

**Clarifications Regarding Toxicity Reduction and  
Identification Evaluations in the National Pollutant  
Discharge Elimination System Program**

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**U.S. Environmental Protection Agency  
Office of Wastewater Management  
Office of Regulatory Enforcement  
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## **NOTICE AND DISCLAIMER**

This document provides guidance to NPDES authorities and persons interested in toxicity reduction evaluations and toxicity identification evaluations (TRES/TIEs) as they relate to whole effluent toxicity (WET) testing. This document is designed to reflect national guidance on conducting TRES/TIEs. This document does not, however, substitute for any aspect of the Clean Water Act, an NPDES permit, or EPA or state regulations applicable to permits or WET testing; nor is this document a permit or regulation itself. This document does not and cannot impose any legally binding requirements on EPA, states, NPDES permittees, and/or laboratories conducting WET testing or TRES/TIEs for permittees or evaluations of ambient water quality for states. EPA and state officials retain discretion to adopt approaches on a case-by-case basis that differ from this guidance based on analysis of site-specific circumstances. This guidance may be revised without public notice to reflect changes in EPA policy.

## EXECUTIVE SUMMARY

This document provides additional clarification on existing EPA guidance on toxicity reduction evaluations (TREs) and toxicity identification evaluations (TIEs). It also discusses how NPDES permittees and permitting authorities should address some technical issues that have been raised to EPA on conducting TREs/TIEs. While this document contains no new guidance on permitting or compliance, it responds to a July 1997 settlement agreement, which required EPA to issue technical guidance that would provide clarification on conducting TREs/TIEs under the NPDES permitting program. The settlement agreement was with the City of San Bernardino (CA), United Water Florida, and the City of Washington (GA).

The technical issues addressed in this guidance include: when and under what circumstances a permittee should conduct a TRE and/or TIE; technical limitations on the TIE process; consideration of persistence and magnitude of toxicity events; ionic imbalances of effluents; the applicability of compliance schedules; and inconclusive TREs/TIEs.

The following points summarize important conclusions of this document:

- A TRE is defined as a methodical, stepwise investigation of the cause(s) of, and appropriate control(s) for, an effluent that has demonstrated acute or chronic whole effluent toxicity (WET). Several options are available to the regulatory authority for requesting a TRE. Whenever a toxic discharge occurs, however, a permittee should consider initiating preliminary TRE procedures to reduce or eliminate toxicity.
- All technically reasonable actions taken to resolve WET should be considered TRE activities. Because TIEs are powerful and effective tools for identifying the source of WET, EPA recommends that permittees consider conducting TIEs early in the TRE process. Likewise, toxicity treatability testing can be effective when trying to resolve WET limit noncompliance and other problems with effluent toxicity.
- Permittees should seek technical review and comment from their regulatory authority when developing TRE plans that outline investigative and problem resolution techniques, including reasonable time lines and milestones, in order to avoid delays and maximize consideration of relevant factors that may affect toxicity. The regulatory authority should then approve the TRE schedule and completion date. The authority should either concur with the technical merit of the plan or recommend modifications that would improve its technical merit. A close cooperative relationship should be established among the permittee (and, if applicable, the permittee's technical consultant) and the permitting authority early in the TRE process. This relationship should be maintained until the TRE is successfully completed and any controls necessary to prevent unacceptable levels of toxicity are fully implemented. This process allows all parties to understand the requirements and expectations, and encourages evolution of the plan toward the most effective resolution. Collaboration among the parties throughout the TRE process will add to its effectiveness and assist in course corrections.

## Technical Limitations of the Procedures

A TIE is a set of procedures that uses physical and chemical treatments to identify or classify the specific chemical compounds causing toxicity in an effluent sample.

- EPA has found that TIEs have been highly successful when aggressively implemented.
- There are no significant distinctions between the acute or chronic WET methodology that would restrict the use of either approach in a TIE. Both acute and chronic TIE procedures are reliable means of resolving toxicity issues and lead to appropriate controls.
- The most common causes of unsuccessful TIEs are (1) poorly qualified laboratories or staff (e.g., laboratories that do not use the TIE guidance appropriately or lack the tools or skills for comprehensive TIE interpretation); (2) wastewater complexity; or (3) both.
- Permittees should carefully consider using individuals who are qualified and experienced in TREs/TIEs. Laboratories should clearly demonstrate past experience and timeliness of their results.
- The difficulty in conducting a TIE, and the time required to complete it, will likely increase in direct proportion to the complexity of toxicants in wastewater. As the number of chemical constituents in wastewater increases, the interactions of those chemicals (e.g., with biological and analytical systems and with each other in the wastewater) can increase the difficulty of identifying toxicants.
- When multiple toxicants are present in a sample, identifying and resolving the toxicants serially may be necessary due to masking or confounding influences. Additionally, the complexity of a TIE may increase as the toxicity magnitude and frequency decrease (less toxic, less often). Conversely, as the toxicity magnitude and frequency increase (more toxic, more often), wastewater components causing toxicity may be more readily identifiable. Despite these difficulties, successful TIE completion is generally possible if proper TIE procedures are conducted by appropriately trained and experienced staff.

## Consideration of Persistence and Magnitude of Toxicity Events

For the purpose of this document, “persistence” is defined as the tendency for toxicity within a single effluent sample to remain over time. “Consistency” is defined as the tendency for toxicity to be expressed in an effluent at similar levels across samples and over time. “Magnitude” refers to the degree of toxicity demonstrated in a test, or how large the effect was in the test. EPA acknowledges that intermittent or marginal toxicity may be more difficult to detect, but that a properly conducted TIE has a reasonable expectation of successfully finding the toxicant(s) responsible for the effluent noncompliance. One definition of marginal toxicity is a very slight (but statistically significant) effect at a dilution that contains a high proportion of effluent (e.g., above 80 percent). For a TIE to successfully identify and confirm toxicants as part of a TRE, toxicity must be present in a sample. Expecting a TIE to immediately follow a single or infrequent event of WET noncompliance is unrealistic. Rather, the persistence (duration, frequency) and magnitude of components of the exposure should be characterized immediately through additional testing

to evaluate whether a TIE would help identify the source of toxicity. Intermittent or marginal toxicity may be addressed by increasing WET testing frequency and always obtaining sufficient sample volume to perform TIE procedures on toxic samples. An alternative to performing a TIE is to conduct treatability tests that use bench-scale treatment units to identify process changes that reduce toxicity through changes in treatment type, arrangement, or method. These tests, however, may not identify which toxicant is being removed or reduced.

### **Ionic Imbalance**

Ionic imbalance is defined as a combination of salts in an effluent that (1) are present at levels or in ratios inconsistent with those normally found in most naturally occurring waters; and (2) demonstrate toxicity during a WET test. These salts are normally referred to as total dissolved solids (TDS). TDS imbalances that result in toxic discharges are the responsibility of the discharger, as are all other effluent constituents and their effects. Industrial processes that typically produce effluent with high concentrations of ions include textile manufacturing, pickling, and oil refining. Processes that produce effluents with low ionic strength typically include remediation of contaminated groundwater and condensate discharge. In some cases, the imbalance may result solely from an ionic imbalance in the source water. For example, if a facility obtains its source water from the same stream to which it discharges, and the source water has a toxic ionic imbalance therefore causing the discharge to be toxic, regulatory authorities should take this into consideration when evaluating the permittee's compliance status.

### **Permit and Enforcement Mechanisms To Implement a TRE**

The regulatory authority may require a TRE through the permit, an order for information, or an enforcement action. EPA strongly recommends, however, that permittees voluntarily institute initial TRE activities any time a toxic event occurs. In addition to WET testing, the TRE process may include investigation of general housekeeping procedures, a facility review of treatment and process chemicals and uses, TIE procedures, treatability tests, and monitoring of suspected toxicants in the effluent.

- The effectiveness of the National Pollutant Discharge Elimination System (NPDES) permitting process for WET relies on having an adequate amount of valid data acquired using EPA-approved test species and methods (USEPA 1993c, 1994a, 1994b, 1995, 1999b).
- EPA recommends that permittees develop a basic TRE strategy (USEPA 1989a, 1999a) before the need arises to facilitate a rapid response in the event of toxicity. The strategy should include a summary of baseline information; operational efficiencies; evaluation of potential TRE consultants; and a thorough review and update of chemical inventories, uses, and input points for potential effluent toxicants.
- EPA recommends that permittees voluntarily institute initial TRE activities any time a toxic event occurs.

- Once a permittee reduces toxicity to an acceptable level (or eliminates it) and maintains compliance, the TRE goal has been met. The TRE then may be discontinued, if appropriate, and the routine monitoring schedule, if included in the permit, resumed.
- Enforcement decisions should be guided by case- and site-specific consideration of existing and historical toxicity, including toxicity magnitude, duration, and frequency, and the permittee's diligence in resolving and preventing WET noncompliance.

### **Inconclusive TREs and TIEs**

In some rare instances, TREs and TIEs have been unsuccessful or inconclusive. EPA acknowledges that some permittees have aggressively pursued a TRE using highly qualified technical support, but have been unable to resolve the problem. EPA has demonstrated its intent for appropriate discretion and constructive resolution through its established record of working cooperatively with permittees in these cases.

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## **BACKGROUND**

EPA's *Technical Support Document for Water Quality-based Toxics Control* (TSD) (USEPA 1991a) defines a TRE as "a site-specific study conducted in a stepwise process designed to identify the causative agents of effluent toxicity, isolate the sources of toxicity, evaluate the effectiveness of toxicity control options, and then confirm the reduction in effluent toxicity." TREs (USEPA 1989a, 1989b, 1999a) comprise all measures that are taken to reduce observed WET to levels that will meet NPDES WET permit requirements and ensure attainment of water quality standards. The evaluations use various tools, which may include TIEs, to meet NPDES objectives.

Each TRE requires case-specific investigative measures and solutions. Therefore, the permittee should consider all appropriate options when developing and implementing a TRE. The regulatory authority has a critical oversight role in ensuring that the permittee aggressively pursues the TRE to resolve toxicity issues in a timely and effective manner. Compliance, however, is ultimately the permittee's responsibility. The regulatory authority should evaluate the adequacy of the TRE strategy and process from a scientific perspective to help the permittee use resources wisely. The regulatory authority should also track the TRE as reports are submitted to ensure that the TRE is proceeding in a reasonable direction at a reasonable rate. The guidance presented here provides both the NPDES permittee and the regulatory authority flexibility when developing site-specific TRE plans.

The TSD recommends various approaches for conducting TREs and TIEs. Additionally, EPA's *Toxicity Reduction Evaluation Guidance for Municipal Wastewater Treatment Plants* (USEPA 1999a) provides a general framework for conducting TREs, along with guidance on available methods and procedures (see especially Figures 1-1, 3-1, 4-1, and 6-1 of the municipal TRE guidance). EPA encourages NPDES permittees to apply the methods referenced in the TSD for reducing toxicity before the discharge is subject to regulatory review (e.g., before application or reapplication for a permit) and action due to WET.

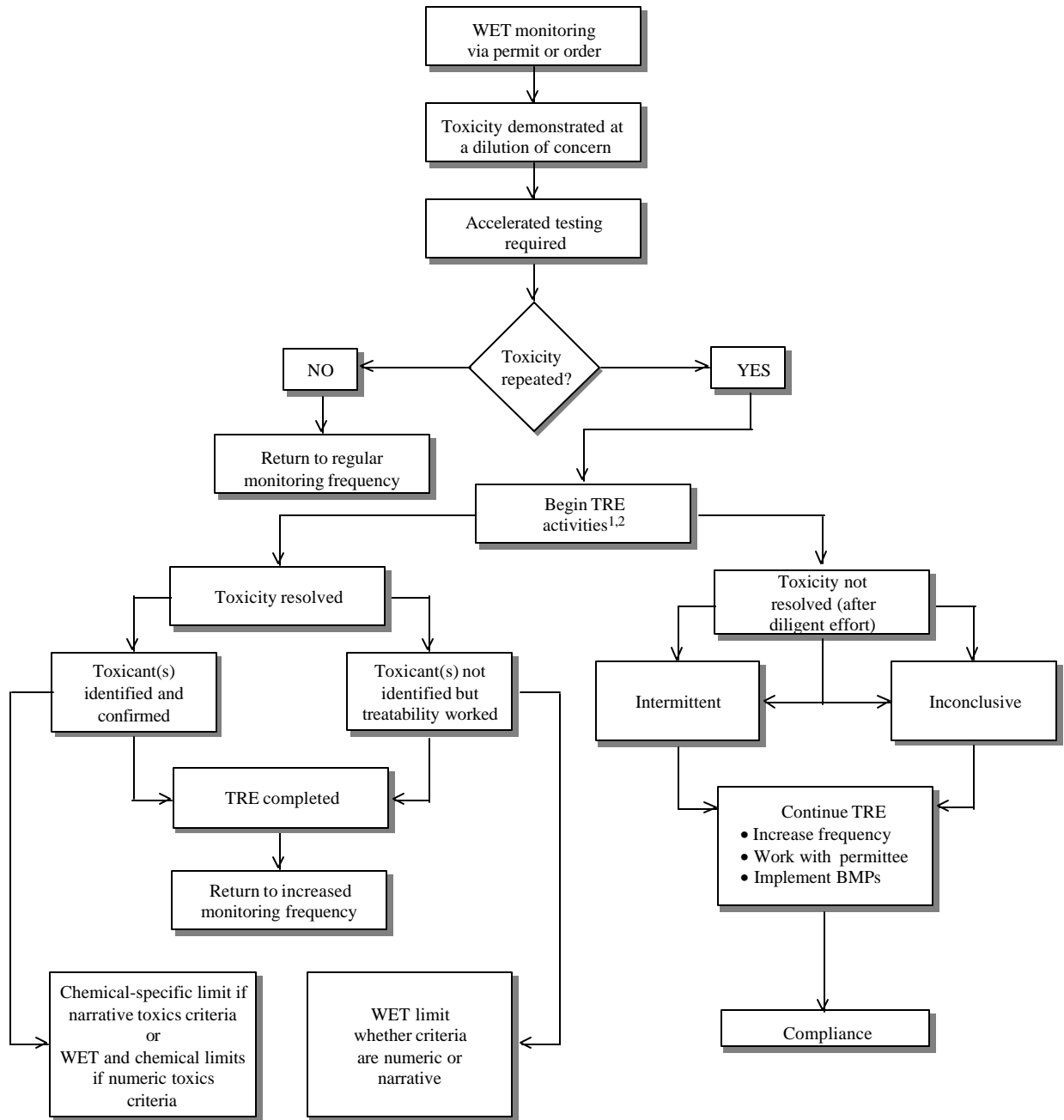
As a component of a TRE, TIE procedures (USEPA 1988, 1991b, 1992, 1993a, 1993b, 1996) are used to characterize and identify the cause(s) of toxicity. EPA strongly recommends conducting TIE procedures, but generally does not require that TIEs be performed as part of a TRE. TIE procedures are commonly performed in three phases—characterization, identification, and confirmation—in either a stepwise fashion or simultaneously, depending on the characteristics of the toxicity and effluent. Based on TIE results, the permittee may decide to conduct treatability tests on the final effluent or conduct source investigations, or both. This may lead to control methods such as chemical substitution, process modification, treatment of process or influent streams (pretreatment), or elimination of processes. Figure 1 illustrates some possible pathways for a TRE.

## **DETERMINING WHEN AND UNDER WHAT CIRCUMSTANCES TO CONDUCT TREs AND TIEs**

Regulation of pollutant parameters, including WET, in the NPDES program is comprised of permitting, compliance assessment, and enforcement. In the permitting phase, the permitting authority determines whether a discharge causes, has reasonable potential to cause, or contributes to an in-stream excursion of



a water quality criterion. When a discharge causes, has reasonable potential to cause, or contributes to an excursion above a *narrative* WET criterion (e.g., no toxics in toxic amounts), a WET limit is generally required. One exception is when the permitting authority demonstrates that chemical-specific limits incorporated into the permit, instead of a WET limit, are sufficient to attain and maintain all applicable numeric and narrative water quality standards [40 CFR § 122.44(d)(1)(v)].



<sup>1</sup> May include chemical use inventory and survey; facility housekeeping, standard operating procedures; operation and maintenance review; process review; review of raw materials/treatment compounds; any recent changes in use, type, or suppliers; and TIEs or treatability studies or both.  
<sup>2</sup> Regular versus Increased—After a TRE, at least quarterly monitoring would ensure that controls were working.

If a discharge causes, has reasonable potential to cause, or contributes to an excursion above a *numeric* WET criterion, the NPDES regulations require establishment of a WET limit [40 CFR § 122.44(d)(1)(iv)]. The NPDES permit will contain a WET limit and requirements to assess WET using prescribed methods (USEPA 1993c, 1994a, 1994b, 1995) at a prescribed frequency. When water quality standards include a numeric criterion for WET and monitoring results indicate that a WET limit is needed to ensure that water quality standards are met consistently, the permitting authority must reopen the permit to incorporate numeric WET limits or establish numeric WET limits when the permit is reissued [40 CFR § 122.44(d)(1)(iv)]. Compliance or noncompliance with a WET limit is determined by comparing the test result to the effluent limit. Whether a permittee uses the correct methods and collects test samples at the location and frequency specified in the permit directly affects compliance status. When a WET limit is not required, a NPDES permit may require WET monitoring only (i.e., monitoring without numeric WET limits).

When the NPDES permit application and screening do not find reasonable potential for toxicity, WET monitoring is recommended to ensure that toxicity is not present due to other factors (e.g., pH, hardness, combined pollutant effects, pollutants not known or suspected to have been present at toxic levels). WET monitoring data may indicate the need for a TRE and WET limits.

The permitting authority should establish in the permit whether (and if so, when) the permittee must initiate a TRE. In the absence of a permit requirement to conduct a TRE, permittees should consider conducting a TRE whenever they anticipate non-compliance with the permit limit. Generally, this will be when toxicity testing results obtained during scheduled monitoring indicate failure at a specific level of dilution, either at or near the low-flow dilution.

Depending on the specific circumstances, a single toxic event could trigger a TRE. For example, poor operation and maintenance at an industrial user may result in a spill of toxic cleaning materials that are washed down through floor drains into the wastewater system, causing a treatment plant upset and a toxicity test failure. In this case, the TRE may be relatively easy to complete, because the source of toxicity could be easily identified. Resolving the toxicity issue(s) may be as straightforward as improving housekeeping procedures to prevent recurrences. In cases of more persistent and elusive toxicity, the TRE may be triggered by failure in a test and in subsequent required testing. Several TIE analyses may be needed to characterize and confirm the sources of the toxicity. In this case, the duration of the TRE may be longer and it may result in chemical-specific permit limits.

EPA's TSD recommends that permits require additional WET testing after any WET test failure. A TRE is appropriate if non-compliant toxicity is demonstrated in any of the additional tests. For all additional tests conducted before and during a TRE, the permittee should obtain a sufficient volume of sample to perform both WET testing and appropriate chemical analysis in the event that the test demonstrates toxicity. Although these additional effluent samples cannot be assumed to be replicates of tests performed on the original sample, they can be used for assessing toxicity persistence and the presence of probable persistent toxic agents.

The permittee may initiate a TRE or the TRE may be required by a regulatory authority when the permittee cannot adequately explain and immediately correct a toxicity problem. For example, when a

permittee cannot readily resolve a toxicity problem, more frequent monitoring can help define the period of noncompliance, thus possibly limiting the duration of the liability for noncompliance. Permittees should independently undertake initial TRE procedures (e.g., additional toxicity testing) whenever a toxic discharge situation exists, whether or not a limit is in effect. Regulatory agencies can require a permittee to conduct a TRE through (1) NPDES permit requirements (e.g., WET monitoring requirements and special conditions of the permit requiring a TRE triggered by observation of unacceptable toxicity); (2) an information request letter issued pursuant to Clean Water Act (CWA) section 308 or analogous state (or tribal) authorities; or (3) an administrative order or consent decree pursuant to authorities in section 309 of the CWA or analogous authorities of state(s) or tribe(s) authorized to administer the NPDES program.

## **CONDUCTING A TRE**

A TRE may be implemented in several ways. A TRE is not necessarily a long-term study, and it does not necessarily require extensive research. Any activities that result in consistently reducing toxicity to an acceptable level may be considered TRE activities. Such activities could include a review of onsite chemicals, chemical use, and housekeeping practices for any permittee determined to be in noncompliance. Some toxicity problems have been resolved by relatively simple means (e.g., sweeping instead of washing down a process area). In other cases, toxic chemicals used in everyday procedures (e.g., toxic flocculent polymers) may be replaced with less toxic compounds (for additional guidance, see USEPA 1989a,b; 1999a). In some cases, correcting or changing chemical use and operational procedures in manufacturing or wastewater treatment processes has an added benefit of cost savings.

EPA recommends that permittees develop a basic TRE strategy before a rapid response to toxicity, such as a complex TRE/TIE or other regulatory action, is needed. The strategy should include a summary of baseline information, including operational efficiencies; evaluation of potential TRE consultants; and a thorough review and update of chemical inventories, chemical uses, and input points for potential effluent toxicants.

TREs usually are triggered by a demonstration of toxicity at an unacceptable level, as defined in the NPDES permit (e.g., a monitoring requirement). Upon demonstrating the need for a TRE, the permittee should immediately undertake two actions, regardless of whether they are required in the permit: (1) temporarily increase the testing frequency for the affected species; and (2) collect additional samples for possible chemical analysis. The testing frequency increase may be necessary to determine if the toxic event is an isolated incident or if a toxicant is being discharged on a regular basis. When an increase in test frequency is required by the regulatory authority, the number of tests required and the period of time over which the tests are to be conducted are at the regulatory authority's discretion. The sample volume collected should be increased to ensure that sufficient sample is available to perform toxicant identification on all samples used for WET testing.

While some alteration of toxicants may occur during storage, every effort should be made to begin TIE analysis as soon as it is apparent that a sample is demonstrating toxicity during the baseline (or scheduled) test. The additional sample volume may be appropriately stored onsite for shipment to laboratories for further testing if toxicity is found. Follow-up chemical analyses of stored samples may be effective only for persistent chemicals or those with persistent characteristics. Results of these follow-up analyses should be

interpreted with caution because the toxicants that are present in the later tests may differ from those causing toxic effects in the original sample. When a suspect toxicant is identified, future samples should always be analyzed for that toxicant and the sampling results should be reported to the regulatory authority. If the toxic event is an isolated incident, demonstrated by lack of toxicity in retesting over an extended period (e.g., during the next three months of accelerated testing), the TRE may be discontinued if the regulatory authority is satisfied that an additional occurrence is unlikely.

The TRE should have a defined start date, plan of execution, and proposed completion date. The regulatory authority and the permittee should meet at the outset of the TRE to identify and understand each other's expectations and limitations. EPA recommends a similar meeting toward the conclusion of the TRE to discuss findings and follow-up. The regulatory authority should be available for consultation at any point in the process.

The regulatory authority typically monitors implementation of the TRE through progress reports submitted by the permittee. EPA recommends that progress reports be submitted at least quarterly. The reports should provide all WET and chemical test data, studies, and other TRE-relevant information that have been developed in the preceding quarter. The regulatory authority is responsible for reviewing TRE progress reports to ensure that data and results submitted by the permittee are reasonable and expected to lead to successful and timely conclusion of the TRE. Therefore, the permittee should document its TRE plan, including expertise used to design and execute the TRE. The permittee is responsible for ensuring that the TRE results in the reduction or elimination of effluent toxicity to the level necessary to prevent adverse in-stream impacts.

The permittee should be allowed an adequate period of time to find several toxic samples to characterize toxicity. A permittee may request and receive permission to terminate the TRE after demonstrating that the toxicity has been adequately characterized and appropriate controls have been selected and properly implemented, resulting in a reduction or elimination of the toxicity. If additional time is needed and technically warranted, the TRE may be extended for a reasonable period of time. At a minimum, the TRE plan should include the WET (and any suspect chemical) testing requirements, testing frequencies, and reporting requirements established by the regulatory authority. For example, full-scale quarterly chronic tests may be required, with low-cost streamlined tests using only the dilution of concern (or 100 percent effluent) and a control during those months when quarterly tests are not otherwise required. This approach allows more frequent testing for a small increase in cost and significantly increases the opportunity of capturing toxic events.

Because many permittees have had more experience with chemical-specific samples, there is a tendency to address WET immediately by analyzing a broad spectrum of chemical parameters (e.g., priority pollutant analyses). Experience indicates that this untargeted approach frequently yields poor results. Unless the potential risks are obviously chemical in nature, EPA recommends that the toxicological methods (i.e., TIE or treatability) be used as the primary means for reducing toxicity.

## REDUCING TOXICITY

Toxicity may be addressed by implementing one of two principal approaches: (1) a TIE or (2) treatability studies. The success of TIEs depends on obtaining an adequate number of toxic samples. EPA recommends that toxic samples be identified using TIE procedures (USEPA 1988, 1991b, 1992, 1993a, 1996) and confirmation testing (USEPA 1993b). Confirmation of the toxicant includes demonstrating that the level of toxicant is high enough to cause the demonstrated level of toxicity at the dilution of concern. Once the toxicant has been identified, levels of the suspect toxicants should be monitored in all future test samples to confirm that similar toxic effects are demonstrated when similar toxicant levels are found. Toxicant reduction at the source should be considered preferable to treating toxicity, but such decisions are heavily influenced by cost factors, energy consumption, and product viability. A TIE and any toxicity treatability tests should be considered early in the TRE process. Regulatory agencies may require TIEs on a case-by-case basis using information-gathering authorities. EPA has developed guidance on how to perform a three-phase TIE, but it does not generally require that these procedures be used (see USEPA TRE and TIE guidance documents listed in the references). Neglecting to identify and control any toxicants responsible for WET test failures, however, may result in a WET permit limit, even if no limit was previously established.

The permittee may choose to use a treatability approach rather than the toxicant identification approach to reduce toxicity. In this case, a WET limit would still be warranted [pursuant to 40 CFR § 122.44 (d)(1)(v)], if the toxicant is not identified, confirmed, and reduced or eliminated through implemented controls. If toxicity is reduced through a treatability test but not identified, a WET limit is required because the specific compound(s) or operational practice(s) that led to toxicity cannot be determined or controlled, and toxicity may recur.

The decision to pursue either the source reduction or treatability path for resolving WET problems depends on the site-specific costs and benefits of both approaches. Permittees and regulatory agencies should discuss the benefits and burdens of both choices. Identification and confirmation of toxicants may lead to chemical-specific limits rather than a WET limit, whereas toxicity reduction using a treatability approach generally results in a WET limit. As noted, a suite of procedures should be used to achieve the most effective and efficient results for each permittee.

When using either the TIE or treatability approach, the toxicity should be reduced to a level that ensures compliance with NPDES permit requirements and attainment of water quality standards. Although a permit may contain chemical-specific limits for some compound(s) and the effluent is meeting those limits, these limits may not be stringent enough to preclude unacceptable levels of toxicity in the WET test. This condition may be due to additive or synergistic effects of compounds or localized conditions of the receiving stream, such as hardness and/or pH. In such cases, imposing WET limits or more stringent chemical-specific limits may be necessary to ensure compliance with numeric WET criteria or narrative toxicity criteria.

## TECHNICAL LIMITATIONS OF THE TIE PROCEDURES

EPA believes that two factors most frequently affect the technical success of characterizing, identifying, and confirming the cause(s) of toxicity: (1) past experience with successful TIE completion; and (2) degree of wastewater complexity and variability (Ausley et al. 1998). Another cause for unsuccessful TIEs is attempting to cut costs by eliminating portions of the TIE effluent characterization process. Permittees conducting TREs and TIEs should closely scrutinize the experience and past successes of potential consultants to ensure that a fully qualified team is conducting the evaluation. Laboratories should demonstrate past experience and clearly show timeliness of their results in proposals. Laboratories should have access to individuals from various disciplines, including aquatic toxicology, chemistry, and wastewater engineering. Throughout the TRE/TIE, permittees should continue to evaluate progress, and, if necessary, make appropriate corrections. Insufficient progress in the TRE/TIE effort should lead to the selection of alternative methods or consultants, or both.

The difficulty in completing a successful TIE, as well as the amount of time required, will likely increase in proportion to the complexity of toxicants in the wastewater. As the number of chemical constituents in the wastewater increases, the interactions of those chemicals (with both biological and analytical systems) compound the difficulty in identifying toxicants. When multiple toxicants are present in a sample, identifying and resolving the toxicants serially may be necessary due to masking or confounding influences. Additionally, the difficulty and complexity of a TIE may increase as the toxicity magnitude and frequency decrease (less toxic, less often). Conversely, as the toxicity magnitude and frequency increase (more toxic, more often), wastewater components causing toxicity may become more readily identifiable. This conclusion has been effectively supported by the research of McCulloch et al. (1998), who found direct relationships between increasing toxicity frequency and magnitude and increased success in toxicant characterization and identification. Despite the potential difficulties, successful TIE completion is generally possible if proper TIE procedures are conducted by trained and experienced staff.

Since EPA published the first editions of the TIE procedures (USEPA 1988, 1989d, 1989e), the commercial, academic, regulatory, and regulated communities have gained additional experience with TIE procedures. These procedures are used routinely and successfully in resolving unacceptable levels of toxicity. To update and provide broad access to these procedures, the Society of Environmental Toxicology and Chemistry (SETAC) is developing a summary report highlighting any TIE procedures that have evolved over the past decade and conducting a comprehensive literature search of TIE-related publications. When SETAC has completed and finalized the summary report and literature search, the information will be available on the Internet at <http://www.setac.org>.

Although conducting chronic toxicity tests and chronic TIE procedures may be more expensive and time-consuming, Ausley et al. (1998) reported no significant distinctions between the methodology or ability of acute and chronic toxicity tests (if properly conducted) to identify toxicants and resolve toxicity issues. Acute and chronic TIE procedures are reliable means of resolving toxicity issues and lead to appropriate controls. The greater time required to conduct chronic TIEs should be considered in evaluating an appropriate compliance schedule.

## CONSIDERATION OF PERSISTENCE AND MAGNITUDE OF TOXICITY EVENTS

Whenever WET monitoring indicates toxicity, initial TRE activities should be instituted by the permittee, whether or not the NPDES permit contains WET limits or TRE requirements. The persistence and magnitude or degree of toxicity, however, should be a primary factor driving the specific procedures that are part of a TRE and the timing of those procedures. Initial TRE activities should include accelerated testing (e.g., weekly testing) in conjunction with an in-plant survey to identify potential causes of toxicity related to changes in processes or chemical use. When WET limits are included in permits, the permittee should pursue methods of reducing toxicity until WET testing indicates that permit limits are being met. Guidelines for initiating TRE activities are identified in the TSD (see Section 5.8.3).

For a TIE to successfully identify and confirm toxicants as part of a TRE, toxicity must be present in a sample. Therefore, increasing the sample volume and testing frequency for the affected test species to obtain samples with sufficient toxicity is prudent to ensure a successful TIE. Expecting a TIE to immediately follow a single or infrequent event of WET noncompliance is unrealistic. Rather, the persistence (duration, frequency) and magnitude of components of the exposure should be characterized immediately through additional testing to evaluate whether a TIE would help reduce toxicity. A TIE should be initiated as soon as toxicity is observed in such follow-up analyses.

Whenever WET monitoring indicates toxicity, EPA recommends additional testing at least once per month for six months. Testing also could be as frequent as once per week for at least two months. During this accelerated testing phase, if more than one sample demonstrates an unacceptable level of toxicity, the facility should evaluate plant housekeeping and conduct TIE characterization steps. When intermittent or marginal toxicity is found, the permittee should routinely obtain and store adequate samples to perform both WET tests and TIE procedures. The permittee should be required to perform sufficient sampling to ensure that toxic samples are obtained in adequate amounts to conduct TIE procedures or treatability tests on the toxic samples. Note that holding time may affect sample toxicity. If samples are held beyond the maximum holding times specified in the WET methods, the potential for toxicity degradation should be factored into the interpretation of the results (e.g., USEPA 1991b, 1992).

An alternative to performing a TIE is to conduct treatability tests that use bench-scale treatment units to identify process changes that reduce toxicity through changes in treatment type, arrangement, or method. These tests, however, may not identify which toxicant is being removed or reduced. As with TIE procedures, toxic samples are necessary to perform treatability tests successfully. Therefore, adequate sampling frequency is required to capture toxic samples. Toxic compounds may be oxidized, reduced, complexed, flocced, or trapped in sludges, resulting in a less toxic discharge. Although treatability tests have been successfully used at publically owned treatment works, these tests have several drawbacks. For example, bench-scale reactors do not always provide an accurate prediction of what will happen in the full-scale plant, and permittees could spend significant amounts of money on inappropriate treatment. (See the attached reference list for documents on treatability guidance and case examples.)

## **IONIC IMBALANCE OF EFFLUENTS**

EPA's TSD (USEPA 1991a) recognizes the presence of excessive levels of total dissolved solids (TDS) in effluent, which can cause an ionic imbalance, as pollution. Several studies have produced a better understanding of the role of ionic imbalance in WET, which has led EPA to confirm its earlier conclusion that wastewater discharges that create physiologically intolerable conditions for sensitive aquatic species should generally be considered toxic. Toxicity caused by TDS or an imbalance of ions can be characterized through various published methods including, for example, the EPA TIE procedures (USEPA 1991b; 1992; 1993a,b; 1996), Ingersoll et al. (1992), McCulloch et al. (1993), Gorsuch et al. (1993), Tietge et al. (1994), Goodfellow et al. (2000), Pillard et al. (2000), and Kline and Stekoll (2000).

A significant imbalance in the ionic content of wastewater relative to receiving streams creates conditions harmful to aquatic populations (Goodfellow et al. 2000, Ausley et al. 1998). This is true whether single ions are excessive, major ions are imbalanced, or even if physiologically necessary ions are lacking or imbalanced. EPA's opinion is that these characteristics, if intolerable to resident aquatic populations, are toxic. Industrial processes that typically produce high ionic-strength effluents include textile manufacturing (salts used in dye leveling), pickling (brines used for preserving), chemical production and metal finishing (chemical manipulation of pH), and boiler blow down (concentration by evaporation). Drinking water filtration facilities and other facilities that use reverse osmosis also produce high TDS waste streams. Waste streams with very low ionic concentrations can come from cooling water, condensate discharge, and contaminated groundwater remediation. In some cases, a TDS imbalance in the discharge may result solely from an ionic imbalance in the source water. For example, if a facility obtains its source water from the same stream to which it discharges, and the source water has a toxic ionic imbalance therefore causing the discharge to be toxic, regulatory authorities should take this into consideration when evaluating the permittee's compliance status.

In assessing potential toxic effects of the ionic makeup of wastewater, making appropriate assumptions regarding dilution and using appropriate test species are essential. Regulatory agencies should consider appropriate dilution scenarios for each discharge (i.e., when allowed under state regulations or applicable water quality standards) to determine the appropriate degree of mixing before measuring the acute and/or chronic effects of the discharge. The TSD (USEPA 1991a) provides guidance on determining these dilution scenarios. Although choices of test organisms to assess toxicity in TIEs may vary, the species tested to assess compliance with WET limits of the NPDES permit should be surrogates of equivalent sensitivity to the EPA-approved species appropriate for the resident community (i.e., marine and estuarine species in a marine and estuarine environment and freshwater species in a freshwater environment).

## **PERMIT AND ENFORCEMENT MECHANISMS TO IMPLEMENT A TRE**

When WET testing shows continued toxicity or toxicity at unacceptable levels, NPDES permits should require an increase in the WET testing frequency for affected species. A TRE may not be necessary as a permit requirement when the permit contains WET limits. When a WET limit is in effect and has been exceeded, the permittee should meet with the regulatory authority to establish a plan and schedule for returning to compliance, similar to the practices currently in use for other permit limit violations. When the permit includes WET monitoring but does not include limits on WET, EPA believes it is appropriate to



include permit conditions that require the permittee to submit a plan and schedule to address toxicity if toxicity is demonstrated by WET monitoring. In addition, the permitting authority may reopen the permit to include WET limits, if additional monitoring indicates toxicity and the need for WET limits. For permits without WET limits but with WET monitoring requirements, EPA recommends increasing WET testing frequency for the affected species upon demonstration of toxicity to at least one WET test per month for at least three months to determine whether toxicity is consistent or intermittent.

EPA supports both TIE and treatability as acceptable approaches to resolving toxicity. For this reason, EPA does not believe specifying a particular methodology for TREs is necessary (e.g., TIEs or treatability testing). EPA recommends identifying the specific toxicant through TIE analysis for several reasons. First, this approach allows the permittee to reduce toxicants at the source rather than treat and pass them to the environment. Second, this approach allows for the possibility of a chemical-specific limit to be used in the permit in lieu of a WET limit in which a narrative toxicity criterion applies. Finally, if treatment is necessary, the specific treatment to control the toxicant can be implemented, resulting in a significant cost savings to the permittee.

As noted above, a TRE may be initiated by a permittee or required by a regulatory authority when the permittee cannot adequately explain and immediately correct a toxicity problem. Regulatory agencies can require a permittee to conduct a TRE through permit requirements, a CWA section 308 letter, or an enforcement action. When allowable under applicable water quality standards, the regulatory authority can establish a compliance schedule for a WET limit as part of the permit. This compliance schedule also allows the permittee more time to collect additional WET monitoring data, to implement or continue a TRE to resolve its toxicity problem, and to come into compliance with a WET limit.

The permittee must comply with all conditions of its NPDES permit. If a permittee violates the permit, the permittee is subject to enforcement action and is expected to return to compliance as soon as possible. When a permit contains a WET limit and the permittee violates the limit, EPA's 1989c national "Enforcement Management System" recommends an immediate, professional review of that violation, which may or may not result in a formal enforcement action. EPA's national enforcement guidance also recommends an escalating enforcement response to continuing violations of any parameter. The enforcement response depends on the circumstances surrounding the violation and can range from an informal action, such as a phone call or warning letter, to a formal administrative action (with or without a penalty), such as a civil judicial enforcement action. EPA considers factors such as nature, severity, and frequency of the violation; human health and environmental impacts; and compliance history of the facility when determining an appropriate enforcement response.

An administrative order requiring a permittee in violation of a WET limit to conduct accelerated WET testing is one enforcement mechanism used to implement a TRE. The administrative order typically would include requirements to implement corrective actions identified by the TRE and to comply with the permit WET requirements by a certain date, according to a schedule established in the order. The enforcement authority also may initiate an administrative penalty order or a civil judicial action if the permittee's WET violations cause known environmental harm or if the permittee neglects to complete routine or accelerated WET testing.

## INCONCLUSIVE TREs/TIEs

EPA's 1989c *Whole Effluent Toxicity Basic Permitting Principles and Enforcement Strategy* states that "In a few highly unusual cases where the permittee has implemented an exhaustive TRE plan, applied appropriate influent and effluent controls, maintained compliance with all other effluent limits, compliance schedules, monitoring, and other permit requirements, but is still unable to attain or maintain compliance with toxicity-based limits, special technical evaluation may be warranted and civil penalty relief granted. Solutions in these cases could be pursued jointly with expertise from EPA and/or the States as well as the permittee." EPA's WET control policy reiterates this statement (USEPA 1994c). EPA and authorized states have already participated in such cases and demonstrated their willingness to work with permittees to arrive at reasonable and protective solutions. Below are some specific examples:

- EPA's laboratory in Duluth, Minnesota, has helped characterize effluents from the private and governmental sectors in several cases where commercial laboratories have been unable to determine the cause of toxicity. Other federal laboratories have developed new or modified procedures to address toxicity attributable to TDS. At the same time, when the permittee has demonstrated that it is aggressively pursuing the reduction or elimination of toxicity, enforcement discretion has enabled the permittee to concentrate greater efforts toward resolving the problem.
- In North Carolina, intensive TIE analyses indicated a key process compound in an industrial wastewater discharge was problematic. Providing a substitute for the compound in the manufacturing process without negatively affecting the facility's product was not possible using available technology. After the facility took progressive steps to identify the toxicant, it was allowed regulatory relief under an agreement to fund industry-wide research into less toxic substitute compounds and the technology the facility needs to operate properly. Resolution in this case was directed at reducing WET for one facility, while extending the possibility of toxicity reduction to the industry as a whole.
- EPA has worked with local governments and developed public education programs to inform citizens about pollution prevention where organo-phosphate pesticide use has been found to cause toxicity in publically owned treatment works. When public education was used aggressively, publically owned treatment works were able to demonstrate compliance with their NPDES permit limits. When public education programs ended, in some cases, WET testing detected the toxicity again.

TIEs that fail to characterize toxicants effectively frequently do so for one of two reasons (Ausley et al. 1998). The first is the inability of inexperienced individuals to interpret results correctly and to follow observed clues through to identification and confirmation. The second is difficulty in applying TIE techniques to samples with intermittent toxicity or with toxicity caused by changing toxicants. Effluents with these characteristics pose challenges even to experienced analysts. These cases present the best possible argument for developing a comprehensive TRE plan that emphasizes stabilizing and optimizing process and treatment options at a facility to minimize "moving" targets.

TREs that fail to resolve noncompliance with WET limits or TREs that do not meet established targets (e.g., an action level for the permit when the permit does not have a WET limit) are incomplete and should continue until all goals are met. Also, inconclusive TIE results should lead investigators to seek assistance from others with additional experience, such as experts at EPA Headquarters or Regional offices or state or local experts. Although alternative approaches are generally site-specific and may be considered trade secrets, some laboratories and trade organizations may share some techniques.

## CONCLUSIONS

TREs and TIEs have been used to successfully resolve toxicity under the NPDES program. Although regulatory authorities can request that a permittee conduct a TRE, permittees should consider initiating preliminary TRE procedures to reduce or eliminate toxicity whenever a toxic discharge occurs that results in noncompliance. Although TIEs are not required to be part of a TRE plan, TIEs should be considered early in the TRE process because EPA has found that TIEs have been highly successful in resolving WET when aggressively implemented. While more difficult to detect, intermittent toxicity may be addressed by increasing WET testing frequency and ensuring that sufficient sample volumes are taken to perform all the necessary TIEs.

Each TRE requires case-specific investigative measures and solutions. Therefore, the permittee should consider all appropriate options when developing and implementing a TRE. For example, a TRE need not be a long-term study or require extensive research. Any activities that result in consistently reducing toxicity to an acceptable level may be considered TRE activities.

Once a permittee reduces toxicity to an acceptable level (or eliminates it) and maintains compliance, the TRE goal has been met. The TRE then may be discontinued, if appropriate, and the routine monitoring schedule, if included in the permit, resumed. Enforcement decisions should be guided by case- and site-specific consideration of existing and historical toxicity, including toxicity magnitude, duration, and frequency, and the permittee's diligence in resolving and preventing WET noncompliance.

Finally, permittees and permitting authorities should establish early in the TRE process a cooperative and communicative relationship that should be maintained until the TRE is successfully completed and any necessary controls are fully implemented. Good communication and a well-conceived TRE plan can ensure that all parties understand the requirements and expectations and can result in a more effective and faster resolution of the observed toxicity.

Additional sources of information on conducting TREs/TIEs are listed in the reference section of this document. The names of Headquarters and Regional contacts for EPA's Water Quality Program can be obtained on the Internet at <http://www.epa.gov/npdes>. The names of SETAC's Expert Advisory Panel on WET are listed at <http://www.setac.org/wetPanels.html>. Also, permittees should stay in close contact with their Regional or state regulatory authority when working through the TRE process.

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