METHOD TNTP-W10900

TOTAL NITROGEN AND TOTAL PHOSPHORUS IN PULP AND PAPER BIOLOGICALLY TREATED EFFLUENT BY ALKALINE PERSULFATE DIGESTION

NCASI
West Coast Regional Center
Aquatic Biology Program
June 2011

Acknowledgements

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National Council for Air and Stream Improvement, Inc. (NCASI). 2011. *NCASI Methods Manual*. Method TNTP-W10900: Total nitrogen and total phosphorus in pulp and paper biologically treated effluent by alkaline persulfate digestion. Research Triangle Park, NC: National Council for Air and Stream Improvement, Inc.

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NCASI Method TNTP-W10900

Total Nitrogen and Total Phosphorus in Pulp and Paper Biologically Treated Effluent by Alkaline Persulfate Digestion

1.0 Scope and Application

1.1 This method is applicable to determinations of total nitrogen (TN) and total phosphorus (TP) in acid preserved pulp, paper, and paperboard mill biologically treated effluent (PP-BTWW). The applicable range of this method is 0.01 to 3.0 mg P/L and 0.1 to 6.0 mg N/L. The range may be extended with sample dilution. Sample size is 7 mL.

1.2 Laboratories intending to use this procedure should establish method detection limits for each sample matrix type. Changes in technology may influence chemical compositions of these matrices, requiring evaluation of samples for digestion efficiency and method performance. Any modification of the method beyond those expressly permitted is subject to application and approval of alternative test procedures under 40 CFR Parts 136.4 and 136.5 (Section 16.1).

2.0 Summary of Method

2.1 The method simultaneously converts ammonia and inorganic (excluding N_2) and organic nitrogen to nitrate and inorganic and organic forms of phosphorus to orthophosphate by means of an alkaline to acid digestion. Native dissolved nitrate and nitrite are unaffected by the digestion.

2.2 Digestion is accomplished by heating acidified, unfiltered samples in the presence of persulfate (a strong oxidizer) at 120^{o}C and 15 psi positive pressure for 30 minutes. The digestion is strongly alkaline at the outset, which promotes oxidation of nitrogen to nitrate. As the persulfate decomposes the solution becomes acidic, allowing oxidation of phosphorus to orthophosphate (Section 16.2). Following digestion, separate semi-automated or manual colorimetric determinative methods of the single digestate are conducted for analysis of TN and TP.

2.3 TN measurement occurs by means of nitrite/nitrate determinative techniques. TP measurements occur by means of orthophosphate determinative techniques (Section 6.5).

3.0 Definitions

3.1 The definitions and purposes herein are specific to this method but conform to common usage as much as possible.

3.1.1 May – this action, activity, or procedural step is neither required nor prohibited

3.1.2 Must – this action, activity, or procedural step is required

3.1.3 Must not – this action, activity, or procedural step is prohibited

3.1.4 Should – this action, activity, or procedural step is suggested but not required

3.2 Total nitrogen (TN) consists of inorganic (ammonia, nitrite, nitrate, excluding N_2) and organic forms of nitrogen. Organic nitrogen may be comprised of biological, synthetic, or natural nitrogen-containing compounds.

3.3 Total phosphorus (TP) consists of inorganic (orthophosphates, polyphosphates) and organically-bound phosphorous that may consist of biological, synthetic, or natural phosphorus-containing compounds.

3.4 Calibration standards are made to encompass the applicable range of the analytical method. They must contain both method analytes and be acidified prior to digestion and analysis.

3.5 Continuing calibration verifications (CCV) consist of calibration points periodically reanalyzed within the test cycle to verify that the initial calibration is applicable throughout the sample run. Calibration verifications are required after every tenth sample and at the end of the analytical sequence.

3.6 The independent calibration verification (ICV) is a solution containing a known concentration of each method analyte derived from externally prepared test materials and is used to verify the quality of the calibration curve. Standards are purchased with certifications of purity, concentration, and authenticity sourced from certified NIST-traceable references. Two separate ICV solutions, one for each target analyte, are mixed and acidified to produce a working standard (Section 7.2.6).

3.7 A laboratory fortified blank (LFB) consists of an aliquot of reagent water to which a known quantity of method analyte has been added in the laboratory. The LFB is processed and analyzed exactly as a sample. Its purpose is to determine if the method is within accepted control limits. In this method, a nitrate LFB is processed to confirm that there is no analytical bias between nitrate-sourced and ammonia-sourced digestates (Section 7.2.8).

3.8 Method blanks (MB) consist of volumes of reagent water that are carried through the entire preparation and analysis exactly as a sample, including exposure to all glassware, equipment, and reagents. They are used to document and correct for any systematic interference from reagents, containers, or the laboratory environment.

3.9 A sample duplicate (DUP) consists of a second aliquot of a sample, identical to the first and processed through the entire method. Sample duplicates are used to assess process reproducibility and method precision (Section 9.4.5).

3.10 A matrix spike (MS) consists of an aliquot of sample to which a known quantity of each method analyte is added in the laboratory. It is then processed through the entire method. The purpose of the MS is to determine whether the sample matrix contributes bias to the analytical results. The bias, or accuracy, is expressed as percent recovery (%R) and is calculated using Equation 4 (Section 9.4.6).

3.11 A matrix spike duplicate (MSD) is prepared using a second aliquot of the same sample used for the MS. The relative percent difference (RPD) between the MS and MSD is used to assess analytical precision and is calculated as shown in Equation 3 (Section 9.4.5).

3.12 A digestion check consists of an aliquot of reagent water or sample to which a known quantity of organic method analyte has been added in the laboratory. Digestion checks may be used in this method and are useful in validating digestion of organic nitrogen and phosphorus in complex matrices (Section 9.4.8). Digestion check solutions can be added to reagent water (LFB) or samples (MS).

4.0 Interferences

4.1 Persulfate digestion for nitrogen species may be less effective in water samples with high organic or nitrogenous compound loading (>20 mg N/L). For these matrices it is recommended that the oxidative capacity of the digestion be confirmed by completing digestion of a spiked sample according to Sections 9.4.6 and 9.4.8.

4.2 Over-acidification of samples at collection sites can result in low recoveries of inorganic and organic nitrogen levels (Section 16.3). This can be avoided by judicial addition of dilute acid and use of narrow-range pH indicator strips at the time of collection. Other methods employed in the field include sample refrigeration, addition of pre-measured acid aliquots in collection vessels, and methods with similar effect.

4.3 Chloride concentrations greater than 10,000 mg/L react with persulfate to form oxychlorides or chlorine that might deplete persulfate required to oxidize inorganic and organic nitrogen species to nitrate. Results will be biased low as a consequence (Section 16.3); however, these levels of chloride have not been encountered in PP-BTWW matrices.

4.4 Phosphorus concentrations may be underestimated under high suspended sediment concentrations (>150 mg/L). The method is only suitable for very turbid samples when turbidity is due to organic matter (algal cells, plant detritus) (Section 16.4).

4.5 Ionic forms of calcium, iron, barium, lead, and silver in the presence of sufficiently high concentrations of phosphorus can interfere by forming phosphate precipitates that may lead to under-reporting of TP (Section 16.3).

4.6 Arsenate [AsO_4^{3-}] forms a color similar to that of phosphate above 1.0 mg As/L and may produce a positive interference. Arsenite [AsO_3^{2-}] at levels >2.0 mg As/L may be oxidized to arsenate (Section 16.3).

4.7 Method interferences may be caused by contaminants in reagents, glassware, and other sample processing equipment. All process materials must be routinely demonstrated to be free from interferences under the conditions of the analyses by running method blanks as described in Section 3.8.

5.0 Safety

5.1 Potassium persulfate must be kept separate from flammable and easily oxidizable substances such as combustibles and many solvents. It is harmful when swallowed and may be harmful if inhaled or absorbed through the skin. It is irritating to mucous membranes, skin, and eyes. Fume hood, eye protection, chemical-resistant gloves, and protective clothing are strongly recommended when using this compound.

5.2 This method does not address all safety issues associated with its use. The laboratory is responsible for maintaining a safe work environment and a current awareness of OSHA regulations regarding safe handling of chemicals used in this method. Material safety data sheets (MSDS) should be available to all personnel involved in these analyses. Prudent and vigilant practices for handling chemicals must be employed at all times.

6.0 Equipment and Supplies

6.1 Autoclave or pressure cooker

6.1.1 Operating and maintenance instructions for high-pressure equipment and vessels should be read and understood before use. Inspect seals, vents, and tubes before use.

6.1.2 Autoclaves and pressure cookers must be capable of developing a steady, relative positive pressure of 15 psi. If using a pressure cooker, models with built-in pressure gauges are recommended. Ensure that the hot plate used with the pressure cooker has adequate heating surface area.

6.2 Heat-resistant and pressure-resistant test tube racks

6.3 Glassware

6.3.1 Class-A volumetric flasks, precision pipettes and micro-volume syringes as required

6.3.2 Borosilicate screw-top glass culture tubes, 16 x125 mm

6.3.3 Polyphenolic tube caps with cemented-in PTFE-faced 14B rubber liners

6.3.4 Glassware should be washed with hot tap water followed by copious rinsing with de-ionized water, then undergo a thorough 5% or 10% (v/v) HCl soak before a final reagent water rinse. Store glassware inverted after air or oven drying. Commercial detergents must never be used for cleaning glassware utilized in this method.

6.4 Top-loading balance capable of weighing to 10 ±0.1 mg

6.5 Analytical equipment and supplies

6.5.1 Instrumentation, equipment, and supplies for determination of nitrite/nitrate and orthophosphate are found within the approved methods for wastewater analysis (Section 16.5). Due to sample volume constraints, manual techniques are not recommended; however, they are permitted.

6.5.1.1 Approved methods for nitrite/nitrate analyses consist of EPA 353.2 (Section 16.6), ASTM D3867-90 (A and B) (Section 16.7), Standard Method 4500-NO_3^- (E and F) (Section 16.8), and USGS I-4545-85 (Section 16.9).

6.5.1.2 Approved methods for orthophosphorus detection include EPA 365.1 (Section 16.10), EPA 365.3 (Section 16.11), Standard Methods 4500-P (E and F) (Section 16.8), and USGS I-4601-85 (Section 16.9).

7.0 Reagents and Standards

7.1 Reagents

7.1.1 Ultrapure reagent water (UPW) such as that produced by commercial purification systems or deionized water (DIW) conforming to Type II reagent water (Section 16.12) and free of the analytes of interest.

7.1.2 1.84 M sulfuric acid solution (10% v/v) – Slowly add 10 mL of concentrated sulfuric acid (H_2SO_4, CAS 7664-93-9) to approximately 50 mL of cold reagent water. Cool and dilute to 100 mL.

7.1.3 Persulfate digestion solution – Dissolve 5.0 g of reagent grade potassium persulfate ($K_2S_2O_8$, CAS 7727-21-1, FW 270.32) and 1.2 g of reagent grade sodium hydroxide (NaOH, CAS 1310-73-2, FW 40.00) in approximately 200 mL of reagent water. Once fully dissolved, transfer to a 250 mL volumetric flask with a reagent water rinse and bring to volume. Reagents with known low nitrogen assays must be used.

7.1.4 Refer to the determinative methods (Section 6.5) for reagent lists and preparation instructions used in analyzing nitrite-nitrate and orthophosphate.

7.2 Standards

7.2.1 Ammonium primary stock, 1000 mg N/L – Add 3.818 g of reagent grade anhydrous ammonium chloride (NH_4Cl, CAS 12125-02-9, FW 53.49) to about 500 mL of reagent water. Quantitatively transfer to a 1 L volumetric flask and dilute to the mark.

7.2.2 Phosphorus primary stock, 500 mg P/L – Add 2.197 g of reagent grade anhydrous potassium dihydrogen orthophosphate (KH_2PO_4, CAS 7778-77-0, FW 136.09) to

about 500 mL of reagent water. Quantitatively transfer to a 1 L volumetric flask and dilute to the mark.

7.2.3 Ammonium working stock, 100 mg N/L – Dilute a portion of the ammonium primary stock (Section 7.2.1) by a factor of 10 using reagent water. Use this solution for spiking samples or quality control solutions at discrete analyte levels.

7.2.4 Phosphorus working stock, 100 mg P/L – Dilute a portion of the phosphorus primary stock (Section 7.2.2) by a factor of 5 using reagent water. Use this solution for spiking samples or quality control solutions at discrete analyte levels.

7.2.5 Combined nitrogen/phosphorus working stock solution – Add equal amounts of ammonium primary stock (Section 7.2.1) and phosphorus primary stock (Section 7.2.2) to an acid-washed beaker. One milliliter (1 mL) of solution is equal to 0.50 mg N + 0.25 mg P. Prepare at least 20 mL of combined solution to reduce volumetric errors. Use this solution for making up the calibration curve (Section 7.3).

7.2.6 ICV combined nitrogen/phosphorus stock solution – Combine purchased ammonia and phosphate reference standards in appropriate amounts to achieve a final concentration of 1.00 mg N/L + 1.00 mg P/L in reagent water. Acidify the solution by incorporating 1.0 mL of 10% sulfuric acid (Section 7.1.2) into every 100 mL of final volume.

7.2.7 Nitrate primary stock, 100 mg N/L – Dissolve 0.072 g of reagent grade potassium nitrate (KNO_3, CAS 7757-79-1, FW 101.1) in about 50 mL of reagent water. Quantitatively transfer to a 100 mL volumetric flask and dilute to the mark.

7.2.8 Nitrate LFB, 1.00 mg N/L – Dilute a portion of nitrate primary stock (Section 7.2.7) by a factor of 10 using reagent water. Acidify the solution by adding 1 mL of 10% sulfuric acid (Section 7.1.2) into every 100 mL of final volume.

7.2.9 Digestion check stock solution – Prepare digestion check solutions and add to reagent water or samples in the TN and TP ranges expected for BT-PPWW. Several organic compounds are suitable for preparing digestion check solutions, including urea (CAS 57-13-6), nicotinic acid, (CAS 59-67-6), sodium phenyl phosphate (CAS 66778-08-03), and others (Section 16.2).

7.3 Calibration standards

Use Table 1 (Section 17.0) as a guide to prepare a dual calibration curve. Spike reagent water with the working combined nitrogen/phosphorus working stock (Section 7.2.5), then acidify with 10% sulfuric acid (Section 7.1.2). The calibration set must include a calibration blank.

8.0 Sample Collection, Preservation, and Storage

8.1 Collection

Samples should be collected in glass or HDPE containers that have been acid washed and thoroughly rinsed (Section 6.3.4).

8.2 Preservation

Samples must be preserved at the time of collection by adding dilute sulfuric acid to attain $1.6 < pH < 2$. Ensure that sample pH is within the correct range using narrow-range pH color strips or a pH meter to avoid over-acidification (Section 4.2).

8.2 Storage

Refrigerate samples at $4 \pm 2^{o}C$ until digested. Maximum holding time is 28 days after collection and preservation. Digested samples may be stored at room temperature for up to 28 days (Section 17.0, Table 5). If either holding time is exceeded a new sample must be collected and digested.

9.0 Quality Control

9.1 Each laboratory that uses this method should operate a formal quality assurance program and maintain records that define the quality of the data that are generated. Data should be compared to established criteria in order to determine if the results of analyses meet the performance characteristics of the method. The requirements of this program consist of an initial demonstration of laboratory capability (Section 9.3) and routine analysis of quality assurance/quality control (QA/QC) solutions as a continuing check on method performance (Section 9.4).

9.2 Analytical performance

9.2.1 Prior to analyzing any samples or standards, equipment performance must be verified by means appropriate to the determinative method (Section 6.5). All QA/QC specifications listed in the associated determinative method must be met.

9.2.2 Calibrations must demonstrate linearity within 10%, as defined by the percent relative standard deviation (%RSD) of the response factors (Equation 2). Otherwise, a coefficient of determination (r^2) equal to or better than 0.995 is required. If the calibration fails, instrument solutions and/or digestion systems should be examined prior to preparation of new calibration and digestion solutions.

9.3 Initial demonstration of method proficiency

9.3.1 Initial precision and recovery (IPR)

9.3.1.1 The laboratory shall make an initial demonstration of its ability to generate acceptable results with this method. Conduct an IPR assessment by digesting and analyzing four aliquots of the combined nitrogen and phosphorus working stock solution (Section 7.2.5) in the range of expected BT-PPWW process effluents.

9.3.1.2 Using the results of the IPR analyses, compute average %R (Equation 1) and %RSD (Equation 2) of the recoveries for each analyte. Compare the %R and %RSD to the corresponding ranges determined during the multi-laboratory validation of the method (Section 17.0, Table 2). Laboratories should be able to achieve IPR values within the ranges stated prior to performing this method.

Equation 1

$$\%\ R = \frac{100 \times \text{Concentration Measured}}{\text{Concentration Spiked}}$$

Equation 2

$$\%\ RSD = \frac{100 \times \text{Standard Deviation of Measured}}{\text{Average of Measured}}$$

9.3.2 Matrix spike precision and recovery

Each laboratory should establish quality control charts and recovery criteria for specific matrices to prove method performance. This is accomplished by documenting %R (Equation 1) of MS samples and %RPD (Equation 3) for DUP and MS/MSD pairs. These results should be compared to the %R and %RPD values determined during the multi-laboratory study (Section 17.0, Table 3). If laboratory results for both TP and TN meet the criteria, method performance is verified.

9.3.3 Method detection limit (MDL)

To establish the capability to detect the analytes in this method, the laboratory should determine the MDL in accordance with the procedure in 40 CFR Part 136, Appendix B (Section 16.5) using the apparatus, reagents, and standards used in practice of this method. Prior to performing this method laboratories should be able to achieve MDLs equal to or less than 0.05 mg N/L for TN and 0.01 mg P/L for TP.

9.4 Method performance

9.4.1 Frequency

The laboratory must prepare DUP and MS sets from a minimum of 10% of routine samples. In addition, representative samples from each new or untested source or sample matrix should be allocated for DUP and MS quality control samples.

9.4.2 Method blank (MB)

One reagent blank should be prepared for every sample batch or for every 20 samples, whichever is more frequent. If any of the compounds of interest or any potentially interfering compounds are found in a blank at greater than the MDL determined in Section 9.3.3, the analysis should be halted until the source of contamination is eliminated. Blanks should be analyzed following initial calibration and prior to any sample analyses. Results should be kept on file with sample analysis data.

9.4.3 Continuing calibration verification (CCV)

CCV solutions must be run before sample analyses, after every tenth sample, and at the end of the sample sequence. The measured concentration for each CCV point must be within ±10% of the expected concentration. If the calibration cannot be verified within the specified limits, reanalyze the CCV. If the second analysis is still outside the limits, discontinue analyses and correct the source of the problem before resuming. All samples following the last acceptable CCV solution must be reanalyzed. Recovery data for all CCV measurements must be kept on file with sample analysis data.

9.4.4 Independent calibration verification (ICV)

One ICV standard solution consisting of mid-range concentrations of N and P (Section 7.2.6) should be digested with each digestion batch. Recovery must be within ±10% of expected concentrations. If it is not, reanalyze the solution. If the second analysis is still outside the limits, discontinue analyses and correct the source of the problem before resuming. Analyze an ICV at least once during sample analysis following calibration. Recovery data for all ICV measurements should be kept on file with sample analysis data.

9.4.5 Sample duplicate (DUP)

One DUP must be digested with every batch or one for every ten samples from the same sample group or matrix, whichever is more frequent. Precision is documented by %RPD of duplicate measurements as calculated using Equation 3.

Equation 3

$$\%\ RPD = \frac{100 \times (S1 - S2)}{\text{Mean concentration}}$$

where:
S1 = Sample 1 concentration
S2 = Sample 2 concentration

9.4.6 Matrix spike (MS)

One MS sample must be prepared and digested for every ten samples from the same sample matrix or one per digestion batch, whichever is more frequent. Using the estimated background concentration, use the appropriate stock solutions (Sections 7.2.3 and 7.2.4) to spike the sample at a level that will give one to five times the background concentration. Digest the MS along with an aliquot of unspiked sample. Calculate %R for the MS using Equation 4. If an MSD was also analyzed calculate the %RPD of the recoveries according to Equation 3. Limits for MS and MSD %R and %RPD should be within those determined during the multi-laboratory study (Section 17.0, Table 3) until sufficient internal performance data and control limits can be established.

Equation 4

$$\%\ R = \frac{100 \times (Cs - Cn)}{S}$$

where:
R = MS or MSD sample recovery
C_s = MS or MSD sample concentration
C_n = native sample concentration
S = concentration equivalent of analyte added to sample

9.4.7 Laboratory fortified blank (LFB)

A nitrate LFB must be digested and analyzed at least once during TN sample analysis. Calculate %R using Equation 1. Recovery must be within ±10% of the expected value. If it is not, reanalyze; if the measurement is still outside the limit, discontinue analyses and determine the source of error. Correct before continuing.

9.4.8 Digestion checks

Digestion checks are not required in the normal operation of this method. However, if they are employed, performance criteria are assessed against the required recovery of ±10% until sufficient internal performance data and control limits can be established. If %R of the checks falls outside established warning limits, method performance is unacceptable for the compound in the matrix.

Limits for digestion check solutions using 2-phenyl glycine (organic nitrogen) and calcium glycerylphosphate (organic phosphorus) were determined during single- and multi-laboratory validations of this method (Section 17.0, Tables 4 and 5).

10.0 Calibration and Standardization

10.1 Analytical equipment operating conditions

Analyze sample digestates using EPA-approved determinative methods for nitrite/nitrate and orthophosphate (Section 6.5). If an auto-analyzer is used follow the manufacturer's instructions to establish operating conditions. Once the system is optimized the same operating conditions must be used to analyze all samples. Instrument response checks (e.g. carryover) may be performed using unacidified, undigested nitrate and orthophosphate standards prior to analysis of digested samples.

10.2 Prepare a digested calibration curve by plotting instrument response against concentration values (Section 9.2.2). The calibration curve may be fitted to calibration solutions concentration/response data using automated operating software or calculator-based regression curve fitting techniques.

11.0 Procedure

11.1 Carry out the digestion in a pressure cooker or autoclave at 121^{o}C and 15 psi positive pressure. All solutions used in a single determinative analysis must be digested in the same batch. The instructions herein are written for digestion using a pressure cooker. If using an autoclave, follow manufacturer's operating instructions to affect the same operating conditions.

11.1.1 Remove samples and standards from refrigerated storage and allow them to come to room temperature. Gather and label a sufficient number of culture tubes and caps.

11.1.2 Add enough deionized water to the pressure cooker to provide a water level roughly equal to a 10 mL volume inside the digestion tubes. Place the uncovered pressure cooker on a hot plate and set the temperature control to low.

11.1.3 Make up acidified calibration solutions (Section 17.0, Table 1) and all required acidified quality control solutions according to Sections 7.2 and 9.4. Select and prepare DUP, MS, and MSD quality control samples as directed in Sections 9.4.5 and 9.4.6. Ensure that calibration solution quantities are adequate for the number of CCV measurements that must be analyzed for the batch size. Because sample size is limited to 10 mL, additional MB and CCV quantities may be prepared as contingent samples in case reanalysis becomes necessary.

11.1.4 Transfer 7.00 ±0.05 mL of all samples into assigned culture tubes. Shake the sample containers vigorously before transferring to ensure homogeneity.

11.1.5 Dispense 3.00 ±0.05 mL of potassium persulfate digestion solution (Section 7.1.3) into each sample tube. Cap and mix gently. Repeat for each tube.

11.1.6 Load the tubes into a heat-proof rack and securely place the rack in the pressure cooker. Ensure that the water level is approximately the same as the sample level inside the tubes. Secure the cover and set the hot plate temperature control to high. After heating for several minutes a steady stream of steam will begin to escape from the top vent. Allow the steam to escape for an additional 5 minutes to affect a complete seal.

11.1.7 Place the regulator on the vent pipe and monitor pressure. After several minutes the pressure will begin to rise, ultimately reaching 15 psi. Once the pressure is stable begin timing the 30 minute digestion. Pressure may slowly increase a few psi during this time without adverse effects.

11.1.8 Turn off the heat after 30 minutes has elapsed. Do not remove the pressure cooker from the hot plate nor remove the regulator or cover until the pressure cooker has returned to atmospheric pressure and is sufficiently cooled to allow safe handling, after which time the racks may be removed and caps may be tightened.

11.1.8.1 The digestion reaction effectively progresses during the pre-heating and cooling phases. Autoclave settings may or may not automatically include these intervals. Thus the 30 minute digestion timing should begin only after the temperature and pressure parameters are reached. Other means of applying thermal energy to the digestion have also been described (Section 16.4).

12.0 Data Analysis and Calculations

12.1 Sample analysis

Analyze the samples using the appropriate determinative method (Section 6.5) and specific instructions for assembly, analytical technique, reagent preparation, step succession, and manifold proportionalities provided by equipment manufacturers.

12.1.1 If suspended particles remain in the digestates they must be removed by sedimentation and decantation or filtration prior to TN analysis to avoid possible clogging of the cadmium column.

12.1.2 Calculate sample concentrations based on the calibration curve. When a calibration blank is used as a zero-point, the value of the MB must not be subtracted from experimental results.

12.1.2 Calculate %R for ICV, CCV, and LFB quality control samples (Equation 1). MS and MSD %R must be corrected for native concentrations as measured in an unfortified sample (Equation 4).

12.1.3 Calculate %RPD (Equation 3) for DUPs and for MS/MSD pairs.

12.2 Data review requirements

12.2.1 Ensure that all sample concentrations are within the calibration range of the instrument. Samples exceeding the highest standard should be quantitatively diluted to fall within the range of the calibration curve and reanalyzed.

12.2.2 Manually review all results to confirm that %R and %RPD of spiked samples are within established control boundaries. If MS or MSD recoveries fall outside the expected recovery range (Section 9.3.2) but laboratory performance for that analyte is shown to be in control, the recovery problem encountered with the spike is judged to be either matrix- or solution-related, not system-related.

12.2.3 Sample concentrations determined for each analyte, range of concentrations, and %RPD determined by DUPs should be tabulated in a method precision log. Record MS %R along with %RPD of all MS/MSD pairs.

12.2.4 Generate a report that includes calculated concentrations of TN in mg N/L and TP in mg P/L and for all samples. Report MB levels and recovery values for CCV, ICV, and LFB samples.

13.0 Method Performance

13.1 Verify that MB levels are below the MDL specified in Section 9.3.3 to confirm that laboratory processes are in control.

13.2 If matrix interference and bias are significant, samples should be assessed for interfering compounds (Section 4) and re-evaluated accordingly.

14.0 Pollution Prevention

No federal or state requirements are known to exist for hazard containment or remediation of laboratory quantities of reagents and solutions used in this method.

15.0 Waste Management

15.1 It is the laboratory's responsibility to comply with all federal, state, and local regulations governing waste management, and to protect the air, water, and land by minimizing releases into the environment. Compliance with all sewage discharge permits and regulations is also required.

15.2 Instructions for sample and waste handling and disposal

15.2.1 All samples and solutions, including process waste and excess reagent solutions, must be neutralized and disposed of in accordance with applicable federal, state, and local regulations.

15.2.2 For further information on waste management, EPA recommends consulting *The waste management manual for laboratory personnel* and *Less is better: Laboratory chemical management for waste reduction*. Both are available from the American Chemical Society's Department of Government Relations and Science Policy, 1155 16th Street NW, Washington DC, 20036.

16.0 References

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17.0 Tables, Diagrams, Flowcharts, and Validation Data

Table 1. Calibration Standard Solution Makeup Scheme

Calibration Point	Standard Volume[a] (µL)	Acid Volume[b] (mL)	Final Volume (mL)	Nominal Concentrations TN (mg/L)	TP (mg/L)
Cal 0	0	2.5	250	0.00	0.00
Cal 1	20	5.0	500	0.02	0.01
Cal 2	50	2.5	250	0.10	0.05
Cal 3	250	2.5	250	0.50	0.25
Cal 4	1,500	2.5	250	3.00	1.50
Cal 5	3,000	2.5	250	6.00	3.00

[a] standard contains 0.50 mg N/L and 0.25 mg P/L as described in Section 7.2.5
[b] 10% sulfuric acid; constitutes 1% of final volume to achieve required pH

Table 2. Initial Precision and Recovery[a]

Study	n	Recovery and Precision (%)		
		Average[b]	Range[c]	SD[d]
		Total Nitrogen		
Single laboratory	4	98.9	97.9 - 100	0.2
Multi-laboratory	12	98.8	94.4 - 105	4.0
		Total Phosphorus		
Single laboratory	4	93.0	91.1 - 94.9	1.6
Multi-laboratory	12	97.1	93.0 – 99.8	1.9

[a] determined using combined standards 5.00 mg N/L ammonium chloride (NH_4Cl) for TN and 1.00 mg P/L potassium dihydrogen ortho-phosphate (KH_2PO_4) for TP in reagent water

[b] average of IPR recoveries from four replicates in reagent water analyzed during single laboratory validation of NCASI Method TNTP-W10900 July 23, 2008, at NCASI West Coast Regional Laboratory, Corvallis, Oregon; multi-laboratory average from pooled IPR results obtained during multi-laboratory validation of NCASI Method TNTP-W10900 July 2009.

[c] range taken from lowest and highest individual IPR recoveries from laboratory validations

[d] precision expressed as standard deviation of pooled percent recoveries

Table 3. Matrix Spike and Matrix Spike Duplicate Recovery and Precision[a]

Study	n	Recovery and Precision (%)		
		Average[b]	Rangec[c]	RPD[d]
		Total Nitrogen		
Single laboratory	2	98.3	92.6 - 104	11.6
Multi-laboratory	6	90.8	84.4 - 96.5	0.2 – 4.4
		Total Phosphorus		
Single laboratory	2	98.8	97.5 - 100	2.5
Multi-laboratory	6	101	97.0 - 105	0.0 – 4.2

[a] determined using combined standards 5.00 mg N/L ammonium chloride (NH_4Cl) for TN and 1.00 mg P/L potassium dihydrogen ortho-phosphate (KH_2PO_4) for TP in PP-BTWW

[b] average of recoveries from MS/MSD pairs in PP-BTWW analyzed during single laboratory validation of NCASI Method TNTP-W10900 July 23, 2008, at NCASI West Coast Regional Laboratory, Corvallis, Oregon; multi-laboratory average from pooled MS/MSD results obtained during multi-laboratory validation of NCASI Method TNTP-W10900 July, 2009

[c] range taken from lowest and highest individual MS/MSD pair results from laboratory validations

[d] precision expressed as percent relative standard deviation of percent recoveries; RPD is absolute value of difference between MS and MSD expressed as a percent: $\%RPD = 100 \times [|MS-MSD| / \frac{1}{2} \times (MS+MSD)]$

Table 4. Organic Analyte Digestion Check Initial Precision and Recovery[a]

Study[b]	n	Recovery and Precision (%)		
		Average[c]	Range[d]	SD[e]
		Total Nitrogen		
Single laboratory A	4	93.3	92.2 - 94.7	1.2
Single laboratory B	4	99.8	98 - 101	2
		Total Phosphorus		
Single laboratory A	4	85.2	84.7 - 85.6	0.7
Single laboratory B	4	91	90 - 92	1

[a] determined using combined standards 1.00 mg N/L 2-phenyl glycine ($C_8H_9NO_2$) for TN and 1.00 mg P/L calcium glycerophosphate ($C_3H_7O_6P{\cdot}Ca$) for TP combined standards in reagent grade water

[b] data from two single laboratory validations of NCASI Method TNTP-W10900: Lab A NCASI West Coast Regional Laboratory, Corvallis, Oregon, July 23, 2008; Lab B Columbia Analytical Services, Kelso, Washington, July 24, 2009

[c] average of IPR recoveries from four replicates in reagent water analyzed during single laboratory validations

[d] range of lowest and highest IPR replicate recoveries

[e] precision expressed as standard deviation of percent recoveries

Table 5. Digested Sample Holding Time Study[a]

Holding Time[b]	n	Recovery and Precision (%)		
		Average[c]	Range[d]	RPD[e]
		Total Nitrogen		
4 days	2	98.4	92.8 - 104	5.7
28 days	2	104	103 - 105	1.9

[a] determined using combined standards 5.00 mg N/L ammonium chloride (NH_4Cl) for TN and 1.00 mg P/L potassium dihydrogen ortho-phosphate (KH_2PO_4) for TP in PP-BTWW; TP determination not performed

[b] holding time is number of days elapsed since digestion

[c] average of recoveries from MS/MSD pair in PP-BTWW on September 22, 2008 and October 16, 2008 at NCASI West Coast Regional Laboratory, Corvallis, Oregon

[d] range of lowest and highest MS/MSD recoveries

[e] relative percent difference; RPD is absolute value of difference between MS and MSD expressed as a percent: %RPD = 100 x [|MS-MSD| / ½ x (MS+MSD)]