

FERTILIZER METHODS

Chapter

PHOSPHATE ANALYSIS

Subject

Ortho-Phosphate – P
Surface and Waste Water – Flow Injection

Adapted from QuickChem Method 10-115-01-1-A
By: Lachat Instruments 6645 West Mill Road, Milwaukee, WI 53218

*Determination of Ortho Phosphate – P in Surface and Waste Waters
by Flow Injection Analysis Colorimetry*

0.02 to 2.0 mg P/L

PRINCIPLE

The OrthoPhosphate ion (PO_4^{3-}) reacts with Ammonium Molybdate and Antimony Potassium Tartrate under acidic conditions to form a complex. This complex is reduced with Ascorbic Acid to form a blue complex that absorbs light at 880 nm. The absorbance is proportional to the concentration of orthophosphate in the sample.

SCOPE & APPLICATION

This method covers the determination of orthophosphate in drinking, ground, and surface waters, and domestic and industrial wastes. This method determines total orthophosphate or if the sample is filtered through a 0.45 micron pore size filter, the result is termed dissolved orthophosphate. The difference between the result of a sample determined directly and filtered is termed insoluble orthophosphate. **The applicable range is 0.02 to 2.00 mg P/L. The method detection limit is 0.02 mg P/L. The method throughput is 80 injections per hour..**

DEFINITIONS

See: Bureau of Feed, Seed & Fertilizer Laboratories, Standard Operating Procedures, and Lachat Definitions

INTERFERENCES

Only orthophosphate forms a blue color in this test. Polyphosphates and Organic Phosphorus compounds are not recovered. The Sulfuric Acid in the Molybdate reagent does not have enough contact time with Polyphosphates to hydrolyze them. The PO_4^{3-} reacts with Ammonium Molybdate and Antimony Potassium Tartrate under acidic conditions to form an Antimony-Phosphomolybdate complex. This complex is reduced with Ascorbic Acid to form a blue

complex that absorbs light at 880 nm. The absorbance is proportional to the concentration of PO_4^{3-} in the digested sample.

Silica forms a pale blue complex that also absorbs at 880 nm. This interference is generally insignificant as a silicate concentration of approximately 30 mg SiO_2/L would be required to produce a 0.005 mg P/L positive error in orthophosphate.

For dissolved orthophosphate, sample turbidity must be removed by filtration prior to analysis. Sample color that absorbs at 880 nm will also interfere. When in doubt about background absorbance, the background concentration should be determined. See the System Notes section for more information.

SAFETY

Each laboratory is responsible for maintaining a current awareness file of the Occupational Health and Safety Act (OSHA) regulations regarding the safe handling of the chemicals specified in this method. A reference file of Material Safety Data Sheets (MSDS) should be made available to all personnel involved in the chemical analysis. The preparation of a formal safety plan is also advisable.

EQUIPMENT & SUPPLIES

Balance - analytical, capable of accurately weighing to the nearest 0.0001 g.

Glassware - Class A volumetric flasks and pipettes or plastic containers as required. Samples may be stored in plastic or glass containers.

Flow injection analysis equipment designed to deliver and react sample and reagents in the required order and ratios.

Auto-Sampler

Multichannel proportioning pump

Reaction unit or manifold

Colorimetric detector

Data system

Acid-washed glassware: All glassware used in the determination of phosphate should be washed with hot 1:1 HCl and rinsed with distilled water. Preferable, this glassware should be used only for the determination of phosphate and after use it should be rinsed with distilled water and kept covered until needed again. If this is done, the treatment with 1:1 HCl is only required occasionally. Commercial detergent should never be used.

10 mm flow cell

PVC Pump tubes **NOTE: PVC PUMP TUBES MUST BE USED FOR THIS METHOD.**

REAGENTS AND STANDARDS

PREPARATION OF REAGENTS

Use deionized (D.I.) water (10 megohm) for all solutions.

Degassing with helium:

To prevent bubble formation, degas all solutions except the standards with helium. Use He at 140kPa (20 lb/in²) through a helium degassing tube (Lachat Part # 50100.) Bubble He through the solution for one minute.

Stock Ammonium Molybdate Solution

By Volume:

In a **1 L** volumetric flask dissolve **40.0 g Ammonium Molybdate Tetrahydrate** [(NH₄)₆Mo₇O₂₄·4H₂O] in approximately **800 mL DI Water**. Dilute to the mark and stir for four hours. Store in plastic and refrigerate. May be stored up to two months when kept refrigerated.

By Weight:

To a tared **1 L** container add **40.0 g Ammonium Molybdate Tetrahydrate** [(NH₄)₆Mo₇O₂₄·4H₂O] and **983 g DI water**. Stir for four hours. Store in plastic and refrigerate. May be stored up to two months when kept refrigerated.

Stock Antimony Potassium Tartrate Solution

By Volume:

In a **1 L** volumetric flask, dissolve **3.0 g Antimony Potassium Tartrate** (Potassium Antimonyl Tartrate Hemihydrate K(SbO)C₄H₄O₆·1/2H₂O) or dissolve **3.22 g Antimony Potassium Tartrate** (Potassium Antimonyl Tartrate Trihydrate - C₈H₄O₁₂K₂Sb₂·3H₂O) in approximately **800 mL of DI Water**. Dilute to the mark and invert three times. Store in a dark bottle and refrigerate. Maybe stored up to two months when kept refrigerated.

By Weight:

To a **1 L** dark, tared container add **3.0 g Antimony Potassium Tartrate** (Potassium Antimonyl Tartrate Hemihydrate K(SbO) C₄H₄O₆·1/2H₂O) or dissolve **3.22 g Antimony Potassium Tartrate** (Potassium Antimonyl Tartrate Trihydrate C₈H₄O₁₂K₂Sb₂·3H₂O) and **995 g DI water**. Stir or shake until dissolved. Store in a dark bottle and refrigerate. Maybe stored up to two months when kept refrigerated.

Molybdate Color Reagent

By Volume:

To a **1 L** volumetric flask add about **500 mL DI Water**, then add **35.0 mL Concentrated Sulfuric Acid** (CAUTION: The solution will get very hot!). Swirl to mix. When it can be comfortably handled, add **72.0 mL Stock Antimony Potassium Tartrate Solution** and **213 mL Stock Ammonium Molybdate Solution**. Dilute to the mark and invert three times. Degas with helium. Prepare fresh weekly.

By Weight:

To a tared **1 L** container add **680 g DI water**, then **64.4 g Concentrated Sulfuric Acid** (CAUTION: The solution will get very hot!). Swirl to mix. When it can be comfortably handled, add **72.0 g Stock Antimony Potassium Tartrate Solution** and **213 g Stock Ammonium Molybdate Solution**. Shake and degas with helium. Prepare fresh weekly.

Ascorbic Acid Reducing Solution, 0.33 M

By Volume:

In a **1 L** volumetric flask dissolve **60.0 g granular Ascorbic Acid** in about **700 mL of DI Water**. Dilute to the mark and invert to mix. Add **1.0 g Dodecyl Sulfate** [$\text{CH}_3(\text{CH}_2)_{11}\text{OSO}_3\text{Na}$]. Prepare fresh weekly. Discard if the solution becomes yellow.

By Weight:

To a tared **1 L** container, add **60.0 g granular Ascorbic Acid** and **975 g DI Water**. Stir or shake until dissolved. Add **1.0 g Dodecyl Sulfate** [$\text{CH}_3(\text{CH}_2)_{11}\text{OSO}_3\text{Na}$]. Prepare fresh weekly. Discard if the solution becomes yellow.

Sodium Hydroxide - EDTA Rinse:

Dissolve **65 g Sodium Hydroxide (NaOH)** and **6 g Tetrasodium Ethylenediamine Tetraacetic Acid (Na₄EDTA)** in **1.0 L or 1.0 kg DI Water**.

PREPARATION OF STANDARDS

Stock Standard 250.0 mg P/L

In a **1 L** volumetric flask dissolve **1.099 g primary standard grade Anhydrous Potassium Phosphate Monobasic (KH₂PO₄)** that has been dried for one hour at 105°C in about **800 mL DI Water**. Dilute to the mark with **DI Water** and invert to mix.

Working Stock Standard Solution 5.00 mg P/L

By Volume:

In a **250 mL** volumetric flask, dilute **5.0 mL Stock Standard** to the mark with **DI Water**. Invert to mix.

By Weight:

To a tared **250 mL** container add about **5 g Stock Standard**. Divide the actual weight of the solution added by **0.02** and make up to this resulting total weight with **DI Water**. Shake to mix.

Working Standards (Prepare Daily)	A	B	C	D	E	F
Concentration mg P/L	2.00	0.50	0.20	0.05	0.01	0.00

By Volume

Volume (mL) of stock standard 2 diluted to 250 mL with DI water .	100.0	25.0	10.0	2.5	0.50	0.00
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By Weight

Weight (g) of stock standard 2 diluted to final weight (~250 g) divided by factor below with DI water .	100	25	10	2.5	0.5	0
Division Factor Divide exact weight of the standard by this factor to give the final weight.	0.4	0.1	0.04	0.01	0.002	---

SAMPLE PRESERVATION AND STORAGE

Samples should be collected in plastic or glass bottles. All bottles must be thoroughly cleaned and rinsed with reagent water. Volume collected should be sufficient to insure a representative sample, allow for replicate analysis (if required), and minimize waste disposal.

The USEPA recommends that samples be filtered immediately upon collection, with a maximum holding time of 48 hours.

CALIBRATION PROCEDURE

Pump D.I. water through all reagent lines and check for leaks and smooth flow. Switch to reagents and allow the system to equilibrate until a stable baseline is achieved.

Place samples and/or standards in the sampler. Input the information required by the data system, such as concentration, replicates and QuikChem scheme.

Calibrate the instrument by injecting the standards. The data system will then associate the concentrations with the instrument responses for each standard.

SYSTEM NOTES

Pump DI water through all reagent lines and check for leaks and smooth flow. Switch to reagents and allow the system to equilibrate until a stable baseline is achieved. Place samples and/or standards in the sampler. Input the information required by data system, such as concentration, replicates and QC scheme.

Calibrate the instrument by injecting the standards. The data system will then associate the concentrations with the instrument responses for each standard.

Glassware contamination is a problem in low level phosphorus determinations. Glassware should be washed with 1:1 HCl and rinsed with deionized water. Commercial detergents should rarely be needed but, if they are used, use special phosphate-free detergents for lab glassware.

Allow 15 min for heating unit to warm up to 37°C.

If necessary, at end of run place the color reagent and Ascorbic Acid transmission lines into the NaOH - EDTA solution. Pump this solution for approximately 5 minutes to remove any precipitated reaction products. Then place these lines in water and pump for an additional 5 minutes. Then pump dry all lines.

DATA ANALYSIS AND CALCULATIONS

Prepare a calibration curve by plotting instrument response against standard concentration. Compute sample concentration by comparing sample response with the standard curve.

Report only those values that fall between the lowest and the highest calibration standards. Samples exceeding the highest standard should be diluted and reanalyzed.

Report results in mg P/L.

METHOD PERFORMANCE

See: Quikchem method 10-115-01-1-A from Lachat Instruments.

FLOWCHARTS AND VALIDATION DATA

See: Quikchem method 10-115-01-1-A from Lachat Instruments.

TABLES AND DIAGRAMS

DATA SYSTEM PARAMETERS FOR QUIKCHEM 8000

The timing values listed below are approximate and will need to be optimized using graphical events programming.

Sample throughput: 80 samples/h, 45 s/sample
Pump Speed: 35
Cycle Period: 45 seconds.

Analyte Data:

Concentration Units: mg P/L
Peak Base Width: 25 s
% Width Tolerance: 100
Threshold: 10000
Inject to Peak Start: 10 s
Chemistry: Direct

Calibration Data:

Level	1	2	3	4	5	6	
Concentration mg P/L	2.0	0.5	0.2	0.05	0.01	0	

Calibration Rep Handling: Weighted Average
Calibration Fit Type: 1st Order Polynomial
Weighting Method: None
Force through zero: No

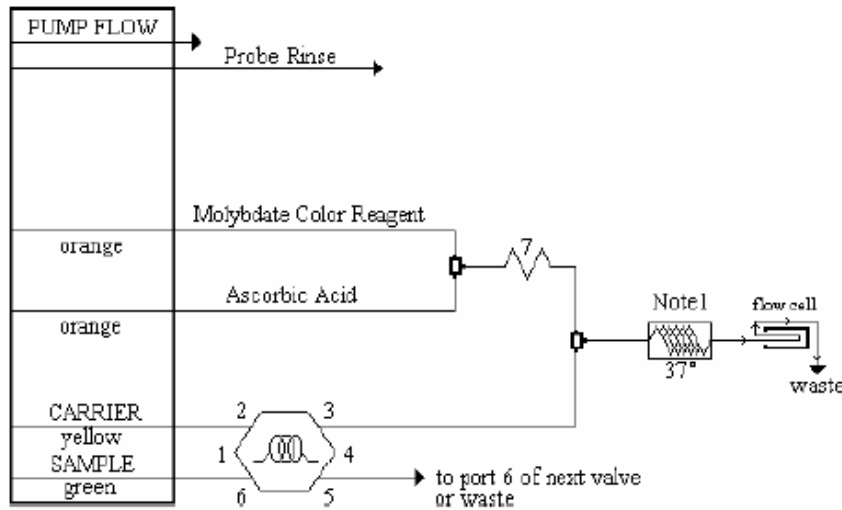
Sampler Timing:

Min. Probe in Wash Period: 9.0 s
Probe in Sample Period: 20 s

Valve Timing:

Load Time: 0 s
Load Period: 15 s
Inject Period: 25 s

ORTHOPHOSPHATE MANIFOLD DIAGRAM




Carrier: DI water

Manifold Tubing: 0.8 mm (0.032 in) i.d. This is 5.2 $\mu\text{L}/\text{cm}$.

AE Sample Loop: 70 cm x 0.8 mm i.d.

QC8000 Sample Loop: 75.5 cm x 0.8 mm i.d.

Interference Filter: 880 nm

Apparatus: An injection valve, a 10 mm path length flow cell, and a colorimetric detector module is required. The  shows 175 cm of tubing wrapped around the heater block at 37°C.

7: 135 cm of tubing on a 7 cm coil support

Note 1: 175 cm of tubing on the heater.

APPROVAL:

Approved by: Weldon Collier Date: 6/8/06
Signature
Bureau Chief
Title

METHOD REVISION HISTORY:

Version	Date	Description	Author
Original	5/8/03		J. Vyas
Revised	6/1/06		J. Vyas

REFERENCE:

U.S. Environmental Protection Agency, Methods for the Determination of Inorganic Substances in Environmental Samples, EPA-600/R-93/100, August 1993, Method 365.1.

Quikchem method 10-115-01-1-A from Lachat Instruments.