



✓ Method 8048

PhosVer 3 (Ascorbic Acid) Method*

Powder Pillows or AccuVac® Ampuls

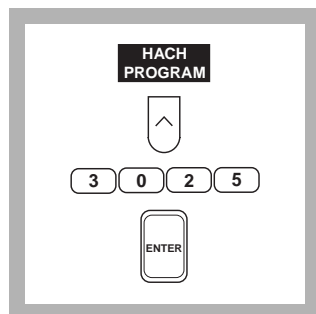
(0 to 2.500 mg/L PO₄³⁻)

Scope and Application: For water, wastewater, seawater; USEPA Accepted for reporting for wastewater analyses**. The estimated detection limits for program numbers 3025 and numbered 3030 are 0.045 and 0.031 mg/L PO₄³⁻ respectively.

* Adapted from *Standard Methods for the Examination of Water and Wastewater*

** Procedure is equivalent to USEPA method 365.2 and Standard Method 4500-P-E for wastewater.

Using Powder Pillows



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

Select the stored program number for phosphorus, ascorbic acid method by pressing **3025** with the numeric keys.

Press: **ENTER**

Note: If samples cannot be analyzed immediately, see *Sample Collection, Storage and Preservation* following these steps.

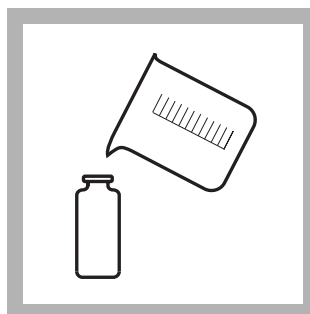
Note: The Flow Cell and Sipper Modules can be used with this procedure. Use a 25-mL sample and reagents with the Flow Cell Module.



2. The display will show: **HACH PROGRAM: 3025 P React. As. LR**

The wavelength (λ), **890 nm**, is automatically selected.

Note: For best results, determine a reagent blank for each new lot of reagent as follows. Prepare a reagent blank by repeating steps 3 through 8, using deionized water as the sample. Zero the instrument on deionized water by pressing the soft key under **ZERO**. Insert the reagent blank and the blank value will be displayed. Correct for the reagent blank by pressing the soft keys under **OPTIONS, (MORE)**, and then **BLANK:OFF**. Enter the reagent blank value and press **ENTER**. Repeat for each new lot of reagent.



3. Fill a sample cell with 10-mL of sample.

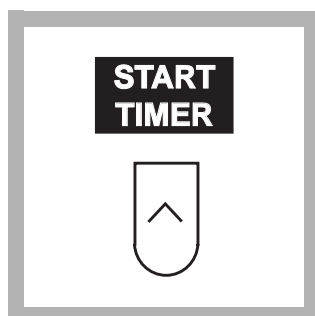
Note: For proof of accuracy, use a 1.0 mg/L Phosphate (0.33 mg/L P) Standard Solution listed under **OPTIONAL REAGENTS AND STANDARDS** in place of the sample.



4. Add the contents of one PhosVer 3 phosphate Powder Pillow to the cell (the prepared sample). Swirl immediately to mix.

Note: A blue color will form if phosphate is present.

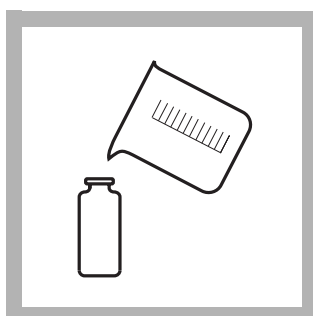
PHOSPHORUS, Reactive (Orthophosphate), continued



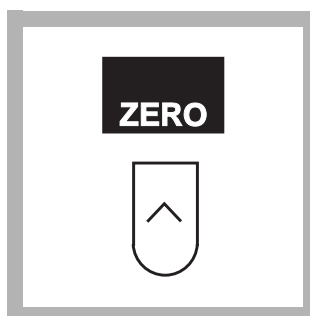
5. Press the soft key under **START TIMER**.

A 2-minute reaction period will begin.

Note: If the sample has been digested using the Acid Persulfate digestion in this manual, this step requires 10 minutes.



6. Fill another sample cell (the blank) with 10 mL of sample. Place it into the cell holder.



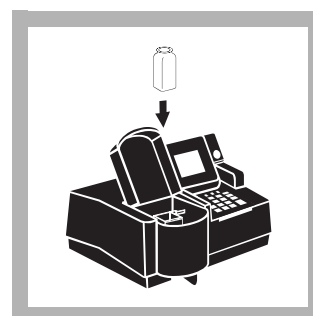
7. When the timer beeps press the soft key under **ZERO**.

The display will show:

0.000 mg/L PO₄³⁻

Note: If you are using a reagent blank correction, the display will show the correction.

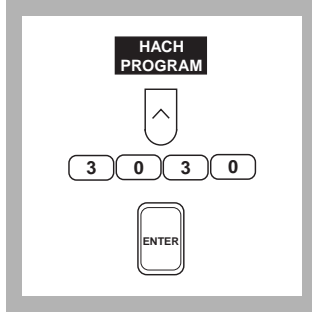
Note: For alternate concentration units press the soft key under **OPTIONS**. Then press the soft key under **UNITS** to scroll through the available options. Press **ENTER** to return to the read screen.



8. Place the prepared sample into the cell holder. Close the light shield. Results in mg/L PO₄³⁻ (or chosen units) will be displayed.

Note: The results can be expressed as PO₄³⁻, P or P₂O₅. Press the soft keys under **OPTIONS**, and then **FORM**: to scroll through the available options.

Using AccuVac Ampuls



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

Select the stored program number for phosphorus, ascorbic acid method by pressing **3030** with the numeric keys.

Press: **ENTER**

Note: If samples cannot be analyzed immediately, see *Sample Collection, Storage and Preservation* following these steps.

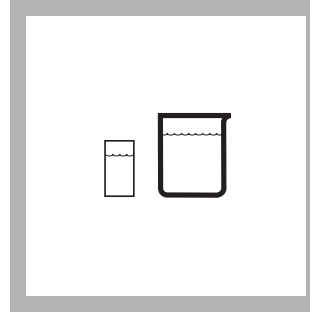
Note: The Flow Cell and Sipper Modules can be used with this procedure.



2. The display will show:
HACH PROGRAM: 3030
P React. As. LR AV

The wavelength (λ), **890 nm**, is automatically selected.

Note: For best results, determine a reagent blank for each new lot of reagent as follows. Prepare a reagent blank by repeating steps 3 through 10, using deionized water as the sample. Zero the instrument on deionized water by pressing the soft key under **ZERO**. Insert the reagent blank and the blank value will be displayed. Correct for the reagent blank by pressing the soft keys under **OPTIONS, (MORE)**, and then **BLANK:OFF**. Enter the reagent blank value and press **ENTER**. Repeat for each new lot of reagent.

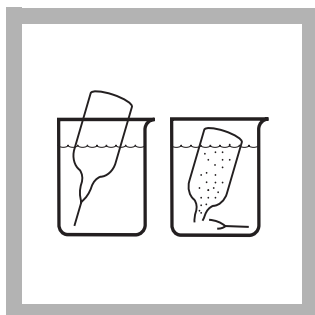


3. Fill a zeroing vial (the blank) with at least 10 mL of sample. Collect at least 40 mL of sample in a 50-mL beaker.



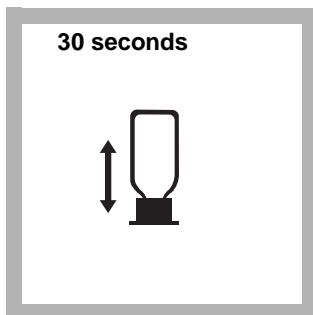
4. Insert the AccuVac Ampul Adapter into the sample cell module by sliding it under the thumb screw and into the alignment grooves. Fasten with the thumb screw.

PHOSPHORUS, Reactive (Orthophosphate), continued



5. Fill a PhosVer 3 Phosphate AccuVac Ampul with sample.

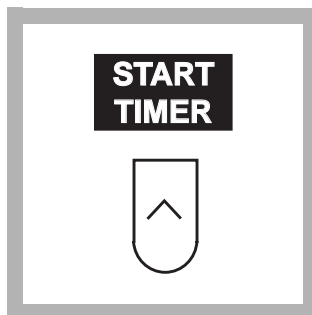
Note: Keep the tip immersed while the ampul fills completely.



6. Place an ampul cap securely over the tip of the ampul. Shake the ampul for approximately 30 seconds. Wipe off any liquid and fingerprints.

Note: A blue color will form if phosphate is present.

Note: Accuracy is unaffected by undissolved powder.



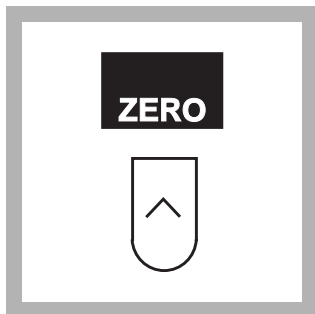
7. Press the soft key under **START TIMER**.

A 2-minute reaction period will begin.

Note: If the sample has been digested using the Acid Persulfate digestion in this manual, this step requires 10 minutes.



8. When the timer beeps, place the zeroing vial into the cell holder. Close the light shield.



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

The display will show:

0.000 mg/L PO₄³⁻

Note: If you are using a reagent blank correction, the display will show the correction.

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.



10. Place the AccuVac Ampul into the cell holder. Close the light shield. Results in mg/L PO₄³⁻ (or chosen units) will be displayed.

Note: The results can be expressed as PO₄³⁻, P or P₂O₅. Press the soft keys under **OPTIONS**, and then **FORM**: to scroll through the available options.

Interferences

Interfering Substance	Interference Levels and Treatments
Aluminum	Greater than 200 mg/L
Arsenate	Interferes at any level
Chromium	Greater than 100 mg/L
Copper	Greater than 10 mg/L
Hydrogen Sulfide	Interferes at any level
Iron	Greater than 100 mg/L
Nickel	Greater than 300 mg/L
pH, excess buffering	Highly buffered samples or extreme sample pH may exceed the buffering capacity of the reagents and require sample pretreatment. pH 2–10 is recommended.
Silica	Greater than 50 mg/L
Silicate	Greater than 10 mg/L
Turbidity (large amounts) or color	May cause inconsistent results because the acid in the powder pillow may dissolve some of the suspended particles and because of variable desorption of orthophosphate from the particles. For highly turbid or colored samples, add the contents of one Phosphate Pretreatment Powder Pillow to 25 mL of sample. Mix well. Use this solution to zero the instrument.
Zinc	Greater than 80 mg/L

Store the PhosVer 3 Phosphate Reagent Powder Pillows and AccuVac Ampuls in a cool, dry environment.

Sample Collection, Storage and Preservation

Collect sample in plastic or glass bottles that have been cleaned with 1:1 Hydrochloric Acid Solution and rinsed with deionized water. Do not use commercial detergents containing phosphate for cleaning glassware used in phosphate analysis.

Most reliable results are obtained when samples are analyzed as soon as possible after collection. If prompt analysis is impossible, preserve samples up to 24 hours by storing at or below 4 °C.

Accuracy Check

Standard Additions Method

- a. Leave the unspiked sample in the sample compartment. Verify that the units displayed are in mg/L. Select standard additions mode by pressing the soft keys under **OPTIONS, (MORE)** and then **STD ADD**.
- b. Press **ENTER** to accept the default sample volume (mL), 25.
- c. Press **ENTER** to accept the default standard concentration (mg/L) 50.
- d. Press the soft key under **ENTRY DONE**.
- e. Snap the neck off a Phosphate 2-mL Ampule Standard, 50-mg/L PO₄³⁻.
- f. Use the TenSette Pipet to add 0.1, 0.2 mL and 0.3 mL of standard, respectively to three 25-mL samples and mix each thoroughly (for AccuVac Ampuls, use 50-mL beakers).

PHOSPHORUS, Reactive (Orthophosphate), continued

- g. Analyze each standard addition sample as described above (use 10-mL aliquots of the standard addition samples for the powder pillow method). Accept the standard additions reading by pressing the soft key under **READ** each time. Each addition should reflect approximately 100% recovery.
- h. After completing the sequence, the display will show the extrapolated concentration value and the “best-fit” line through the standard additions data points, accounting for matrix interferences.
- i. See Section 1.4.1 *Standard Additions* for more information.

Method Performance

Precision

Standard: 1.000 mg/L PO₄³⁻

Program	95% Confidence Limits
3025	0.979–1.021 mg/L PO ₄ ³⁻
3030	0.985–1.014 mg/L PO ₄ ³⁻

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

Estimated Detection Limit

Program	EDL
3025	0.045 mg/L PO ₄ ³⁻
3030	0.031 mg/L PO ₄ ³⁻

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 the 40 CFR part 136, appendix B, see Section 1.5.1.

Sensitivity

Program Number: 3025

Portion of Curve	ΔAbs	ΔConcentration
0.010 Abs	0.010	0.0168 mg/L
1.25 mg/L	0.010	0.0177 mg/L
2.25 mg/L	0.010	0.0185 mg/L

Program Number: 3030

Portion of Curve:	ΔAbs	ΔConcentration
0.010 Abs	0.010	0.0160 mg/L
1.1 mg/L	0.010	0.0193 mg/L
1.98 mg/L	0.010	0.0184 mg/L

See Section 1.5.3 *Sensitivity Explained* for more information.

Calibration Standard Preparation

To perform a phosphate calibration using the ascorbic acid method, use a 10-mg/L Phosphate Standard Solution (Cat. No. 14204-16).

Prepare calibration standards containing 0.300, 1.500, and 2.400 mg/L Phosphate as follows:

- a. Into three different 100-mL Class A volumetric flasks, pipet 3, 15, and 24 mL of the 10-mg/L Phosphate Standard Solution using Class A glassware.
- b. Dilute to the mark with deionized water. Mix thoroughly.
- c. Using the ascorbic acid method and the calibration procedure described in the *User-Entered Programs* section of the *DR/4000 Spectrophotometer Instrument Manual*, generate a calibration curve from the standards prepared above.

Summary of Method

Orthophosphate reacts with molybdate in an acid medium to produce a Phosphomolybdate complex. Ascorbic acid then reduces the complex, giving an intense molybdenum blue color.

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

Please see *Section 1* for more information on proper disposal of these materials.

PHOSPHORUS, Reactive (Orthophosphate), continued

REQUIRED REAGENTS AND STANDARDS (Using Powder Pillows)

Description	Quantity Required Per Test	Unit	Cat. No.
PhosVer 3 Phosphate Reagent Powder Pillows, 10-mL.....	1 pillow	100/pkg.....	21060-69

REQUIRED REAGENTS AND STANDARDS (Using AccuVac Ampuls)

PhosVer 3 Phosphate Reagent AccuVac Ampuls.....	1 ampul.....	25/pkg.....	25080-25
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REQUIRED EQUIPMENT AND SUPPLIES (Using Powder Pillows)

DR/4000 1-Inch Cell Adapter	1	each.....	48190-00
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REQUIRED EQUIPMENT AND SUPPLIES (Using AccuVac Ampuls)

DR/4000 AccuVac Ampul Adapter.....	1	each.....	48187-00
Beaker, 50-mL.....	1	each.....	500-41
Cap, ampul, blue.....	1	25/pkg.....	1731-25
Sample Cell, 10-mL with cap (zeroing vial).....	1	each.....	21228-00

OPTIONAL REAGENTS AND STANDARDS

Hydrochloric Acid Standard Solution, 6.0 N (1:1)	500 mL.....	884-49
Phosphate Pretreatment Powder Pillows	50/pkg.....	14501-66
Phosphate Standard Solution, 1-mg/L as PO ₄	500 mL.....	2569-49
Phosphate Standard Solution, 10-mg/L as PO ₄	946 mL.....	14204-16
Phosphate Standard Solution, 2-mL PourRite Ampul, 50-mg/L as PO ₄	20/pkg.....	171-20H
Phosphate Standard Solution, 10-mL Voluette Ampul, 50-mg/L as PO ₄	16/pkg.....	171-10
Sodium Chloride, ACS.....	454 g.....	182-01
Sodium Hydroxide Standard Solution, 5.0 N.....	100 mL MDB*.....	2450-32
Water, deionized	4 liters.....	272-56

OPTIONAL EQUIPMENT AND SUPPLIES

AccuVac Snapper Kit	each.....	24052-00
Ampule Breaker Kit	each.....	21968-00
DR/4000 Carousel Module Kit	each.....	48070-02
DR/4000 Flow Cell Module Kit, 1-inch.....	each.....	48070-04
DR/4000 Flow Cell Module Kit, 1-cm.....	each.....	48070-05
DR/4000 Sipper Module Kit, 1-inch.....	each.....	48090-03
Flask, volumetric, Class, 100-mL	each.....	14574-42
pH Paper, pH 1.0 to 11.0	5 rolls/pkg.....	391-33
pH Meter, <i>sensio</i> TM 1, portable	each.....	51700-00
Pipet, 2-mL, serological	each.....	532-36
Pipet, TenSette, 0.1 to 1.0 mL	each.....	19700-01
Pipet Tips, for 19700-01.....	50/pkg.....	21856-96
Pipet, volumetric, Class A, 3.00-mL.....	each.....	14515-03
Pipet, volumetric, Class A, 6.00-mL.....	each.....	14515-06
Pipet, volumetric, Class A, 9.00-mL.....	each.....	14515-09
Pipet Filler, safety bulb.....	each.....	14651-00
PourRite Ampule Breaker	each.....	24846-00
Spoon, measuring, 0.1-g.....	each.....	511-00

* Larger sizes available from Hach.



FOR TECHNICAL ASSISTANCE, PRICE INFORMATION AND ORDERING:
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Outside the U.S.A. – Contact the HACH office or distributor serving you.
On the Worldwide Web – www.hach.com; E-mail – techhelp@hach.com

HACH COMPANY
WORLD HEADQUARTERS
Telephone: (970) 669-3050
FAX: (970) 669-2932