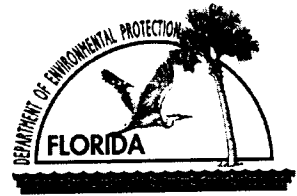


# Interoffice Memorandum



TO: Ms. Ligia Mora-Applegate  
THROUGH: Dr. William Coppenger *W.C.*  
FROM: David Whiting *DW*  
SUBJECT: Toxicity Test Methods  
DATE: June 24, 2004

Attached you will find a revised list of EPA and/or ASTM toxicity test methods for the various media which may need to be tested at a hazardous waste site. I have updated the list to include the most recent versions of EPA methods. In addition, the recommended tests (those tests in bold) have been updated to include the chronic tests when available. Given that the toxicity testing associated with a hazardous waste site does not typically get repeated over and over again, it makes sense to try to obtain as much information as possible in the initial toxicity tests. The chronic tests, while more expensive, will provide both acute and chronic effect level data which can be used in determining the size of the contaminated area which must be remediated and the level to which it must be remediated to reduce the potential for effects below either acute or chronic levels.

Let me know if you have any questions or need further assistance.

DDW/fm

BUREAU OF WASTE CLEANUP

JUN 29 2004

Program & Technical  
Support Section

Available Methods for Hazardous Waste Site Toxicity Testing (Recommended Methods in Bold) Revised 4/17/2004

Test Sample Type	Sample Salinity	Test Species	Test Type	Test Method
Soil		<b>Earthworm, Eisenia foetida</b> <b>Earthworm, Eisenia foetida</b> <b>White Worm, Enchytraeus sp.</b> <b>Springtail, Folsomia candida</b> <b>Nematode, Caenorhabditis elegans</b> Lettuce seed, Lactuca sativa Lettuce root, Lactuca sativa Corn root, Zea mays	Acute, survival Chronic, reproduction Chronic, reproduction Chronic, reproduction Acute, survival Acute, germination Chronic, elongation Chronic, elongation	EPA/600/3-88/029; A.8.5, ASTM E1676-97 ISO 11268-2:1998 ISO/CD 16387 ISO 11267:1999 ASTM E2172-01 EPA/600/3-88/029; A.8.6 EPA/600/3-88/029; A.8.7 Sunderland et al. 1991
Whole Sediment w/overlying culture water	Freshwater	Amphipod, Hyalella azteca <b>Amphipod, Hyalella azteca</b> Midge, Chironomus tentans <b>Midge, Chironomus tentans</b>	Acute, survival Chronic, survival, growth, reproduction Acute, survival Life-Cycle	EPA/600/R-99/064; 100.1 <b>EPA/600/R-99/064 (42-day); 100.4</b> EPA/600/R-99/064; 100.2 <b>EPA/600/R-99/064; 100.5</b>
	Salt water	Amphipod, Ampelisca abdita Amphipod, Leptochirus plumulosus <b>Amphipod, Leptochirus plumulosus</b> Amphipod, Euhastorius estuarius Amphipod, Rhepoxynius abronius Amphipod, Hyalella azteca (1-15 ppt) <b>Amphipod, Hyalella azteca (1-15 ppt)</b> Coot Clam, Mulinia lateralis	Acute, survival Acute, survival Chronic, growth, reproduction Acute, survival Acute, survival Acute, survival Acute, survival Chronic, survival, growth, reproduction Chronic, development	EPA/600/R-94/025; 100.4 EPA/600/R-94/025; 100.4 <b>EPA-600-R-01-020</b> EPA/600/R-94/025; 100.4 EPA/600/R-94/025; 100.4 EPA/600/R-99/064; 100.1 <b>EPA/600/R-99/064 (42-day); 100.4</b> EPA ERL-N
Elutriate and Pore Water	Freshwater	Bannerfin Shiner, Cyprinella leedsi Fathead Minnow, Pimephales promelas <b>Fathead Minnow, Pimephales promelas</b> Water Flea, Daphnia pulex Water Flea, Daphnia magna Water Flea, Ceriodaphnia dubia <b>Water Flea, Ceriodaphnia dubia**</b> <b>Green Alga, Selenastrum capricornutum</b>	Acute, survival Acute, survival Chronic, growth Acute, survival Acute, survival Acute, survival Chronic, reproduction Chronic, growth	EPA-821-R-02-012* EPA-821-R-02-012* <b>EPA-821-R-02-013*; 1000.0</b> EPA-821-R-02-012* EPA-821-R-02-012* EPA-821-R-02-012* <b>EPA-821-R-02-013*; 1002.0</b> <b>EPA-821-R-02-013*; 1003.0</b>
	Salt water	Inland Silverside, Menidia beryllina <b>Inland Silverside, Menidia beryllina</b> Opossum Shrimp, Americamysis bahia <b>Opossum Shrimp, Americamysis bahia</b> Coot Clam, Mulinia lateralis <b>Sea Urchin, Arbacia punctulata</b>	Acute survival Chronic, growth Acute, survival Chronic, growth Chronic, reproduction Chronic, fertilization	EPA-821-R-02-012* <b>EPA-821-R-02-014*; 1006.0</b> EPA-821-R-02-012* <b>EPA-821-R-02-014*; 1007.0</b> EPA ERL-N <b>EPA-821-R-02-014*; 1008.0</b>
Water	Freshwater	Bannerfin Shiner, Cyprinella leedsi Fathead Minnow, Pimephales promelas <b>Fathead Minnow, Pimephales promelas</b> Water Flea, Daphnia pulex Water Flea, Daphnia magna Water Flea, Ceriodaphnia dubia <b>Water Flea, Ceriodaphnia dubia**</b> <b>Green Alga, Selenastrum capricornutum</b>	Acute, survival Acute, survival Chronic, growth Acute, survival Acute, survival Acute, survival Chronic, reproduction Chronic, growth	EPA-821-R-02-012 EPA-821-R-02-012 <b>EPA-821-R-02-13; 1000.0</b> EPA-821-R-02-012 EPA-821-R-02-012 EPA-821-R-02-012 <b>EPA-821-R-02-13; 1002.0</b> <b>EPA-821-R-02-13; 1003.0</b>

Test Sample Type	Sample Salinity	Test Species	Test Type	Test Method
	Salt water	Inland Silverside, <i>Menidia beryllina</i> <b>Inland Silverside, <i>Menidia beryllina</i></b> Opossum Shrimp, <i>Americamysis bahia</i> <b>Opossum Shrimp, <i>Americamysis bahia</i></b> Coot Clam, <i>Mulinia lateralis</i> <b>Sea Urchin, <i>Arbacia punctulata</i></b>	Acute, survival <b>Chronic, growth</b> Acute, survival <b>Chronic, growth</b> Acute and Chronic <b>Chronic, fertilization</b>	EPA-821-R-02-012 <b>EPA-821-R-02-014; 1006.0</b> EPA-821-R-02-012 <b>EPA-821-R-02-014; 1007.0</b> EPA ERL-N <b>EPA-821-R-02-014; 1008.0</b>

\*Use EPA/600/3-88/029 for preparing elutriates from soils. Use EPA-823-B-98-004 for preparing elutriates from sediments

\*\* Not suitable for use with some groundwater.

ASTM E1676-97. Standard Guide for Conducting Laboratory Soil Toxicity or Bioaccumulation Tests With the Lumbricid Earthworm *Eisenia Fetida*

ASTM E2172-01. Standard Guide for Conducting Laboratory Soil Toxicity Tests with the Nematode *Caenorhabditis elegans*.

EPA/600/3-88/029 Protocols for short-term toxicity screening of hazardous waste sites.

EPA-823-B-98-004 Evaluation of dredge material proposed for discharge in waters of the U.S. - Testing manual.

EPA/600/R-99/064 Methods for measuring the toxicity and bioaccumulation of sediment-associated contaminants with freshwater invertebrates, 2nd ed.

EPA/600/R-94/025 Methods for measure the toxicity and bioaccumulation of sediment-associated contaminants with estuarine and marine amphipods

EPA-821-R-02-013 Short-term methods for estimating the chronic toxicity of effluents and receiving waters to freshwater organisms, 2nd Edition.

EPA-821-R-02-014 Short-term methods for estimating the chronic toxicity of effluents ad receiving waters to marine and estuarine organisms, 2nd Edition.

EPA-821-R-02-012 Methods for measuring the acute toxicity of effluents and receiving waters to freshwater and marine organisms, 5th Ed.

EPA ERL-N Embryo-larval test using the bivalve *Mulinia lateralis* - SOP Rev. 0, March 1993. Narragansett ERL.

ISO 11268-2:1998. 1998. Soil Quality – Effects of Pollutants on Earthworms (*Eisenia fetida*) – Part 2: Determination of Effects on Reproduction.

ISO/CD 16387. Soil Quality – Effects of Pollutants on Enchytraeidae (*Enchytraeus* sp.) – Determinations of effects on reproduction and survival.

ISO 11267:1999. Soil Quality – Inhibition of Reproduction of *Collembola* (*Folsomia candida*) by Soil.

Sutherland, S.L., P.W. Santelmann, and T.A. Baughman. 1991. A rapid, sensitive soil bioassay for sulfonylurea herbicides. Weed Science 39, 296-298.