4500-NO_3^- NITROGEN (NITRATE)*#(1)

4500-NO_3^- A. Introduction

1. Selection of Method

Determination of nitrate (NO_3^-) is difficult because of the relatively complex procedures required, the high probability that interfering constituents will be present, and the limited concentration ranges of the various techniques.

An ultraviolet (UV) technique (Method B) that measures the absorbance of NO_3^- at 220 nm is suitable for screening uncontaminated water (low in organic matter).

Screen a sample; if necessary, then select a method suitable for its concentration range and probable interferences. Nitrate may be determined by ion chromatography (Section 4110) or capillary ion electrophoresis (Section 4140). Applicable ranges for other methods are: nitrate electrode method (D), 0.14 to 1400 mg NO_3^--N/L; cadmium reduction method (E), 0.01 to 1.0 mg NO_3^--N/L; automated cadmium reduction methods (F and I), 0.001 to 10 mg NO_3^--N/L. For higher NO_3^--N concentrations, dilute into the range of the selected method.

Colorimetric methods (B, E) require an optically clear sample. Filter turbid sample through 0.45-µm-pore-diam membrane filter. Test filters for nitrate contamination.

2. Storage of Samples

Start NO_3^- determinations promptly after sampling. If storage is necessary, store for up to 2 d at 4°C; disinfected samples are stable much longer without acid preservation. For longer storage of unchlorinated samples, preserve with 2 mL conc H_2SO_4/L and store at 4°C. NOTE: When sample is preserved with acid, NO_3^- and NO_2^- cannot be determined as individual species.

4500-NO_3^- E. Cadmium Reduction Method

1. General Discussion

a. Principle: NO_3^- is reduced almost quantitatively to nitrite (NO_2^-) in the presence of cadmium (Cd). This method uses commercially available Cd granules treated with copper sulfate ($CuSO_4$) and packed in a glass column.

© Copyright 1999 by American Public Health Association, American Water Works Association, Water Environment Federation

The NO_2^- produced thus is determined by diazotizing with sulfanilamide and coupling with N-(1-naphthyl)-ethylenediamine dihydrochloride to form a highly colored azo dye that is measured colorimetrically. A correction may be made for any NO_2^- present in the sample by analyzing without the reduction step. The applicable range of this method is 0.01 to 1.0 mg NO_3^--N/L. The method is recommended especially for NO_3^- levels below 0.1 mg N/L where other methods lack adequate sensitivity.

b. Interferences: Suspended matter in the column will restrict sample flow. For turbid samples, see ¶ A.1. Concentrations of iron, copper, or other metals above several milligrams per liter lower reduction efficiency. Add EDTA to samples to eliminate this interference. Oil and grease will coat the Cd surface. Remove by pre-extraction with an organic solvent (see Section 5520). Residual chlorine can interfere by oxidizing the Cd column, reducing its efficiency. Check samples for residual chlorine (see DPD methods in Section 4500-Cl). Remove residual chlorine by adding sodium thiosulfate ($Na_2S_2O_3$) solution (Section 4500-NH_3.B.3*d*). Sample color that absorbs at about 540 nm interferes.

2. Apparatus

a. Reduction column: Purchase or construct the column*#(2) (Figure 4500-NO_3^-:1) from a 100-mL volumetric pipet by removing the top portion. The column also can be constructed from two pieces of tubing joined end to end: join a 10-cm length of 3-cm-ID tubing to a 25-cm length of 3.5-mm-ID tubing. Add a TFE stopcock with metering valve[1] to control flow rate.

b. Colorimetric equipment: One of the following is required:

1) *Spectrophotometer,* for use at 543 nm, providing a light path of 1 cm or longer.

2) *Filter photometer,* with light path of 1 cm or longer and equipped with a filter having maximum transmittance near 540 nm.

3. Reagents

a. Nitrate-free water: See ¶ B.3*a*. The absorbance of a reagent blank prepared with this water should not exceed 0.01. Use for all solutions and dilutions.

b. Copper-cadmium granules: Wash 25 g new or used 20- to 100-mesh Cd granules†#(3) with 6*N* HCl and rinse with water. Swirl Cd with 100 mL 2% $CuSO_4$ solution for 5 min or until blue color partially fades. Decant and repeat with fresh $CuSO_4$ until a brown colloidal precipitate begins to develop. Gently flush with water to remove all precipitated Cu.

c. Color reagent: Prepare as directed in Section 4500-NO_2^-B.3*b*.

d. Ammonium chloride-EDTA solution: Dissolve 13 g NH_4Cl and 1.7 g disodium ethylenediamine tetraacetate in 900 mL water. Adjust to pH 8.5 with conc NH_4OH and dilute to 1 L.

© Copyright 1999 by American Public Health Association, American Water Works Association, Water Environment Federation

e. Dilute ammonium chloride-EDTA solution: Dilute 300 mL NH_4Cl-EDTA solution to 500 mL with water.

f. Hydrochloric acid, HCl, 6*N*.

g. Copper sulfate solution, 2%: Dissolve 20 g $CuSO_4.5H_2O$ in 500 mL water and dilute to 1 L.

h. Stock nitrate solution: Prepare as directed in ¶ B.3*b*.

i. Intermediate nitrate solution: Prepare as directed in ¶ B.3*c*.

j. Stock nitrite solution: See Section 4500-NO_2^-.B.3*e*.

k. Intermediate nitrite solution: See Section 4500-NO_2^-.B.3 *f*.

l. Working nitrite solution: Dilute 50.0 mL intermediate nitrite solution to 500 mL with nitrite-free water; 1.00 mL = 5 µg NO_2^--N.

4. Procedure

a. Preparation of reduction column: Insert a glass wool plug into bottom of reduction column and fill with water. Add sufficient Cu-Cd granules to produce a column 18.5 cm long. Maintain water level above Cu-Cd granules to prevent entrapment of air. Wash column with 200 mL dilute NH_4Cl-EDTA solution. Activate column by passing through it, at 7 to 10 mL/min, at least 100 mL of a solution composed of 25% 1.0 mg NO_3^--N/L standard and 75% NH_4Cl-EDTA solution.

b. Treatment of sample:

1) Turbidity removal—For turbid samples, see ¶ A.1.

2) pH adjustment—Adjust pH to between 7 and 9, as necessary, using a pH meter and dilute HCl or NaOH. This insures a pH of 8.5 after adding NH_4Cl- EDTA solution.

3) Sample reduction—To 25.0 mL sample or a portion diluted to 25.0 mL, add 75 mL NH_4Cl-EDTA solution and mix. Pour mixed sample into column and collect at a rate of 7 to 10 mL/min. Discard first 25 mL. Collect the rest in original sample flask. There is no need to wash columns between samples, but if columns are not to be reused for several hours or longer, pour 50 mL dilute NH_4Cl-EDTA solution on to the top and let it pass through the system. Store Cu-Cd column in this solution and never let it dry.

4) Color development and measurement—As soon as possible, and not more than 15 min after reduction, add 2.0 mL color reagent to 50 mL sample and mix. Between 10 min and 2 h afterward, measure absorbance at 543 nm against a distilled water-reagent blank. NOTE: If NO_3^- concentration exceeds the standard curve range (about 1 mg N/L), use remainder of reduced sample to make an appropriate dilution and analyze again.

© Copyright 1999 by American Public Health Association, American Water Works Association, Water Environment Federation

c. Standards: Using the intermediate NO_3^-- N solution, prepare standards in the range 0.05 to 1.0 mg NO_3^--N/L by diluting the following volumes to 100 mL in volumetric flasks: 0.5, 1.0, 2.0, 5.0, and 10.0 mL. Carry out reduction of standards exactly as described for samples. Compare at least one NO_2^- standard to a reduced NO_3^- standard at the same concentration to verify reduction column efficiency. Reactivate Cu-Cd granules as described in ¶ 3*b* above when efficiency of reduction falls below about 75%.

5. Calculation

Obtain a standard curve by plotting absorbance of standards against NO_3^--N concentration. Compute sample concentrations directly from standard curve. Report as milligrams oxidized N per liter (the sum of NO_3^--N plus NO_2^--N) unless the concentration of NO_2^--N is separately determined and subtracted.

6. Precision and Bias

In a single laboratory using wastewater samples at concentrations of 0.04, 0.24, 0.55, and 1.04 mg NO_3^- + NO_2^--N/L, the standard deviations were ±0.005, ±0.004, ±0.005, and ±0.01, respectively. In a single laboratory using wastewater with additions of 0.24, 0.55, and 1.05 mg NO_3^- + NO_2^--N/L, the recoveries were 100%, 102%, and 100%, respectively.[2]

7. References

1. WOOD, E.D., F.A.J. ARMSTRONG & F.A. RICHARDS. 1967. Determination of nitrate in sea water by cadmium-copper reduction to nitrite. *J. Mar. Biol. Assoc. U.K.* 47:23.
2. U.S. ENVIRONMENTAL PROTECTION AGENCY. 1979. Methods for Chemical Analysis of Water and Wastes, Method 353.3. U.S. Environmental Protection Agency, Washington, D.C.

8. Bibliography

STRICKLAND, J.D.H. & T.R. PARSONS. 1972. A Practical Handbook of Sea Water Analysis, 2nd ed. Bull. No. 167, Fisheries Research Board Canada, Ottawa, Ont.

NYDAHL, F. 1976. On the optimum conditions for the reduction of nitrate by cadmium. *Talanta* 23:349.

AMERICAN SOCIETY FOR TESTING AND MATERIALS. 1987. Annual Book of ASTM Standards, Vol. 11.01. American Soc. Testing & Materials, Philadelphia, Pa.

© Copyright 1999 by American Public Health Association, American Water Works Association, Water Environment Federation

Endnotes

1 (Popup - Footnote)
* APPROVED BY STANDARD METHODS COMMITTEE, 1997.

2 (Popup - Footnote)
* Tudor Scientific Glass Co., 555 Edgefield Road, Belvedere, SC 29841, Cat. TP-1730, or equivalent.

3 (Popup - Footnote)
† EM Laboratories, Inc., 500 Exec. Blvd., Elmsford, NY, Cat. 2001, or equivalent.

© Copyright 1999 by American Public Health Association, American Water Works Association, Water Environment Federation