6. Using Toxicity Data as Indicators of Water Quality

Contents

6. Using Toxicity Data as Indicators of Water Quality

The whole-effluent approach to toxics control for the protection of aquatic life involves the evaluation of substances using acute and chronic tests to measure the toxicity of wastewater and ambient waters. Whole-effluent toxicity (WET) testing is an important component of the U.S. Environmental Protection Agency's (EPA's) integrated approach for controlling the discharge of toxic chemicals and other materials into surface waters. Such WET tests are typically conducted in concert with other types of monitoring such as chemical, physical, and biological assessments. Toxicity is a valuable indicator for assessing and protecting against impacts on water quality and designated uses caused by the aggregate toxic effect of pollutants. Like chemical-specific limitations and standards, WET tests provide some predictive capability to assess the occurrence of toxicity under predefined conditions. Instream evaluation of populations of aquatic species (bioassessments) provides information on past exposures of organisms to toxic conditions, but this approach only estimates reactions to exposures that have already occurred. Instream biomonitoring procedures are therefore always reactive to an insult rather than predictive and protective. Contaminants may flow directly from industrial and municipal waste dischargers, may come from polluted runoff in urban and agricultural areas, or may collect in the sediments. Toxicity evaluations can be used to assess the type and extent of degraded water quality.

Toxicity tests used for whole-effluent and surface waters include surrogate freshwater or marine (depending on the mixture of effluent and receiving water) plants, invertebrates, and vertebrates (U.S. EPA 1991, EPA/505/2-001). EPA has published CFR part 136 methods for WET tests (60 Fed Reg 53529, Oct. 16, 1995) and the manuals are incorporated by reference, so in effect the methods are regulations. These methods are contained in the following three documents:

- Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms (EPA/600/4-90/027F) (U.S. EPA 1993)

- Short-Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Water to Freshwater Organisms (EPA/600/4-91/002) (U.S. EPA 1994a)

- Short-Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Water to Marine and Estuarine Organisms (EPA/600/4-91/003) (U.S. EPA 1994b)

In WET tests, aquatic organisms (plants, invertebrates, and vertebrates) are exposed to samples of effluent in the laboratory. These exposures are conducted under controlled conditions and the response(s) of the organisms recorded. Acute toxicity is generally measured using a multiconcentration test, termed a definitive test, that consists of a control (clean water) and several (generally five) effluent concentrations. The tests are designed to provide concentration-response information, expressed as the percentage of effluent concentration that is lethal to 50% of the test organisms (LC50) within the prescribed period. The tests can also be used to determine the highest effluent concentration at which survival is not statistically significantly different from the control. Acute tests (U.S. EPA 1993, 1999) generally use death as the measured effect of a given effluent over 24 to 96 hours. Sublethal WET tests (often described as chronic tests) use longer durations of exposure (up to 9 days) to ascertain the adverse effects of an effluent on survival, growth, and/or reproduction of the organisms. For freshwater ecosystems, EPA has focused on short-term tests for three species, designed to estimate the

chronic toxicity in a water sample (U.S. EPA 1994a, 1999). These methods include a fish, larval fathead minnow (*Pimephales promelas*), a zooplankton (*Ceriodaphnia dubia*), and an alga (*Selenastrum capricornutum*). The marine and estuarine short-term tests estimate chronic toxicity (U.S. EPA 1994b, 1999) with two fish species, sheepshead minnow (*Cyprinodon variegatus*) and the inland silverside (*Menidia berylina)*, a red alga (*Champia parvula*), an East Coast mysid (*Mysidopsis bahia*), and a sea urchin (*Arbacia punctulata*). The EPA toxicity tests and other single-species tests were intended to be screening tools (i.e., to indicate the potential for wastewater or ambient water samples to cause biological community impacts, characterizing relative ecosystem effects) and "early warning" signals (a measurement that indicates the potential for aquatic ecosystem impairment prior to actual damage to biological communities (U.S. EPA 1991, 1994a). The toxicity tests are applicable to ambient water samples regardless of the sources (i.e., point or nonpoint) of contaminants. The results of all tests may be used to make quantitative estimates of the degree of toxicity of the test material.

Sediment contamination is a widespread environmental problem that can pose a threat to aquatic ecosystems. Sediment acts as a reservoir for common chemicals such as pesticides, herbicides, polychlorinated biphenyls (PCBs), polycyclic aromatic hydrocarbons (PAHs), and metals such as lead, mercury, and arsenic. Contaminated sediments may be directly toxic to aquatic life (organisms found in the water and in or near the sediment) or can be a source of contaminants for bioaccumulation (where a substance is taken up by an organism) in the food chain. Protecting sediment quality is an important part of restoring and maintaining the biological integrity of our nation's waters. Sediment is an integral component of aquatic ecosystems, providing habitat, feeding, spawning, and rearing areas for many aquatic organisms. Because sediment serves as a reservoir for contaminants, it is a source of contaminants to the water column and organisms. The extent and severity of sediment contamination in the United States, as documented in the National Sediment Inventory (U.S. EPA, 1997) and through contaminated site histories, emphasize the need for better tools for reducing and preventing sediment contamination.

Whole-sediment toxicity tests are an important tool for sediment quality assessment. They directly measure sediment toxicity to a test species under laboratory conditions, and are especially valuable because they account for interactive effects of chemical mixtures. Benthic community analyses are also useful for sediment assessment, because they account for instream conditions. Sediment toxicity testing should be conducted to characterize the nature and extent of contamination.

EPA has published the following guidance documents, which provide laboratory methods for measuring the toxicity of whole sediments:

- Methods for Assessing the Toxicity of Sediment-Associated Contaminants with Estuarine and Marine Amphipods (U.S. EPA 1994c)

- Methods for Measuring the Toxicity and Bioaccumulation of Sediment-Associated Contaminants with Freshwater Invertebrates, second edition (U.S. EPA 2000)

- Methods for Assessing the Chronic Toxicity of Marine and Estuarine Sediment-Associated Contaminants with the Amphipod *Leptocheirus plumulosus*, March 2001. EPA-823-F-01-008

When the effluent toxicity tests, receiving water toxicity tests, and whole-sediment toxicity tests are effectively used as the primary water quality indicator, the sample locations can be further characterized by the use of Toxicity Identification Evaluations (TIEs) to help identify the causative pollutants or pollutant categories.

First, appropriate effluent, receiving water, and sediment toxicity tests should be performed. EPA provides guidance on the sample size to allow for concurrent or subsequent chemical testing. Testing should be conducted to characterize the nature and extent of contamination, with the appropriate lethal and sublethal toxicity tests. There may be limitations in relying on whole-effluent and/or sediment toxicity tests to determine attainment of water quality standards (WQS). Because test organisms are selected on the basis of overall sensitivity to chemical pollutants and ecological relevance, toxicity tests are conducted with a limited range of species whose sensitivity may not be known when the chemicals of concern are unknown. On the other hand, toxicity tests have strengths in being able to detect effects from unknown or unmeasured chemicals and interactive toxicity of multiple chemicals.

6.1 How Are Toxicity Data Used Within the Context of the State's WQS?

Typically, toxicity data are used to interpret a state's narrative WQS of "no toxics in toxic amounts." Narrative criteria can be the basis for limiting toxicity in waste discharges where a specific pollutant can be identified as causing or contributing to the toxicity but there are no numeric criteria in the state standards, or where toxicity cannot be traced to a particular pollutant. Section 131.11(a)(2) requires states to develop implementation procedures that explain how the state will ensure that narrative criteria are met.

The state should describe what types of toxicity data it is requiring or considering requiring, and the appropriate test methods. WET data are generated when effluent samples are tested. Tests with receiving water samples and ambient water tests using the promulgated WET test methods should also be documented. Typically, tests with effluent are conducted with a specific discharger's effluent, using either ambient or standard laboratory water as dilution water. Receiving water toxicity tests are conducted with ambient water alone, compared with a standard laboratory dilution water, and may be done with a dilution series effluent test. Sediment toxicity test data may be required to assess contaminated sites.

Some states, however, have adopted numeric criteria for WET. EPA regulations (40 CFR 122.44) cover the national surface water toxics control program. These regulations are linked to WQS requirements and specifically address the control of pollutants with and without numeric criteria. For example, section 122.44 (d)(1)(v) provides the permitting authority with several options for establishing effluent limits when a state does not have chemical-specific numeric criteria for a pollutant at a concentration that causes or contributes to a violation of the narrative criteria. Where a state, territory, or authorized tribe adopts narrative criteria for toxic pollutants

to protect designated uses, the state, territory or authorized tribe must provide information identifying the method by which it intends to regulate point source discharges of toxic pollutants on water quality limited segments based on such narrative criteria. Such information may be included as part of the standards or may be included in documents generated by the state, territory, or authorized tribe in response to the Water Quality Planning and Management Regulations (40 CFR part 35). If a state standard is not in attainment, the water should be listed. Information regarding how the state, territory or authorized tribe intneds to regulate point source discharges of toxic pollutants on water quality limited segments based on narrative criteria is discussed in a December 1988 EPA policy and guidance, Guidance for State Implementation of Water Quality Standards for CWA section 303(c)(2)(B).

The regulatory basis for requiring WET is found in EPA regulations at 40 CFR 122.44(d)(1)(v). These regulations require NPDES permits to contain WET limits where a permittee has been shown to cause, have the reasonable potential to cause, or contribute to an instream excursion of a narrative criterion. State implementation procedures should, at a minimum, specify or reference methods to be used in implementing WET controls.

The choice of species depends on the type of regulatory WET test to be conducted and the most representative test species (e.g., for a freshwater vs. marine discharger); the test method manuals stipulate species-specific test conditions. Effluent samples are typically collected as 24-hour composite samples and tested in static renewal or static toxicity tests. The type and duration of the tests and species required must be documented. Procedures for the collection of effluent and ambient water samples, selection of the test method, and species to use under different testing conditions are described in the three promulgated WET methods (U.S. EPA 1993, 1994a,b). Similarly, the sediment tests are described in guidance (U.S. EPA 1994c, 2000, 2001).

6.2 What Actions Does the State Take To Assess and Document Data Quality?

If the state is using WET data, receiving water data, and sediment toxicity test data, it should document the relevant QA/QC procedures to document data quality. The manuals that provide the methods for the promulgated WET test methods (U.S. EPA 1993, Methods for Measuring the Acute Toxicity of Effluents and Receiving Water to Freshwater and Marine Organisms; U.S. EPA 1994a, Short-Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Water to Marine and Estuarine Organisms; and U.S. EPA 1994b, Short-Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Water to Freshwater Organisms) describe the QA/QC specific to these WET test methods. Specifically, Chapter 4 of each of the test manuals contains the QA/QC specifications for the promulgated WET test methods. When the WET test methods are used, Chapter 4 (Quality Assurance) should be strictly adhered to in order to ensure that the outcome of the test results is of the best quality.

EPA has developed sediment methods for aquatic invertebrates (U.S. EPA 2000, Methods for Measuring the Toxicity and Bioaccumulation of Sediment-Associated Contaminants with Freshwater Invertebrates; U.S. EPA 1994c, Methods for Assessing the Toxicity of Sediment-Associated Contaminants with Estuarine and Marine Amphipods; and U.S. EPA 2001, Methods for Assessing the Chronic Toxicity of Marine and Estuarine Sediment-Associated Contaminants

with the Amphipod *Leptocheirus plumulosus* - First Edition, EPA/600/R-01/020). These manuals also have QA guidance for obtaining organisms, water, and sediment samples, and conducting the toxicity test.

6.2.1 How Does the State Define Data Quality?

EPA encourages states, territories, interstate commissions, and authorized tribes to use the data quality objectives (DQO) process to define minimum-quality data requirements. The DQO process is a systematic procedure for defining the criteria that the data collection design should satisfy, including when to collect samples, where to collect samples, how many samples to collect, and the tolerable level of decision error for the project.

States should also document required procedures that ensure the quality of the data collected. These include information on sample collection and handling protocols, use of standardized methods, QC procedures, and data management. These procedures are generally included in a QA project plan or SOP. It is becoming increasingly clear that these procedures should be available to other organizations such as tribal, interstate, state, Federal, academic, and volunteer citizen groups that also monitor water quality. These stakeholders may agree to meet state data quality requirements if the agency clearly spells out these requirements in its assessment and listing methodology or other readily available and well-publicized documents.

6.2.2 How Does the State Review and Evaluate Data Quality?

The term "data quality assessment" means the scientific and statistical evaluation to determine if data obtained from toxicity monitoring operations are appropriate and of sufficient quality and quantity to support water quality attainment decisions. As discussed in Chapter 3, the user must know in what context a data set is to be used in order to determine whether the data set is adequate. Guidance for assessing the quality of available data sets is provided in detail in Practical Methods for Data Quality Assessment (EPA/600/R-96/084).

For assessing whether toxicity data are acceptable, EPA recommends a tiered approach (as described in Chapter 3). In this approach, the first step is to screen all reports to determine whether appropriate procedures were used and QA/QC measures were in place (e.g., whether SOPs were in place describing each step). The second step is to evaluate whether samples were collected under the appropriate conditions. The next step is to review the sample collection and any associated analytical methods to determine compatibility with the agency's QA/QC requirements and SOPs. Then the evaluator should determine whether the sample collection and analytical methods were actually followed in creation of the data set. The final step is to assess whether the metadata accompanying the data set meet the agency's requirements.

Upon determining that the data meet basic documentation requirements, the evaluator should decide whether additional screening of the actual data sets is needed. A state may consider reviewing reference toxicant values outside the control chart values and dose-response data for each test. If the data reveal potential problems or errors in the collection or analysis, an in-depth analysis of QA/QC procedures may be appropriate. This screening could include reviews of

QA/QC reports to determine whether the data set meets QA/QC requirements regarding measurement systems, the approach to handling missing data and problem data sets, or any deviations from SOPs.

6.2.3 How Does the State Document the Quality of the Data Used To Support WQS Attainment Decisions?

The 305(b) Consistency Workgroup developed a table to assist in documenting the quality of toxicity data and information used to support WQS attainment decisions (U.S. EPA 1997). This table is reproduced as Table 6-1.

6.3 How Does the State Analyze and Interpret Toxicity Data To Determine WQS Attainment/Impairment?

A state should document the methods used to analyze effluent, receiving water, and sediment toxicity test methods. The state's procedures should be specific about the method used; that is, LC50s must be calculated following the methods in the promulgated WET manuals. The state should also define the control used and the receiving water or reference site sediment and how significantly different comparisons were determined.

The most important element of the state, territory or authorized tribe's assessment and listing methodology is documentation of how the state analyzes and interprets data to determine WQS attainment and identify impaired waters. This documentation should be consistent with the state's, territory's, or authorized tribe's implementation procedures that are described either in the WQS or alternatively in other implementing regulations or policies and procedures documents such as the continuous planning process or consolidated assessment and listing methodology. An assessment methodology should take into account the balance between desired minimum data requirements from a strict scientific perspective and the practical realities of the availability of information and the strength of the available evidence. For example, a state's methodology could require a minimum level of decision errors for making an attainment decision, except in cases where overwhelming evidence of impairment is found. An example of overwhelming evidence would be repeated sampling events showing very high toxicity values for each species, regardless of the known cause of the toxicity.

6.3.1 What Statistical Analyses for Interpreting Toxicity Data Are Relevant to State WQS?

The statistical methods to analyze the promulgated WET testing methods, as well as the sediment and bioaccumulation methods, are part of the testing methods themselves and should be strictly followed. One problem states have had with analyzing data is related to not following the correct statistical procedures described in the methods. The state's DQOs and QA/QC procedures should clearly define adequate statistical and other implementation procedures to ensure that all parties are aware of the minimum data set and statistical analysis requirements.

Table 6-1. Hierarchy of toxicological approaches and levels for evaluation of aquatic life use attainment

Level of info[a]	Technical components	Spatial/temporal coverage	Data quality[b]	WBS codes[c]
1	Any one of the following: • Acute or chronic WET • Acute ambient • Acute sediment	1-2 WET tests/yr or 1 ambient or sediment sample tested in a segment or site	Unknown/low; minimal replication used; laboratory quality or expertise unknown	510, 520, 530, 550
2	Any of the following: • Acute or chronic ambient • Acute sediment • Acute and chronic WET for effluent-dominated system	3-4 WET tests/yr or 2 ambient or sediment samples tested in a segment or site at different times	Low/moderate; little replication used within a site; laboratory quality or expertise unknown or low	510, 520, 530, 540, 550
3	Any of the following: • Acute and chronic WET for effluent-dominated system • Chronic ambient or acute or chronic sediment	Monthly WET tests or total of three tests based on samples collected in a segment at three different times	Moderate/high; replication used; trained personnel and good laboratory quality	510, 520, 540, 550
4	Both of the following: • Acute and chronic ambient and • Acute or chronic sediment	≥ 4 tests in total based on samples collected in a segment at four different times including low-flow conditions	High; replication used; trained personnel and good laboratory quality	530, 540, 550

[a] Level of information refers to rigor of toxicity testing, where 1 = lowest and 4 = highest.

[b] Refers to ability of the toxicity testing endpoints to detect impairment or to differentiate along a gradient of environmental conditions.

[c] WBS Assessment Type Codes from Table 1-1.

The procedures should describe how the state uses trend analysis for toxicity monitoring or requirements.

6.3.2 How Does the State Make Attainment/Impairment Decisions in the Absence of a "Perfect" Data Set?

State assessment and listing methodologies should describe the state's efforts to make water quality attainment/impairment decisions in the absence of complete data sets that meet all their data quality requirements. A state should develop procedures for looking for overwhelming evidence of water quality impairment, such as why a single sample (with well-documented QC methods) shows high toxicity.

6.4 References

U.S. Environmental Protection Agency (U.S. EPA). 1988. Guidance for state implementation of water quality standards for CWA section 303(c)(2)(B). Office of Water Regulations and Standards, Criteria and Standards Division, Washington, DC.

U.S. EPA. 1991. Technical support document for water quality-based toxics control. Washington, DC. EPA/505/2-90-001.

U.S. EPA. 1993. Methods for measuring the acute toxicity of effluents and receiving water to freshwater and marine organisms. 4th ed. Office of Research and Development, Washington, DC. EPA/600/4-90/027F.

U.S. EPA. 1994a. Short-term methods for estimating the chronic toxicity of effluents and receiving water to marine and estuarine organisms. 2nd ed. July. Office of Research and Development, Washington, DC. EPA/600/4-91/003.

U.S. EPA. 1994b. Short-term methods for estimating the chronic toxicity of effluents and receiving water to freshwater organisms. 3rd ed. Office of Research and Development, Washington, DC. EPA/600/4-91/002.

U.S. EPA. 1994c. Methods for assessing the toxicity of sediment-associated contaminants with estuarine and marine amphipods. Office of Research and Development, Duluth, MN. EPA-600-R-94-025.

U.S. EPA. 1997. The incidence and severity of sediment contamination in the United States. Volume 1: National sediment quality survey (EPA-823-R-97-006); Volume 2: Data summaries for areas of probable concern (EPA-823-R-97-007); Volume 3: Sediment contaminant point source inventory. (EPA-823-R-97-008). Office of Water, Office of Science and Technology, Washington, DC.

U.S. EPA. 1999. Errata for effluent and receiving water toxicity test manuals: Acute toxicity of effluents and receiving waters to freshwater and marine organisms; Short-term methods for estimating the chronic toxicity of effluents and receiving waters to freshwater organisms; and Short-term methods for estimating the chronic toxicity of effluents and receiving waters to marine and estuarine organisms. January 1999. Office of Research and Development, Duluth, MN. EPA-600/R-98/182.

U.S. EPA. 2000. Methods for measuring the toxicity and bioaccumulation of sediment-associated contaminants with freshwater invertebrates. 2nd ed. Office of Research and Development, Duluth, MN. EPA 600/R-99/064.

U.S. EPA. 2001. Methods for assessing the chronic toxicity of marine and estuarine sediment-associated contaminants with the amphipod *Leptocheirus plumulosus*. March 2001. Office of Water, Office of Science and Technology, Washington, DC. EPA-823-F-01-008.