Condensed Version of NR 149 *

* ***NOTE: This condensed version is offered only to help facilitate understanding of NR 149 requirements. This is not an official version of administrative code requirements. For actual, specific requirements, always refer back to an official version of NR 149.***

Laboratory Certification and Registration
{The previous version was repealed and recreated}

Effective Date: September 1, 2008

Why was the new code necessary? Laboratory operations have undergone significant advances since the last version of our code (1996) was approved. Other state and national programs have implemented new rules that reflect these advances. This version of NR 149 incorporates many of those changes, and where appropriate, moderates them by incorporating suggestions expressed by our regulated community. This version of NR 149 specifically

- Improves the structure of the program
- Clarifies the certification and registration process
- Establishes a more equitable fee structure
- Clarifies PT requirements
- Stipulates procedures for on-site evaluations
- Clarifies quality system requirements by adding specificity, flexibility and definitions.
- Makes Wisconsin's drinking water program current with the US EPA's and adjacent states.
- Brings Wisconsin's general laboratory regulations in line with national standards and those adopted by other states.

Laboratory Certification and Registration

Subchapter I: General Provisions

149.01 - Purpose

The purpose of this code (NR149) is to establish a program for the certification and registration of laboratories performing testing under Wisconsin state statute 299.11.

149.02 - Applicability

NR149 applies to your laboratory if you are submitting data for a covered program. If you are not sure if the program that your data is submitted to is a covered program - consult the program. Plant process control samples are not compliance samples.

Administrative codes and programs requiring analyses to be performed by a certified or registered laboratory are chs. NR

- 110 – Sewerage Systems,
- 113 – Servicing Septic Systems,
- 123 – Well Compensation Program,
- 131 – Metallic Mineral Prospecting,
- 132 – Metallic Mineral Mining,
- 140 – Groundwater Quality,
- 145 – Private Wells,
- 150 – Environmental Analysis and Review Procedures,
- 157 – Management of PCBs,
- 158 – Hazardous Substance Discharge Notification,

- 182 – Metallic Mining Waste,
- 206 – Land Disposal of Municipal and Domestic Wastewaters,
- 210 – Sewage Treatment Works,
- 211 – General Pretreatment Requirements,
- 212 – Wasteload Allocated Effluent Limits,
- 214 – Land Treatment of Industrial Liquid Wastes,
- 216 – Stormwater Management,
- 219 – Analytical Test Methods and Procedures,
- 347 – Sediment Sampling and Analysis,
- 507 – Environmental Monitoring for Landfills,
- 661 – Hazardous Waste Identification and Listing,
- 662 – Hazardous Waste Generator Standards,
- 664 – Hazardous Waste Treatment, Storage and Disposal Facility Standards,
- 665 – Interim License Hazardous Waste Treatment, Storage and Disposal Facility Standards,
- 700 – General Requirements for Investigation and Remediation of Environmental Contamination,
- 712 – Environmental Response Actions,
- 716 –Site Investigations,
- 809 – Safe Drinking Water,
- 811 – Design of Community Water Supplies,
- 845 – County Administration of NR 812 (Private Wells),
- HFS 46 – Group Day Care Centers for Children.

A covered program includes the programs listed above and includes any department program, project, permit, contract or site investigation that requires analytical work to be performed by a certified or registered laboratory.

NR149 requirements are the minimum requirements that must be followed by all laboratories.

In addition to NR149 requirements, all laboratories must follow the requirements set forth in the methods they use unless NR149 grants explicit, alternative allowances.

When determining what requirements to follow, the strictest requirement of all requirements must be followed when the requirement is listed in the method, and Code, and Statute. If the method is silent on a requirement then the Code requirement must be followed if one exists.

Legal priority of requirements
1. Statute (299.11)
2. Code (NR149)
3. Method

Example: Method 123 requires that 2 standards and a blank be used for initial calibration. NR149 requires that 3 standards and a blank be used for initial calibration. Statute 299.11 is silent on initial calibration. The strictest requirement of all three is NR149 and therefore 3 standards and a blank must be used for initial calibration even though the method only requires 2.

149.03 - Definitions

New definitions were added and old definitions were updated. Definitions of note are listed below.

Term	Definition
Chain of Custody	Custody must be documented from collection through disposal. This requirement applies to all labs except for those that collect their own samples. Certified WWTP's will have to document sample receipt through disposal for all samples that are not from their plant.

Chain of Custody Form	Documents a signature, date and time, the transfer of the sample from collector to transport to the laboratory and laboratory sample receipt.
Laboratory control sample (LCS)	Also commonly called blank spike or lab fortified blank. This a QC sample that has a "clean" matrix (reagent water or Ottawa sand) that is spiked with the analytes of interest and this sample is processed through the method just like normal samples and the method blank. The purpose of an LCS is to demonstrate that the laboratory is able to obtain accurate results in a clean matrix. This establishes that your procedure is in control.
Limit of Detection (LOD)	For our purposes is equal to the Method Detection Limit (MDL).
Proficiency Testing (PT) samples	Are the equivalent to Performance Evaluation (PE) samples or reference samples. PT samples are required for each analyte or analyte group for each matrix where they are commercially available.
Quality Control Standard (QCS)	Is equivalent to what was called Blind samples.
Quality System	Is a documented management plan to ensure quality throughout the laboratory.
Quality System Matrix	Defines the different matrix types that the department considers to be unique. Each quality system matrix will require their own set of quality control limits. The quality system matrices are ➢ Biosolids (sludge) ➢ Chemical Wastes ➢ Drinking water ➢ Groundwater ➢ Leaching procedure extracts ➢ Oil ➢ Soil ➢ Wastewater effluent ➢ Wastewater influent
Registered laboratory	Does not perform any analyses for hire or any drinking water analyses. A registered laboratory may be an industrial lab, a lab owned by a municipality, or a lab owned by more than one municipality performing tests solely for the owning municipalities.
Second source standard	A standard from a vendor that is different than the laboratory's calibration standard. It may also be a different lot number from the same vendor as the calibration standards.
Traceability	Is the ability to relate a result or measurement to the appropriate state, national or international standards through an unbroken chain of documentation.

Matrix	Percent Settleable (or Total) Solids
Aqueous	< 15%
Either Aqueous or Solid	10-15%
Solid	> 15%

Batch	Definition (QC samples are not counted as samples)
Analytical	Samples analyzed together as a group without interruption. - No maximum limit on number of samples. -

Preparation (batches of 8 or more samples)	Samples prepared together as a group within a 24 hr period.
Preparation (batches of 7 or less samples)	Samples prepared together within a 1 week period.

Blank	Definition
Instrument	Used to "zero" the response of an analytical instrument.
Calibration	Used as a zero concentration standard in the calibration. The instrument response is used; the response of a zero concentration standard is not automatically assigned a zero response.
Method	A blank sample (with clean matrix) prepared and analyzed exactly as the samples that are associated to it.
Temperature	A sample container filled with at least 40 mL of water transported with the samples from sample collection to arrival at the laboratory. This blank is used to determine the temperature of the samples during shipment and at receipt.

Tiers of Accreditation: Aqueous & Solid (non-drinking water)	Tiers of Accreditation (drinking water)
Matrix	Matrix
Analytical Technology	Method
Analyte or Analyte Group	Analyte or Analyte Group

149.04 - Disclaimer

Certification or Registration status does not imply that the department endorses the quality of the data generated by the laboratory.

Subchapter II: Program Administration

149.05 - Required certification or registration

Indicates when certification or registration is required.

149.06 - Certificates

Indicates when and to whom certificates shall be issued, who maintains ultimate ownership of the certificates and modifications of the certificates. Certificates must be displayed somewhere in the laboratory where they are clearly visible.

149.07 - Transfer of certification and registration

Indicates the conditions upon which the department may approve certificate transfers.

149.08 - Recognition of other certifications, registrations, accreditations, licenses or approvals

Indicates which government or private organizations the department will recognize the agreements they have made regarding laboratory approvals. The private organization agreement does not imply that the department will accept laboratories as accredited based solely on their NELAC credentials.

149.09 - Certification Standards Review Council

Indicates how the certification standards review council interacts with the department.

149.10 - Enforcement

Indicates specific actions that can lead the department to implement enforcement.

149.11 - Discretionary Acceptance

Indicates the department's policy regarding discretionary acceptance of data from laboratories that are neither certified nor registered.

149.12 – Variances

Indicates the department's policy in granting and approving variances. In addition, the steps required to request variances are provided.

Subchapter III: Program Structure

149.13 - Fields of accreditation (certification and registration)

Accreditation is now granted based on a 3-tier structure. The department grants accreditation as certification or registration. The biggest change here is that in the past both aqueous and solids could be performed under one certification or registration. Now you will have to maintain specific certification or registration on the solid matrix in addition to the aqueous one.

Determine accreditation type:

- Registration
- Certification

Determine matrices (Tier 1):

- Aqueous
- Solid
- Drinking Water

Determine Analytical Technology or Method (Tier 2):

- For aqueous and solid matrices there are 23 different analytical technologies to choose from - they are listed in NR 149.13.
- For drinking water choose an approved method.

Determine which analyte or analyte group (Tier 3)

- The analytes or analyte groups that the department will certify or register are listed in Appendices I and II of NR149.

The five major categories of accreditation and matrix are listed below. In each of these categories use Tiers II and III to be specific.

Registered	Registered	Certification	Certification	Certification
Aqueous	Solid	Aqueous	Solid	Drinking Water
Analytical Technology	Analytical Technology	Analytical Technology	Analytical Technology	Method
Analyte or Analyte Group	Analyte or Analyte Group	Analyte or Analyte Group	Analyte or Analyte Group	Analyte or Analyte Group

Accreditation is not specific to preparatory steps.

Waste characterization extractions include only the extraction procedure (TCLP, SPLP, ASTM, …). Laboratories must also maintain certification or registration for any analyte to be determined in the resulting extract.

NR 149 also defines "classes" of analyte groups for aqueous and solid matrices (NR 149.13Table 3) and "classes" of analyte groups for the drinking water matrix (NR 149.13 Table 4).

For a small wastewater lab analyzing BOD, TSS, NH3-N (by ISE) and TP (colorimetric) the new certificate would look something on the order of:

REGISTRATION

Matrix: Aqueous

Technology: Oxygen Demand assays (BOD or cBOD)

- **Analyte:** Biochemical Oxygen Demand (BOD)

Technology: Electrometric Assays

- **Analyte:** Ammonia as N

Technology: Colorimetric or Nephelometric

- **Analyte:** Total Phosphorus

Technology: Gravimetric Assays – Residue

- **Analyte:** Totals Suspended Solids

For a commercial lab analyzing lead in both aqueous and solid matrices using more than one analytical technique the new certificate would look something on the order of.

CERTIFICATION
Matrix: Aqueous
Class: Metals
Technology: FLAA
- **Analyte**: Lead

Technology: GFAA
- **Analyte**: Lead

Technology: ICP
- **Analyte:** Lead

CERTIFICATION
Matrix: Solid
Class: Metals
Technology: FLAA
Analyte: Lead
Technology: ICP
Analyte: Lead

Subchapter IV: Certification and Registration Process

149.14 - Application for certification or registration

Indicates the steps required to apply for an initial certification or registration. Initial applications are required for laboratories seeking certification or registration for the first time, wanting certification or registration in additional matrices, or changing their status from a registered to a certified laboratory.

Indicates the steps required to apply for a revised certification or registration. Revised applications are used by certified or registered laboratories to apply for additional techniques for a certified or registered matrix or additional analytes in a certified or registered analytical technique.

Indicates the criteria required to apply for transfer of certification or registration.

Indicates the steps required to apply for reciprocal agreement certification or registration.

Indicates the department's timetable for issuing certificates.

149.15 - Period, renewal and expiration of certification or registration

The certification and registration period starts on September 1 and ends on August 31 of the following year for all laboratories certified or registered directly by the Department.

Indicates the steps required for the annual renewal process.

149.16 - Notification of relocation

Indicates the requirements for laboratory relocation.

149.17 – Laboratory name change

Indicates the requirements for laboratory name change.

149.18 - Subcontracting of analyses by certified or registered laboratories

Indicates the requirements to subcontract analyses.

149.19 – Requirements for certification in the drinking water matrix

Indicates that labs must follow the minimum criteria and procedures listed in Chapters III and IV or the "Manual for the Certification of Laboratories Analyzing Drinking Water" 5th Edition.

Replicates (or matrix spike duplicates) – required 1/batch or 1/20 samples if more than 20 samples.

MDL requirements are presented.

Certification is not required for

- Fluoride under NR 809
- Free residual chlorine or total residual chlorine under NR 809.705
- pH under NR 809.14
- Turbidity under NR 809.725, Table A

Laboratories must notify water supply facilities within 48 hours after completing analysis whenever compliance samples exceed the MCL.

149.20 – Requirements for certification or registration in the whole effluent toxicity analyte class

This section applies to laboratories perform whole effluent toxicity testing.

149.21 - Fees

The section describes the schedule, structure and formula for assessing fees to laboratories.

As with most other aspects of NR 149, changes related to program fees are most directly related to program structure changes. The Laboratory Certification & Registration Program operates under a simple balanced budget. Fees are collected to equal the cost of running the program each year. Each test category is assigned a number of "relative value units" (RVU) that roughly equates to the technical difficulty associated with performing tests under that test category. The cost to run the program for a given certification period is simply divided by the total number of RVUs for the labs in the program to obtain an individual RVU cost. The DNR's Natural Resources Board approves the Lab Certification & Registration Program's budget annually in March. Lab fees are then assessed each May by multiplying the number of lab RVUs by the cost per RVU.

While the basic RVU process for determining fees is not changing, the change from category—test structure to a matrix—technology—analyte system required us to establish RVUs for each technology, rather than each test category. This allowed us to build in greater equity. One example was the BOD test. BOD has historically been considered an "entry-level" test in many labs, whereby entry level analysts are assigned BOD testing. In practice, however, BOD is more of a bioassay than a "bucket chemistry" type of test, and the Standard Methods procedure requires a significant level of detail and procedure to perform the test correctly. Consequently, we have increased BOD from 1 RVU to 3 RVU to reflect the difficulty level associated with performing this test correctly.

Transitioning: Fees for FY09 (9/1/08 - 8/31/09)
Since the revisions to NR149 do not take effect until September 1, 2008 and fees for the September 1, 2008 to August 31, 2009 certification period must be invoiced in May of 2008 (due July 1, 2008) the fees associated with the new structure will not be implemented until May of 2009 for the 2010 certification period.

Subchapter V: Proficiency Testing

149.22 – Required analyses of proficiency testing samples.

Proficiency testing samples is the new way to described reference samples. The same PT sample can be analyzed by more than one technique.

A PT sample is required per certification or registration period for each analyte or analyte group identified by the department.

For non-DW matrices an aqueous PT sample is required for each combination of
- Technology
- Analyte/Analyte Group

For DW matrix an aqueous PT sample is required for each combination of
- Method
- Analyte/Analyte Group

A solid matrix PT is not required for solid matrices; instead an aqueous PT is used for solids.

The department will provide a list of required testing samples and approved PT sample providers on our website.

PT samples are not required for the following
- Metals analyses in aqueous and solid matrices using Flame atomic absorption Spectrophotometry technique.
- Metals analyses in aqueous and solid matrices using colorimetric techniques (hexavalent chromium is not exempted)
- Ultra-low level metals analyses in aqueous and solid matrices.

Instead these analyses require analysis of QCS three times per year at evenly spaced intervals.

149.23 - Approval of proficiency testing sample providers

This section presents some of the criteria that are used when evaluating approval for PT providers.

149.24 - Schedule of Analysis

For renewals, PT results shall be reported no sooner than January 1 and no later than August 15 of the same calendar year as the effective date of the renewal.

For applications, PT samples shall not be analyzed more than 6 months prior to the date of the application.

Labs with 3 consecutive PT sample failures in a year, for any technique and analyte or analyte group combination, shall submit 2 consecutive acceptable PT samples.

149.25 - Treatment of proficiency testing samples by laboratories

PT samples must be subjected to the same preparatory and analytical steps as the samples unless the preparation instructions submitted by the provider specifically instruct omitting a step. Preparatory steps include digestions, distillations, extractions, concentrations and dilutions.

This section further describes how laboratories are to deal with PT samples.

149.26 - Submittals

This sections indicates how PT sample results are to be submitted to the department.

149.27 – Proficiency testing sample acceptance limits and grading

If the laboratory reports a result for an analyte that is not present in the PT sample (multiple analyte PTs) then that analyte counts as a failure towards the 80% acceptance rule count.

If the laboratory does not report a result for an analyte that is present in the PT sample (multiple analyte PTs) then that analyte counts as a failure towards the 80% acceptance rule count.. For example if the PT sample for VOCs contains benzene, toluene and carbon tetrachloride only, yet the lab reports acceptable results for only benzene and toluene then the PT results for the lab would fail because carbon tetrachloride was present in the sample but it wasn't reported.

This section also describes the PT samples grading criteria.

149.28 - Procedure for correcting unacceptable PT sample results

This section describes the procedures to follow if the laboratory has unacceptable PT sample results.

Subchapter VI: On-site Laboratory Evaluations

149.29 - Purpose, type and frequency

This section describes the purpose of laboratory evaluations, when they are announced or unannounced evaluations and how often they are required.

149.30 - Evaluation procedures and appraisals

The program promotes consistency in on-site evaluations. In addition, the department will provide a mechanism for the laboratories to voluntarily appraise the evaluation process.

149.31 - Evaluation reports

The department will issue reports to the evaluated laboratory within 30 days of the conclusion of the on-site visit that documents the deficiencies observed during the evaluation.

149.32 - Evaluation corrective action

This section describes the department and laboratories responsibilities in regards to the corrective actions taken to resolve the on-site deficiencies that were discovered.

149.33 - Conflicts of interest

The department has a written policy that describes situations where evaluator conflicts would exist. In those cases, the evaluator would not be able to evaluate the laboratory and instead a different evaluator, one devoid of any conflicts of interest would be assigned. The intent is to use auditors that can perform an objective and unbiased evaluation of the laboratory.

149.34 – Evaluator qualifications

The department has documentation on file that demonstrates the evaluator's education, experience and credentials to perform evaluations competently.

Subchapter VII: Quality Systems

149.35 - General requirements

This subchapter contains the core of operational requirements that laboratories need to follow. This section requires that the laboratory name and the department be informed of, the one individual that is responsible for the laboratory's quality system.

149.36 - Laboratory personnel

The section does not specify requirements for education, experience or training of analytical personnel. It does require that all analysts complete an initial demonstration of capability (IDC) or continuing demonstration of capability for any tests they perform in which the referenced methodology specifies a requirement that they be performed. If the referenced methods do NOT specify a procedure for demonstrating capability (e.g. Standard Methods procedures for BOD, TSS, ammonia by ISE, and manual colorimetric phosphorus), then the laboratory is required to develop and document their own internal procedures used to determine that a given analyst has demonstrated the ability to produce quality analytical results.

149.37 - Quality Manual

The laboratory's quality system must be defined in a quality manual. The quality manual was known formerly as the laboratories quality assurance manual. All policies and procedures governing the laboratories quality system shall be documented or referenced in the quality manual. NR149 allows a lab to name the manual however they choose, but it will be referenced as the "quality manual" in any program related documents. The single most significant change is that the that labs must not only have a Quality Manual, but it must contain specific content at a minimum and the lab is required to follow it. There is flexibility in the format of the manual as long as it addresses a set of content elements. Manuals must be kept current and the revision dates must be documented within the manual. The department has created a template quality manual for laboratories to use as a starting point (see the department website for this information).

The minimum elements must be addressed or referred to in the quality manual.

- Organization and management structure of the laboratory.

- Procedures for retention, control and maintenance of documents used in or associated with analyses.
- Procedures for achieving traceability of standards, reagents and reference materials used to derive any results or measurements.
- Procedures for handling samples.
- Lists of major analytical instruments and support equipment.
- Procedures for calibration, verification and maintenance of major analytical instruments and support equipment.
- Procedures for evaluating quality control samples, including, but not limited to, method blanks, laboratory control samples, matrix fortified samples and replicates.
- Procedures for initiating, following up on and documenting corrective action addressing quality assurance and quality control failures, discrepancies or nonconformance.
- Procedures for reviewing analytical data and reporting analytical results.

149.38 - Corrective action for quality system and quality control samples

Laboratories must take corrective action when

- Lab policies or procedures are not followed.
- Any quality control sample fails to meet their acceptance limits. Example QC samples are
  - Calibration curve standards
  - Calibration curves
  - Initial calibration verification standards
  - Continuing calibration verification standards
  - Proficiency testing samples
  - Quality control standard samples
  - LOD studies
  - Method blanks
  - Instrument blanks
  - Laboratory control samples and their duplicates
  - Matrix spike samples and their duplicates
  - Sample duplicates
  - Internal standards
  - Surrogate standards
  - Initial and continuing demonstration of capability samples
  - Tune standards

Corrective actions must identify (document) the following

- What is the problem (source)
- What was done to correct the problem (what was fixed)
- Was the problem corrected (did the fix work)
- If the problem was not corrected, then a new fix must be tried and it's effectiveness evaluated

Corrective actions taken to fix quality control sample failures must be taken before the affected results are reported by the laboratory.

149.39 - Records and documents

This section clarifies the types of administrative, analytical, and technical records laboratories need to maintain to demonstrate compliance with the Chapter. Laboratories still must retain records for a minimum of 3 years after the generation of the last entry in a record or document. If the data that is maintained for 3 years has any associated records that are necessary to reconstruct the data that is older than 3 years, then those records also must be retained.

Administrative Records *(that must be retained)*

- Certificate of certification or registration
- Reciprocal state/agency certificates

- Personnel qualifications, experience and training
- Initial demonstration of capabilities for each analyst
- Copies or access to the regulations, methods or documents necessary to comply with NR149.

Analytical and Technical Records *(that must be retained)*

- **Any data necessary to allow historical reconstruction of all lab activities that contributed to generating reported results (e.g. raw data, derived data, original observations).**
- Collection, arrival, processing, and analysis dates of samples received for analysis.
- Collection and analysis time for tests with ≤ 48 hr hold time.
- Preservation status of samples on arrival at the laboratory.
- Identity of lab personnel preparing and testing samples.
- Identification of the analytes (*groups*) analyzed in samples.
- Preparatory techniques, such as digestions, extractions and clean-ups, to which samples are submitted
- Methods of analysis used for samples.
- Results of sample analysis.
- Traceability of standards and reagents used to in analysis (certificates of analysis and prep records)
- Calibration verification information and measurements of lab support equipment associated with sample analysis and storage.
- Initial and continuing calibration data associated with samples analyzed.
- Raw data for analytical instrument calibrations and samples. *EXEMPTION: ICP & ICP/MS & spectrophotometers*
- Results of QC samples associated with samples analyzed.
- Corrective actions associated with samples analyzed.
- Maintenance performed on lab support equipment and analytical instruments.
- Environmental conditions crucial to tests performed at lab facilities at the time samples are analyzed.
- Reports of final results submitted to clients or the DNR.

149.40 - Standard operating procedures (SOPs)

Written SOPs are required that document or reference the activities needed to maintain their quality systems and that enable performing or reproducing an analysis in its entirety as performed at the laboratory. Laboratories SOPs can consist of copies of published documents (manuals, procedures, methods – if followed exactly), annotated published documents with modifications, or in-house written procedures. SOPs must indicate their date of issue or revision. SOPs that described the analytical methods that the laboratory uses for testing will be called "analytical methods" and will be maintained in an analytical methods manual. At a minimum, each "analytical method" (method SOP) must address or refer to the following minimum elements:

1. Identification of the test method.
2. Applicable analytes.
3. Applicable matrices.
4. Method sensitivity.
5. Potential interferences.
6. Equipment and analytical instruments.
7. Consumable supplies, reagents and standards.
8. Sample preservation, storage and hold time.
9. Quality control samples and frequency of their analysis.
10. Calibration and standardization.
11. Procedure for analysis.
12. Data assessment and acceptance criteria for quality control measures.
13. Corrective actions and contingencies for handling out of control or unacceptable data.

Listed below is a distillation of specific SOPs which are now required under NR149. This list should be considered minimum requirements. Certainly other SOPs can and should be developed; but these are SOPs clearly specified by rule and ones which your auditor will be looking for once audits under the new rule begin in September 2008.

SUBJECT	SOP Required	NR149 ref.
CALIBRATION	Procedures for calibration, verification and maintenance of major analytical instruments and support equipment.	149.37(3)(f)
EQUIP MAINTENANCE	The laboratory shall establish procedures for the maintenance of analytical instruments to prevent contamination or deterioration that may affect reported results.	149.44(4)(b)
ANALYTICAL METHOD SOPS	Laboratories shall maintain written standard operating procedures that document or reference activities needed to maintain their quality systems and that enable performing or reproducing an analysis in its entirety as performed at the laboratory.	149.40(1)(a)
QC: BLANKS & ZEROING	The laboratory shall establish procedures for zeroing an instrument and the treatment of calibration blanks, when the referenced analytical method used by the laboratory requires the response of a calibration blank to be part of a calibration function.	149.44(6)(h)
QC: CONFIRMATION OF ORGANIC ANALYTES	The laboratory shall establish procedures to confirm the results of organic analytes determined by techniques that, unlike mass spectrometry, do not provide a positive unique identification when… *1. The history of a sample source does not suggest the likely presence of the detected analyte. 2. A client or approved project plan requires it.)*	149.48(9)(a)
QC: CORRECTIVE ACTION	Procedures for initiating, following up on and documenting corrective action addressing quality assurance and quality control failures, discrepancies or nonconformance.	149.37(3)(h)
QC: LOD & LOQ	Laboratories shall establish procedures to relate limits of detection to limits of quantitation.	149.48(2)(f)
QC SAMPLES	Procedures for evaluating quality control samples, including, but not limited to, method blanks, laboratory control samples, matrix fortified samples and replicates.	149.37(3)(g)
QC SAMPLE BATCH REQUIREMENTS	Laboratories shall establish procedures for identifying and documenting preparation batches that facilitate determining compliance with the frequencies of quality control samples required by this subchapter.	149.48(1)(d)
RECORDS	Procedures for retention, control and maintenance of documents used in or associated with analyses.	149.37(3)(b)
RECORDS	Procedures to control and manage all records and documents that form part of its quality system and that are required to demonstrate compliance with this chapter. The procedures shall ensure that documents required to perform analyses and to ensure the quality of generated data are available to laboratory personnel, and that records and documents are reviewed periodically for continuing suitability and, when necessary, revised to facilitate compliance with the requirements of this chapter.	149.39(1)(a)
RECORDS	Procedures to prevent unauthorized access or amendments to records and documents.	149.39(1)(g) 4.
REPORTING	Procedures for reviewing analytical data and reporting analytical results. (Required to be in Quality Manual)	149.37(3)(i)

REPORTING	The laboratory shall establish procedures and rules for reporting results for samples analyzed by dual column and dual detector systems. These procedures must declare: (1)Under what conditions a presumptive identification is confirmed. (2) Under what conditions a presumptive identification is reported. (3) The value that will be reported when the dual systems both provide quantitative confirmed results	149.48(9)(b)
SAMPLE HANDLING	Sample Acceptance Policy. The laboratory shall have and follow a written policy that clearly outlines the conditions under which samples will be accepted or rejected for analysis, or under which associated reported results will be qualified.	149.46(2)(a)
SAMPLE HANDLING	Procedures for handling samples. (Required to be in Quality Manual)	149.37(3)(d)
SAMPLE HANDLING	The laboratory shall establish and follow procedures for identifying samples uniquely. (The procedures shall ensure that the identity of samples cannot be confused physically or when referenced in records or other documents.)	149.46(3)(a)
SAMPLE STORAGE	The laboratory shall have procedures and appropriate facilities for avoiding deterioration, contamination, loss or damage of samples during storage.	149.46(6)(a)
SAMPLE CONTAINERS	When the laboratory provides containers and preservatives for sample collection, including bulk sampling containers such as "carboys", the laboratory shall have standard operating procedures in place which address concerns that the containers are adequately cleaned and not contributing to contamination of samples, do not contain analytes of interest at levels which will affect sample determinations and that the preservatives used are sufficiently pure to maintain the validity of reported results. NOTE: The laboratory should establish procedures to ensure and document that the sample containers it provides do not contribute contaminants before they are used for collecting samples.	149.46(1)(b)
TRACEABILITY	Procedures for achieving traceability of standards, reagents, and reference materials used to derive any results or measurements. (Required to be in Quality Manual)	149.37(3)(c)

149.41 - Method selection

Laboratories must choose methods that are approved by covered programs under this code and that are suitable for the matrix, parameter, concentration level, regulatory limit and potential interferences in the samples to be tested. When methods are not prescribed by the covered programs the laboratory shall consult with the Department for an appropriate choice.

149.42 - Alternative methods

Laboratories may use methods other than those approved by the covered programs if the EPA has already approved the method and if the laboratory requests approval to use this alternate method. This section describes the procedures that laboratories must follow to obtain approval to use an alternative method.

149.43 - Laboratory facilities

This section does not impose specific physical design requirements. It does have general statements requiring laboratories to ensure separation of incompatible analytical activities, and to monitor environmental conditions when they influence test results (e.g., BOD, TCLP).

149.44 - Laboratory equipment

This section introduces the concept of splitting equipment into two types: support equipment (e.g., ovens, balances, thermometers) and analytical equipment (e.g., DO meters, ion meters, spectrophotometers, GCs).

Procedures for verifying the accuracy of support equipment are included, such as verifying the calibration status of balances, thermometers, and auto-pipettes.

If support equipment or analytical instruments leaves the direct control of the laboratory for any reason, the laboratory shall ensure that the function and calibration status of that equipment is checked before using it.

Requirements for calibration of analytical instruments are discussed in great detail, something that was lacking in previous versions of the rule. Calibration is broken down into initial (full) calibrations and then the process of verifying that an existing initial calibration remains valid. A maximum one-year time limit is established for initial calibrations. The initial calibration sequence is laid out logically: select a calibration function, determine the number of calibration levels and specific concentrations for them,

This section also introduces the concept of using second source standards to evaluate an initial calibration

- Support equipment
  - All support equipment must be calibrated or verified over its range of use using reference materials traceable to NIST.
  - The acceptance criteria used for calibration checks must be those criterion that are established in approved methods, department guidance or tolerances established by manufacturers.
  - When the results of a calibration or calibration verification do not meet the acceptance criteria then the equipment can not be used until repaired. If the deviation is a consistent bias and a correction factor is employed, the equipment may be used.
  - Devices used to measure temperature must be calibrated or verified at least annually.
  - The operating temperature of autoclaves, incubators, ovens and water baths used as part of a method must be measured on each day that they are used.
  - Refrigerators, freezers, ovens and incubators holding samples continuously must be checked on each day that the lab conducts analyses. The lab must set equipment settings and conditions to maintain the required temperatures on days that the lab does not conduct analyses.
  - Analytical balances must be check at least once a month with a gram weight and a milligram weight.
  - The weights used to verify the analytical balance must be certified for accuracy at least every 5 years by a metrology service outside of the laboratory.
  - Mechanical and automatic volumetric dispensing devices (pipettes, micro-pipettes, burettes, automatic dilutors and dispensers) must be checked for accuracy at least quarterly when they are used.

- Analytical instruments
  - Operators must be trained
  - Instructions on the instrument use and maintenance must be available to operators
  - All instruments must have a documented maintenance program
  - Analytical instruments must be calibrated at least once in a year.
  - Calibrations must be verified before using the instrument to determine quantitative results.

- **Initial (full) calibration**
  - Laboratories shall perform an initial calibration after instruments undergo non-routine maintenance, when repeated use or other conditions change their expected behavior, and when their continuing calibration cannot be verified.
  - The minimum number of standard concentrations required to establish calibration shall be 3 except for:
    - DO – 1
    - ICP and ICPMS – 1
    - ISE and pH meters - 2
    - Quadratic calibrations - 5
    - Cubic calibrations – 7
  - One of the standards in the initial calibration must be at a concentration near the LOQ

- The laboratory shall provide the equations and coefficients or parameters necessary to characterize the calibration function unless the instrument is a DO, ISE or pH meter.
- The laboratory must use the simplest linear calibration function unless it has documentation that proves a non-linear or weighted function provides a statistically improved definition for the calibration range.
- Calibrations may not be forced through zero.
- Laboratories shall establish acceptability criteria for the initial calibration. Some of the accepted criteria are as follows:
  - Response factors relative standard deviation must be ≤ 20% [unless the method specifies otherwise]
  - Linear regression, for inorganic analyses, correlation coefficient, $r \geq 0.995$
  - Linear regression, for organic analyses, correlation coefficient, $r \geq 0.990$
  - Quadratic regression, for inorganic analyses, coefficient of determination, $r^2 \geq 0.995$
  - Quadratic regression, for organic analyses, coefficient of determination, $r^2 \geq 0.990$
- Laboratories shall establish procedures for zeroing an instrument and the treatment of the calibration blank when the referenced method requires the response of a calibration blank to be part of the calibration curve
- Results must be calculated from initial calibrations unless otherwise allowed by regulation, method or program.
- The instrument response must be within the response range of the initial calibration or samples must be diluted such that the diluted sample response is within the range.
- When a dilution is required the dilution must be the lowest dilution required to obtain a response within a the calibration. So if 5 analytes in a sample require a dilution and all 5 are at different concentrations, you must dilute the sample so that the lowest result is within the range of the curve. You cannot dilute the sample once for just the highest concentration analyte. Exceptions do exist for ICP and ICPMS where LDR studies are well defined, see code for details.

- **Initial calibration verification (ICV)**
  - All initial calibrations must be verified using a second source standard before quantitating any samples, unless
    - The instrument is a DO, ISE or pH meter.
    - The laboratory analyzes quality control standards as required.
  - The acceptance criteria for the ICV will be the same as the CCV unless otherwise required by regulation, method or program.

When more stringent instrument calibration verification (ICV or CCV) requirements are required in mandated test methods or regulations they must be followed except when:

- The method requires analyzing more than 3 standards for linear calibration, and the lab narrows the calibration to no more than 2 orders of magnitude and uses at least 3 standards.
- The method requires analyzing more than one CCV for a linear calibration and the lab has narrowed the calibration to no more than 2 orders of magnitude and uses at least one standard to verify continued calibration.

- **Continuing calibration verification (CCV)**

When an initial (full) calibration is not performed on the day of analysis, the validity of the initial calibration shall be verified prior to quantitating samples by analyzing a continuing calibration verification (CCV) standard.

A CCV standard shall also be analyzed after the consecutive analysis of each group of 20 samples.

CCV standards are not required for analyses that cannot be spiked, such as BOD, cBOD and TSS, or those analyses that do not involve a calibration, such as titrations.

The CCV standards can be from the same source used to generate the initial calibration. The number and concentration of calibration standards required to demonstrate continuing instrument calibration is outlined below.

CALIBRATION FUNCTION	# OF STANDARDS REQUIRED FOR VERIFICATION	CONCENTRATION OF VERIFICATION STANDARD
Tuning an instrument to conform to a universally accepted scientific law or scale (i.e. electrometric techniques)	The laboratory shall analyze at least a single verification standard	The concentration of the standard shall be within the range established during the initial calibration
Average response/ calibration factor, linear regression, least squares analysis, or otherwise obeys a linear model	The laboratory shall analyze at least a single verification standard	The concentration of the standard may be varied within the established calibration range.
Quadratic regression, 2nd order polynomial, or other quadratic model	The laboratory shall analyze at least 2 verification standards	One of the standard concentrations shall be chosen to verify continuing calibration near the point of inflection of the calibration function
Cubic regression, 3rd order polynomial, or other cubic model	The laboratory shall analyze at least 3 verification standards	Two of the standard concentrations shall be chosen to verify continuing calibration near the points of inflection of the calibration function
Discrete or non-smooth segments	The laboratory shall analyze one standard per calibration segment	The concentrations of the standards shall be different from the ones used to establish each segment.

The acceptance criteria for CCV standards must be those defined in the referenced method. If the referenced method does not contain acceptance criteria for CCV standards use the following for all reported analytes:

Inorganic CCVs: 90 – 110%
Organic CCVs: 85 – 115%

If any of the analytes fail in the CCV the lab can re-analyze the CCV. If the reanalyzed CCV passes you can continue with normal reporting. If the reanalyzed CCV fails again then the lab must take corrective action before analyzing any samples. Once corrective action has been performed, the lab must pass 2 consecutive CCVs before analyzing samples or an initial (full) calibration must be performed.

If samples must be reported with failing CCV results the samples must be qualified appropriately.

149.45 - Measurement Traceability

This section indicates that the follow measures of traceability be taken.

- All results must be traceable to the standards, reagents, and reference materials used to derive results.
- All analytical and technical records (raw data, derived data or original observations) must be maintained such that historical reconstruction of all lab activities can be made as they contributed to the generation of results.
- Labs must certify the accuracy of all reference materials used to calibrate or verify methods.

- ❑ The identity, source, lot number, receipt date, expiration date, and purity of all standards and reagents used must be documented.
- ❑ Original containers of standards and reagents must be labeled with receipt and expiration dates.
- ❑ Preparation records must be kept that detail how working standards and reagents are prepared.

149.46 - Handling of samples

Sample Collection

- Labs must maintain records supplied by the sample collectors.
- If labs provide bottles for sample collection, the lab must have SOPs in place to address cleaning of said bottles.

Sample Acceptance Policy

- Labs must have and follow a written policy that clearly outlines the conditions under which samples will be accepted or rejected or qualified results

Sample Handling

- All samples must be identified uniquely.
- The unique sample id must be placed on a sample container as a durable label.
- Chain of custody documentation must take place when a facility does not perform their own sample collection, transport and analysis.

Sample Preservation and Holding Time

- Sample preservation and holding times established by State and Federal regulations take precedence over those listed in referenced methods. When this information is not listed in State or Federal regulations the method requirements shall be followed.

Sample Receipt Documentation

- Sample receipt and condition must be documented in chronological order.
- The following information must be documented
  - The entity submitting the samples
  - Collect date
  - Collect time (if test being analyzed has a holding time of 48 hrs or less)
  - Received date
  - Lab unique identification number
  - Sample preservation status upon receipt
  - Other sample conditions upon receipt
  - A unique link between the field sample id and the lab sample id
  - The requested analyses
  - The requested methods when the sample collector specifies them
  - Any comments if samples did not meet the lab sample acceptance policy

Sample Storage (also applies to sample extracts, digestates, leachates or concentrates)

- Procedures must be in place for avoiding sample deterioration, contamination, loss or damage during storage.
- Thermal preservation must be maintained.
- Samples must be stored separately from all standards, reagents, food and other potentially contaminating sources.

149.47 - Laboratory test reports

Laboratory test reports must include at least the following information:

- Name, address, contact name and telephone number of the lab who performed the test

- Lab certification or registration number
- The name and address of the entity whose samples were analyzed
- Field Id
- Analysis methods used
- Collect date
- Receipt date
- Preparation date (if applicable)
- Analysis date
- Parameter name, final result and unit of measure
- Dilution factor (if applicable)
- If samples are reported on a dry weight basis (adjusted for moisture content) then the report must include a statement to this end and the solids result (along with the LOD and LOQ)
- If the parameter reported requires that a LOD be reported, then the LOD and LOQ must be reported.
- If results required adjustment for dilution, there must be some indication that the LOD and LOQ have also been adjusted accordingly
- Names and signatures of the responsible party authorizing the report.
- Description of any deviations encountered by the lab
- Test report date

Amended Test Reports

- Amended test reports must clearly identify the reason for the amendment and reference the original report.

Subcontracted Test Results

- The subcontractors facility identification code and any qualifiers identified by the subcontracting lab must be reported along with the associated results.

149.48 - Quality control requirements for chemical testing

Laboratories may not adjust or correct the sample results by the recoveries of associated LCLS, MS or surrogates unless a method or project plan approved by the department requires it.

Laboratories may not subtract results found in method blanks from sample results unless a method or project plan approved by the department requires it.

Labs must establish procedures for identifying and documenting preparation batches that facilitate determining compliance with the frequency of QC samples required by this code.

LOD

- Must be determined for all parameters reported except
  - BOD
  - Tests for which analyzing a spike sample is impossible
  - Titrimetric tests
  - Gravimetric tests (except oil and grease)
- All steps in the method must be part of the LOD study.
- Must be determined at least annually.
- Must be determined each time there is a change in the test method or instrumentation such that sensitivity is affected.
- LOQs must be determined (via an established procedure) such that they are greater than the LOD.

Calibration blank means a sample containing insignificant or undetectable levels of target analytes used to establish the analytical zero of a calibration function.

Method Blank (MB)

- The method blank must go through all steps in the method, the same as the samples.
- Not required for pH, alkalinity, conductivity or solids determination.
- One per preparation batch required
- Must be in addition to a calibration blank when a CB is required.
- If any parameters are detected at concentrations greater than the LOD, the lab shall evaluate the nature of the contamination and its effect on the associated samples.
- Samples in a batch must be reanalyzed, reprepped or qualified if the method blank exceeds the highest of the following:
    - LOD
    - 5% of the regulatory limit for that parameter
    - 10% of the concentration of the sample (evaluate each sample)

Laboratory control sample (LCS)

- One per preparation batch required
- The LCS must go through all steps in the method, the same as the samples (except leaching).
- Not required for tests where spiking the sample is not possible (pH, solids, chlorophyll a, color, odor)
- Matrix spikes or certified reference material may be processed for all reported analytes in place of LCS, if the acceptance criteria used on the MS is the LCS acceptance criteria (and the lab must take corrective action if the MS fails)
- Must calculate the percent recovery for each reported analyte
- The LCS for BOD and CBOD tests.
    - Use GGA
    - One per prep batch if more than 20 samples analyzed per week
    - One per week if less than 20 samples analyzed per week
- The LCS for PCBs
    - At least one Aroclor must be spiked per prep batch
- The LCS for other multi-peaked parameters (i.e. toxaphene, chlordane)
    - Spike with only one of the mulit-peak analytes
    - Each one of the parameters must be spiked at least once a year.
- Acceptance criteria for LCS is as follows:
    - If published by the department, can use that criteria
    - If not published by the department use:
        - Method defined criteria
        - In-house laboratory limits if the method does not define criteria
        - Project plan criteria
- If the LCS fails
    - Reprocess LCS and associated samples
    - Reanalyze LCS and associated samples
    - Qualify associated samples

Quality control standard (QCS)

- If second source standards are not used to verify the initial calibration then QCSs are required.
- QCS required 3 times per year at evenly spaced intervals for all certified or registered parameters (that are amenable to fortification and are commercially available)
- Not required for pH, ISE, BOD, TSS, DO meters
- Acceptance criteria are those provided by the vendor.
- If failed, need to do another one within 30 days.
- The QCS must go through all steps in the method, the same as the samples.

Matrix Spike (MS) and Matrix Spike Duplicate (MSD)

- Required only when the test method requires them and sufficient sample has been received.
- Required when listed in project plans
- Required when used in place of LCS samples
- Not required for BOD, CBOD, pH, solids, alkalinity, acidity, chlorophyll a, color, odor

- The MS and MSD must go through all steps in the method, the same as the samples (except leaching).
- One per preparation batch required for each quality system matrix
- Spike with all analytes as specified in the method, project plan or client or all reported analytes
- Spiked with all analytes when used in lie of LCS
- Must calculate recovery and RPD
- Acceptance criteria for MS and MSD is as follows:
    - If published by the department, can use that criteria
    - If not published by the department use:
        - Method defined criteria
        - In-house laboratory limits if the method does not define criteria
        - Project plan criteria
- If the MS fails
    - Reprocess MS and associated parent sample
    - Reanalyze MS and associated parent sample
    - Qualify associated parent sample

Sample Replicate

- May be analyzed in place of MSD when there is a high probability that the sample will contain the analytes of interest at or above the LOQ
- Required only when the test method requires them and sufficient sample has been received.
- Required when listed in project plans
- The sample replicate must go through all steps in the method
- One per preparation batch required for each quality system matrix
- Must calculate RPD or absolute range
- Acceptance criteria for sample replicates is as follows:
    - If published by the department, can use that criteria
    - If not published by the department use:
        - Method defined criteria
        - In-house laboratory limits if the method does not define criteria
        - Project plan criteria
- If the sample replicate fails
    - Reprocess MS and associated parent sample
    - Reanalyze MS and associated parent sample
    - Qualify associated parent sample

Surrogate Spikes

- When required in methods or project plans add to all samples and quality control samples in the prep batch.
- Must calculate recovery for each surrogate added to each sample.
- Acceptance criteria for surrogates is as follows:
    - If published by the department, can use that criteria
    - If not published by the department use:
        - Method defined criteria
        - In-house laboratory limits if the method does not define criteria
        - Project plan criteria
- If the surrogate fails
    - Determine if the failure is a matrix effect or not, if so, qualify the results.
    - If not matrix effect, reprocess and reanalyze the affected sample or qualify results

Selectivity

- Lab must establish procedures to confirm results of organic analytes when the technique used does not provide a unique identification (i.e GCMS) in the following cases
    - History of the sample does not suggest the likely presence
    - Client or project plan require it
- When a dual column or dual detector is used, the lab must establish rules and procedures for reporting these results

  - When is presence considered confirmed
  - When is presence reported
  - What value is reported
- Lab must develop and document acceptance criteria for retention time windows
- Lab must develop acceptance criteria for MS tuning

149.49 - Quality control requirements for whole effluent toxicity testing

Laboratories analyzing whole effluents for acute and chronic toxicity shall follow the QC requirements referenced in the "State of Wisconsin Aquatic Life Toxicity Methods Manual", 2nd edition. Chemical testing in support of whole effluent toxicity testing shall follow the QC requirements specified in NR149.48 except that laboratories do not need to analyze matrix spikes or matrix spike duplicates for ammonia or hardness.