



## Method 10071

## Persulfate Digestion Method

### Test 'N Tube™ Vials

(0.0 to 25.0 mg/L N)

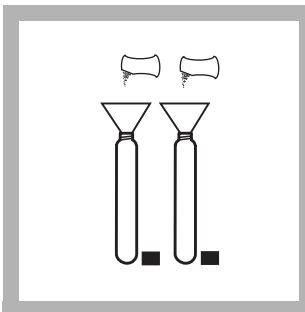
**Scope and Application:** For water and wastewater; digestion is required for determining total nitrogen.  
Digestion procedure included in method.



**1.** Turn on the COD Reactor. Heat to 103–106 °C (best temperature is 105 °C). Place the plastic shield in front of the reactor.

**Note:** Ensure safety devices are in place to protect the analyst from splattering due to leakage.

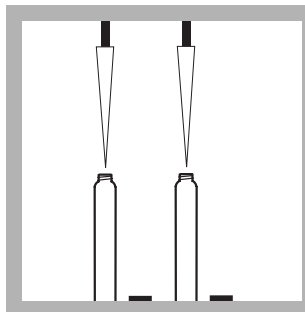
**Note:** For proof of accuracy, run a 20-mg/L  $\text{NH}_3\text{-N}$  standard through digestion and analysis.



**2.** Using a funnel, add the contents of one Total Nitrogen Persulfate Reagent Powder Pillow to each of two Total Nitrogen Hydroxide Reagent vials.

**Note:** Wipe reagent from the lid and the tube threads.

**Note:** One reagent blank is sufficient for each set of samples.



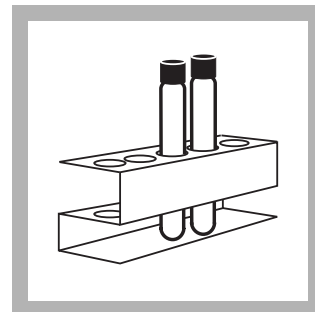
**3.** Add 2 mL of sample to a vial. This is the prepared sample.

Add 2 mL of the deionized water included in the kit to a second vial. This is the reagent blank.

Cap both vials, shake vigorously to mix (more than 30 seconds), and place the vials in the COD Reactor. Heat for 30 minutes.

**Note:** Use only water that is free of all nitrogen-containing species as a substitute for the deionized water provided.

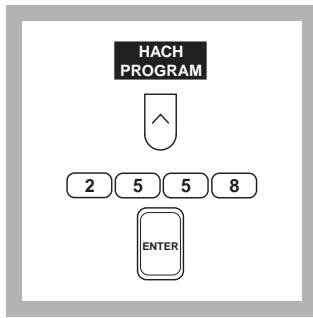
**Note:** The persulfate reagent may not dissolve completely after shaking. This will not affect accuracy.



**4.** Using finger cots or gloves, remove the hot vials from the reactor and allow to cool to room temperature.

**Note:** It is important to remove the vials from the COD Reactor after exactly 30 minutes.

# NITROGEN, Total, continued



5. Press the soft key under **HACH PROGRAM**.

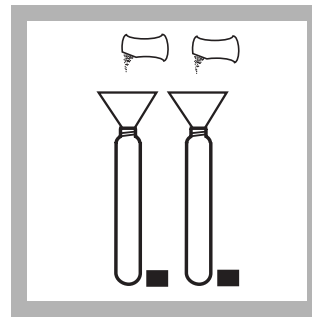
Select the stored program for Test 'N Tube Total Nitrogen by pressing **2558** with the numeric keys.

Press: **ENTER**



6. The display will show: **HACH PROGRAM:2558 N, Total, TNT**

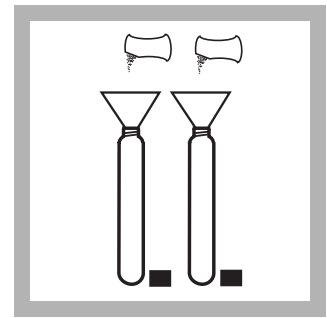
The wavelength ( $\lambda$ ), **410 nm**, is automatically selected.



7. Remove the caps from the digested vials and add the contents of one TN Reagent A Powder Pillow to each vial. Cap tubes and shake for 15 seconds.

Press the soft key under **START TIMER** after shaking.

A 3-minute reaction period will begin.

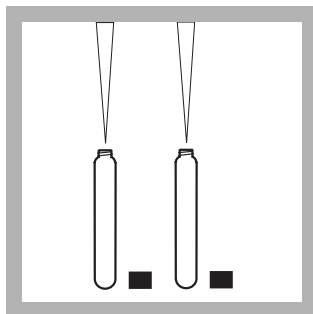


8. After the timer beeps, remove the caps from the vials and add one TN Reagent B Powder Pillow to each vial. Cap the tubes and shake for 15 seconds.

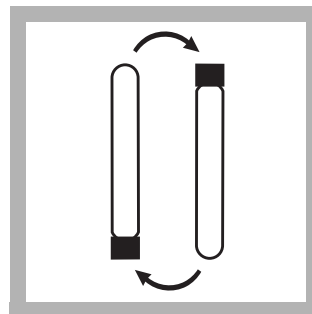
Press the soft key under **START TIMER** after shaking.

A 2-minute reaction period will begin.

**Note:** The reagent will not completely dissolve. The solution will begin to turn yellow.

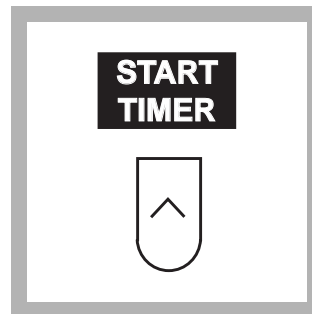


9. After the timer beeps, remove the caps from two TN Reagent C Vials and add 2 mL of digested, treated sample to one vial. Add 2 mL of the digested, treated reagent blank to the second TN Reagent C Vial.



10. Cap and invert 10 times to mix. Use slow, deliberate inversions for complete recovery. The tubes will be warm.

**Note:** Hold the tube vertical with the cap up. Invert the vial and wait for all of the solution to flow to the cap end. Return the vial to the upright position and wait for all of the solution to flow to the vial bottom. This is one inversion (10 inversions = 30 seconds).



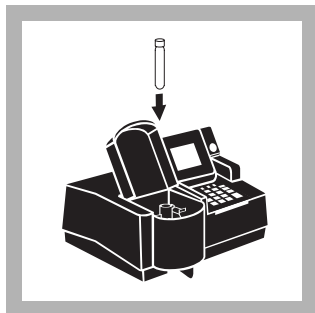
11. Press the soft key under **START TIMER**.

A 5-minute reaction period will begin.

**Note:** The yellow color will intensify.



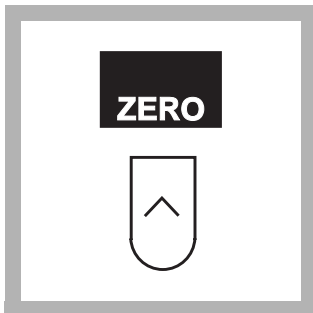
12. Insert the Test 'N Tube Adapter into the sample cell module by sliding it under the thumb screw and into the alignment grooves. Fasten with the thumb screw.



**13.** When the timer beeps, clean the outside of the TN Reagent C Vial containing the reagent blank. Place the vial in the adapter and close the light shield.

**Note:** Wiping with a damp towel followed by a dry one will remove fingerprints or other marks.

**Note:** The reagent blank is stable when stored in the dark; see *Blanks For Colorimetric Measurement* following these steps.



**14.** Press the soft key under **ZERO**.

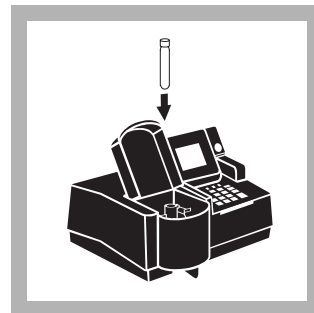
The display will show:

**0.0 mg/L N**

**Note:** For alternate concentration units, press the soft key under **OPTIONS**. Then press the soft key under **UNITS** to scroll through the available options. Press **ENTER** to return to the read screen.



**15.** Wipe the TN Reagent C Vial containing the sample.



**16.** Place the prepared sample into the cell holder and close the light shield. The result in mg/L total nitrogen will be displayed.

**Note:** Multiple samples may be read after zeroing on one reagent blank.

**Note:** Results may be expressed as N, NH<sub>3</sub> or NO<sub>3</sub><sup>-</sup>. Press the soft keys under **OPTIONS** and then **FORM**: to scroll through the available options. Press **ENTER** to return to the read screen.

**Note:** If the test overranges, repeat the digestion and measurement with diluted sample. The digestion must be repeated for accurate results.

## Interferences

The substances in the following table have been tested and found **not** to interfere up to the indicated levels (in mg/L):

Substance	Maximum Level Tested (mg/L)
Barium	2.6
Calcium	300
Chromium (3+)	0.5
Iron	2
Lead	6.6 ppb
Magnesium	500
Organic Carbon	150
pH	13 pH units
Phosphorus	100
Silica	150
Silver	0.9
Tin	1.5

# NITROGEN, Total, continued

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Interfering substances that resulted in a concentration change of  $\pm 10\%$ :

Substance	Level and Effect
Bromide	>60ppm; positive interference
Chloride	>1000 ppm; positive interference

Hach chemists tested this chemistry on standard nitrogen solutions prepared from the following compounds and obtained 95% recovery:

- Ammonium chloride
- Ammonium sulfate
- Ammonium acetate
- Urea
- Glycine

Ammonium chloride or nicotinic-PTSA spikes in domestic influent, effluent and the ASTM standard specification for substitute wastewater (D 5905-96) also resulted in  $\geq 95\%$  recovery.

The large amounts of nitrogen-free organic compounds in some samples may decrease digestion efficiency by consuming some of the persulfate present. Samples known to contain high levels of organics should be diluted and re-run to verify digestion efficiency.

## Sample Collection, Storage and Preservation

Collect samples in clean plastic or glass bottles. Best results are obtained with immediate analysis.

Preserve the sample by reducing the pH to 2 or less with concentrated sulfuric acid (at least 2 mL). Store at 4 °C (39 °F) or less. Preserved samples may be stored up to 28 days. Warm samples to room temperature and neutralize with 5 N sodium hydroxide before analysis. Correct the test result for volume additions; see Section 1.2.2 *Correcting for Volume Additions*.

## Method Performance

### Precision

Hach analysis of two independent nutrient standards shows the lowest average percent recovery at 95% with a standard deviation of  $\pm 2\%$ .

### Precision

Standard: 15 mg/L NH<sub>3</sub>-N

Program	95% Confidence Limits
2558	14.2–15.8 mg/L N

For more information on determining precision data and method detection limits, refer to Section 1.5.

**Estimated Detection Limit**

Program	EDL
2558	2 mg/L N

For more information on derivation and use of Hach’s estimated detection limit, see Section 1.5.2. To determine a method detection limit (MDL) as defined by 40 CFR part 136, appendix B, see Section 1.5.1.

**Sensitivity**

Program Number: 2558

Program	ΔAbs	ΔConcentration
Entire Range	0.010	0.52 mg/L N

See Section 1.5.3 *Sensitivity Explained* for more information.

**Accuracy Check**

This method generally yields 95–100% recovery on organic nitrogen standards. For proof of accuracy Hach offers a set of 3 Primary Standards for Kjeldahl Nitrogen.

1. Prepare one or more of the following 3 solutions. Each preparation is for an equivalent 25-mg/L N standard. Use the deionized water included in the kit or water that is free of all organic and nitrogen-containing species.
  - a. Weigh 0.3379 g of Ammonium p-Toluenesulfonate (PTSA). Dissolve in a 1000-mL volumetric flask with deionized water. Add deionized water to the 1000-mL mark.
  - b. Weigh 0.4416 g of Glycine p-Toluenesulfonate. Dissolve in a 1000-mL volumetric flask with deionized water. Add deionized water to the 1000-mL mark.
  - c. Weigh 0.5274 g of Nicotinic p-Toluenesulfonate. Dissolve in a 1000-mL volumetric flask with deionized water. Add deionized water to the 1000-mL mark.
2. Analyze each of these solutions using the test procedure above. Calculate the percent recovery for each using this formula:

$$\% \text{ recovery} = \frac{\text{measured concentration}}{25} \times 100$$

The percent recovery should be:

Compound	Lowest Expected % Recovery
Ammonia-PTSA	95%
Glycine-PTSA	95%
Nicotinic-PTSA	95%

Hach analysts have found Ammonia-PTSA to be the most difficult to digest.

Other compounds may yield different percent recoveries.

# NITROGEN, Total, continued

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## Standard Solution Method

For proof of accuracy, substitute 2 mL of a 20-mg/L ammonia nitrogen standard solution for the sample in the procedure\*. A single analyst should obtain less than 5% variation on replicates.

To adjust the calibration curve using the reading obtained with the 20-mg/L N standard solution, press the soft keys under **OPTIONS, (MORE)** then **STD: (OFF)**. Press **ENTER** to accept the default concentration. If an alternate concentration is used, enter the actual concentration and press **ENTER** to return to the “read” screen. See Section 1.5.5 *Adjusting the Standard Curve*, for more information.

## Standard Additions Method

- a. Fill three 25-mL graduated mixing cylinders with 25 mL of sample.
- b. Snap the neck off an Ammonia Nitrogen Voluette® Ampule Standard Solution, 160-mg/L as NH<sub>3</sub>-N
- c. Use the TenSette® Pipet to add 0.3, 0.6, and 0.9 mL of standard, respectively, to the three mixing cylinders.
- d. Stopper each cylinder and mix thoroughly.
- e. Add 2 mL of each prepared solution, respectively, to three Total Nitrogen Hydroxide Reagent Vials.
- f. Analyze each standard addition sample as described in the procedure. The nitrogen concentration should increase by approximately 1.9, 3.8, and 5.6 mg/L N, respectively.
- g. If these increases do not occur, see Section 1.4.1 *Standard Additions* for troubleshooting information.

## Blanks For Colorimetric Measurement

The reagent blank may be used up to seven days for measurements using the same lots of reagents. Store it in the dark at room temperature (18–25 °C). If a small amount of white floc appears prior to the end of one week, discard the reagent blank and prepare a new one.

## Calibration Standard Preparation

To perform a total nitrogen calibration using the Test ‘N Tube Persulfate Digestion method, prepare calibration standards containing 5.00, 10.00, 15.00, and 20 mg/L nitrogen (NH<sub>3</sub>-N) as follows:

- a. Into three different 100-mL Class A volumetric flasks, pipet 5.00, 10.00, 15.00, and 20 mL of a 100-mg/L Ammonia Nitrogen Standard Solution using Class A glassware.
- b. Using the Basic Persulfate Digestion method and the calibration procedure described in the *User-Entered Programs* section of the *DR/4000*

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\* To prepare a 20-mg/L ammonia nitrogen standard, use a 20-mL Class A pipet to transfer 20 mL of a 100-mg/L Ammonia Nitrogen Standard Solution (see *OPTIONAL REAGENTS AND STANDARDS*) to a 100-mL Class A volumetric flask. Dilute to the line with deionized water.

*Spectrophotometer Instrument Manual*, generate a calibration curve from the standards prepared above.

## Summary of Method

An alkaline persulfate digestion converts all forms of nitrogen to nitrate. Sodium metabisulfite is added after the digestion to eliminate halogen oxide interferences. Nitrate then reacts with chromotropic acid under strongly acidic conditions to form a yellow complex with an absorbance maximum at 410 nm.

## Safety

Good safety habits and laboratory techniques should be used throughout the procedure. Consult the *Material Safety Data Sheet* for information specific to the reagents used. For additional information, refer to Section 1.

## Pollution Prevention and Waste Management

For information on pollution prevention and waste management, refer to Section 1.

### REQUIRED REAGENTS AND STANDARDS

Test 'N Tube Total Nitrogen Reagent Set (50 sets of vials).....26722-45  
 Includes: (50) Total Nitrogen Hydroxide Reagent Vials\*, (50) TN Reagent C Vials\*, (1) 26718-46, (1) 26719-46, (1) 26720-46, (1) 272-42

Description	Quantity Required		Cat. No.
	Per Test	Unit	
Total Nitrogen Hydroxide Reagent Vials, 0.1 N .....	2 vials .....	50/pkg .....	*
Total Nitrogen Persulfate Reagent Powder Pillows .....	2 pillows .....	50/pkg .....	26718-46
TN Reagent A Powder Pillows .....	2 pillows .....	50/pkg .....	26719-46
TN Reagent B Powder Pillows.....	2 pillows .....	50/pkg .....	26720-49
TN Reagent C Vials.....	2 vials .....	50/pkg .....	*
Water, deionized .....	4 mL .....	100 mL.....	272-42f

### REQUIRED EQUIPMENT AND SUPPLIES

COD Reactor, 115/230 VAC, North American Plug.....	1 .....	each.....	45600-00
COD Reactor, 230 VAC, European Plug.....	1 .....	each.....	45600-02
DR/4000 Test Tube Adapter.....	1 .....	each.....	48189-00
Funnel, micro .....	1 .....	each.....	25843-35
Safety Shield, laboratory bench, 38 x 40 cm.....	1 .....	each.....	23810-00
Test Tube Cooling Rack .....	1-3 .....	each.....	18641-00

### OPTIONAL REAGENTS AND STANDARDS

Description	Unit	Cat. No.
Ammonia Nitrogen Standard Solution as N, 100-mg/L.....	500 mL.....	24065-49
Ammonia Nitrogen Standard Solution, 10-mg/L NH <sub>3</sub> -N.....	500 mL.....	153-49
Ammonia Nitrogen Standard Solution, 100-mg/L NH <sub>3</sub> -N.....	500 mL.....	24065-49
Ammonia Nitrogen Standard Solution, 10-mL Voluette Ampule, 160-mg/L NH <sub>3</sub> -N .....	16/pkg .....	21091-10
Primary Standard Set, for Kjeldahl Nitrogen.....	set of 3.....	22778-00
Sodium Hydroxide Standard Solution, 5.0 N.....	59 mL SCDB.....	2450-26
Sulfuric Acid, ACS, concentrated .....	500 mL.....	979-49
Water, organic-free .....	500 mL.....	26415-49

\* These items are not sold separately. Please reorder the complete set (Cat. No. 26722-45).

# NITROGEN, Total, continued

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## OPTIONAL EQUIPMENT AND SUPPLIES

Balance, analytical, 110 VAC.....	each.....	26103-00
Balance, analytical, 220 VAC.....	each.....	26103-02
Cylinder, mixing, graduated, 25-mL (3 required) .....	each.....	20886-40
Flask, volumetric, Class A, 100-mL .....	each.....	14574-42
Flask, volumetric, Class A, 1000-mL (3 required) .....	each.....	14574-53
pH Paper, pH 1.0 to 11.0 .....	5 rolls/pkg.....	391-33
Pipet, volumetric, Class A, 20-mL .....	each.....	14515-20
Pipet, TenSette, 1.0- to 10.0-mL .....	each.....	19700-10
Pipet Tips, for 19700-10 TenSette Pipet .....	50/pkg.....	21997-96
Pipet, TenSette, 0.1- to 1.0-mL .....	each.....	19700-01
Pipet Tips, for 19700-01 TenSette Pipet .....	50/pkg.....	21856-96



**FOR TECHNICAL ASSISTANCE, PRICE INFORMATION AND ORDERING:**

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Outside the U.S.A. – Contact the HACH office or distributor serving you.

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