

PhosVer® 3 with Acid Hydrolysis Method

Method 8180

0.06 to 3.50 mg/L PO₄³⁻

Test 'N Tube™ Vials

Scope and application: For water, wastewater and seawater.



Test preparation

Instrument-specific information

Table 1 shows all of the instruments that have the program for this test. The table also shows adapter and light shield requirements for the instruments that use them.

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

Table 1 Instrument-specific information for test tubes

Instrument	Adapters	Light shield
DR 6000, DR 5000	—	—
DR 3900	—	LZV849
DR 3800, DR 2800, DR 2700	—	LZV646
DR 1900	9609900 (D ¹)	—
DR 900	4846400	Cover supplied with the instrument

¹ The D adapter is not available with all instrument versions.

Before starting

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

DR 3900, DR 3800, DR 2800 and DR 2700: Install the light shield in Cell Compartment #2 before this test is started.

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.

Clean all glassware with 6.0 N (1:1) hydrochloric acid, then fully rinse with deionized water to remove contaminants.

The reagent that is used in this test is corrosive. Use protection for eyes and skin and be prepared to flush any spills with running water.

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

Description	Quantity
Total and Acid Hydrolyzable Phosphorus Reagent Set	1
DRB200 Reactor	1
Funnel, micro	1
Light shield or adapter (For information about sample cells, adapters or light shields, refer to Instrument-specific information on page 1.)	1

Items to collect (continued)

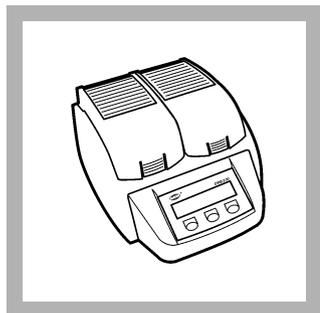
Description	Quantity
Pipet, TenSette [®] , 1.0- to 10.0-mL, with pipet tips	1
Test tube rack	1
Water, deionized	varies

Refer to [Consumables and replacement items](#) on page 5 for order information.

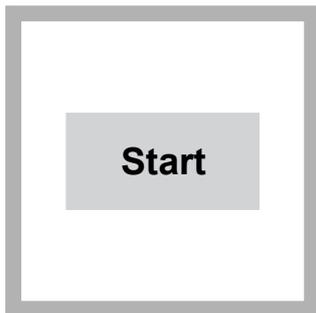
Sample collection and storage

- 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.

Acid hydrolysis, Test 'N Tube procedure



1. Start the DRB200 Reactor. Preheat to 150 °C. Refer to the DRB200 manual.

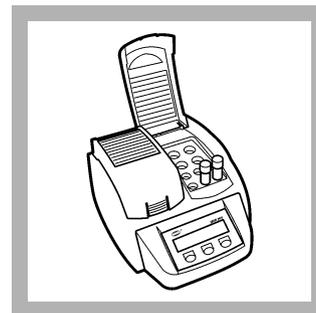


2. Start program **536 P Total/AH PV TNT**. For information about sample cells, adapters or light shields, refer to [Instrument-specific information](#) on page 1.

Note: Although the program name can be different between instruments, the program number does not change.



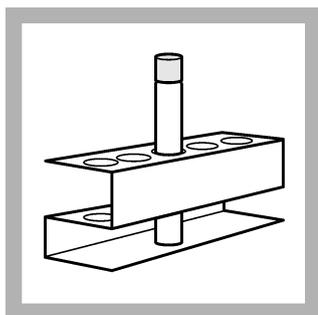
3. Add 5.0 mL of sample to the Total and Acid Hydrolyzable Test Vial. Close the vial and mix.



4. Insert the vial into the reactor. Close the reactor.



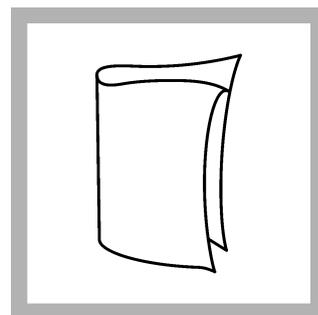
5. Start the instrument timer. A 30-minute reaction time starts.



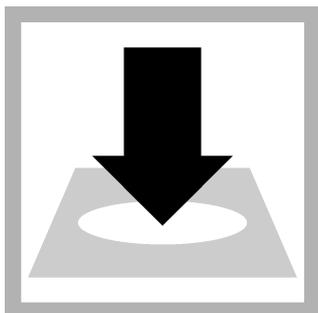
6. When the timer expires, carefully remove the vial from the reactor. Set the vial in a test tube rack. Let the vial cool to room temperature.



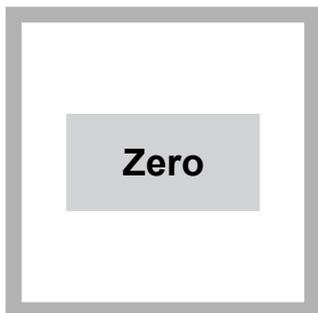
7. Add 2 mL of 1.00 N Sodium Hydroxide to the vial. Cap the vial and shake to mix.



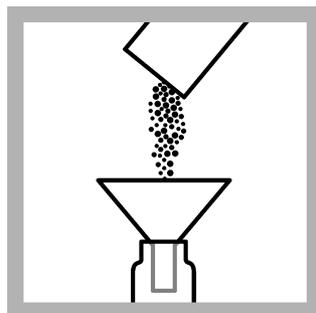
8. Clean the vial.



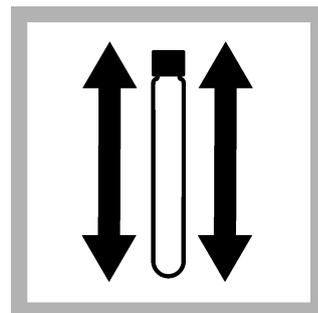
9. Insert the vial into the 16-mm cell holder.



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



11. Add the contents of one PhosVer 3 Powder Pillow to the vial.

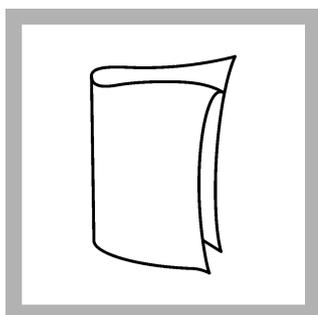


12. Put the cap on the vial. Shake for 20 to 30 seconds. The powder will not completely dissolve.

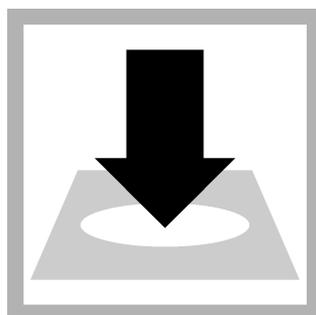


13. Start the instrument timer. A 2-minute reaction time starts.

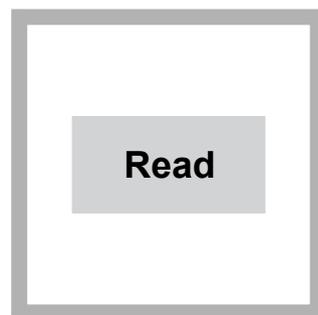
Read the results within two to eight minutes after adding the PhosVer 3 reagent.



14. Clean the vial.



15. Insert the vial into the 16-mm cell holder.



16. 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

Interfering substance	Interference level
Sulfide	More than 9 mg/L sulfide. Remove sulfide interference as follows: <ol style="list-style-type: none"> 1. Measure 25 mL of sample into a 50-mL beaker. 2. Add Bromine Water drop-wise with constant swirling until a permanent yellow color remains. 3. Add Phenol Solution drop-wise with constant swirling until the yellow color just disappears. Use this sample in the test procedure.
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 pre-treatment may be necessary.
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 2-mL Ampule Standard, 50-mg/L as PO_4^{3-}
 - 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.
 5. 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 25-mL portions of fresh sample. Mix well.
 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:

- 3.0-mg/L phosphate standard solution

1. Use the test procedure to measure the concentration of the standard solution.
2. 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 slight 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
536	3.00 mg/L PO ₄ ³⁻	2.93–3.07 mg/L PO ₄ ³⁻	0.06 mg/L PO ₄ ³⁻

Summary of method

Phosphates in condensed inorganic forms (meta-, pyro-, or other polyphosphates) are converted to reactive orthophosphate before measurement. The sample is pretreated with acid and heat to hydrolyze the condensed inorganic forms to orthophosphate. 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 (DR 1900: 710 nm) for spectrophotometers or 610 nm for colorimeters.

Pollution prevention and waste management

Reacted samples contain molybdenum and must be disposed of as a hazardous waste. Dispose of reacted solutions according to local, state and federal regulations.

Consumables and replacement items

Required reagents

Description	Quantity/test	Unit	Item no.
Water, deionized	varies	100 mL	27242
Total and Acid Hydrolyzable Phosphorus Reagent Set	—	50 tests	2742745
Includes:			
PhosVer [®] 3 Phosphate Reagent Powder Pillow, 10-mL	1	50/pkg	2106046
Potassium Persulfate Powder Pillow	1 pillow	50/pkg	2084766
Sodium Hydroxide, 1.54 N	varies	100 mL	2743042
Sodium Hydroxide Standard Solution, 1.00 N	2 mL	100 mL	104542
Total and Acid Hydrolyzable Test Vials (not sold separately)	1	50/pkg	—

Required apparatus

Description	Quantity/test	Unit	Item no.
DRB 200 Reactor, 110 VAC option, 15 x 16-mm wells	1	each	LTV082.53.40001
DRB 200 Reactor, 220 VAC option, 15 x 16-mm wells	1	each	LTV082.52.40001
Funnel, micro, poly	1	each	2584335
Pipet, volumetric, Class A, 2.00-mL	1	each	1451536
Pipet, volumetric, Class A, 5.00-mL	1	each	1451537
Pipet filler, safety bulb	1	each	1465100

Required apparatus (continued)

Description	Quantity/test	Unit	Item no.
Pipet, TenSette [®] , 1.0–10.0 mL	1	each	1970010
Pipet Tips, for TenSette [®] Pipet, 1.0–10.0 mL	2	250/pkg	2199725
Test tube rack	1	each	1864100

Recommended standards

Description	Unit	Item no.
Drinking Water Standard, Mixed Parameter, Inorganic for F ⁻ , NO ₃ -N, PO ₄ ³⁻ , SO ₄ ²⁻	500 mL	2833049
Phosphate Standard Solution, 50-mg/L, 10-mL Voluette [®] Ampules	16/pkg	17110
Phosphate Standard Solution, 1-mg/L as PO ₄ ³⁻	500 mL	256949
Phosphate Standard Solution, 3-mg/L as PO ₄ ³⁻	946 mL	2059716
Wastewater Effluent Standard Solution, Mixed Parameter, for NH ₃ -N, NO ₃ -N, PO ₄ ³⁻ , COD, SO ₄ ²⁻ , TOC	500 mL	2833249

Optional reagents and apparatus

Description	Unit	Item no.
Bromine Water, 30 g/L	29 mL	221120
Mixing cylinder, graduated, 25-mL	each	189640
Hydrochloric Acid Solution, 6.0 N (1:1)	500 mL	88449
Phenol Solution, 30-g/L	29 mL	211220
Paper, pH, 0–14 pH range	100/pkg	2601300
Filter paper, folded, 3–5-micron, 12.5-cm	100/pkg	69257
Funnel, poly, 65-mm	each	108367
Thermometer, non-mercury, –10 to +225 °C	each	2635700
Bottle, sampling, with cap, low density polyethylene, 250-mL	12/pkg	2087076
Pipet tips for TenSette [®] Pipet, 1.0–10.0 mL	50/pkg	2199796
Beaker, 50-mL	each	50041H

Optional standards

Description	Unit	Item no.
Ampule Breaker, 10-mL Voluette [®] Ampules	each	2196800
Phosphate Standard Solution, 3-mg/L as PO ₄ ³⁻	946 mL	2059716
Phosphate Standard Solution, 10-mg/L as PO ₄ ³⁻	946 mL	1420416
Phosphate Standard Solution, 15-mg/L as PO ₄ ³⁻	100 mL	1424342
Phosphate Standard Solution, 30-mg/L as PO ₄ ³⁻	946 mL	1436716
Phosphate Standard Solution, 50-mg/L, 10-mL Voluette [®] Ampules	16/pkg	17110
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



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