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Total Nitrogen Analysis by Low Alkaline Persulfate Digestion

Use with NECi's Nitrate Reductase Products, either: NTK-TTSR-25/100 or a Discrete Analyzer Reagent Pack

Introduction

This method follows a paper by Halstead et al. (1999), Potential for chlorate interference in ion chromatographic determination of total nitrogen in natural waters following alkaline persulfate digestion. This method is for nitrate only, and cannot be used for phosphate determination. It is intended to be used in conjunction with **NECi Standard Range Nitrate Test Kit** to provide a means of assessing the total amount of nitrogen in a water sample. Total nitrogen=organic nitrogen + ammonium + nitrate + nitrite. Alternatively, Nitrate + Nitrite can be determined by the NECi Discrete Analyzer Nitrate Analysis Method (to use this method, order the **NECi Discrete Analyzer Reagent Pack** for your instrument).

Required Chemicals

- Potassium nitrate
 - Sigma P6083
- Sodium Persulfate
 - Sigma S6172
- Sodium Hydroxide
 - Fisher S318
- HPLC Water
 - Spectrum HP902

Required Labware

- Reactor/Cuvette Tubes.
 - Hach 25831-25
- Various Volumetric Flasks, Pyrex.
- Autoclave
- Ultrasonic Bath
- NECi Standard Range Nitrate Test Kit.
 - NECi NTK-TTSR-25/100
 - Alternatively, order the NECi Discrete Analyzer Reagent Pack for your instrument.

Stock Solutions

Preparation of Stock Solutions is explained in detail on the reverse of this sheet

1. 25 mL Alkaline Digesting Solution (0.22 M $\text{Na}_2\text{S}_2\text{O}_8$)
1.31 g $\text{Na}_2\text{S}_2\text{O}_8$ + 25 mL 0.5 M NaOH
2. 1000 ppm Potassium Nitrate Standard (Dilute 1:100 before use)
1.44 g KNO_3 + 200 mL HPLC H_2O

Sample composition

5.0 mL Standard or sample
1.0 mL Alkaline Digesting solution

Follow all local regulations with regard to disposal of all toxic waste generated from using this method.

Reagent Preparation:

- **Alkaline Digesting Solution:** Add 1.31 g of sodium persulfate ($\text{Na}_2\text{S}_2\text{O}_8$, MW: 238.03) to 20 mL of 0.5 M NaOH. Place into ultrasonic bath to dissolve the $\text{Na}_2\text{S}_2\text{O}_8$ (~2 minutes). Bring volume to 25 mL with additional 0.5 M NaOH. *Prepare fresh solution before every set of assays.*
- **1000 ppm Stock Standard:** Prepare stock solutions by dissolving 1.44 g KNO_3 in 200 mL of HPLC water using a volumetric flask. *The stock solution is stable for 6 months when stored at 4°C.* To create a 10 ppm working solution, dilute stock solution 100x with HPLC water: add 0.1 mL of stock solution to 9.90 mL HPLC water. Further dilute working solution for the standard solutions, using volumes listed in the chart below:

Chart 1. Preparation of Nitrate Calibrants

$\text{NO}_3\text{-N}$ mg N/L (or ppm)	10 ppm working Standard (in mL)	HPLC water (in mL)
0.025	0.025	4.975
0.10	0.050	4.950
0.50	0.250	4.750
0.75	0.375	4.625
1.25	0.625	4.375
2.50	1.250	3.750
5.00	2.500	2.500

Procedure:

1. Mix 5 mL of sample and 1.0 mL alkaline digesting solution in autoclavable culture tubes with loose caps.
2. Autoclave the mixture at 250°F (121°C) and 17lb/in² (117.2 kPa) for one hour. Set the autoclave for slow release of pressure, to ensure that no samples rupture due to a rapid change in pressure.
3. Remove samples from the autoclave and allow samples to cool to room temperature. These samples can be stored at room temperature for several days.
4. Analyze samples for total nitrate concentration using one of these methods:
 - a. NECi Standard Range Nitrate Test Kit: follow the test kit instructions using the Calibrants shown in Chart 1.
 - b. Discrete Analyzer (DA) Nitrate Analysis Method: Use the NECi DA Reagent Pack for your instrument and apply standard QC/QA practices for Nitrate analysis.

Calculations:

1. Calculate the Total Nitrogen in the sample using a Standard Curve and the linear regression fit to the line. The values for the Total Nitrogen in the samples will be in units of mg N/L.
2. The Kjeldahl Method reports Total Nitrogen as mg N/L. Since the NECi Nitrate Test Kit is calibrated as mg N/L, the results of the analysis done here will be in the same units as the traditional Kjeldahl Method. Thus, there is no need to convert the results to any other form.

Alternate Procedure to Correct for Nitrate and Nitrite in the Samples

If the initial Nitrate + Nitrite, as mg N/L, is determined before the sample is digested, then the Total Nitrogen in the form of Organic Nitrogen and Ammonium can be calculated.

1. In a portion of the sample, analyze for Nitrate + Nitrite using either the NECi Nitrate Test Kit or the NECi Discrete Analyzer Analysis Method. This will yield the Total Nitrate+ Nitrite in mg N/L.
2. In the other portion of the sample, carry out the persulfate digestion as described above. Determine the Nitrate in the digested sample by either the NECi Nitrate Test Kit or the NECi Discrete Analyzer Nitrate Analysis Method. This will yield the Total Nitrogen (Organic Nitrogen + Ammonium + Nitrate + Nitrite) in mg N/L.
3. To calculate the NET Total Nitrogen as Organic Nitrogen + Ammonium in mg N/L, subtract the Total Nitrate + Nitrite in mg N/L in the sample from the Total Nitrogen + Ammonium + Nitrate + Nitrite) in mg N/L in the sample.

Please call NECi to learn about adapting our method or visit www.nitrate.com