DOC316.53.01119

Phosphorus, Reactive (Orthophosphate)

USEPA^{1,2} PhosVer 3[®] (Ascorbic Acid) Method³ Method 8048 0.02 to 2.50 mg/L PO₄³⁻ Powder Pillows or AccuVac[®] Ampuls

Scope and application: For drinking water, wastewater and seawater.

- USEPA Accepted for reporting for wastewater analyses. Procedure is equivalent to USEPA and Standard Method 4500-P-E for wastewater.
- ² USEPA Accepted for reporting for drinking water analysis. Procedure is an acceptable version of EPA Method 365.1, approved at 40 CFR part 141 NPDWR compliance monitoring.
- ³ Adapted from Standard Methods for the Examination of Water and Wastewater.



Test preparation

Instrument-specific information

Table 1 shows sample cell and orientation requirements for reagent addition tests, such as powder pillow or bulk reagent tests. Table 2 shows sample cell and adapter requirements for AccuVac Ampul tests. The tables also show all of the instruments that have the program for this test.

To use the table, select an instrument, then read across to find the applicable information for this test.

Table 1 Instrument-specific information for reagent addition

Instrument	Sample cell orientation	Sample cell
DR 6000	The fill line is to the right.	2495402
DR 3800		
DR 2800		10 mL
DR 2700		
DR 1900		
DR 5000	The fill line is toward the user.	
DR 3900		
DR 900	The orientation mark is toward the user.	2401906

Table 2 Instrument-specific information for AccuVac Ampuls

Instrument	Adapter	Sample cell
DR 6000	_	2427606
DR 5000		
DR 900		— 10 mL
DR 3900	LZV846 (A)	
DR 1900	9609900 or 9609800 (C)	
DR 3800	LZV584 (C)	2122800
DR 2800		
DR 2700		_ 10 mL

Before starting

Install the instrument cap on the DR 900 cell holder before ZERO or READ is pushed.

For the best results, measure the reagent blank value for each new lot of reagent. Replace the sample with deionized water in the test procedure to determine the reagent blank value. Subtract the reagent blank value from the sample results automatically with the reagent blank adjust option.

Review the Safety Data Sheets (MSDS/SDS) for the chemicals that are used. Use the recommended personal protective equipment.

Dispose of reacted solutions according to local, state and federal regulations. Refer to the Safety Data Sheets for disposal information for unused reagents. Refer to the environmental, health and safety staff for your facility and/or local regulatory agencies for further disposal information.

Items to collect

Powder pillows

Description	Quantity
PhosVer® 3 Phosphate Reagent powder pillow, 10-mL	1
Sample cells. (For information about sample cells, adapters or light shields, refer to Instrument-specific information on page 1.)	2

Refer to Consumables and replacement items on page 6 for order information.

AccuVac Ampuls

Description	Quantity
PhosVer® 3 Phosphate Reagent AccuVac® Ampul	1
Beaker, 50-mL	1
Sample cells (For information about sample cells, adapters or light shields, refer to Instrument-specific information on page 1.)	1
Stopper for 18-mm tubes and AccuVac Ampuls	1

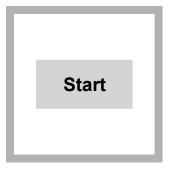
Refer to Consumables and replacement items on page 6 for order information.

Sample collection and storage

 Collect samples in clean glass or plastic bottles that have been cleaned with 6 N (1:1) hydrochloric acid and rinsed with deionized water.

- Do not use a detergent that contains phosphate to clean the sample bottles. The phosphate in the detergent will contaminate the sample.
- Analyze the samples as soon as possible for best results.
- If immediate analysis is not possible, immediately filter and keep the samples at or below 6 °C (43 °F) for a maximum of 48 hours.
- Let the sample temperature increase to room temperature before analysis.

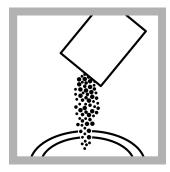
Powder pillow procedure



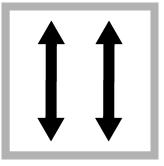
1. Start program 490 P React. PP. For information about sample cells, adapters or light shields, refer to Instrument-specific information on page 1.



2. Prepare the sample: Fill a sample cell with 10 mL of sample.



3. Add the contents of one PhosVer 3 Phosphate Reagent Powder Pillow to the cell. A blue color develops if phosphorus is in the sample.

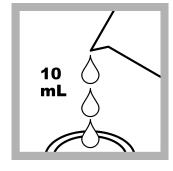


4. Immediately close the sample cell. Shake vigorously for 20–30 seconds.

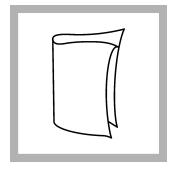


5. Start the instrument timer. A 2-minute reaction time starts.

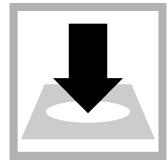
If the sample was digested using the Acid Persulfate digestion, a 10-minute reaction period is necessary.



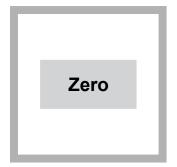
6. Prepare the blank: Fill a second sample cell with10 mL of sample.



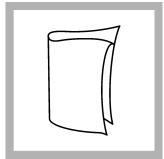
7. When the timer expires, clean the blank sample cell.



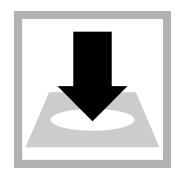
8. Insert the blank into the cell holder.



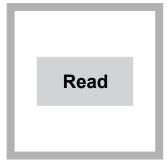
9. Push **ZERO**. The display shows 0.00 mg/L PO_4^{3-} .



10. Clean the prepared sample cell.

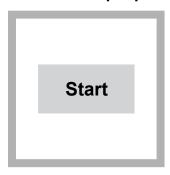


11. Insert the prepared sample into the cell holder.

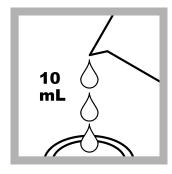


12. Push **READ**. Results show in mg/L PO_4^{3-} .

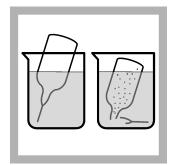
AccuVac Ampul procedure



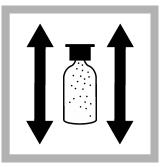
1. Start program 492 P React. PV AV. For information about sample cells, adapters or light shields, refer to Instrumentspecific information on page 1.



2. Prepare the blank: Fill the sample cell with 10 mL of sample.



3. Prepare the sample:
Collect at least 40 mL of sample in a 50-mL beaker.
Fill the AccuVac Ampul with sample. Keep the tip immersed while the AccuVac Ampul fills completely.

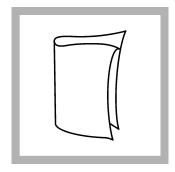


4. Close the AccuVac Ampul. Shake for approximately 30 seconds. Accuracy is not affected by undissolved powder.



5. Start the instrument timer. A 2-minute reaction time starts.

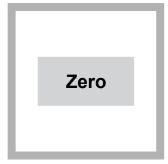
If the sample was digested using the Acid Persulfate digestion, a 10-minute reaction period is necessary.



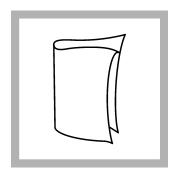
6. When the timer expires, clean the blank sample cell.



7. Insert the blank into the cell holder.



8. Push **ZERO**. The display shows $0.00 \text{ mg/L PO}_4^{3-}$.



9. Clean the AccuVac Ampul.



10. Insert the prepared sample AccuVac Ampul into the cell holder.



11. Push **READ**. Results show in mg/L PO_4^{3-} .

Interferences

Interfering substance	Interference level
Aluminum	More than 200 mg/L
Arsenate	Interferes at any level
Chromium	More than 100 mg/L
Copper	More than 10 mg/L
Hydrogen Sulfide	Interferes at any level
Iron	More than 100 mg/L
Nickel	More than 300 mg/L
Highly buffered samples or extreme sample pH	Can prevent the correct pH adjustment (of the sample) by the reagents. Sample pretreatment may be necessary. A pH range of 2–10 is recommended.
Silica	More than 50 mg/L
Silicate	More than 10 mg/L
Turbidity or color	Samples with a high amount of turbidity can give inconsistent results. The acid in the reagents can dissolve some of the suspended particles and variable desorption of orthophosphate from the particles can occur.
Zinc	More than 80 mg/L

Accuracy check

Standard additions method (sample spike)

Use the standard additions method (for applicable instruments) to validate the test procedure, reagents and instrument and to find if there is an interference in the sample. Items to collect:

- Phosphate standard solution, 50 mg/L PO₄³⁻ ampule
- Ampule breaker
- Pipet, TenSette[®], 0.1–1.0 mL and tips
- Mixing cylinders, 25-mL (3)
- 1. Use the test procedure to measure the concentration of the sample, then keep the (unspiked) sample in the instrument.
- **2.** Go to the Standard Additions option in the instrument menu.
- 3. Select the values for standard concentration, sample volume and spike volumes.
- 4. Open the standard solution.
- Prepare three spiked samples: use the TenSette pipet to add 0.1 mL, 0.2 mL and 0.3 mL of the standard solution, respectively, to three 10-mL portions of fresh sample. Mix well.

Note: For AccuVac[®] Ampuls, add 0.2 mL, 0.4 mL and 0.6 mL of the standard solution to three 50-mL portions of fresh sample.

- **6.** Use the test procedure to measure the concentration of each of the spiked samples. Start with the smallest sample spike. Measure each of the spiked samples in the instrument.
- 7. Select **Graph** to compare the expected results to the actual results.

Note: If the actual results are significantly different from the expected results, make sure that the sample volumes and sample spikes are measured accurately. The sample volumes and sample spikes that are used should agree with the selections in the standard additions menu. If the results are not within acceptable limits, the sample may contain an interference.

Standard solution method

Use the standard solution method to validate the test procedure, the reagents and the instrument.

Items to collect:

- 50 mg/L phosphate standard solution
- 100-mL volumetric flask, Class A
- 4-mL volumetric pipet, Class A and pipet filler safety bulb
- Deionized water
- 1. Prepare a 2.00-mg/L phosphate standard solution as follows:
 - **a.** Use a pipet to add 4.00 mL of a 50-mg/L phosphate standard solution into the volumetric flask. (Alternately, use one of the available mixed parameter standards. These standards contain 2.0 mg/L phosphate.)
 - **b.** Dilute to the mark with deionized water. Mix well. Prepare this solution daily.
- **2.** Use the test procedure to measure the concentration of the prepared standard solution.
- 3. Compare the expected result to the actual result.

Note: The factory calibration can be adjusted slightly with the standard adjust option so that the instrument shows the expected value of the standard solution. The adjusted calibration is then used for all test results. This adjustment can increase the test accuracy when there are small variations in the reagents or instruments.

Method performance

The method performance data that follows was derived from laboratory tests that were measured on a spectrophotometer during ideal test conditions. Users can get different results under different test conditions.

Program	Standard	Precision (95% Confidence Interval)	Sensitivity Concentration change per 0.010 Abs change
490	2.00 mg/L PO ₄ 3-	1.98–2.02 mg/L PO ₄ ^{3–}	0.02 mg/L PO ₄ ³⁻
492	2.00 mg/L PO ₄ ³⁻	1.98–2.02 mg/L PO ₄ ^{3–}	0.02 mg/L PO ₄ ^{3–}

Summary of method

Orthophosphate reacts with molybdate in an acid medium to produce a mixed phosphate/molybdate complex. Ascorbic acid then reduces the complex, which gives an intense molybdenum blue color. The measurement wavelength is 880 nm for spectrophotometers (DR 1900: 710 nm) or 610 nm for colorimeters.

Consumables and replacement items

Required reagents

Description	Quantity/Test	Unit	Item no.
PhosVer® 3 Phosphate Reagent Powder Pillow1, 10 mL	1	100/pkg	2106069
OR			
PhosVer® 3 Phosphate Reagent AccuVac® Ampul	1	25/pkg	2508025

Required apparatus

Description	Quantity/Test	Unit	Item no.
Beaker, 50 mL	1	each	50041H
Stoppers for 18-mm tubes and AccuVac Ampuls	2	6/pkg	173106

¹ PhosVer is a registered trademark of Hach Company.

Recommended standards

Description	Unit	Item no.
Phosphate Standard Solution, 10-mL Voluette® Ampule, 50 mg/L as PO ₄	16/pkg	17110
Phosphate Standard Solution, 50 mg/L as PO ₄ ³⁻	500 mL	17149
Phosphate Standard Solution, 1 mg/L as PO ₄ ³⁻	500 mL	256949
Drinking Water Standard, Mixed Parameter, Inorganic for F-, NO ₃ –N, PO ₄ ^{3–} , SO ₄ ^{2–}	500 mL	2833049
Wastewater Effluent Standard Solution, Mixed Parameter, for NH ₃ -N, NO ₃ -N, PO ₄ ³⁻ , COD, SO ₄ ²⁻ , TOC	500 mL	2833249
Water, deionized	4 L	27256

Optional reagents and apparatus

Description	Unit	Item no.
AccuVac [®] Drainer	each	4103600
AccuVac® Ampul Snapper	each	2405200
AccuVac [®] Ampul vials for sample blanks	25/pkg	2677925
Ampule Breaker, 10-mL Voluette [®] Ampules	each	2196800
Bottle, sampling, with cap, low density polyethylene, 250 mL	12/pkg	2087076
Mixing cylinder, graduated, 50 mL	each	189641
Flask, volumetric, Class A, 100 mL, glass	each	1457442
Hydrochloric Acid Solution, 6.0 N (1:1)	500 mL	88449
Paper, pH, 0–14 pH range	100/pkg	2601300
Phosphate Treatment Powder Pillows	100/pkg	1450199
Phosphate Standard Solution, 10 mg/L as PO ₄	946 mL	1420416
Phosphate Standard Solution, 15-mg/L as PO ₄ ³⁻	100 mL	1424342
Phosphate Standard Solution, 100-mg/L as PO ₄	100 mL	1436832
Phosphate Standard Solution, 10-mL ampule, 500 mg/L as PO ₄	16/pkg	1424210
Phosphate Standard Solution, 500-mg/L as PO ₄	100 mL	1424232
Pipet, TenSette [®] , 0.1–1.0 mL	each	1970001
Pipet, TenSette [®] , 1.0–10.0 mL	each	1970010
Pipet tips for TenSette® Pipet, 0.1–1.0 mL	50/pkg	2185696
Pipet tips for TenSette® Pipet, 0.1–1.0 mL	1000/pkg	2185628
Pipet tips for TenSette® Pipet, 1.0–10.0 mL	50/pkg	2199796
Pipet tips for TenSette® Pipet, 1.0–10.0 mL	250/pkg	2199725
Pipet, volumetric, Class A, 4.00 mL	each	1451504

