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M E M O R A N D U M

***** N O T I C E *****

This document has been developed to provide Department staff with guidance on how to ensure compliance with statutory and regulatory requirements, including case law interpretations, and to provide consistent treatment of similar situations. This document may also be used by the public to gain technical guidance and insight regarding how the department staff may analyze an issue and factors in their consideration of particular facts and circumstances. This guidance document is not a fixed rule under the State Administrative Procedure Act section 102(2)(a)(I). Furthermore, nothing set forth herein prevents staff from varying from this guidance as the specific facts and circumstances may dictate, provided staff's actions comply with applicable statutory and regulatory requirements. This document does not create any enforceable rights for the benefit of any party.

Revised
3/22/07

TO: Regional Water Engineers, Bureau Directors, Section Chiefs

SUBJECT: Division of Water Technical and Operational Guidance Series (1.3.2) ACUTE AND CHRONIC TOXICITY TESTING IN THE SPDES PERMIT PROGRAM
(Originator: Edward Kuzia/Nicole Wright)

I. PURPOSE

This guidance document describes the procedures which should be followed when determining whether to include toxicity testing in a SPDES permit and how to implement a toxicity testing program.

II. DISCUSSION

A toxicity test includes the following:

1. Exposure of test organisms to a representative effluent sample appropriately diluted with upstream water, along with necessary controls and quality assurance.

2. Observations on the effect of the exposure to the test organisms and calculation of certain parameters to define the toxicity of the effluent.

3. Conclusions as to whether the effluent, when diluted with the receiving water, may cause unacceptable harm to aquatic life.

Detailed descriptions of the tests and how the results should be interpreted are included in the Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms Fifth Edition, EPA-821-R-02-012 (2002), Short Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Freshwater Organisms Fourth Edition EPA-821-R-02- 013 (2002), and Short Term Methods for Estimating the Chronic Toxicity of Effluents to Marine and Estuarine Organisms Third Edition, EPA-821-R-02-014(2002). The instructions in these manuals must be followed carefully by the permittee or contractor. The manuals are available from the U.S. Environmental Protection Agency (www.epa.gov/waterscience/WET).

III. GUIDANCE

Refer to the attached document as Acute and Chronic Toxicity Testing in the SPDES Permit Program.

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Sandra Allen
Director, Division of Water

Attachment

cc: TOGS Distribution Attached

TECHNICAL AND OPERATIONAL GUIDANCE SERIES 1.3.2

ACUTE AND CHRONIC TOXICITY TESTING IN THE

SPDES PERMIT PROGRAM

GUIDANCE

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ACUTE AND CHRONIC TOXICITY TESTING IN THE SPDES PERMIT PROGRAM

INTRODUCTION

Division of Water Technical and Operational Guidance Series 1.3.1 (TOGS 1.3.1), Total Maximum Daily Loads And Water-Quality Based Effluent Limits, includes guidance for determining when aquatic toxicity testing should be included in SPDES permits. The authority to require toxicity testing is in Part 702.16(b) of Chapter X, Title 6 of the New York State Codes, Rules and Regulations. This document now incorporates methods for determining and placing a whole effluent toxicity (WET) limit in a permit. This limit is based on performing a Reasonable Potential Determination (RPD) on WET data. The RPD is done to test if the effluent has the statistical probability to exceed a WET limit. If so a WET limit is placed in the permit. The purpose of toxicity testing is to ensure that no chemicals are discharged to surface waters in amounts toxic to aquatic life. The water quality engineer makes the determination to include toxicity testing in a permit based on any one of the following criteria.

1. The presence of substances in the effluent for which ambient water quality criteria do not exist. All facilities that have potential for discharging a chemical for which no criteria exist should be required to have effluent toxicity testing.

2. Uncertainties in the development of total maximum daily loads (TMDLs), wasteload allocations (WLAs) and water quality-based effluent limits (WQBELs), caused by inadequate ambient and/or discharge data, high natural background concentrations of pollutants, available treatment technology, and other such factors.

3. The presence of substances for which water quality-based effluent limits are below analytical detectability.

4. The possibility of complex synergistic or additive effects of chemicals. Toxicity Testing should be required when the number of metals or organic compounds discharged by a permittee equals or exceeds five. Even though the individual metals or organic compounds may be limited by the permit, the combined effects of all the metals and organic compounds may produce toxicity in the receiving water, and therefore should be evaluated by toxicity tests of the effluent.

5. Observed detrimental effects on the receiving water biota.

6. Toxicity testing done by DEC or EPA indicates a problem.

7. Waste treatment plants which exceed a discharge of 1 MGD. Facilities of less than 1MGD may be required to test. Municipalities which are managing industrial waste pretreatment programs should be considered for toxicity testing monitoring requirements. The number and type of industrial discharges to the municipal system should be reviewed in making a final toxicity testing monitoring determination. Effluent

toxicity testing at municipal facilities should be performed on wastewater prior to disinfection practices using chlorine.

The term "Whole Effluent Toxicity Testing" (WET) refers to the process whereby aquatic organisms are exposed to a control treatment of upstream receiving water and a geometric series of at least five effluent concentrations, with a dilution factor of not greater than 0.50. This produces an endpoint in percent effluent. The results of these tests are used to determine whether any toxicity exhibited is acceptable, given the Instream Waste Concentration (IWC) of effluent. All testing and monitoring is carried out by the permittee or its contractor. The sampling schedule for toxicity testing should conform as closely as possible to sampling for chemical analysis and coincide with such sampling whenever possible. A report of all testing activity and results is to be sent to the Department and should contain the specific data outlined in Appendix I. These include, but are not limited to discharge flow rate, temperature, dissolved oxygen, hardness, total alkalinity, specific conductance, pH, un-ionized ammonia and chemical/physical parameters which are limited in the permit. Testing is to be done according to the following: Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms Fifth Edition, EPA-821-R-02-012 (2002), Short Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Freshwater Organisms Fourth Edition, EPA-821-R-02-013 (2002), and Short Term Methods for Estimating the Chronic Toxicity of Effluents to Marine and Estuarine Organisms Third Edition, EPA-821-R-02-014 (2002), herein referred to as the EPA Manuals. These methods are listed in the Federal Register 40CFR Part 136.

Toxicity testing is a method of evaluating the potential for impact to aquatic life in the receiving water. If, after appropriate testing and monitoring, the tests determine the effluent has the potential for toxicity in the receiving water, a toxicity reduction evaluation (TRE) may be required. The requirement for the TRE is based on a 50 % frequency of test failure which suggest that toxic conditions may exist in the receiving water. Once toxicity is identified the TRE program is designed to identify the cause of toxicity and develop a program to eliminate the toxicity of the discharge. After the TRE is completed, a whole effluent toxicity limit may be placed in the permit to ensure continued compliance.

Only toxicity to aquatic life is addressed in this document. Other procedures, such as bioaccumulation testing, pose a number of scientific problems which remain to be solved before they can be used on a routine basis as part of a SPDES toxics control strategy. Also, protection of water as a drinking water supply cannot be assured through toxicity testing with aquatic life.

Toxicity Testing Requirements

Aquatic toxicity tests are used to determine if an effluent will be toxic in the receiving water. The toxicity testing strategy is based on two types of tests. Both tests use an invertebrate and a fish species. The Tier I test is a 48-hour acute test which measures lethality or immobilization effects. The Tier II test is a 7-day chronic test which measures reproduction, growth, and survival. The Tier I test is applied to measure acute toxicity at the edge of the acute mixing zone. In cases when the chronic dilution factor is greater than 10:1, the acute test can be used to measure the potential for chronic toxicity at the edge of the chronic mixing zone with the use of an application factor. The Tier II test should be used in all waters (except D and SD) when the chronic dilution factor is less than 10:1. This is because the acute test does not adequately predict chronic toxicity at low dilutions. The Tier II test may also be used to follow up on acute tests to more exactly determine the potential for chronic toxicity in the receiving water.

Review of toxicity test data may result in 3 primary outcomes:
1) Toxicity is predicted in the receiving water and a Toxicity Reduction Evaluation (TRE) is required.
2) Toxicity is not predicted in the receiving water, but statistical analysis of the test data predicts a potential for exceeding a toxicity limit, and therefore a WET limit must be established in the permit. This is termed a Reasonable Potential Determination (RPD).
3) Toxicity in the receiving water is not predicted and statistical analyses do not predict the potential to exceed a toxicity limit. After permit monitoring requirements are satisfied, further testing may be deferred until the next permit cycle.

Toxicity Reduction Evaluation (TRE)

A TRE is done when unacceptable toxicity is evident from the results of acute or chronic toxicity tests. The TRE includes a Toxicity Identification Evaluation (TIE) which determines the cause of toxicity and a plan to reduce the toxicity of the discharge once the toxic component is identified. The TRE should result in a WET limit or a chemical specific limit if a specific chemical is linked to the cause of toxicity. The TRE requirement will also include a WET limit recommendation in the Environmental Benefit Permit Strategy (EBPS), but this limit may be deferred if the permittee clearly demonstrates progress towards the removal of toxicity from the effluent. This recommendation may also be removed if the permittee demonstrates that the effluent has reduced toxicity below what is predicted by the RPD for requirement of a WET limit.

Reasonable Potential Determination (RPD)

A toxicity-based limit may be established for facilities without requiring a TRE. The difference between a TRE and an RPD is that a TRE is triggered by an identified toxicity problem, while an RPD is applied to determine if there could be a toxicity problem due to effluent variability. After four or more toxicity tests, the data may be analyzed to determine if a WET limit should be required in the permit due to effluent variability. The procedure for this statistical analysis, RPD, is presented in Appendix VI. EPA requirements in the Great Lakes Water Quality Agreement (GLWQA) require this analysis for Great Lakes Waters, but for

regulatory consistency it is being applied statewide.

Low Toxicity

When toxicity is not sufficient to require a TRE, or if the RPD does not trigger a requirement for a WET limit, after permit monitoring requirements are satisfied, no further toxicity testing may be required until the next permit cycle.

Determination of Effluent and Receiving Water Toxicity

With regard to aquatic toxicity, the goal of the SPDES permit program is to ensure that the effluent toxicity, after dilution with the receiving waters, is below the Instream Waste Concentration (IWC), as defined in glossary, that will have a detrimental effect on aquatic life (see calculations below). For D and SD water classes, this effect is an impact on fish survival. Toxicity testing for D and SD waters is discussed in Appendix V. All other water classes, which are the preponderance of NYS waters, must also be protected against an impairment on fish propagation. All Great Lakes Waters and their tributaries should be considered Class C for toxicity testing purposes. The highest concentration of effluent in the receiving water that will not impair its best uses is termed the Maximum Allowable Waste Concentration (MAWC). The MAWC is defined toxicologically as not exceeding 0.3 Toxic Units Acute (TUa) or 1 Toxic Unit Chronic (TUc) at the edge of the respective acute and chronic mixing zones (See TOGS 1.3.1 for discussion and definition of mixing zones).

Toxicity is reported in Toxic Units (TU), which are the reciprocals of acute(LC 50 or EC50) and chronic toxicity (NOEC or IC25) endpoints as percent effluent. The Toxic Units Acute (TUa) and Toxic Units Chronic (TUc) are calculated for Tier I and Tier II tests respectively as described below:

Toxic Units Acute (TUa)

$$TUa = 100/EC50 \text{ or } 100/LC50$$

where the EC50 is defined as the percent effluent concentration which causes an observable adverse effect (such as immobilization, or serious incapacitation) on 50% of the test organisms, or where the LC50 is defined as the percent effluent concentration which is lethal to 50% of the organisms tested. In all cases the endpoints of the more sensitive fish or invertebrate organism tested will be used to calculate the TUa.

Toxic Units Chronic (TUc)

$$TUc = 100/NOEC \text{ or } 100/IC25$$

where the No Observable Effect Concentration (NOEC) is defined as the highest tested concentration of an effluent at which no adverse effects on survival, growth, or reproduction are observed on the test organisms at a specific time of observation (e.g. 7 days) or where the Inhibition Concentration 25 (IC25) is defined as the effluent concentration that would cause a 25% reduction in a non-lethal biological measurement of the test organism such as reproduction or growth. The most sensitive endpoint of the more sensitive organism tested will be used to calculate the TUc. For protection of the survival of aquatic organisms, the actual TUa at the edge of the acute mixing zone must be less than or equal to 0.3 TUa. That is

$$TUa/\text{acute DF} + 1 \leq 0.3 \qquad (1)$$

Where TUa equals the Toxic Units calculated from the more sensitive test species in a 2 species effluent test, divided by the acute dilution factor +1 (DF+1). Equation 1 should be the basis for acute limits, if required, or action levels when an effluent is subject to discharge monitoring requirements.

The IWC is inversely proportional to the dilution factor +1 (DF +1) (Equation 2), which is the ratio of receiving stream flow to effluent flow at the point of mixing, or

$$\text{IWC (in \% effluent)} = 100/\text{DF}+1 \quad (2)$$

Dilution Factors

For rivers and streams, the flow used to calculate the chronic DF should be the MA7CD10 (as defined in glossary) flow. In these cases the dilution factor is the ratio of the effluent flow to the MA7CD10 flow. The flow of the effluent is incorporated into the dilution therefore DF+1. For non-stream receiving waters such as lakes, and ponds, where no site-specific data or models are available, a 10:1 dilution factor should be used as an allowable mixing factor (see T.O.G.S. 1.3.1).The acute dilution factor is usually 0.5 x chronic DF except in lakes, estuaries, and estuarine embayments where the acute and chronic DF are identical. In the Niagara and St. Lawrence Rivers, the acute DF and the chronic DF are 50:1 and 100:1 respectively. Lake, estuarine and marine discharges may have specific models to determine the acute and chronic mixing zones and their subsequent dilution factors. The water quality engineer should provide dilution data where models are used to the permit writer and the TTU.

For waters that must be protected for fish propagation, the TUc for an effluent can be determined directly by chronic life-cycle or partial life-cycle Tier II tests, as described in the EPA Manuals. For protection against chronic toxicity, the TUc at the edge of the chronic mixing zone must be ≤ 1. That is

$$\text{TUc/chronic DF}+1 \leq 1 \text{ TUc} \quad (3)$$

Where TUc equals the chronic toxic units calculated from the more sensitive species in a 2 species test. Equation 3 should be the basis for the chronic WET limits when required, or action levels for monitoring requirements in discharges to waters other than Class D and SD.

In the tiered approach described here, the Tier I acute test can be used to eliminate the requirements of a Tier II test if adequate dilution exists in the receiving waters. It should be assumed that 10 x TUa in the receiving waters will predict chronic effects in the receiving water. The chronic test requirement should still be in all permits (except those discharging to D or SD waters) even if the DF+1 is substantially >10:1. Consequently the permittee may be given the option to directly measure their chronic toxicity rather than estimate it from acute endpoints. The screening condition for determining the potential for chronic toxicity from acute data can be expressed as

$$10 \text{ x TUa/chronic DF}+1 \leq 1\text{TUc} \quad (4)$$

If the undiluted effluent causes an effect on less than 50% of the specimens (EC50 >100%), then no EC50 can be calculated from the test data. This means that the maximum TUa value on

the left-hand side of Eqn.4 is <1. For Eqn. 4, if the dilution factor+1 is less than or equal to 10, the equation cannot be used to determine whether the chronic toxicity in the receiving water is, or is not acceptable. When this situation occurs, it is recommended that chronic toxicity testing be done in the first instance because acute and chronic toxicity can be determined from the chronic tests. Potential for acute toxicity can be determined by calculating an LC50 from the chronic data directly. In class D waters in situations when the acute DF+1 is <3.3:1, the evidence for any degree of morbidity or mortality in the test specimens relative to the controls should be examined to decide if a Toxicity Reduction Evaluation by the permittee is required.

Overall Strategy for A, B, & C Waters

1. The Tier I test is a 48-hour acute test on both a vertebrate and an invertebrate species. It should follow the protocols described in the EPA Manual. The acute MAWC is 0.3 TUa at the edge of the acute mixing zone. The sampling schedule for toxicity testing should conform as closely as possible to sampling for chemical analysis and should coincide with such sampling when possible. In some cases, for instance, where range-finding acute tests or low dilution indicate the discharger may need to go to Tier II anyway, the permit writer may require Tier II testing in the first instance, or the discharger may elect to bypass the Tier I test and go directly to a Tier II test. In cases where the IWC exceeds 10% effluent, it is recommended that Tier II testing be considered first.

2. All surface waters (Class A, B, and C) must be protected for fish propagation. When the dilution is less than 10:1, one cannot assure, even with an EC50 or LC50 of 100% (TUa = 1), that toxicity to fish propagation will not occur in the receiving water (per Equation 4). "Any evidence of toxicity" includes any statistically significant differences in mortality or morbidity between the test specimens and controls. When the dilution is less than 10:1, the acute test (Tier I) does not allow chronic toxicity to be predicted from acute data; consequently, a Tier II test should be required..

3. If the TUa is less than 1 when the dilution factor is ≤ 10, chronic toxicity may occur in the receiving water (see discussion under 2 and Equation 4).

4. The Tier II test is a chronic test on both a vertebrate and invertebrate species. The test should follow the protocols described in the EPA Manuals. If Tier I tests demonstrate that either the fish or invertebrate species is clearly more sensitive to the effluent, the more sensitive of the two species may be used solely for the Tier II test upon written approval from the Department. TUc can be calculated from the Tier II test results and this should be ≤ 1 in the receiving water if the water is to be protected against chronic toxicity.(Equation 3)

5. If it is determined that the discharge is likely to be toxic based on the evaluation criteria (Appendix II), a Toxicity Reduction Evaluation may be required. WET Limits based upon Reasonable Potential Determination may have already required a toxicity-based limit. When an effluent is found to likely be toxic following application of the test evaluation criteria in Appendix II, a program to evaluate the source and nature of the toxicity should be required. This Toxicity Reduction Evaluation program should be developed by the

discharger and submitted, along with all the test data supporting the proposed program, to the Department for approval. The outcome of this evaluation should be the determination of a suitable pollutant monitoring parameter, and a plan to reduce toxicity, all of which must be acceptable to the Department.

6. When the appropriate toxicity tests indicate that toxicity is not present, there should be no need for a Toxicity Reduction Evaluation. However, when the permit is renewed or when modifications or changes are made to the permittee's operating procedures, further testing may be required by the Department. Generally if the initial reason for including a facility in the WET program remains, the testing requirement should remain on the cyclic basis originally established in the permit. (See example of calculations in a hypothetical case in Appendix III).

Guidance for Reasonable Potential Determination

The Reasonable Potential Determination (RPD) is applied to results of effluent toxicity data (minimum of 4 tests) to determine if a toxicity-based limit is required in the permit because of effluent variability. Toxicity testing should be placed in permits when chemical-based limitations are not adequate to regulate the discharge. A TRE will be required of the permittee if action levels or limits are exceeded at a 50% rate. During a TRE procedure, the limit may be deferred until the completion of the TRE to a time not to exceed five years from the initiation of the TRE. The permittees will be subjected to an RPD of their discharge data to determine if the effluent is likely to exceed an action level. This action level is based on a TUa of 0.3 and a TUc of 1 at the edge of the acute and chronic mixing zones respectively. If the RPD predicts an exceedance of the action level, the action level becomes the permit limit. Once the reasonable potential to exceed an action level is determined, the permit will be prioritized for modification (EBPS) and a WET limit (former action level) will be placed in the permit. In determining if a WET Limit can be established for an effluent, a situation may arise when the effluent has no acute or chronic toxicity. In cases when there is little dilution the multiplying factor may mathematically suggest a need for a WET limit. Since no toxicity in 100% effluent is essentially a non-detect, these data cannot be used to establish a WET limit. However, it is not recommended in these low dilution non toxic situations to drop toxicity testing from the permit because if any toxicity does occur in the discharge it may have serious impacts on the receiving water, and therefore the discharge requires monitoring. The action level would remain as such if the RPD indicated no need for a limit. The RPD multipliers are found in Table 1.

The evaluation for determining reasonable potential is as follows-

1. The action levels are determined for acute and chronic toxicity in the permit .

2. The permittee does a minimum of 4 tests in one year.

3. If no TRE is indicated, the data are subjected to an RPD.
   The most toxic result is identified.

4. NYSDEC Toxicity Testing Unit (TTU) uses the reasonable potential multiplier

appropriate for the number of tests run to determine if the action level (potential limit) may be exceeded by the permittee. The RPD is done after 4 WET tests are submitted. Any single WET test may determine the need for a WET limit.If 10 or fewer tests are done, the RP multiplier is taken from Table 1. If more than10 tests have been done the coefficient of variation is calculated and the mutiplier is taken from Table 3.2 in the Technical Support Document for Water Quality-based Toxics Control EPA/505/2-90-001 March1991.

5. If the action level is exceeded, the TTU recommends to the Permit writer that the permit be prioritized for modification to incorporate a WET limit into the permit. If the action level is not exceeded after application of the Reasonable Potential multiplier, no limit is required.

6. Note that all other requirements such as TREs apply. A limit may be deferred while the permittee is conducting a TRE. An example of how the data for a determination of reasonable potential appears in Appendix VI.

APPENDIX I

PERMIT REQUIREMENTS

Frequency of Testing Guidelines

Toxicity testing should be carefully tied into the known variability of plant processes. Generally, quarterly testing should be required when it is known that the effluent is fairly uniform over the course of the year. Where little is known about variability, or where it is known that the effluent is highly variable, a more frequent testing program should be required. When there are seasonal increases in effluent flow and/or expected loadings of waste, then the samples for toxicity testing should be taken during these critical times. Testing should range from weekly, for highly variable dischargers, to quarterly for more uniform discharges. Frequency might be high at the beginning of a permit period when little is known about the toxicity of the effluent and would be expected to decrease once the effluent has been shown to be non-toxic over a reasonable period of time.

Balanced against effluent variability should be the variability in the composition of the ambient water which may have an important influence on toxicity. In an attempt to test under varying ambient conditions, quarterly effluent toxicity monitoring is typically required.

If an individual toxicity test has failed, and the discharger believes the failure was due to an infrequent event such as a spill, the discharger should present evidence of this together with a plan for preventing future occurrences.

Persistence of Toxicity

In some cases, toxicity limits may be set to protect waters downstream from the discharge. In such situations, the persistence of the toxicity during the time the effluent takes to reach a critical downstream segment is important. Persistence can be measured in the laboratory or can be evaluated by instream measurements. The permittee is responsible for such determinations, and the results after evaluation by the Department, may lead to a modification in the toxicity limit, the testing procedure, or both. Methods for measuring persistence are discussed in the EPA Manuals.

Permit Requirement for Tier I and Tier II Tests

When a decision is made to require toxicity testing in a permit, the following should be specified: (See sample permit pages which follow for details.)

1. The test level is either Tier I (acute) or Tier II (chronic).

2. The reporting requirements.

3. The frequency of testing.

Considerable discretion is left to the Department regarding exactly what is to be included in each permit since the requirements will vary from one site to another. Some flexibility should

also be left for the discharger to tailor the tests to its particular situation. In some cases, a discharger may elect to by-pass the Tier I test and go directly to a Tier II test. In other cases, particularly where persistence of toxicity is questioned by the permittee, the permittee should be allowed to perform in-stream measurements. Evaluation of multiple acute (Tier I) tests may result in the requirement for a TRE proposal. The tests should follow the procedures described in the EPA toxicity testing manuals. For non-saline waters, freshwater species (*Pimephales promelas* and *Ceriodaphnia dubia*) are specified. For saline waters, marine organisms (*Cyprinodon variegatus* and *Mysidopsis bahia*) are specified. Other test species may be approved by the TTU. (see acceptable test species in the glossary). The saline waters are all S classified tidal waters including the lower Hudson River south of the northern Rockland and Westchester County lines.

Testing Program Changes

Where it has been determined that a permittee must proceed to chronic (Tier II) testing, the following language should be incorporated in the notification letter. These progam changes are at the recommendation of the Toxicity Testing Unit and sent to the permittee by the Regional Water Manager:

> "The review of Tier I toxicity test data submitted thus far indicates _____ of _____ tests submitted have failed after application of appropriate uncertainty factors. Therefore, Tier II (chronic) tests are necessary as required in Part I, page _____ of your SPDES permit. Test methods should follow those described in the EPA Acute and Chronic Manuals. Tests should be performed on a quarterly basis (4/year) commencing (date) and unless otherwise specified should be performed on a fish and an invertebrate species. A calendar quarter is a 3 month period. After one year (4 tests) has been completed, these data will be evaluated. Review of Tier II test data may result in the requirement for a TRE.

Toxicity Reduction Evaluation

If the TTU determines that a Toxicity Reduction Evaluation (TRE) study is necessary, the Toxicity Reduction Evaluation Compliance Schedule will be invoked by letter notification to the permittee from the Regional Water Engineer. The type and complexity of a TRE program is highly site dependent. The permittee will develop a TRE which should adequately answer the questions raised by the toxicity found in the effluent. It is recommended that TRE procedures developed by the EPA be used by the permittee, unless the permittee develops an alternative procedure acceptable to the TTU. The TRE study proposal should be submitted to the TTU within 45 days following the effective date of the letter notification. This schedule may also be used to set up a schedule for compliance with the recommendations resulting from the approved TRE study. The TRE should include test schedules, reporting requirements, and anticipated completion dates subject to approval of TTU. Specific guidance and procedures should follow, but not necessarily be limited to the documents cited as follows:

U.S. EPA .1988. Methods for Toxicity Identification Evaluations:

Phase 1, Toxicity Characterization Procedures (EPA/600/6-91/003);
Phase 2, Toxicity Identification Procedures (EPA/600/3-88/035);
Phase 3, Toxicity Confirmation Procedures (EPA/600/3-88/036).
Toxicity Identification Evaluation: Charaterization of Chronically Toxic Effluents Phase 1 (EPA/600/6-91/005F May 1992).

Sample Permit Language

Inststructions to permit writers for including toxicity testing in a permit, and sample permit language follow.

WHOLE EFFLUENT TOXICITY (WET) TESTING IN SPDES PERMITS

INSTRUCTIONS TO PERMIT WRITERS

(A) The previous WET Testing format has been discontinued in favor of including WET Testing requirements in the effluent limits table with the other parameters. Unlike the previous system in which WET Testing was "monitor only" and reported separately from DMRs, numerical Action Levels are now typically specified and results reported on DMRs. These changes should facilitate an improved understanding of WET Testing. Another consequence is a reduction in permit length by ~ 2 pages. If Reasonable Potential Determination demonstrates that permit limits are required, the action level becomes the permit limit.

(B) Action Levels will be provided by the Toxicity Testing Unit (TTU) or Water Quality Engineer who will also indicate the type of testing required (see C below) and the dilution factors. Action Levels are calculated as follows: Acute Action Level = (acute dilution factor +1) X (0.3); and Chronic Action Level = (chronic dilution factor +1). However, Acute Action Level = 0.3 if the dilution factor +1 <3.3. The acute dilution factor is generally ½ the chronic dilution factor +1 for streams and equal to the chronic dilution factor for lakes and estuaries. Results are reported in Toxic Units (TU) which are calculated as described in the permit footnote (see D below).

(C) There are four possible requirements:

1. "**Acute and if necessary Chronic**" is usually specified if dilution factor +1>10:1 and discharge is outside Great Lakes basin;
2. "**Chronic only**" is specified if discharge is to Great Lakes basin (all surface water classes) or outside Great Lakes basin if dilution factor +1 <10:1 (Class C or higher), including lakes with assigned dilution factor of 10:1;
3. "**Acute only**" is specified if the receiving water is Class D (unless tributary to a nearby stream of Class C or higher) except in Great Lakes basin (see 2 above); and
4. "**Simultaneous Acute and Chronic**" is very uncommon and additional special conditions may be provided by the TTU.

Acute test results are generated directly from both Acute and Chronic testing. Chronic test results are generated directly from Chronic testing or predicted from Acute test data using the formula in the permit footnote, with the exception of the "Acute only" test regime where it is not necessary to predict chronic results. Therefore in most instances, both Acute and Chronic test results should be reported and subsequently obtained either directly from the test reports themselves or calculated (predicted) as necessary.

(D) Whenever WET Testing is required, an explanatory footnote must be included in the permit. The permit writer must edit the footnote to specify permit specific information: test regime, fresh or marine test species, dilution ratios (provided by the WQ engineer), and monitoring period. Also, any red typeface permit writer instructions must be deleted. The permit writer will determine the monitoring period based on his or her knowledge of the facility and after consideration of any recommendations provided by the WQ Engineer or TTU. For the majority of the facilities, the preferred requirement is a minimum of one year of quarterly toxicity testing once every five years.

(E) Examples of each of the three common WET Testing scenarios are provided below. Note - these pages are not "stand alone" pages and are not to be inserted in permits, rather applicable table rows and footnotes should be cut and pasted into permits. Using this new format WET Testing is displayed in the same format as all other parameters in the PERMIT LIMITS, LEVELS AND MONITORING table.

(F) Questions on these procedures should be directed to the TTU or BWP.

EXAMPLES OF THE THREE COMMON WET TESTING SCENARIOS

PARAMETER	EFFLUENT LIMIT		PQL	MONITORING ACTION LEVEL		UNITS	SAMPLE FREQUENCY	SAMPLE TYPE	FN
	Monthly Avg.	Daily Max.	Daily Max.	TYPE I	TYPE II				
EXAMPLE #1 "Acute and if necessary Chronic" - discharge to a medium-size or large river where the site specific dilution factors + 1 are 20:1 for acute and 40:1 chronic.									
WET - Acute Invertebrate				6.0		TUa	Quarterly	seefootnote	1
WET - Acute Vertebrate				6.0		TUa	Quarterly	seefootnote	1
WET - Chronic Invertebrate				40		TUc	Quarterly	seefootnote	1
WET - Chronic Vertebrate				40		TUc	Quarterly	seefootnote	1
EXAMPLE #2 "Chronic only" - discharge to a small stream with a chronic dilution factor +1 <10. Note that for discharges to a lake, including Erie and Ontario, with 10:1 dilution the corresponding action levels would be 3.0 and 10. Note that for discharges to an intermittent stream of Class C or higher, or Class D in the Great Lakes basin, the corresponding action levels would be 0.3 and 1.0. Assume chronic df+1 for example #2 is 9									
WET - Acute Invertebrate				1.5		TUa	Monthly	seefootnote	1
WET - Acute Vertebrate				1.5		TUa	Monthly	seefootnote	1
WET - Chronic Invertebrate				9		TUc	Monthly	seefootnote	1
WET - Chronic Vertebrate				9		TUc	Monthly	seefootnote	1
EXAMPLE #3 - "Acute only" discharge to an intermittent Class D stream outside the Great Lakes basin where dilution factors are zero.									
WET - Acute Invertebrate				0.3		TUa	Monthly	seefootnote	1
WET - Acute Vertebrate				0.3		TUa	Monthly	seefootnote	1

The Footnote applicable to all WET Testing scenarios is provided on the following page.

WET TESTING FOOTNOTE TO BE INCLUDED IN EACH PERMIT WHERE WET TESTING IS REQUIRED

1. Whole Effluent Toxicity (WET) Testing:

Testing Requirements - WET testing shall consist of PICK ONE OF THE FOLLOWING 4 CHOICES AND DELETE THE OTHER 3 **Acute and if necessary Chronic / Chronic only / Acute only / Acute and Chronic simultaneously**. WET testing shall be performed in accordance with 40 CFR Part 136 and TOGS 1.3.2 unless prior written approval has been obtained from the Department. FOR DISCHARGES TO FRESH WATERS INCLUDE THE FOLLOWING TWO SENTENCES & DELETE THOSE APPLICABLE TO MARINE WATERS The test species shall be *Ceriodaphnia dubia* (water flea - invertebrate) and *Pimephales promelas* (fathead minnow - vertebrate). Receiving water collected upstream from the discharge should be used for dilution. FOR DISCHARGES TO MARINE WATERS INCLUDE THE FOLLOWING TWO SENTENCES & DELETE THE PREVIOUS TWO WHICH ARE APPLICABLE TO FRESH WATERS The test species shall be *Mysidopsis bahia* (mysid shrimp - invertebrate) and *Cyprinodon variegatus* (sheepshead minnow - vertebrate). Artificial salt water should be used for dilution. All tests conducted should be static-renewal (two 24 hr composite samples with one renewal for Acute tests and three 24 hr composite samples with two renewals for Chronic tests). The appropriate dilution series bracketing the IWC and including one exposure group of 100% effluent should be used to generate a definitive test endpoint, otherwise an immediate rerun of the test is required. WET testing shall be coordinated with the monitoring of chemical and physical parameters limited by this permit so that the resulting analyses are also representative of the sample used for WET testing. The ratio of critical receiving water flow to discharge flow (i.e. dilution factor) is _____:1 for acute, and _____:1 for chronic. Discharges which are disinfected using chlorine should be dechlorinated prior to WET testing or samples shall be taken immediately prior to the chlorination system.

Monitoring Period - WET testing shall be performed at the specified sample frequency PICK ONE OF THE FOLLOWING 4 CHOICES AND DELETE THE OTHER 3 OR IF APPROPRIATE CREATE UNIQUE REQUIREMENT **for the duration of the permit / during calendar years ending in ___ and ___ , beginning in January and lasting for a period of one full year / for a period of one full year beginning _____**.

Reporting - Toxicity Units shall be calculated and reported on the DMR as follows: TUa = (100)/(48 hr LC50) or (100)/(48 hr EC50) (note that Acute data is generated by both Acute and Chronic testing) and TUc = (100)/(NOEC) when Chronic testing has been performed or TUc = (TUa) x (10) when only Acute testing has been performed and is used to predict Chronic test results, where the 48 hr LC50 or 48 hr EC50 and NOEC are expressed in % effluent. This must be done for both species and using the Most Sensitive Endpoint (MSE) or the lowest NOEC and corresponding highest TUc. In cases when theacute dilution factor + 1 is less than 3.3, and there is statistically significant mortality in 100% effluent as compared to the control in acute tests, a TUa of 1is reported. Report a TUa of 0.3 if there is no statistically significant toxicity in 100% effluent as compared to control.

The complete test report including all corresponding results, statistical analyses, reference toxicity data, daily average flow at the time of sampling and other appropriate supporting documentation, shall be submitted within 60 days following the end of each test period to the Toxicity Testing Unit. A summary page of the test results for the invertebrate and vertebrate species indicating TUa, 48 hr LC50 or 48 hr EC50 for Acute tests and/or TUc, NOEC, IC25, and most sensitive endpoints for Chronic tests, should also be included at the beginning of the test report.

WET Testing Action Level Exceedances - If an action level is exceeded then the Department may require the permittee to conduct additional WET testing including Acute and/or Chronic tests. Additionally, the permittee may be required to perform a Toxicity Reduction Evaluation (TRE) in accordance with Department guidance. If such additional testing or performance of a TRE is necessary, the permittee shall be notified in writing by the Regional Water Engineer. The written notification shall include the reason(s) why such testing or a TRE is required. If an action level is not exceeded the Toxicity Testing Unit will perform a Reasonable Potential Determination to identify if a WET limit is required.

APPENDIX II

CRITERIA FOR EVALUATION OF TOXICITY TEST RESULTS

Test Failure

TOGS 1.3.2 outlines the criteria for failure of a single effluent toxicity test. The failure of a single test indicates the potential for toxic conditions to occur in the receiving waters and is cause for concern. However, the requirement for a permittee to increase the sensitivity of testing, or to conduct a toxicity reduction evaluation, should not be based on a single test result. A 50% failure rate during a one year period will usually result in a recommendation to proceed to Tier II tests or a TRE. Frequency and cause (if known) of test failure must be factored into any decision. Other factors, such as severity of toxicity reported or anticipated treatment improvements will also influence the course of action taken following review of toxicity test results. Nevertheless, the following guidelines should be applied in the evaluation of toxicity test results. Any significant variance from these general guidelines require the endorsement of the TTU.

1. Severity of Toxicity

When a chronic application factor of 10 results in a Tier I test failure 50% of the time, some judgement should still be applied in considering the appropriate response. For instance, this might occur if, after application of the factor, it is evident that the discharge is only slightly more toxic than that which would result in a passed Tier I test. Further, Tier II or TRE requirements may not be required if it appears that the discharge can meet Tier I requirements with corrective measures. However, Tier I tests should still be required to be passed after corrective measures are taken. Conversely, if LC50 values demonstrate high toxicity relative to IWC or if 0.3 TUa is exceeded in the acute mixing zone, then a more rapid response in the form of Tier II testing or TRE may occur. That is, a recommendation for Tier II tests or TRE may be made to the Region by the Toxicity Testing Unit without waiting for a full year's toxicity tests.

2. Other Considerations on Whether or Not Tier II or TRE is Required:

   a. Tier II or recommendation for TRE should be needed when:
      ° Confirmed unhealthy biota in receiving stream as listed in Priority Waterbody List (PWL).
      ° Probable productivity impacts upon stream biota which could be due to toxic discharge

   b. Tier II or recommendation for TRE possibly not needed when:
      ° Additional treatment planned (direct)
      ° Additional indirect (pre)treatment planned
      ° Production changes that would reduce discharge loading are planned

Test Passage

Toxicity testing requirements may be terminated by the Department if completed tests have demonstrated that the effluent is clearly non-toxic and/or can be best regulated by chemical-specific means. However, toxicity testing may be continued in a permit if, in the best professional judgement of the Department, toxicity testing remains an appropriate method for monitoring an effluent. Testing should remain in the permit if the reason for requiring testing in the permit still applies.

Although other factors may also warrant the continuation of toxicity testing, such continuation is normally based on the following conditions:

1. Failures have occurred, but at less than 50% frequency. Any single test failure may result in continued toxicity testing.

2. Chemical-specific criteria cannot be used to monitor the effluent. Examples of such conditions would be:

    a. Mixed complex effluents
    b. Analytical detectability problems for chemicals of concern

3. Factors determining the need for toxicity testing on page 2 of this document still apply to the SPDES permit under review.

APPENDIX III

EXAMPLES OF CALCULATIONS

Assume a plant discharged 10 MGD of wastewater into a river whose MA7CD10 is 4,000 cfs. The river is classified as B and toxicity testing is required.

Samples were collected quarterly for one year and tested using the Tier I 48-hour acute test with *Ceriodaphnia dubia* The most sensitive EC_{50}'s reported were 34, 36, 38 and 40%.

1. First calculate the dilution of the wastewater in the receiving stream. The cfs is converted to mgd using:

   mgd = cfs x 0.646

2. This gives the receiving stream flow as 4000 x 0.646 = 2584 mgd. The dilution factor for the discharge is then:

   DF = 2584/10 = 258.4

3. It is evident that the effluent will receive > 10:1 dilution i.e. DF > 10.

4. The most toxic EC50 is 34% or 2.9 TUa (100/34).

5. The condition for passing the Tier I acute test with respect to protecting for chronic toxicity is that the TUa x 10 /chronic DF + 1 ≤ 1

6. Substituting in the data 2.9TUa x 10/258.4 + 1 = 0.11

7. Therefore the acute test is projecting no more than 0.11TUc at the edge of the chronic mixing zone.

8 Since the most toxic acute test does not project chronic toxicity no chronic testing is necessary since 0.11 ≤ 1

9. The data must also be tested to determine if acute toxicity may occur at the edge of the acute mixing zone.

10. Since the acute mixing zone is defined as having one half the dilution of the chronic mixing zone, the DF = 129.2

11. The greatest toxicity of any effluent test was 2.9 TUa.

12. The condition for protecting against acute toxicity at the edge of the acute mixing

zone is that the TUa/acute DF+1 $\leq$0.3

13. Substituting the data 2.9TUa/129.2 + 1 = 0.02 TUa.

14. Since 0.02TUa is less than 0.3TUa no acute toxicity should be expected at the edge of the acute mixing zone.

APPENDIX IV

TOXICITY TESTING REQUIREMENTS IN SPDES PERMITS

(1) Water Quality Management Section (WQM) Bureau of Water Assessment and Management (BWAM) recommends toxicity testing in the permit.

(2) Testing and reporting requirements and action levels are written into permit or rejected by the Bureau of Water Permits (BWP) after consultation with the permit writer, Regional Office, WQM and Toxicity Testing Unit (TTU). TTU, assists with permit language as necessary.

(3) Permittee submits test results to Region, TTU, and Compliance Assurance Section(CA), Bureau of Water Compliance(BWC) for review. TTU responds to technical questions from permittee/consultant.

(4) TTU reviews test results and makes determination regarding further testing, permit limit or TRE; recommendations are sent to BWP and Region. Copies to BWC.

To Step 5a,b, or c

(5a) If TTU determination is made that no additional testing or TRE is required: BWP and Region are notified. If limit is required a recommendation is placed in the EBPS .

(5b) If region confirms TTU determination, that additional testing or TRE is required: Region invokes Tier II testing or TRE by letter notification to permittee.

(5c) If Region does not accept TTU recommendations, it resolves differences with TTU.

To Step 3 (additional testing) or Step 6 (TRE)

(6) TRE Plan submitted to Region and reviewed concurrently by BWP, Region and TTU with coordinated comments to permittee from Region.

(7) TRE approved or rejected by BWP. Schedule for implementation incorporated into modified permit by BWP and Region. BWP and Region monitor schedule and reporting requirements.

(8) Permittee conducts toxicity tests to demonstrate effectiveness of treatment.

To Step 4

BWAM = Bureau of Water Assessment and Management
BWC = Bureau of Water Compliance
BWP = Bureau of Water Permits
CA = Compliance Assurance Section
WQM = Water Quality Management Section
TTU = Toxicity Testing Unit
TRE = Toxicity Reduction Evaluation

APPENDIX V

TOXICITY TESTING REQUIREMENTS FOR CLASS D AND SD WATERS

The toxicity testing strategy is based on a simple, relatively inexpensive acute test (Tier I test). This 48-hour acute test, on both a vertebrate and invertebrate species, should be the only test required for Class D and SD waters where only fish survival is the best use. For D and SD waters, a Toxicity Reduction Evaluation should be required when the results of the Tier I test show that the best uses of the receiving waters will be contravened. The Toxicity Reduction Evaluation should require the permittee to locate the source of the toxicity and develop suitable monitoring and permit limit parameters. If toxicity can be traced to a specific chemical, then this chemical can be used as such a parameter. The permittee is also required to submit a plan to reduce its effluent toxicity for Departmental review.

For D and SD waters that must be protected for fish survival only, toxicity in the receiving water must not exceed 0.3 TUa. Therefore the 0.3 x [(DF/2) +1] must not be exceeded in any acute toxicity test performed on a discharge to class D waters. An example of how tests are evaluated follows below.

Four tests are performed with LC 50 values of 25%, 35%, 40% and 75 % for the most sensitive species tested. The most toxic LC50 value was 25% which equals 4 TUa (100/25) . The receiving water provides a DF of 30. The formula for determining the TUa in the receiving water is TUa/[DF/2)+1], and is calculated below.

$$4 \text{ TUa} /[(30/2)+ 1] = 0.25 \text{ TUa}$$

Since 0.25TUa is less than 0.3 TU this is a passing test . Because all tests would result in a TUa that is less than 0.3, there is no failed test in the example given above.

If an effluent action level or limit is required for the above discharge, it would be calculated by multiplying 0.3 x the [(DF/2)+1] or 0.3 x [15+1]. The acute effluent limit would be 4.8 TUa.

Appendix VI

REASONABLE POTENTIAL DETERMINATION

If an effluent exhibits some level of acute or chronic toxicity a Reasonable Potential Determination will be conducted to determine if the discharge may exceed the action level. If after application of the reasonable potential multiplier the action level is exceeded, a limit should be required in the permit. The multiplier is selected by determining the number of tests that the discharger has done, and then multiplying the results of the most toxic test results times the reasonable potential multiplier determined from Table 1 if 10 or fewer tests have been done. Once it has been determined that the permit has potential for exceeding action levels, the permit will be prioritized for modification and a limit be established. An example of a Reasonable Potential Determination is described herein.

1) The action level for TUc of the effluent has been set at 25 TUc. This is determined as follows: The chronic DF for this discharge is 24. A TUc of $\leq$1TUc must be met at the edge of the chronic mixing zone.. The action level is calculated by multiplying the (dilution factor +1) x 1. Therefore (24+1) x1= 25.

2) Assume that four chronic tests have been done on a SPDES discharge. The NOECs for the most sensitive species tested were 12.5 %, 25%, 50%, and 12.5 %.

3) The most toxic effluent sample were the two which produced an NOEC of 12.5%. Note that the lower the NOEC the more toxic the effluent sample.

4) The NOEC is then converted to toxic units using the equation 100/NOEC = TUc. Therefore 100/12.5 = 8 TUc.

5) Since 4 tests have been run the reasonable potential multiplier from Table 1 is 2.6. The equation for determining the reasonable potential is RPM(reasonable potential multiplier) x TUc) therefore 2.6 x 8 = 20.8.

6) The action level of 25 TUc is compared to the Reasonable Potential Determination of 20.8 TUc from the toxicity data. Since 20.8 is < 25 no limit is required for this discharge.

7) The reasonable potential for acute toxicity is similarly determined. A TUa of 0.3 must be maintained at the edge of the acute mixing zone. In the above case the acute dilution factor is one half of the chronic DF or 12.0.

8) The following LC 50 values determined from acute toxicity tests on the most sensitive species were 70%, 100%, 80% and 75%. Therefore the most toxic test resulted in an LC 50 of 70%. This results in a toxic unit value of 1.4TUa (100/70).

9) A value of 0.3 TUa must be met at the edge of the acute mixing zone. The action level set for this discharge is calculated as the [(chronic dilution factor/2)+1] x0.3 or [(25/2)+1] x 0.3 = 3.9.

10) Calculating the reasonable potential for acute toxicity for this discharge is done by using the reasonable potential multiplier of 2.6 for 4 tests (Table 1) and multiplying it by the TUa of the most toxic acute test (1.4 TUa) or 2.6 x1.4 = 3.6 TUa. Since 3.6 is < 3.9 (acute action level set in the permit), there is no need for an acute toxicity limit.

Table 1: Reasonable Potential Multiplying Factors: 95% Confidence Level and 95% Probability Basis (Source: Table 3.2, Technical Support Document for Water Quality-based Toxics Control EPA/505/2-90-001 March1991)

Number of Samples	Multipliers with Coefficient of Variation set at 0.6
1	6.2
2	3.8
3	3.0
4	2.6
5	2.3
6	2.1
7	2.0
8	1.9
9	1.8
10	1.7

For data sets with n> 10 the coefficient of variation (CV) is calculated from the data (CV = standard deviation/mean). Multiplying factor can then be determined from Table 3.2 as cited above in the Technical Support Document for Water Quality-based Toxics Control EPA/505/2-90-001 March1991.

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GLOSSARY

Aceptable Test Species - Freshwater Invertebrates: *Ceriodaphnia dubia, Daphnia magna,* and *Daphnia pulex*. Freshwater Fish: *Pimephales promelas, Oncorhynchus mykiss*, and *Salvelinus fontinalis.* Estuarine/Marine Invertebrate: *Mysidopsis bahia.* Estuarine/Marine Fish: *Cyprinodon variegatus, Menidia beryllina, Menidia menidia,* and *Menidia peninsulae.*

ACR - **A**cute to **C**hronic **R**atio or the ratio of the acute toxicity of an effluent to its chronic toxicity. It is used as a factor for estimating chronic toxicity on the basis of acute toxicity data, or for estimating acute toxicity on the basis of chronic toxicity data. Calculated as the LC50/NOEC for the same effluent. For the purposes of Reasonable Potential Determinations, EPA recommends an ACR=10.

Action Level - The level of toxicity in toxic units of a discharge which may cause unacceptable toxicity at the edge of the acute or chronic mixing zone. The action level is identical numerically to a WET limit, when required, of an effluent.

Acute toxicity - a relatively short-term lethal or other (e.g., immobilization, equilibrium loss) effect, usually defined as occurring within 48 hours or 96 hours

Alkalinity - a measure of the buffering capacity of a water

Ambient monitoring - all forms of monitoring conducted beyond the immediate influence of a discharge pipe, including sediment and biological sampling.

Bioaccumulation - chemicals are taken up by aquatic organisms from water directly and through consumption of food containing the chemicals.

Bioconcentration - chemicals are taken up directly from water with uptake exceeds excretion.

cfs - cubic feet per second

Chronic toxicity - effects are measured in terms of reduced reproduction, growth, and/or survival from life cycle or partial life cycle tests with aquatic organisms; see also *effluent toxicity testing*.

Classification or **stream classification** - a regulatory description of the designated use/best usage of a water.

Conductivity - a measure of the amount of dissolved salts in a water, based on the ability of an electric current to pass through a water sample.

Dissolved oxygen (DO) - a measure of the amount of oxygen gas dissolved in water. Fish and other aquatic organisms need dissolved oxygen to survive and propagate. A minimum concentration of 4 mg/l is generally necessary for survival of warm water fish species, while cold water species require a minimum of 5 mg/l.

EC50 - Median effect concentration or the concentration of effluent, expressed as a percent volume, that causes an adverse effect other than mortality to 50% of the exposed test organisms under the conditions of the test in a specified time.

Effluent toxicity test - a procedure to determine the toxicity of an effluent using living organisms. (usually juvenile fish or aquatic macroinvertebrates). A toxicity test measures the degree of effect on exposed test organisms of a specific chemical or effluent.

EPA - Environmental Protection Agency

Instream Waste Concentration (IWC) - the concentration of the effluent in the receiving water.

Hardness - the sum of the calcium and magnesium concentrations of a water expressed in terms of parts per million (mg/l) of calcium carbonate.

LC50 - Median lethal concentration or the statistically estimated concentration of effluent, expressed as a percent volume, that is expected to be lethal to 50% of the exposed test organisms under the conditions of the test in a specified time.

LOEC - **L**owest **O**bserved **E**ffect **C**oncentration or the lowest concentration of effluent, expressed as a percent volume, that causes a statistically significant adverse effect on the exposed test organisms as compared to the controls under the conditions of the test in a specified time.

Macroinvertebrates - the larger-than-microscopic invertebrate organisms that inhabit aquatic habitats; freshwater forms are primarily aquatic insects, worms, clams, snails and crustaceans.

MA7CD10 - minimum average 7-consecutive-day flow at a recurrence interval of 10 years.

MAWC - **M**aximum **A**llowable **W**aste **C**oncentration or the highest concentration of an effluent in the receiving water,\ expressed as a percent volume, that if maintained over time will cause no adverse biological impact. The acute MAWC is0.3 TUa and the chronic MAWC is 1.0 TUc.

MGD - million gallons per day

NOEC - **N**o **O**bserved **E**ffect **C**oncentration or the highest concentration of effluent, expressed as a percent volume, that causes no statistically significant effect on the exposed test organisms as compared to the controls under the conditions of the test in a specified time.

NYSDEC - New York State Department of Environmental Conservation

pH - a measure of the acidity (less than 7) or alkalinity (greater than 7) of a water sample on a scale of 0 to 14. Specifically, the logarithm of the reciprocal of the hydrogen ion concentration. The desirable pH of natural waters should generally fall within the range of 6.5 to 8.5.

RP - Reasonable Potential is the statistical potential to exceed an effluent limit.

RPMF - Reasonable Potential Multiplying Factor or a numerical value that multiplies the maximum observed effluent value expressed as TUs in an effluent data set.

SPDES - State Pollutant Discharge Elimination System

STP - sewage treatment plant.

Tier I testing - Acute toxicity testing

Tier II testing - Chronic toxicity testing

Total maximum daily load (TMDL) - is the sum of the individual wasteload allocations and load allocations. A margin of safety is included with the two types of allocations so that any additional loading, regardless of source, would not produce a violation of water quality standards.

TRE - Toxicity Reduction Evaluation; 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.

TUa - **T**oxic **U**nits$_{acute}$ is the reciprocal of the effluent concentration that causes 50% of the test organisms to die in an acute toxicity test. Calculated as 100/LC50 or 100/EC50.

TUc - **T**oxic **U**nits$_{chronic}$ is the reciprocal of the effluent concentration that causes no observable effect on the test organisms in a chronic toxicity test. Calculated as 100/NOEC.

Wasteload allocation (WLA) - is the portion of a receiving water's total maximum daily load that is allocated to one of its existing or future point sources of pollution.

Water quality based effluent limit - a limit which is necessary to protect the receiving water with regard to its mandated best usages.

Water quality standard - scientifically derived values based upon bioassays of aquatic organisms or estimated risks to human health and adopted pursuant to Title 6, Chapter X, Parts 700-705 of New York State Codes, Rules and Regulation.

Whole Effluent Toxicity (WET)- the toxicity of an effluent to aquatic organisms

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