INTERNATIONAL STANDARD

ISO 15681-1

First edition
2003-12-15

Water quality — Determination of orthophosphate and total phosphorus contents by flow analysis (FIA and CFA) —

Part 1: Method by flow injection analysis (FIA)

Qualité de l'eau — Dosage des orthophosphates et du phosphore total par analyse en flux (FIA et CFA) —

Partie 1: Méthode par analyse avec injection en flux (FIA)

Reference number
ISO 15681-1:2003(E)

ISO copyright office
Case postale 56 • CH-1211 Geneva 20
Tel. + 41 22 749 01 11
Fax + 41 22 749 09 47
E-mail copyright@iso.org
Web www.iso.org

Published in Switzerland

Contents

Page

Foreword

ISO (the International Organization for Standardization) is a worldwide federation of national standards bodies (ISO member bodies). The work of preparing International Standards is normally carried out through ISO technical committees. Each member body interested in a subject for which a technical committee has been established has the right to be represented on that committee. International organizations, governmental and non-governmental, in liaison with ISO, also take part in the work. ISO collaborates closely with the International Electrotechnical Commission (IEC) on all matters of electrotechnical standardization.

International Standards are drafted in accordance with the rules given in the ISO/IEC Directives, Part 2.

The main task of technical committees is to prepare International Standards. Draft International Standards adopted by the technical committees are circulated to the member bodies for voting. Publication as an International Standard requires approval by at least 75 % of the member bodies casting a vote.

Attention is drawn to the possibility that some of the elements of this document may be the subject of patent rights. ISO shall not be held responsible for identifying any or all such patent rights.

ISO 15681-1 was prepared by Technical Committee ISO/TC 147, *Water quality*, Subcommittee SC 2, *Physical, chemical and biochemical methods*.

ISO 15681 consists of the following parts, under the general title *Water quality — Determination of orthophosphate and total phosphorus contents by flow analysis (FIA and CFA)*:

— *Part 1: Method by flow injection analysis (FIA)*

— *Part 2: Method by continuous flow analysis (CFA)*

Introduction

Methods of determining water quality using flow analysis automated wet chemical procedures, and are particularly suitable for the processing of many analytes in water in large sample series at a high analysis frequency.

Analysis can be performed by flow injection analysis (FIA) [1], [2] or continuous flow analysis (CFA) [3]. Both methods share the feature of an automatic dosage of the sample into a flow system (manifold) where the analyte in the sample reacts with the reagent solutions on its way through the manifold. The sample preparation may be integrated in the manifold. The amount of reaction product is measured in a flow detector (e.g. flow photometer). This part of ISO 15681 describes the FIA method.

The user should be aware that particular problems could require the specification of additional marginal conditions.

Water quality — Determination of orthophosphate and total phosphorus contents by flow analysis (FIA and CFA) —

Part 1:
Method by flow injection analysis (FIA)

WARNING — Persons using this part of ISO 15681 should be familiar with normal laboratory practice. This part of ISO 15681 does not purport to address all of the safety problems, if any, associated with its use. It is the responsibility of the user to establish appropriate safety and health practices and to ensure compliance with any national regulatory conditions. Molybdate and antimony waste solutions should be disposed of properly. It is absolutely essential that tests conducted according to this part of ISO 15681 be carried out by suitably qualified staff.

1 Scope

This part of ISO 15681 specifies flow injection analysis (FIA) methods for the determination of orthophosphate in the mass concentration range from 0,01 mg/l to 1,0 mg/l (P), and total phosphorus by manual digestion in accordance with ISO 6878 [5], [6] for the mass concentration range from 0,1 mg/l to 10 mg/l (P). The range of application can be changed by varying the operating conditions.

This part of ISO 15681 is applicable to various types of water (such as ground, drinking, surface, leachate and waste waters).

This method is also applicable to the analysis of seawater, but with changes in sensitivity, by adaptation of the carrier and calibration solutions to the salinity of the samples.

2 Normative references

The following reference documents are indispensable for the application of this document. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendments) applies.

ISO 3696, *Water for analytical laboratory use — Specification and test methods*

ISO 5667-1, *Water quality — Sampling — Part 1: Guidance on the design on sampling programmes*

ISO 5667-2, *Water quality — Sampling — Part 2: Guidance on sampling techniques*

ISO 5667-3, *Water quality — Sampling — Part 3: Guidance on the preservation and handling of water samples*

ISO 6878:— [1)], *Water quality — Determination of phosphorus — Ammonium molybdate spectrometric method*

ISO 8466-1, *Water quality — Calibration and evaluation of analytical methods and estimation of performance characteristics — Part 1: Statistical evaluation of the linear calibration function*

1) To be published.

3 Interferences

3.1 General interferences

ISO 6878:—, Annex B gives a list of general interferences. In addition, or contrary to the cited standard, the following guidelines apply.

a) Arsenate causes serious interference. 100 µg/l As, present as arsenate, results in a response comparable to approximately 30 µg/l P.

b) If the silicate concentration in samples is not greater than 60 times the phosphorus concentration, interferences by silicate can be neglected.

c) Fluoride interference is significant above 50 mg/l.

d) Nitrite interference is significant above 5 mg/l. The interference can be eliminated by acidifying samples after collection.

e) For samples containing high concentrations of oxidizing agents, the amount of added reducing reagent can be insufficient. In this case it is advisable to remove the oxidizing material prior to digestion.

f) The self-absorption of the sample can be compensated by measuring, in addition to the sample signal (8.6), the signal of the sample without the admixture of the reagents. In this case, the difference of the two responses is used for the evaluation (see Clause 9).

3.2 Interferences in the determination of total-P

The interferences from silicate, nitrite, fluoride and iron described for the determination of orthophosphate are generally not observed, due to the pre-digestion and the higher analytical range.

The efficiency of the digestion can be affected for water samples with a chemical oxygen demand (COD) value of more than 10 times the highest concentrations of the calibration solutions (5.16). In this case the sample should be diluted.

4 Principle

4.1 Determination of orthophosphate

The sample is injected into a carrier stream, which is merged with an acidic ammonium molybdate solution.

The resulting molybdophosphoric acid is reduced by tin(II) chloride to molybdenum blue [4], [5].

4.2 Total phosphorus with manual digestion

Phosphorus compounds in the sample are oxidized manually with a potassium peroxodisulfate solution, in accordance with ISO 6878. The resulting orthophosphate is determined by the molybdenum blue reaction as in 4.1 [5], [6].

5 Reagents

Use analytical grade chemicals unless otherwise specified. The phosphate blank value shall be checked (8.3).

Degas carefully all carrier and reagent solutions for the FIA determinations before use, e.g. by vacuum filtration or purging with helium (for at least 10 min).

5.1 Water, complying to grade 1 of ISO 3696.

5.2 Sulfuric acid, H_2SO_4.

5.2.1 Sulfuric acid (I), ρ = 1,84 g/ml; 98 % (mass fraction).

5.2.2 Sulfuric acid (II), $c(H_2SO_4)$ = 2,45 mol/l.

To approximately 800 ml of water (5.1) carefully add 136 ml of sulfuric acid (I) (5.2.1) while stirring. Cool and dilute to 1 000 ml with water (5.1).

5.3 Ammonium heptamolybdate tetrahydrate, $(NH_4)_6Mo_7O_{24} \cdot 4H_2O$.

5.4 Hydrazine sulfate, $N_2H_6SO_4$.

5.5 Tin(II) chloride dihydrate, $SnCl_2 \cdot 2\ H_2O$.

5.6 Potassium peroxodisulfate, $K_2S_2O_8$.

5.7 Potassium dihydrogen phosphate, KH_2PO_4, dried to constant mass at 105 °C ± 5 °C.

5.8 Potassium pyrophosphate, $K_4P_2O_7$.

5.9 Organophosphorus compounds to check the digestion.

5.9.1 Pyridoxal-5-phosphate monohydrate, $C_8H_{10}NO_6P \cdot H_2O$ *or alternatively*:

5.9.2 Disodium phenylphosphate, $C_6H_5Na_2PO_4$.

5.10 Molybdate solution (R1 in Figure A.1).

Dissolve 35 ml of sulfuric acid (I) (5.2.1) and 10 g of ammonium heptamolybdate tetrahydrate (5.3) in about 800 ml water (5.1), cool and dilute to 1 000 ml.

The solution is stable for 3 months if stored at room temperature.

5.11 Tin (II) chloride reagent (R2 in Figure A.1).

Dissolve 28 ml of sulfuric acid (I) (5.2.1), 200 mg of tin(II) chloride (5.5) and 2 g of hydrazine sulfate (5.4) in about 800 ml water (5.1), cool and dilute to 1 000 ml. Store at 4 °C ± 2 °C.

The solution is stable for 1 week if stored at 4 °C ± 2 °C.

NOTE Instead of hydrazine sulfate (5.4), *N,N*-diethylhydroxylamine, DEHA (Annex D) may be used. This change was not part of the validation in the interlaboratory trial cited in Annex B.

5.12 Carrier solutions

5.12.1 Carrier solution I, for orthophosphate determination (C1 in Figure A.1).

The carrier solution I is water (5.1).

5.12.2 Carrier solution II, for total phosphorus (P) determination after manual digestion (C1 in Figure A.1).

Add 5 ml of sulfuric acid (I) (5.2.1) to 1 000 ml of water (5.1) and mix.

5.13 Orthophosphate stock solution I, ρ = 50,0 mg/l P.

Dissolve 220 mg ± 1 mg of potassium dihydrogenphosphate (5.7) in water (5.1) and dilute to 1 000 ml. Store in a tightly closed glass bottle.

The solution is stable for 2 months if stored at 4 °C ± 2 °C.

5.14 Orthophosphate stock solution II, ρ = 10,0 mg/l P.

Dilute 20 ml of orthophosphate stock solution I (5.13) to 100 ml with water (5.1).

Prepare freshly each day of use.

5.15 Orthophosphate stock solution III, ρ = 1,00 mg/l P.

Dilute 2 ml of orthophosphate stock solution I (5.13) to 100 ml with water (5.1).

Prepare freshly each day of use.

5.16 Calibration solutions

Prepare at least 5 calibration solutions, evenly distributed over the working range, by diluting solutions 5.13 to 5.15 according to the range required.

Ranges:

For orthophosphate: Range II: 0,01 mg/l to 0,10 mg/l P
Range I: 0,10 mg/l to 1,00 mg/l P

For total phosphorus: Range II: 0,10 mg/l to 1,00 mg/l P
Range I: 1,00 mg/l to 10,0 mg/l P

Tables 1 to 3 give examples for the preparation of 10 calibration solutions for the above-mentioned ranges.

Table 1 — Example for the preparation of 10 calibration solutions for the orthophosphate range II (0,01 mg/l to 0,10 mg/l P)

Millilitres of orthophosphate stock solution III (5.15) diluted to 100 ml	1	2	3	4	5	6	7	8	9	10
Concentration of calibration solutions, mg/l P	0,01	0,02	0,03	0,04	0,05	0,06	0,07	0,08	0,09	0,10

Table 2 — Example for the preparation of 10 calibration solutions for the orthophosphate range I and total phosphorus range II (0,1 mg/l to 1,0 mg/l P)

Millilitres of orthophosphate stock solution II (5.14) diluted to 100 ml	1	2	3	4	5	6	7	8	9	10
Concentration of calibration solutions, mg/l P	0,10	0,20	0,30	0,40	0,50	0,60	0,70	0,80	0,90	1,00

Table 3 — Example for the preparation of 10 calibration solutions for the total phosphorus range I (1 mg/l to 10 mg/l P)

Millilitres of orthophosphate stock solution I (5.13) diluted to 100 ml	2	4	6	8	10	12	14	16	18	20
Concentration of calibration solutions, mg/l P	1,00	2,00	3,00	4,00	5,00	6,00	7,00	8,00	9,00	10,0

Prepare the calibration solutions immediately before use.

5.17 Standards for verifying hydrolysis and digestion efficiency.

5.17.1 Potassium pyrophosphate stock solution, ρ = 100 mg/l P.

Dissolve 533 mg ± 3 mg of potassium pyrophosphate (5.8) in about 800 ml of water (5.1) and dilute to 1 000 ml. Store in a sealed glass container at 4 °C ± 2 °C.

The solution is stable for 6 months.

5.17.2 Potassium pyrophosphate solution I, to check hydrolysis efficiency, ρ = 0,50 mg/l P, for the total-P working range II (0,10 mg/l to 1,00 mg/l P).

Dilute 0,5 ml of potassium pyrophosphate stock solution (5.17.1) and 100 µl of sulfuric acid (II) (5.2.2) to 100 ml with water (5.1).

The solution is stable for 1 month if stored at 4 °C ± 2 °C.

5.17.3 Potassium pyrophosphate solution II, to check hydrolysis efficiency, ρ = 5,00 mg/l P, for the total-P working range I (1,00 mg/l to 10,0 mg/l P).

Dilute 5 ml of potassium pyrophosphate stock solution (5.17.1) and 100 µl of sulfuric acid (II) (5.2.2) to 100 ml with water (5.1).

The solution is stable for 1 month if stored at 4 °C ± 2 °C.

5.17.4 Organophosphorus stock solution, ρ = 100 mg/l P.

Dissolve 856 mg ± 4 mg of pyridoxal-5-phosphate monohydrate (5.9.1) in about 800 ml of water (5.1) and dilute to 1 000 ml.

The solution is stable for 6 months if stored in a tightly closed glass container at 4 °C ± 2 °C.

Alternatively:

Dissolve 704 mg ± 3 mg of disodium phenylphosphate (5.9.2) in about 800 ml of water (5.1), acidify with sulfuric acid II (5.2.2) to pH ≈ 2 and dilute to 1 000 ml with water (5.1).

The solution is stable for 3 months if stored in the dark at 4 °C ± 2 °C.

5.17.5 Organophosphorus solution I, to check the digestion efficiency, ρ = 0,50 mg/l P for the total P working range II (0,10 mg/l to 1,00 mg/l P).

Dilute 0,5 ml of organophosphorus stock solution (5.17.4) and 100 µl of sulfuric acid (II) (5.2.2) to 100 ml with water (5.1).

The solution is stable for 1 month at 4 °C ± 2 °C.