediment	0
270	Aeromonas hydrophila	Extract test (N/A)	2
320	Panagrellus redivivus	Extract test (N/A)	0
279	Vibrio fischeri	Elutriate test (N/A)	0
308	Alkaline phosphatase (APA)		0
084	Salmonella typhimurium	Extract test (N/A)	0

TABLE D-14. (cont.)

Test No.	Chemical Discrimination Sensitivity
238	Similar sensitivity as Microtox (15-min, pore water) test (Giesy et al. 1988); good discrimination (more than D. magna survival).
237	More sensitive than C. tentans sediment test.
200	
242	
333	
256	
269	
285	More sensitive than Lumbriculus variegatus; more sensitive than D. magna and D. pulex; high sensitivity in general.
330	
304	Similar sensitivity as Microtox (15-min, pore water) test (Giesy et al. 1988); good discrimination (more than D. magna survival).
250	Species similar in sensitivity to Daphnia app. for copper but less sensitive to mixed chemicals in harbor muds (Nebeker et al. 1984).
323	Species similar in sensitivity to Daphnia app. for copper but less sensitive to mixed chemicals in harbor muds (Nebeker et al. 1984).
324	Species similar in sensitivity to Daphnia app. for copper but less sensitive to mixed chemicals in harbor muds (Nebeker et al. 1984).
226	
260	High sensitivity to metals and herbicides (Thomas et al. 1986).
272	Good discrimination among sites (Giesy et al. 1988; 1990); high sensitivity; responsive to PAHs (Giesy et al. 1990).
235	
261	High sensitivity to metals and herbicides (Thomas et al. 1986).
252	
176	Good discrimination among sites (Giesy et al. 1988; 1990); high sensitivity.
211	Sensitive species; less sensitive than Microtox (15-min, pore water test) and C. tentans (10-d, sediment test) (Giesy et al. 1988).
232	
233	
263	High sensitivity to metals and herbicides (Thomas et al. 1986).
302	Natural genotoxicity may occur due to decomposition of plants.
244	
262	High sensitivity to metals and herbicides (Thomas et al. 1986).
195	Teratogenicity endpoint more sensitive than similar test with P. promelas (Dawson et al. 1988).
278	
273	High sensitivity; more sensitive to hydrophobic chemicals than elutriate Microtox.
274	High sensitivity; more sensitive to hydrophobic chemicals than elutriate Microtox.
270	
320	
279	
308	
084	

TABLE D-14. (cont.)

Test No.	Test Species	Particular Chemicals	Chemical Discrimination: Overall Sensitivity	Regulatory Status
238	Chironomus tentans		4	Low
237	Chironomus riparius		4	Low
200	Hyalella azteca		4	Low to Medium
242	Hexagenia limbata		4	Low
333	Hydrilla verticillata		2	Low
256	Echinochloa crusgalli		4	Low
269	Hyalella azteca		4	Low
285	Ceriodaphnia dubia		4	Medium
330	Tubifex tubifex		4	Low
304	Chironomus tentans		4	Low to Medium
250	Lumbriculus variegatus		4	Low
323	Lumbriculus variegatus		4	Low
324	Lumbriculus variegatus		4	Low
226	Pimephales promelas		1	Medium
260	Selenastrum capricornutum		3	Medium
272	Photobacterium phosphoreum		3	Low to Medium
235	Oncorhynchus mykiss (Salmo gairdneri)		4	Low
261	Selenastrum capricornutum		3	Low
252	Pristina leidyi		2	Low
176	Photobacterium phosphoreum		3	Low
211	Daphnia magna	Metals	4	Low
232	Pimephales promelas		1	Low to Medium
233	Pimephales promelas		1	Low
263	Chlorella vulgaris		3	Low
302	Oncorhynchus mykiss (Salmo gairdneri)		4	Low
244	Panagrellus redivivus		2	Low
262	Chlorella vulgaris		3	Low
195	Xenopus laevis		4	Low
278	Vibrio fischeri		3	Low
273	Photobacterium phosphoreum		3	Low
274	Photobacterium phosphoreum		3	Low
270	Aeromonas hydrophila		3	Low
320	Panagrellus redivivus		2	Low
279	Vibrio fischeri		3	Low
308	Alkaline phosphatase (APA)		3	
084	Salmonella typhimurium		3	Low

TABLE D–14. (cont.)

Test No.	General Notes
238	Sensitive to a wide range of contaminants; has short generation time, is easily cultured in the lab.
237	Sensitive to a wide range of contaminants; has short generation time, is easily cultured in the lab; 10–day exposure optimal.
200	Tests >30 days can add potential reproductive capacity as another endpoint, with reproductive behavior, appearance of secondary sex characteristics, egg production, and number of young produced.
242	Important component of fish and waterfowl diets; provide critical link between changing organic detritus into a readily available food source (Burton and Ingersoll 1994).
333	Rooted aquatic plants can be used to evaluate the overlying water, interstitial water, and the sediment (Burton and Ingersoll 1994).
256	Rooted aquatic plants can be used to evaluate the overlying water, interstitial water, and the sediment (Burton and Ingersoll 1994). Test is not yet demonstrated to be reliable for sediment.
269	Tests >30 days can add potential reproductive capacity as another endpoint, with reproductive behavior, appearance of secondary sex characteristics, egg production, and number of young produced.
285	Unlike other daphnid species, C. dubia can be cultured in hard or soft water; cannot survive dissolved oxygen levels <5 mg/L; organisms are delicate and should be handled carefully during testing.
330	Oligochaetes traditionally have been regarded as relatively insensitive to many classes of contaminants (Giesy and Hoke 1989).
304	Sensitive to a wide range of contaminants; has short generation time, is easily cultured in the lab; 10–day exposure optimal.
250	Oligochaetes are relatively insensitive to many classes of contaminants (Giesy and Hoke 1989); play a major role in the processing of organic material and as a food source for benthic feeding fish (Burton and Ingersoll 1994).
323	Oligochaetes are relatively insensitive to many classes of contaminants (Giesy and Hoke 1989); play a major role in the processing of organic material and as a food source for benthic feeding fish (Burton and Ingersoll 1994).
324	Oligochaetes are relatively insensitive to many classes of contaminants (Giesy and Hoke 1989); play a major role in the processing of organic material and as a food source for benthic feeding fish (Burton and Ingersoll 1994).
226	Seven day test has shown an excellent correlation with ecological evaluations of polluted waters; toxic response between pore water and elutriate (relative to sediment test did not differ significantly; not cost effective for screening.
260	DeZwart and Sloof (1993) and LeBlanc (1984) reported that this growth test is more sensitive than other commonly used tests; further development needed correlation with Microtox (Giesy and Hoke 1990).
272	Burton and Ingersoll (1994) reported that Microtox had equal sensitivity to Oncorhynchus mykiss, Pimephales promelas, Lepomis macrochirus, Cyprinidon variegatus, and Daphnia magna.
235	Toxicity tests have been done world–wide with this species; large database.
261	DeZwart and Sloof (1993) and LeBlanc (1984) reported that this growth test is more sensitive than other commonly used tests; further development needed correlation with Microtox (Giesy and Hoke (1990).
252	Oligochaetes are relatively insensitive to many classes of contaminants (Giesy and Hoke 1989); play a major role in the processing of organic material and as a food source for benthic feeding fish (Burton in press).
176	Burton and Ingersoll (1994) reported that Microtox had equal sensitivity to Oncorhynchus mykiss, Pimephales promelas, Lepomis macrochirus, Cyprinidon variegatus, and Daphnia magna.
211	Major group in many zooplankton communities (Burton and Ingersoll 1994); cladocerans are one of the more sensitive groups of organisms used in toxicity testing (Mayer and Ellersieck 1986).
232	Toxic response between pore water and elutriate (relative to the bulk sediment test) did not differ significantly; not cost effective for screening.
233	Fish feed on sediments during test (Burton 1991)
263	Giesy and Hoke (1990) recommended further development.
302	Test depends on a chemical extraction procedure that is specific for neutral nonionic compounds
244	
262	Giesy and Hoke (1990) recommended further development.
195	
278	
273	Burton and Ingersoll (1994) reported that Microtox had equal sensitivity to Oncorhynchus mykiss, Pimephales promelas, Lepomis macrochirus, Cyprinidon variegatus, and Daphnia magna.
274	Burton and Ingersoll (1994) reported that Microtox had equal sensitivity to Oncorhynchus mykiss, Pimephales promelas, Lepomis macrochirus, Cyprinidon variegatus, and Daphnia magna.
270	
320	
279	
084	

TABLE D-14. (cont.)

Test No.	Test Species	References
238	Chironomus tentans	ASTM (1991b) (E1383-90); Giesy et al. (1990); Wentsel et al. (1977a)
237	Chironomus riparius	ASTM (1991b) (E1383-90)
200	Hyalella azteca	ASTM (1991b) (E1383-90)
242	Hexagenia limbata	Krantzberg and Boyd (1992)
333	Hydrilla verticillata	Klaine and Hinman (no date)
256	Echinochloa crusgalli	Walsh et al. (1991)
269	Hyalella azteca	Borgmann et al. (1989)
285	Ceriodaphnia dubia	Environment Canada (1992e)
330	Tubifex tubifex	Reynoldson et al. (1991)
304	Chironomus tentans	Giesy et al. (1990)
250	Lumbriculus variegatus	Dermott and Munawar (1992)
323	Lumbriculus variegatus	Phipps and Ankley (1990)
324	Lumbriculus variegatus	Dermott and Munawar (1992)
226	Pimephales promelas	Environment Canada (1992d)
260	Selenastrum capricornutum	Environment Canada (1992c)
272	Photobacterium phosphoreum	Hoke et al. (1992)
235	Oncorhynchus mykiss (Salmo gairdneri)	Birge et al. (1984); Krantzberg (1989); Krantzberg and Boyd (1992)
261	Selenastrum capricornutum	Burton et al. (1989)
252	Pristina leidyi	Smith et al. (1991)
176	Photobacterium phosphoreum	Jacobs et al. (1992)
211	Daphnia magna	Nebeker et al. (1984); see Ceriodaphnia dubia (Environment Canda 1992e)
232	Pimephales promelas	Dawson et al. (1988)
233	Pimephales promelas	Krantzberg and Boyd (1992)
263	Chlorella vulgaris	Munawar and Munawar (1987)
302	Oncorhynchus mykiss (Salmo gairdneri)	Chapman et al. (1985); U.S. EPA (1991b)
244	Panagrellus redivivus	Burnett et al. (no date); Samoiloff et al. (1980)
262	Chlorella vulgaris	Munawar and Munawar (1987)
195	Xenopus laevis	Dawson et al. (1988)
278	Vibrio fischeri	Microbics (1993)
273	Photobacterium phosphoreum	Microbics (1991)
274	Photobacterium phosphoreum	Brouwer et al. (1990)
270	Aeromonas hydrophila	Dutka and Kwan (1981); Flemming and Trevors (1989)
320	Panagrellus redivivus	Samoiloff et al. (1983)
279	Vibrio fischeri	Microbics (1993)
308	Alkaline phosphatase (APA)	Burton et al. (1989)
084	Salmonella typhimurium	Jarvis and Reilly (1992)

Notes

EL – elutriate
EX – extract
INT – interstitial water
S – sediment

N/A – not applicable
N/S – not specified

EC – Environment Canada (1990, 1992)
GB – U.S. EPA and U.S. COE (1991)
IN – U.S. EPA and U.S. COE (1993)
TT – U.S. EPA (1992)

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