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Welcome to this presentation on the United States Environmental Protection Agency's, hereafter USEPA, National Pollutant Discharge Elimination System, or NPDES, Whole Effluent Toxicity Permit Development. This presentation is part of a Web-based training series on Whole Effluent Toxicity, or WET, sponsored by the USEPA HQ's Office of Wastewater Management's Water Permits Division.

You can review this stand-alone presentation, or, if you have not already done so, you might also be interested in viewing the other presentations in the series, which cover the use of Whole Effluent Toxicity in the USEPA's NPDES permits program.

Before we get started with this presentation, I have two important housekeeping items.



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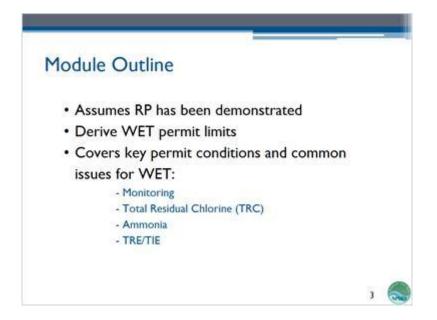
First, let me introduce myself. My name is Laura Phillips, and I'm USEPA's National WET Coordinator with the Water Permits Division within the Office of Wastewater Management at the USEPA HQ in Washington D.C. Second, now for those housekeeping items. You should be aware that all the materials used in this presentation have been reviewed by USEPA staff for technical and programmatic accuracy; however, the views of the speakers are their own and do not necessarily reflect those of USEPA. The NPDES permits program, which includes the use of Whole Effluent Toxicity testing, is governed by the existing requirements of the Clean Water Act and USEPA's NPDES permit implementation regulations. These statutory and regulatory provisions contain legally binding requirements. However, the information in this presentation is not binding. Furthermore, it supplements, and does not modify, existing USEPA policy and guidance on Whole Effluent Toxicity under the NPDES permits program. USEPA may revise and/or update this presentation in the future.

You should also know that this module was developed based on the live USEPA HQ NPDES WET course that the Water Permits Division of the Office of Wastewater Management has been teaching to USEPA regions and states for several years. This course, where possible, has been developed with both the non-scientist and scientist in mind, and while not necessary, it is recommended that a basic knowledge of biological principles and Whole Effluent Toxicity will be helpful to the viewer. Prior to this course, a review of the USEPA's Permit Writer's online course, which is also available at USEPA's

NPDES WET Course Online Training Curriculum

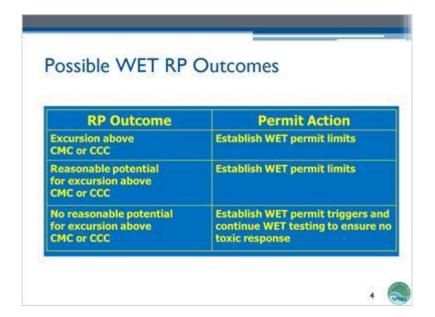
NPDES website, is recommended.

When appropriate a blue button will appear on a slide. By clicking this button, additional slides will present information regarding either freshwater or marine USEPA WET test methods. When these additional slides are finished, you will be automatically returned to the module slide where you left off. The blue button on this slide provides the references for USEPA's WET test methods that will be presented throughout this module. Alright. Let's take a look at the development of USEPA NPDES WET permit limits.



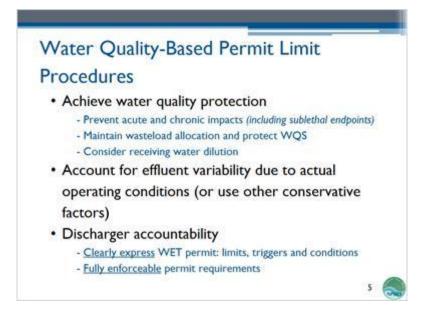
Notes:

For the purposes of this module, it is assumed that a Reasonable Potential, or RP, analysis has already been conducted for the NPDES permitted discharger, and Reasonable Potential has been demonstrated. This module will focus on the process of deriving NPDES WET permit limits, in addition to covering some other key considerations when establishing WET monitoring requirements as part of the required NPDES permit conditions. This module will also discuss some common WET permitting challenges, such as how Total Residual Chlorine and ammonia should be addressed. Also covered are recommendations for appropriate NPDES permit language. When including Toxicity Reduction Evaluations and Toxicity Identification Evaluations, or TREs/TIEs, appropriate language is important to ensure that the expectations are clear and the requirements are enforceable.



Notes:

One outcome is that there has been an excursion of either the narrative or numeric aquatic life protection criteria, commonly referred to as WET criteria, for acute or chronic toxicity, or for both. Another case is where an excursion has not occurred, but based on the RP analysis, there exists the potential of an excursion or the potential to contribute to an excursion of the state's WET water quality standards. For these two RP outcomes, it is required under USEPA's NPDES permit regulations listed at Title 40 of the Code of Federal Regulations, or CFR, Part 122.44(d)(1)(iv) for numeric WET criteria and Part 122.44(d)(1)(v) for narrative WET criteria, to establish a NPDES WET limit. A third possible outcome of RP analysis is that there is no potential of an excursion above either the acute or chronic WET criterion, in which case the permit action is to establish triggers and to continue monitoring to ensure that water quality standards are protected.



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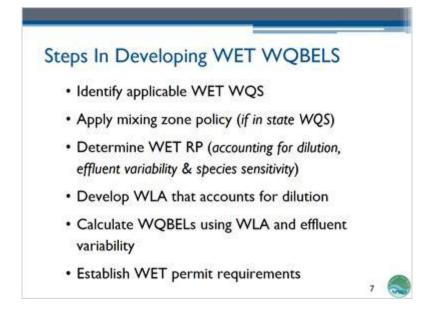
In order to ensure the protection of state WET water quality standards, USEPA developed NPDES permit water quality-based procedures for developing Water Quality-Based Effluent Limits, or WQBELs. These water guality-based procedures are the same for WET as they are for any other pollutant parameter. WET WQBELs are established to prevent acute and chronic toxicity impacts that would result in an excursion of the state's water quality standards, including sub-lethal effects, such as reduced reproduction or growth. The water quality-based procedures calculate the wasteload allocation for an effluent discharge that will not result in an excursion of state water quality standards. The wasteload allocation, or WLA, takes into account effluent dilution, if a mixing zone is allowed under a state's water quality standards or permitting regulations, and also effluent variability. Once the WQBELs and other NPDES permit requirements are established, the permittee is accountable for meeting all the NPDES permit requirements, including any permit conditions and limits. However, if the NPDES permit conditions or requirements are not clearly expressed in the permit, then the permittee may not fully address what was intended, and it may be harder to enforce all of the permit requirements. For WET, this means clearly expressing monitoring conditions, triggers or limits, and other permit requirements to establish clear expectations for the permittee and ensure that the requirements are fully enforceable.

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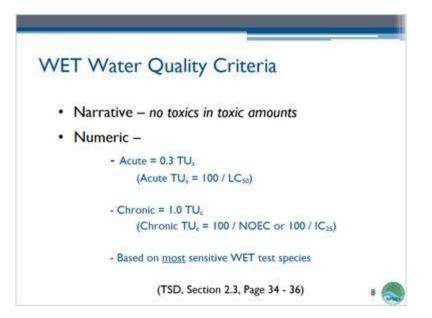
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Therefore, as mentioned in the last slide, there is no substitute for a wellwritten NPDES permit. A well-written permit makes for fewer misunderstandings and miscommunications as to what the permittee is required to do to comply with their permit.



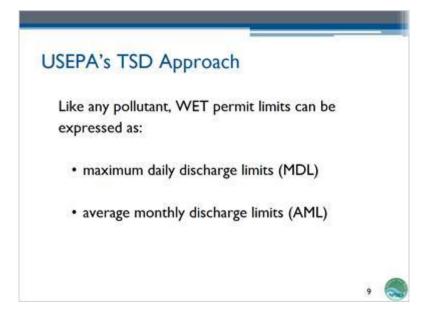
Notes:

There are several steps in developing NPDES WET Water Quality-Based Effluent Limits. These include: identifying the applicable water quality standards for WET; applying a mixing zone if allowed under the state's water quality standards or permitting regulations; determining RP by accounting for allowed effluent dilution, characterizing effluent variability with respect to WET test results and the sensitivity of the test species. The WLA for the effluent is calculated and that WLA is then used, along with a determination of WET variability, to calculate WQBELs. Finally, this information is used to establish NPDES WET permit requirements including WET limits.



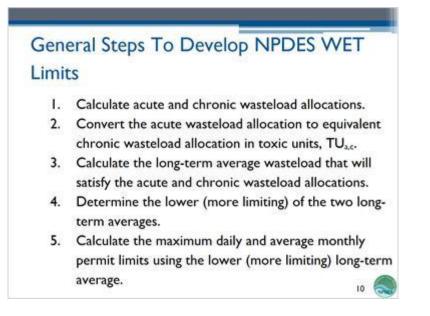
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WET water quality criteria are presented in the form of narrative and numeric criteria. Most states have a WET narrative criterion in their water quality standards similar to "no toxics in toxic amounts." WET testing directly addresses the narrative criterion. In addition to narrative criteria, some states have adopted the numeric acute and chronic WET criteria that are recommended in USEPA's 1991 Technical Support Document for Water Quality-based Toxics Control, commonly referred to as the USEPA's TSD. These numeric WET water quality criteria are 0.3 toxic units acute and 1.0 toxic units chronic. An acute toxic unit, or TU_a, is equal to 100 divided by the LC_{50} , and a chronic toxic unit, or TU_c , is equal to 100 divided by either the NOEC or IC₂₅. Compliance with WET triggers or limits based on these WET criteria is determined using valid WET data generated from WET tests using the most sensitive test species. The most sensitive test species is determined by conducting WET tests with ideally three test species, such as an alga, an invertebrate, and a vertebrate, with an effluent dilution series that brackets the in-stream waste concentration.



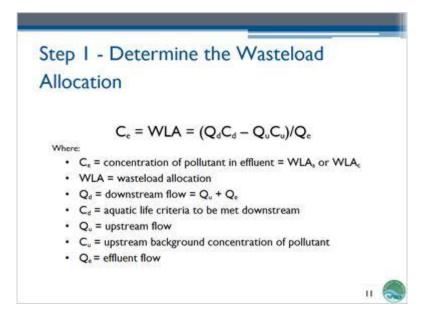
Notes:

In developing NPDES WET permit limits, WET is handled in the same way as other pollutant parameters. Both a Maximum Daily Limit, or MDL, and an Average Monthly Limit, or AML, are calculated for WET as explained in the following slides.



Notes:

To develop NPDES WET limits for a permitted effluent discharge, the first step is to calculate both the acute and chronic wasteload allocations and also an "equivalent chronic wasteload allocation." The equivalent chronic wasteload allocation is needed to calculate both an acute and a chronic Long Term Average, or LTA. The LTA concentration for WET is calculated such that it will satisfy both the acute and chronic WLAs. A determination is then made as to whether the acute or the chronic LTA is more limiting. The result of that determination is then used to calculate maximum daily and average monthly WET limits using the lower or more limiting LTA. In the next several slides, we will go through an example illustrating each of these steps.

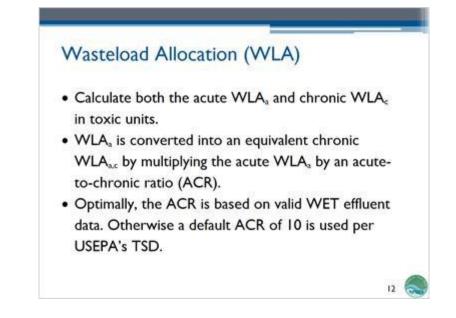


Notes:

In the first step, the Wasteload Allocation, or WLA, for WET is determined. Before starting, some key information will be required for this step. First the receiving water upstream flow under critical conditions is identified, which is denoted as Q_u. Typically, the critical receiving water condition occurs under dry conditions when receiving water flow is at a relatively low level and is usually expressed as cubic feet per second, or cfs. Second, the permitted facility's effluent design flow (also expressed in cfs), denoted here as Q_e, needs to be determined. Third, the state's WET criteria to be met downstream, denoted here as C_d, needs to be identified. The downstream flow, denoted as Q_d, is determined by adding the upstream flow under critical conditions (Q_u) to the facility's design effluent flow (Q_e). The other information needed is the upstream background concentration, or C_u, to account for toxicity, if any, in the upstream receiving water unless it is established that there is no upstream toxicity. The toxicity assessment of the upstream receiving water can be determined through ambient toxicity testing using the most sensitive WET test species for both acute and chronic toxicity using USEPA WET test methods. Once this minimum required information is determined, then the WLA calculations can begin. The WLA is equal to the multiplied product of the downstream flow (Q_d) and the downstream aquatic life protection criteria (Cd), from which is subtracted the multiplied product of the upstream flow (Q_u) and the background concentration (C_u). That calculated result is then divided by the facility's design effluent flow (Qe). As mentioned before, when establishing in-stream

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WET permit limits the upstream background toxicity, or C_u , is factored into the WLA. If it is known that the upstream water is not toxic to the test species selected for evaluating the toxicity of the effluent using USEPA WET test methods, then the C_u would equal zero, and the multiplied product or term Q_uC_u would also be zero in this equation.

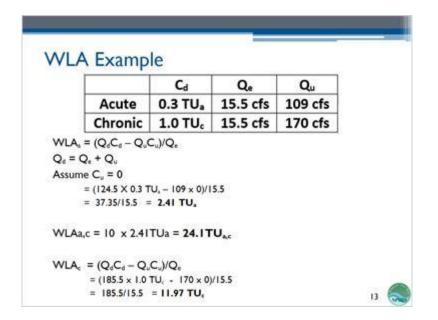


Notes:

When calculating the wasteload allocation, or WLA, used later to calculate both the acute and chronic LTA, it is necessary to consider both acute and chronic toxicity. The first step is to calculate the acute wasteload allocation, or WLA_a, in acute toxic units, or TU_a.

The next step is to convert the WLA_a to what USEPA refers to as an *"equivalent chronic waste load allocation or WLA_{a,c}"* by multiplying the WLA_a by an acute-to-chronic ratio, or ACR. This conversion of the WLA_a is done so that the acute toxicity effects can be compared to the chronic toxicity effects. The WLA_{a,c} will be used later in the LTA calculations to determine which LTA is lower and, therefore, more conservative towards protecting the state's WET water quality standards. Ideally, the ACR would be based on the actual ratio of acute and chronic toxicity units from valid WET test data for the permitted effluent discharge. This site-specific ACR is rarely available however, because of a lack of simultaneous acute and chronic WET test data. Therefore, the USEPA recommended default ACR of 10, provided in the USEPA's TSD, is typically used to obtain the equivalent chronic WLA_{a,c}.

Finally, the last step before calculating the LTA is to calculate the chronic wasteload allocation or WLA_c in chronic toxic units, or TU_c .



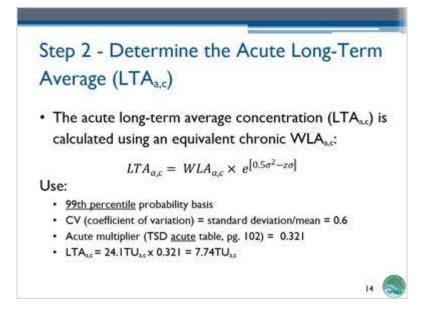
Notes:

In this WLA calculation example, all the necessary information including the acute and chronic WET criteria (C_d) as well as the upstream and facility's effluent design flows are provided. For acute WET, the criterion is 0.3 TU_a, and for the chronic the criterion is 1.0 TU_c. The Q_e is the effluent flow, which in this example is 15.5 cfs. The Q_e, or the facility's effluent design flow, will be used for both acute and chronic toxicity. The Q_u, which is the upstream flow, again under critical conditions, is different for the acute and chronic WLA in this example. That is because the state identified a different mixing zone for acute than for chronic WET for the effluent discharge into the receiving stream. Since the acute upstream flow for the acute mixing zone is less than the chronic upstream flow for the chronic mixing zone, this indicates that the effluent has less dilution available for the acute WLA calculation. The wasteload allocation for acute, WLA_a, for the mixing zone is the formula shown in the previous slide, using the WET acute criterion of 0.3 TU_a. For this example, it is assumed that the upstream background concentration for toxicity, or C_u, is zero, meaning that there is no toxicity upstream. The downstream flow is the sum of the acute upstream flow of 109 cfs and the facility's effluent flow of 15.5 cfs, which is equal to 124.5 cfs. This downstream flow of 124.5 cfs is then multiplied by the acute criterion of 0.3 TU_a. Since the upstream background toxicity or C_u, for the purposes of this example, is assumed to be zero, then the product of Q_u multiplied by C_u in the numerator is also zero. So, what remains in this equation is 124.5 cfs multiplied by 0.3 TU_a which equals 37.35 cfs TU_a divided by the effluent flow

of 15.5 cfs, and the cfs units in the numerator and denominator cancel out, leaving only the unit of TU_a . Therefore, this calculation results in a WLA_a value of 2.41 TU_a.

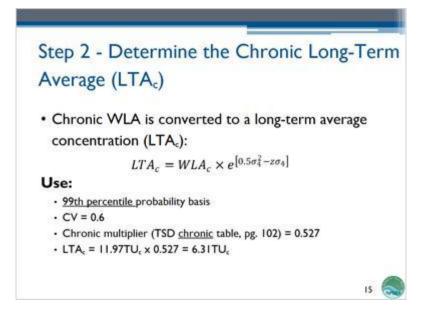
The next step is to convert the WLA_a to an "*equivalent chronic wasteload allocation, or WLA_{a,c}*", by multiplying the WLA_a by an acute-to-chronic ratio, or ACR. For this example, the USEPA TSD's recommended default ACR of 10 is used, yielding an *equivalent chronic WLA_{a,c}* of 24.1 TU_{a,c} which denotes that the WLA_{a,c} is a converted value based on an ACR.

Next, the chronic WLA, or WLA_c, is calculated based on the chronic mixing zone using the same equation as shown in the previous slide with the chronic WET criterion of 1.0 TU_c. Also, the WLA_c uses the chronic upstream flow, which in this example is 170 cfs. Using the same basic WLA formula used to calculate the WLA_a, plugging in the calculated downstream flow, Q_d, of 185.5 cfs multiplied by the downstream chronic criteria, C_d, of 1.0 TU_c equals a product of 185.5 cfs TU_c, from which is then subtracted the product of the chronic upstream flow, Q_u, of 170 cfs and the assumed background concentration, C_u, of zero. This calculation yields a numerator of 185.5 cfs TU_c. Finally, when dividing 185.5 cfs TU_c by the effluent flow of 15.5 cfs, the cfs units in the numerator and denominator cancel out, leaving only the unit of TU_c . Therefore, this calculation results in a WLA_c value of 11.97 TU_c. So, we now have a 24.1 TU_{ac} from converting the acute WLA_a to an *equivalent* chronic WLA_{a,c} and we have a chronic WLA_c of 11.97 TU_c. These WLAs will now be used in the next slides to calculate the acute long-term average concentration, or LTA_{a.c}, and the chronic long-term average concentration or LTA_c.



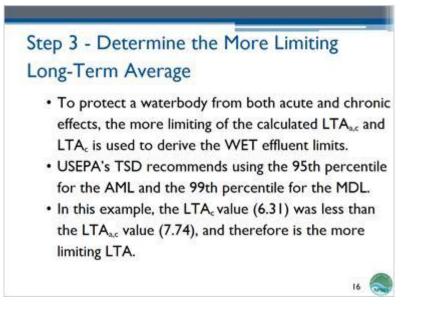
Notes:

Having calculated the wasteload allocation values, the next step is to determine the converted acute long-term average, or LTA_{a,c}, using the *equivalent chronic WLA_{a,c}*. Similar to the way the LTAs are calculated for other pollutant parameters, USEPA's TSD recommends using the 99th percentile probability basis for calculating the acute LTA_{a.c}. Page 102 of the USEPA's TSD has the tables that provide the multiplier term for the equation based on the Coefficient of Variation, or CV, of valid WET test data for the permitted facility. In this example, we use the default CV for WET of 0.6 because there was insufficient WET test data to calculate a facility site-specific CV. Using a CV of 0.6 and the 99th percentile probability basis, the table on page 102 of USEPA's TSD indicates that the acute multiplier is 0.321. Multiplying the converted WLA_{a,c} of 24.1 TU_{a,c} that was calculated in the previous slide by the USEPA's TSD multiplier of 0.321 results in an acute LTA_{a.c} of 7.74 TU_{a.c}, indicating that this acute LTA_{a.c} is an acute to chronic converted LTA. Since the acute LTA_{a.c} has been converted to a chronic basis, this LTA_{a.c} can now be directly compared with the chronic LTA_c, which we will calculate in the next slide.



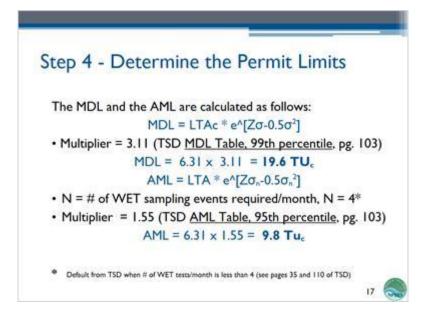
Notes:

To calculate the chronic LTA_c, we again use the 99th percentile probability basis. The CV is the same as before, the default value of 0.6. USEPA's TSD has another table on page 102 for the chronic LTA multipliers. Using the CV of 0.6, this table indicates a chronic multiplier of 0.527. Following the chronic LTA equation, the chronic WLA_c of 11.97 TU_c, calculated in the previous slide, is multiplied by the chronic multiplier of 0.527, which yields a chronic LTA_c of 6.31 TU_c.



Notes:

In Step 3, we need to determine which long-term average, the converted acute $LTA_{a,c}$ or the chronic LTA_c , is lower or more limiting. In this example, the LTA_c , or chronic long-term average, was 6.31 TU_c and the $LTA_{a,c}$, or the converted acute long-term average, was 7.74 TU_{a,c}. Therefore, the chronic long-term average of 6.31 TU_c was lower and is determined to be the more limiting LTA in this example.

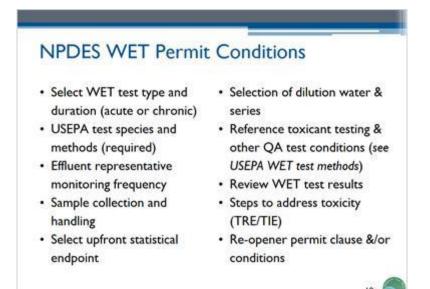


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To determine NPDES WET permit limits in Step 4, the more limiting or lower LTA is used. In our example, this was determined to be the chronic LTA_c of 6.31 TU_c. The Maximum Daily Limit, or MDL, and the Average Monthly Limit, or AML, are calculated using the formulae outlined in USEPA's TSD. The USEPA's TSD recommends using the 99th percentile for calculating the MDL and the 95th percentile for calculating the AML.

For the MDL, the chronic LTA_c is multiplied by a factor which can be found in USEPA's TSD using the 99th percentile table at the CV of 0.6 on page 103. Therefore, the multiplier provided at the intersection of the column for the 99th percentile and a CV of 0.6 is 3.11, which when multiplied by the LTA_c of 6.31 TU_c yields a MDL of 19.6 TU_c.

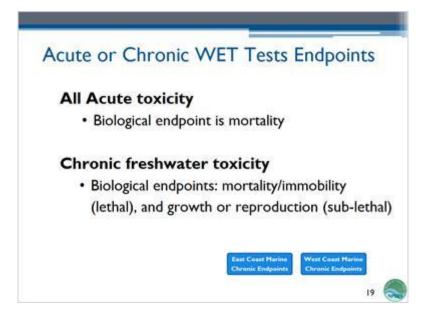
To calculate the Average Monthly Limit, or AML, the more limiting LTA_c is multiplied by a factor which is found using the 95th percentile table on page 103 of USEPA's TSD. This multiplier factor relies on the number of WET sampling events required per month, or "N." USEPA's TSD recommends an "N" value of 4 when there are fewer than four WET test sampling events per month. The multiplier factor listed in the 95th percentile table at the intersection of the CV of 0.6 and an N=4 is 1.55. Therefore, the AML is the chronic LTA_c of 6.31TU_c multiplied by 1.55 which is equal to 9.8 TU_c.



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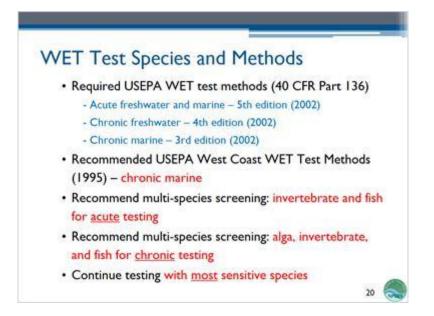
In addition to including WET limits or triggers in the NPDES permit, other WET permit conditions which should be included are the many items covered in the previous modules. For example, the NPDES permit needs to specify the type of WET test, acute or chronic; the specific USEPA WET test method and the USEPA WET test species to be used for WET monitoring, making sure that the most current test methods are specified or incorporated by reference in the permit's general permit conditions. These WET test methods are specified in 40 CFR Part 136 or, in the case of the USEPA West Coast WET test methods, in the document entitled: "Short-Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to West Coast Marine and Estuarine Organisms" (EPA/600/R-95-136). In addition, the NPDES permit should clearly specify the appropriate WET test endpoints to be calculated and the statistical approaches to be used for each type of WET test required. Other important NPDES permit conditions to specify in the NPDES permit include: the dilution water to be used in WET testing, for example, laboratory water or water from the receiving waterbody if found not to be toxic; requested documentation of Quality Assurance/Quality Control performed; WET monitoring requirements should there be an exceedance of the WET trigger or WET limit, such as an accelerated WET monitoring schedule and accompanying regulatory actions depending on the results of accelerated WET monitoring; Toxicity Reduction Evaluation/Toxicity Identification Evaluation plan requirements; and a re-opener clause or additional requirements should the WET test data warrant it, such as developing WET

limits in the permit if such limits were not already required.



Notes:

The USEPA WET Test Methods Module discusses the various USEPA WET test methods, endpoints, and other important information regarding WET testing under the NPDES permit program. With regards to this module on NPDES WET permitting, it is important that the NPDES permit specify the types of WET tests required, acute or chronic, and associated endpoints that need to be reported. The type of WET test endpoints reported should be based on the facility-specific NPDES WET permit limits or triggers developed, the available effluent dilution in the receiving waterbody, and other factors discussed previously in this module.



Notes:

As discussed in more detail in the WET Test Methods Module, there are many USEPA WET test methods and test species available for NPDES WET permit compliance monitoring. For more information on these test methods, please see the WET Test Methods Module. In general, after WET testing has been conducted several times for a permitted facility, monitoring should continue using the USEPA WET test species that was determined to be the most sensitive to the effluent in order to properly characterize the effluent's toxicity for both permit compliance and reasonable potential determinations for the next permit cycle.

Testing Frequency	Volume of Discharge		
Monthly	> 1 MGD		
Quarterly	≤1 MGD		
Other considerations:			
 Case by case basis 			
 Intermittent discharge 			
 Compliance record 			
 Effluent variability 			

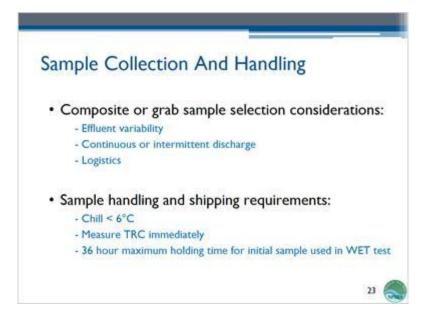
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WET monitoring should be sufficiently frequent to properly characterize the toxicity of the permitted effluent. In general, USEPA recommends monthly WET testing for high flow effluent discharges, for example, greater than 1 million gallons per day; and guarterly WET testing for effluent discharges having less flow. The frequency of WET testing to include in the NPDES permit is determined on a case-by-case basis. Factors to consider when determining the appropriate effluent WET test monitoring frequency include: whether the effluent is discharged intermittently, such as wastewater treatment discharges released into the receiving stream periodically rather than continuously; the NPDES facility's compliance record for other NPDES permit conditions; and the degree of effluent variability in terms of other water quality parameters monitored in their permit or discharge flow rates. If the effluent is discharged intermittently, the timing and frequency of WET monitoring should be specified accordingly. If the facility has a questionable NPDES permit compliance record or is likely to be highly variable due to the facility's operations or type of wastewater treatment used, then frequent monitoring, such as monthly WET testing, may be warranted.

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%	obability (currence is
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# of Tests	10%	20%	30%
1	0.10	0.20	0.30
5	0.41	0.67	0.83
10	0.65	0.89	0.97
	0.88	0.99	0.99

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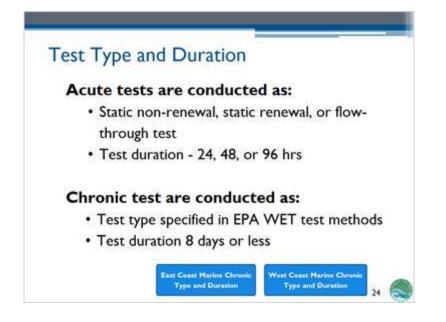
This slide shows the probability of detecting a truly toxic sample from a NPDES facility discharge depending on the actual percent of toxic effluent occurrences (the percentages along the top row of the table) and the number of WET tests conducted on the effluent. What this table shows is that if, for example, an effluent is actually toxic 30% of the time, and the facility's NPDES permit requires one WET test a year, then there is only a 30%, or 0.30, probability that effluent toxicity will be detected, even though the effluent is actually toxic almost a third of the year (see the fourth column, second row of the table). By contrast, note that if the NPDES permit was to require 5 WET tests a year, there would be an 83%, or 0.83, probability of detecting toxicity in this effluent (fourth column, third row of the table). Oftentimes, the actual percentage of toxicity events is not known for a discharge unless there has been adequate WET testing over time. For this reason, USEPA's TSD recommends at least quarterly WET test monitoring in NPDES permits.



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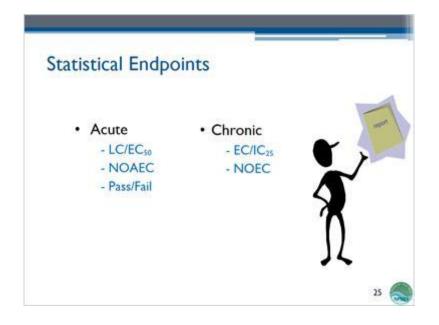
How the permitted effluent should be collected and handled for WET test monitoring is another important aspect that should be specified in the NPDES permit. Oftentimes, states require that either time-weighted or flowweighted 24-hour composite samples are to be collected for WET testing, because these types of samples give a good indication of the average effluent quality over a 24-hour period. However, in some cases, a grab effluent sample may be more appropriate. In cases where there is known to be high effluent variability, even within a 24-hour period, such as with intermittent discharges or periodic batch treatment releases, grab samples rather than 24-hour composite samples may yield a more representative assessment of the effluent toxicity in the receiving waterbody. The inherent logistics in collecting effluent samples may also play a role in determining whether composite or grab samples should be required. If the facility's effluent discharge is in a very remote location, where it is challenging to obtain a 24-hour composite sample, grab samples may be the only option. Effluent sample handling requirements for WET test monitoring are discussed in detail in the WET Test Methods Module and the WET Test QA/QC Module. Effluent samples need to be chilled to less than 6° centigrade if they will not be tested immediately. Total Residual Chlorine should be measured on the effluent sample right after collection and recorded in the sample chain of custody form for later inspection by the laboratory receiving the effluent sample for testing. Unless the logistics for collecting effluent samples prohibit timely effluent sample shipping, the first effluent sample received by

the laboratory for WET testing should be held no longer than 36 hours from the time the effluent sample collection was completed.



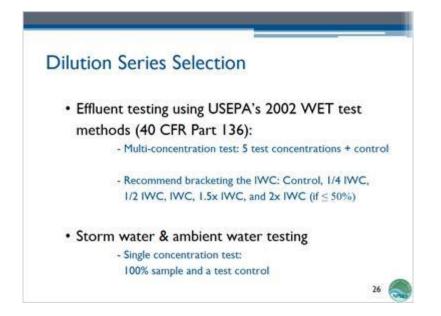
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Many USEPA WET test methods may be conducted using either a static or static-renewal mode, depending on the test method selected. The NPDES permit should specify the WET test type where the method allows for either static or static-renewal exposures. Likewise, the USEPA acute WET test methods allow for some flexibility in the test duration depending on the type of WET test, particularly for acute testing. The NPDES permit should specify the test duration required for a given acute USEPA WET test method. Chronic USEPA WET test methods specify the type of exposure and the duration of the test. Please see the WET Test Methods Module for more details regarding test types and the duration of tests.



Notes:

The NPDES permit should clearly specify the statistical WET test endpoints that are to be reported with the WET monitoring test data required in the NPDES permit. The WET Statistics Module presents a detailed discussion of the different statistical endpoints.

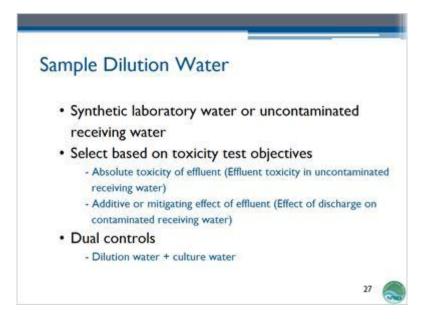


Notes:

USEPA WET test methods at 40 CFR Part 136 require 5 effluent test concentrations and a control treatment. The effluent test concentrations selected for a WET test can impact the statistical analysis used and the confidence one has in the resulting WET test endpoints, as discussed in the WET statistics module. In general, USEPA recommends using an effluent test concentration series that brackets the permitted effluent In-stream Waste Concentration, or IWC. For facilities that have a permitted IWC less than or equal to 50% effluent, the effluent test concentration series could be 0.25 times the IWC as the lowest effluent concentration, 0.5 times the IWC, the IWC, 1.5 times the IWC, and 2 times the IWC, plus a control treatment. For those facilities that have an IWC greater than 50% effluent, the WET test dilution series will need to be adjusted accordingly. For facilities that have an IWC at or near 100%, it may be appropriate to use a general dilution series such as 12.5%, 25%, 50%, 75%, 100% effluent, and a control treatment. The effluent test concentration series should not use closely spaced effluent concentrations. USEPA recommends using a dilution factor greater than or equal to 0.5. If too small a dilution factor is used, for example, control treatment, 70%, 75%, 80%, 85%, 90% effluent, where the IWC is 80% effluent, then precision of the statistical endpoint will be compromised, and therefore, one will have less confidence in the WET endpoint reported. Ambient toxicity testing or collecting samples from the waterbody and testing them in the laboratory using USEPA WET test methods, and storm water toxicity testing do not require conducting the WET test with multiple

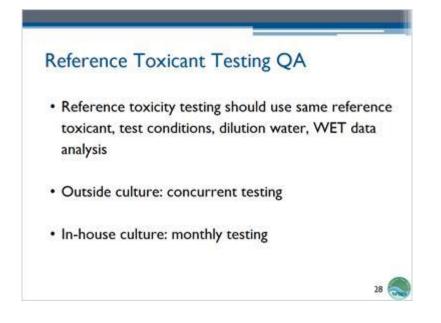
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dilutions of the collected sample. In these cases, the test typically consists of a control treatment and the 100% (undiluted) test sample.



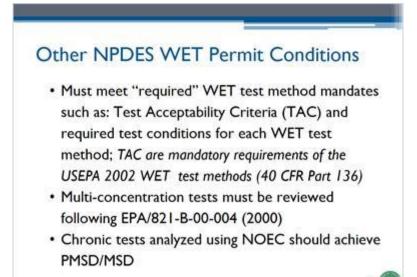
Notes:

The NPDES permit should specify the type of dilution water to be used in WET monitoring. The USEPA WET Test Methods Module discusses dilution water. Generally, most states require the use of synthetic laboratory dilution water using the recipes given in USEPA's WET test methods manuals. If the permit requires the permitted discharger to use dilution water obtained from the field, such as the receiving water, the laboratory should also run a control treatment using laboratory dilution water as a quality control check on the field collected dilution water.



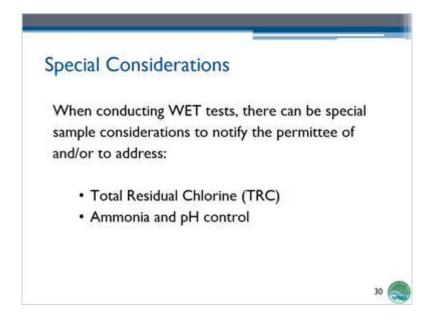
Notes:

As explained in detail in the USEPA WET Testing QA/QC Module, laboratory reference toxicant testing information should accompany WET test results submitted to the NPDES permitting authority. This provides a quality assurance check on the laboratory and the WET test results submitted. If the WET test organisms are not from the laboratory's cultures and were obtained from an external supplier, USEPA requires concurrently challenging a subset of those test organisms in a reference toxicant test to assess the health and sensitivity of the organisms. If the organisms are obtained from in-house cultures, then reference toxicant testing of those organisms should be conducted on a monthly basis to ensure that they are in satisfactory health before initiating a WET test.



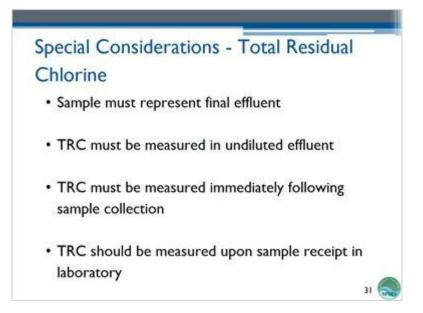
Notes:

The WET Test Methods Module discusses Test Acceptability Criteria, or TAC, for different USEPA WET test methods. There are overall TACs, as well as specific TACs, for each test method which must be met; if not, the WET test is invalid and needs to be conducted again using a fresh effluent sample. The NPDES permit should specify follow-up actions if WET test data are determined to be invalid. USEPA also recommends reviewing the WET test water quality data, for example, dissolved oxygen levels, temperature, as well as sample chain of custody forms, concentration-response WET test data, and other aspects of the data as discussed in USEPA's 2000 WET Test Variability guidance. If an NOEC endpoint is required in chronic WET testing in the NPDES permit, the Percent Minimum Significant Difference, PMSD, or Minimum Significant Difference, MSD, need to be met as well as other required quality assurance checks under the USEPA WET test methods.



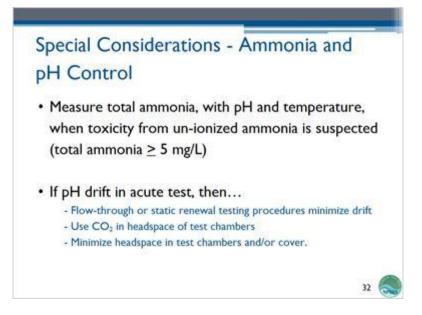
Notes:

Two special considerations regarding NPDES effluent samples collected for WET testing, which have been raised by NPDES permitting authorities and their permittees, will be covered in the next few slides. These considerations involve how to conduct WET testing when there is either elevated Total Residual Chlorine, TRC, or ammonia in the effluent sample.



Notes:

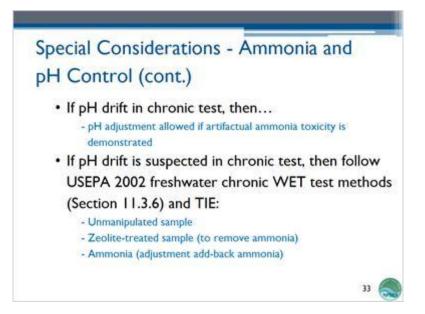
In order to ensure compliance with and protect a state's WET water quality standards, the WET monitoring must represent the permitted effluent discharged to the receiving waterbody. Representative effluent monitoring includes consideration of any chemicals added during the treatment process, including chlorine or other disinfectants, so that the final discharged effluent will be used for WET tests. The USEPA recommends measuring the Total Residual Chlorine, or TRC, in the effluent sample immediately following collection and again upon arrival at the laboratory prior to WET testing. Regardless of whether TRC is observed in the effluent sample, the effluent sample should be tested for WET without making adjustments for TRC. The laboratory should record the effluent TRC concentration in the WET test documentation.



Notes:

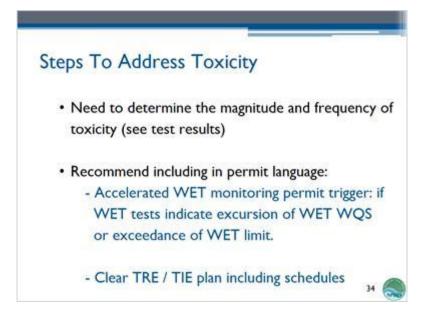
Ammonia can present a different type of WET test challenge. USEPA recommends measuring ammonia in the undiluted effluent sample upon receipt by the laboratory. Since ammonia toxicity is strongly influenced by pH and to a lesser degree temperature, the likelihood of observing toxicity due to ammonia in a WET test will depend on the ammonia concentration and pH of the water in the WET test chambers. A higher pH in the test solutions will convert more of the ammonia to the unionized form of ammonia which is toxic to fish and other aquatic life. In some types of WET tests, the laboratory may observe an increase in the pH in test solutions over the course of the test. This is commonly referred to as pH drift and is often an artifact of the laboratory conditions and is not necessarily a reflection of what happens in the receiving waterbody. The pH drift is often more commonly observed in chronic WET tests using fish as the test organisms rather than invertebrates such as Ceriodaphnia. Also it is more likely to occur during a static WET test where fresh effluent solution is not being added periodically as it is done in flow-through or static-renewal WET tests. The USEPA WET test methods provide several solutions for addressing pH drift, including increasing the exchange of fresh effluent samples into test chambers, either by using more frequent renewals or flow-through WET test procedures; introducing carbon dioxide gas into the headspace of the test chambers; or simply by minimizing the headspace in the test chambers and covering the test chambers to limit exchange with the atmosphere. Each of these USEPA suggested WET test method procedures will tend to stabilize the pH drift without resulting in

other chemical changes to the effluent sample. These procedures therefore maintain the representativeness of the final discharged effluent while correcting the issues associated with pH drift during the WET test.



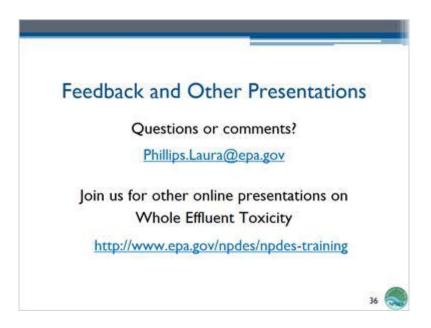
Notes:

If pH drift is observed in a chronic WET test, pH adjustment of the effluent sample may be allowed if it is demonstrated that the pH drift is an artifact of the laboratory WET test conditions and is not occurring in the receiving waterbody. However, one of the recommended USEPA WET test procedures for addressing pH drift, mentioned in the previous slide, should be used prior to considering pH adjustment of the effluent sample. USEPA's freshwater chronic WET test methods provide specific procedures, as noted in this slide, for identifying whether the observed toxicity is due to ammonia. These procedures are also discussed in USEPA's Phase I and Phase II Toxicity Identification Evaluation, or TIE, manuals and USEPA's Phase I chronic TIE manual.



Notes:

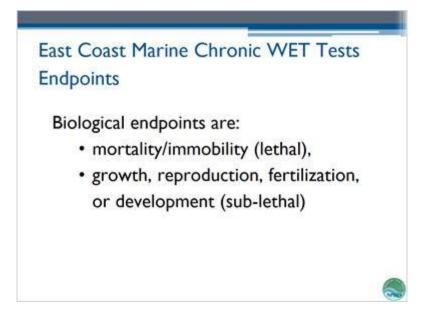
The magnitude and frequency of toxicity as demonstrated by valid WET test data generated from a NPDES permit's WET monitoring schedule should be routinely reviewed by the NPDES permitting authority. In addition, it is a good strategy to include in the NPDES permit, conditions that specify followup or accelerated WET testing requirements if toxicity is observed in a WET test that results in non-compliance with the permit's WET limits or indicates an excursion of the state's WET water quality standards. In addition, the NPDES permit should specify the need for a Toxicity Reduction Evaluation/Toxicity Identification Evaluation, or TRE/TIE, should accelerated WET testing indicate that the effluent is toxic at a level that would result in an excursion of the state's WET water quality standards. The TRE/TIE module in this training series will discuss the use of TRE/TIEs in NPDES permits in more detail.



Notes:

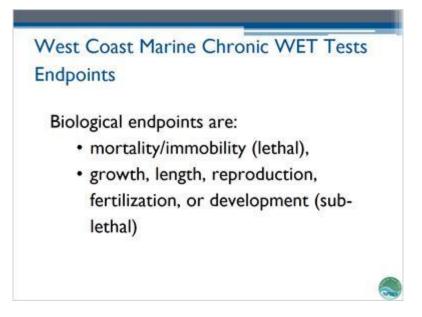
Thank you for joining us for this USEPA's NPDES Whole Effluent Toxicity training presentation. We hope that you have enjoyed it! If you have questions or comments on this or any part of the USEPA's NPDES WET online training curriculum, click on the email address given on this slide to send a message to Laura Phillips, USEPA HQ National WET Coordinator. Remember, you will find all of the USEPA's NPDES WET online training presentations, under the USEPA's NPDES training section found on the Office of Wastewater Management's NPDES website.

See you next time!



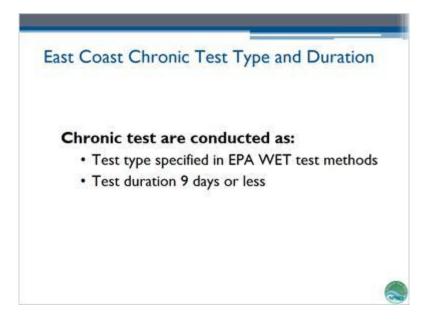
Notes:

For USEPA East Coast chronic WET tests, the test endpoints include lethal endpoints measured as mortality or immobility, as well as sub-lethal endpoints measured as growth, reproduction, fertilization, or development.



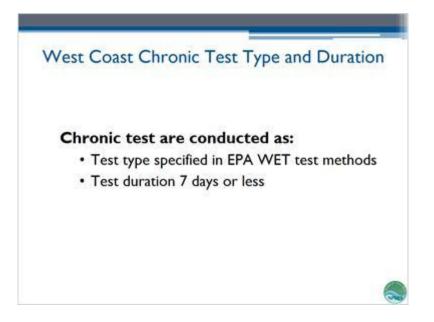
Notes:

For USEPA West Coast chronic WET tests, the WET test endpoints include lethal endpoints measured as mortality or immobility, as well as sub-lethal endpoints, measured as growth, length, reproduction, fertilization, or development.



Notes:

For USEPA East Coast marine chronic WET tests, the test type is specified in the USEPA WET test methods, and the test duration is no longer than 9 days.



Notes:

For USEPA West Coast marine chronic WET tests, the test type is specified in the USEPA WET test methods, and the test duration is no longer than 7 days.