CHAPTER 1.10 - Ammonia & Associated WET Requirements

The Department promulgated new water quality standards for ammonia in s. NR 106.36, Wis. Adm. Code, on March 1, 2004, which adjusts WET requirements in certain situations. The following guidance is given in two parts:

Part One: The first part of this chapter provides guidance for Department staff, WET labs and permittees, regarding requirements in s. NR 106.36(2), which allows effluent samples used in chronic fathead minnow WET tests to be modified to remove ammonia prior to testing when early life stage (ELS) - absent ammonia criteria are in effect.

Part Two: The second part of this chapter gives guidance for staff, to support s. NR 106.36(3), which states that pond systems that have been granted a variance pursuant to s. NR 106.38, Wis. Adm. Code, should not perform WET tests during months when they are not effectively treating ammonia.

NOTICE: This document is intended solely as guidance, and does not contain any mandatory requirements except where requirements found in statute or administrative rule are referenced. This guidance does not establish or affect legal rights or obligations, and is not finally determinative of any of the issues addressed. This guidance does not create any rights enforceable by any party in litigation with the State of Wisconsin or the Department of Natural Resources. Any regulatory decisions made by the Department of Natural Resources in any matter addressed by this guidance will be made by applying the governing statutes and administrative rules to the relevant facts.

Part One: WET Sample Modification When ELS-Absent Criteria Are In Effect

USEPA's *1999 Update of Ambient WQC for Ammonia* contains a provision to adjust (relax) chronic water quality criteria (WQC) for ammonia when water temperatures are colder (< 15º C) and early life stages (ELS) are absent, in order to account for reduced sensitivity to ammonia by juvenile and adult fish at lower temperatures. In Wisconsin, this translates into higher (less stringent) limits for ammonia during winter months. Fathead minnow ELS were used, with other data, to develop the adjustment between ELS present and absent criteria, and the fathead minnow is known to be very sensitive to ammonia. Of the data used to develop WQC for ammonia, only *Hyalella* (an invertebrate), *Musculium* (a mussel), and *Lepomis* (blue gill) ELS were more sensitive to ammonia than the fathead minnow ELS. Like fathead minnow ELS, *Lepomis* ELS would not be expected in receiving waters during the ELS-absent period.

It has been pointed out that a potential conflict may exist between allowing this "relaxation" of the ammonia criteria during ELS-absent periods and requiring chronic WET tests during those same periods, since chronic WET tests are conducted using fathead minnow ELS. Under this scenario, a situation could arise in which a permittee is in compliance with its water quality based effluent limit (WQBEL) for ammonia during an ELS-absent time period, but have a positive (failing) chronic fish WET test due to ammonia during that same period. Due to this perceived conflict, the Department added language to s. NR 106.36(2) "relaxing" WET standards during these ELS-absent periods (i.e., to not consider WET due to ammonia to be a WET "failure"). The argument is that a finding of ammonia toxicity in the WET test would not be applicable to receiving water conditions since the life stage used in testing is not found in the receiving waters during those periods.

Permit Language When ELS-Absent Criteria Are Applied

The following may be added to WPDES permits when WQBELs based on ELS-absent criteria are given:

> *"Effluent samples used in chronic fathead minnow tests may be modified to remove ammonia prior to testing, according to s. NR 106.36(2), Wis. Adm. Code, during periods when ammonia limits based on early life stage-absent criteria are in affect."*

Modification of Samples in Chronic Fathead Minnow Tests During ELS-Absent Periods

The question arises then of how to conduct a chronic fathead minnow WET test when ELS-absent ammonia criteria are in effect in the receiving water. Under certain circumstances, it may be appropriate to conduct the fathead minnow portion of the chronic WET test on effluent samples that have been treated to remove ammonia prior to testing. This will allow the effects of contaminants other than ammonia to still be assessed.

According to s. NR 106.36(2), Wis. Adm. Code, chronic fathead minnow WET test samples may be modified to remove ammonia prior to testing when the permittee can demonstrate to the satisfaction of the department that all of the following conditions are met:

(a) The chronic WET test in question is being conducted during a period when ammonia WQBELs based on early life stage-absent criteria are in affect.

(b) The permittee has demonstrated compliance with applicable acute and chronic WQBELs for ammonia during the testing period.

(c) Total ammonia measured in effluent samples to be used in the WET test are less than the applicable chronic WQBEL given in the permit, but greater than the "ammonia threshold number", as determined below:

1. Find the pH range in the table below that includes the pH measured in the effluent sample after warming to test temperature.

2. Divide 100 by the instream waste concentration (IWC) given in the permit, then multiply that number by the "ammonia threshold number" which corresponds to the appropriate pH range given in the table below.

3. If the total ammonia concentration in the effluent is greater than the final number given in step 2, the lab should remove ammonia from the sample prior to use in the fathead minnow chronic WET test.

Effluent pH (s.u., after warming)	Ammonia Threshold Number* (mg/l total ammonia)
6.0 - 6.5	30
6.6 - 7.0	25
7.1 - 7.5	15
7.6 - 8.0	5
8.1 - 9.0	1

*The “Ammonia Threshold Numbers” given in the table above are based on effluent ammonia levels expected to cause toxicity in FHM chronic tests. These values are based on data collected at the USEPA Environmental Effects Research Laboratory, Mid-Continent Ecology Division/ORD, Duluth. MN (general information available in “*Methods for Aquatic Toxicity Identification Evaluations: Phase II Toxicity Identification Procedures for Samples Exhibiting Acute and Chronic Toxicity*”, EPA/600/R-92/080; specific data unpublished).

Example:

effluent flow = 0.458 mgd
7Q10 = 1.6 cfs
IWC = 64%
Effluent pH is normally in the 7.6-8.0 range, so the “ammonia threshold” number would be 5
WQBEL based on ELS-absent criteria is 16.0 mg/L
100/64 = 1.56
1.56 x 5 = 7.80
If effluent ammonia level was $\geq$ 7.8 and < 16.0, the lab should remove ammonia from WET samples prior to testing.

Acceptable Methods For Treating Effluent Samples to Remove Ammonia

When all of the conditions listed above (as required by s. NR 106.36(2)) are met, effluent samples used in fathead minnow chronic tests may be treated to remove ammonia with zeolite resin prior to testing. Samples should be treated daily, before use in WET tests, rather than batch treated for multiple day usage. Ammonia, pH, hardness, and alkalinity should be measured prior to and after zeolite treatment. A blank (an extra negative control) should also be run through zeolite, to account for toxic artifacts due to the zeolite treatment. Samples used for fathead minnow chronic tests should not be modified in any way other than ammonia removal with the zeolite resin. Samples used in concurrent acute tests with the fathead minnow and *Ceriodaphnia dubia* and chronic tests with *C. dubia* are not to be modified (to remove ammonia or other modifications) prior to testing.

In order to protect against a shift in pH, effluent samples should not be aerated prior to testing unless dissolved oxygen is < 4.0 mg/L. It has been demonstrated that supersaturation of dissolved gases in an effluent sample does not negatively affect fathead minnow or *Ceriodaphnia dubia* in WET tests and therefore removal of supersaturation by aeration of the sample is not necessary. However, induced stress by supersaturated gases should be avoided. If *C. dubia* exposed to test solutions with supersaturated gases are observed to be "caught up" in the bubbles associated with the supersaturated gases, gentle mixing of the sample following warming can be done to reduce supersaturated gases without greatly affecting the sample integrity. In order to protect against artifactual ammonia toxicity caused by a shift in pH during testing, all acute and chronic tests should be conducted in a CO_2 atmosphere or under flow-through conditions that maintain the pH at a level no lower than the measured effluent pH at the time of discharge (for acute) or no lower than the receiving water pH (for chronic).

Decisions regarding WET monitoring frequencies and scheduling should be made according to guidance in Chapter 1.3 of the WET Guidance Document and acute and chronic WET tests should be required during ELS-absent periods, when recommended by the guidance in Chapter 1.3. Continued WET testing during winter months is important, when possible, because wastewater treatment (and, therefore, effluent toxicity) can be significantly different during colder weather. Chronic fathead minnow WET tests conducted during periods when ELS-absent ammonia criteria are in effect will still be used to assess effluent toxicity from other toxicants. Toxicity due to ammonia (and other compounds) will also continue to be assessed with acute WET tests and the *C. dubia* chronic test during periods when ELS-absent ammonia criteria are applied.

Can This "Relaxation" Of WET Requirements Be Applied To Other Substances Where Chemical-Specific Limits Are Being Met?

It is important to note that this is not just a matter of passing a chemical-specific limit for a toxicant and failing a WET test that identifies that chemical as the toxicant. This is a special case only for ammonia because this provision to adjust (relax) the chronic criteria during periods of the year when water temperatures are colder (less than 15º C) and fish ELS are absent, is unique to ammonia. Ammonia is the only chemical for which data showing a difference in sensitivity between ELS and adult fish has been used to allow for an adjustment in the WQC in the absence of fish ELS. This approach is believed to be appropriate for ammonia because the early life stage used in the chronic fathead minnow WET test, and other early life stages that the fathead minnow is being used as a surrogate for, are not found in receiving waters during ELS-absent periods. Therefore, it is believed that a positive chronic fathead minnow WET test result caused by ammonia toxicity would not likely be indicative of negative effects in the receiving water because the life stages that experience those toxic effects would not be present. In addition, any significant chronic toxicity due to ammonia that may be harmful to those species that are present in the receiving water during ELS-absent periods should be detected by the *C. dubia* chronic test.

Will Treating Effluent Samples With Zeolite Remove Toxicants Other Than Ammonia?

According to required WET test methods (ss. NR 149.22 and NR 219.04, Wis. Adm. Code), ammonia must be measured in all effluent samples used in WET tests. Since it is a common constituent of municipal effluents and often a cause of effluent toxicity, ammonia is relatively well known and it's toxicity relationships understood. If the amount of ammonia and pH of a sample is known, one can make an educated guess about the chances of toxicity occurring due to ammonia. However, even if the ammonia concentration present in the sample is sufficient to cause toxicity, other chemicals may be present to cause toxicity if the ammonia is removed. Therefore, it is necessary to complete the chronic fathead minnow portion of the WET test after the ammonia has been removed from the sample, in order to assess the potential for effluent toxicity due to other toxicants.

Zeolite is composed of naturally occurring or synthetically created crystalline, hydrated alkali-aluminum silicates. When zeolite is exposed to an aqueous solution (such as an effluent sample), the positively charged resin removes cations from the solution. Since it is an effective ion exchange resin, zeolite has frequently been used in toxicity identification work (*Methods for Aquatic Toxicity Identification: Phase II TIE Procedures*, EPA/600/R-92/080), used specifically to remove the ammonium ion (NH_4+) from effluent samples. However, because of its ion exchange properties, it also has the ability to remove other cations such as heavy metals. In addition, although the primary action of zeolite is chemical (ion exchange), the physical manipulation of filtration also occurs during the process. Removal of compounds via filtration through zeolite may include surfactants and polymers. Also, changes in the ionic balance of the sample caused by the zeolite treatment may cause chemicals that would not have caused toxic effects before zeolite treatment to be rendered biologically available.

While it is true that modification of samples with zeolite can remove substances other than ammonia and may modify the sample in other ways, the potential for this method to reduce the ability of the WET test to detect toxicity due to other substances should be small in most cases. In addition, substances other than ammonia that may be removed by zeolite, especially heavy metals, are usually more toxic to *C. dubia* than the fathead minnow (exceptions to this can occur, depending on the matrix of a sample - hardness, alkalinity, total organic carbon - which can affect whether a substance is more toxic to a vertebrate or invertebrate). Since samples used in the *C. dubia* test are not being modified, any toxicity due to these substances should still be detected. Therefore, removal of metals from the sample is probably not of relative importance when assessing effluent toxicity because of the battery of organisms required in Wisconsin.

Some surfactants and polymers can be more toxic to fish than *Ceriodaphnia dubia*. Since the presence of ammonia in an effluent sample can mask toxicity caused by other substances, it could also be argued that the removal of ammonia will allow the detection of other substances that may have been missed in the chronic fathead minnow WET test had ammonia been present.

What About WET Data Already Collected During Previous ELS-Absent Periods That Have Failed Due To Ammonia?

In some cases, a permittee may have already collected WET data during previous years when ELS-absent ammonia criteria would have been applicable. The question arises then of how to handle a positive result from a WET test conducted under those conditions. If a fathead minnow chronic test completed prior to this guidance and conducted during ELS-absent periods showed a positive result which was apparently due to ammonia toxicity, if concurrent acute and *C. dubia* chronic tests did not show any toxicity, and the four conditions listed under the section titled "*Modification of Samples in Chronic Fathead Minnow Tests During ELS-Absent Periods*" are met, it may be appropriate to label such a test “not applicable”.

WET tests collected during ELS-absent periods that are subsequently labeled “not applicable” should not be used when assessing a discharge’s WET potential, when determining WET testing frequency, or the need for a WET limit. It is important to keep in mind that this exclusion applies only to ammonia because it is the only water

quality criterion that has an adjustment for the presence or absence of fish ELS. Any WET tests which showed toxicity in an acute test or a *C. dubia* chronic test, or in a fathead minnow chronic test due to any toxicant other than ammonia should not be excluded from WET determinations. Test results generated by acute tests and *C. dubia* chronic tests conducted during periods when ELS-absent ammonia criteria are in effect are still applicable for assessing effluent toxicity from all toxicants, including ammonia. Fathead minnow chronic tests conducted during periods when ELS-absent ammonia criteria are in effect are still applicable for assessing effluent toxicity from all toxicants, except ammonia.

The acceptability and applicability of all WET tests depend on the experience and professional judgment of the Department staff reviewing the WET data. The Biomonitoring Coordinator should be consulted whenever decisions are being made regarding the applicability of WET data during ELS-absent periods.

Part Two: WET Monitoring When a Variance Has Been Given for Ammonia

WET has been monitored in municipal and industrial effluents where toxicity potential was thought to be present since the promulgation of ch. 106, Wis. Adm. Code, in 1989. Ammonia is a significant toxic pollutant and has been regulated accordingly in the WET program. As such, dischargers who have failed WET tests due to ammonia have been held to the same standards as those who failed due to other sources. Many facilities have failed WET tests due to elevated levels of ammonia. Most of these facilities were lagoon systems who performed WET tests during winter months, although other facilities with different types of wastewater treatment have had WET problems which may be associated with ammonia. To date, most facilities with treatment systems other than lagoons that have completed WET tests have not had difficulty meeting WET standards due to ammonia. However, it is believed that stabilization ponds and aerated lagoons that conduct WET tests in winter months are likely to fail due to ammonia, since these types of treatment are not designed to remove ammonia during cold weather.

According to NR 106.36(3), Wis. Adm. Code, lagoon and stabilization pond systems that treat primarily domestic wastewater that have been granted a variance pursuant to s. NR 106.38 are not required to perform WET tests during the months of December through May. Since it has been concluded that lagoon and pond systems will not be effectively treating ammonia during these months, WET failures would be expected due to ammonia. In order to assess the effects of contaminants other than ammonia, WET testing is required during other months of the year (June through November) to identify toxic effects of the discharge.

What About WET Data Already Collected Previously During Cold Weather Conditions That Have Failed Due To Ammonia?

In some cases, a permittee who has received a variance according to s. NR 106.36(3) may have collected WET data in previous years. The question arises then of how to handle a positive result from a WET test conducted during the winter months. WET tests collected under these conditions should not be used when assessing a discharge's WET potential, when determining WET testing frequency, or the need for a WET limit (i.e., in WET reasonable potential determinations), if it has been determined that failures apparently occurred solely due to ammonia. It is important to keep in mind that the variance granted according to NR 106.36(3) applies only to ammonia and test results which indicate toxicity due to substances other than ammonia still apply.

The acceptability and applicability of all WET tests depend on the experience and professional judgment of the Department staff reviewing the WET data. The Biomonitoring Coordinator should be consulted whenever decisions are being made regarding the applicability of WET data collected at lagoon systems during the winter months.

What Can be Done to Demonstrate "Ammonia Only" as a Toxicity Source?

In order to determine whether ammonia was responsible for effluent toxicity, permittees and/or Department staff may have to consider existing WET and ammonia data. There are a number of "clues" to look for in a WET test

which can be used to determine whether ammonia may be the cause of toxicity. Ammonia concentrations can be measured easily and looking at this information is a good first step when deciding whether ammonia may be a problem. If ammonia and pH levels suggest potential for ammonia toxicity, additional evaluations should be done. Sole dependence on ammonia analyses is not advisable in most situations, however, because there is little or no additivity between ammonia and most other toxicants. Even if the ammonia concentration is sufficient to cause toxicity, other compounds may be present to cause toxicity if the ammonia is removed.

The fathead minnow is more sensitive to ammonia than *C. dubia*, so only in tests where the fathead minnow was most sensitive should ammonia be considered as the sole source of toxicity. Since the fathead minnow is also sensitive to other compounds (e.g., some surfactants and dyes), it is necessary to look further once sensitivity to the fathead minnow is noted. Ammonia levels which may cause toxicity to the species used in a WET test are relatively easy to predict when effluent pH is known (consult the Biomonitoring Coordinator if help is needed to determine whether NH3 levels are high enough to cause toxicity).

Ammonia, Toxicity, and pH Drift

Natural processes which act to regulate the pH of natural waters also occur in mixtures of natural surface waters and effluents. With the exception of lagoon systems, effluent pH values are often lower than those in receiving waters, due to the presence of excess carbon dioxide resulting from the artificially high rates of respiration of microorganisms in wastewater treatment plants. When the effluent is discharged to surface waters (or mixed with receiving waters in laboratory settings), respiration rates fall to "natural" levels and excess carbon dioxide is stripped, causing a pH rise or drift upwards. Upwards pH drift can significantly effect ammonia toxicity expressed in WET tests, since higher pHs result in more ammonia toxicity. Due to algal activity, effluent pHs in lagoon systems are often high. Extra care should be taken in tests where ammonia problems are possible. Chapter 2.8 discusses the "CO2 entrapment method", when it is required, and why it is necessary to use this method to control pH drift. The Biomonitoring Team recommends that all WET tests (especially those where relatively high ammonia is possible) be done using the "CO2 entrapment method" in order to control pH drift and eliminate the potential for "false positive" toxicity results (i.e., ammonia toxicity expressed at artificially high pHs).