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Water quality — Determination of phenol index by flow analysis (FIA and CFA)

Qualité de l'eau — Détermination de l'indice phénol par analyse en flux (FIA et CFA)

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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 3.

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.

International Standard ISO 14402 was prepared by Technical Committee ISO/TC 147, *Water quality*, Subcommittee SC 2, *Physical, chemical biochemical methods*.

Annex A of this International Standard is for information only.

Introduction

Methods for determination of water quality using flow analysis and automatic wet chemical procedures are particularly suitable for the processing of large sample series at a high analysis frequency.

Differentiation is needed between flow injection analysis (FIA) [1, 2] and continuous flow analysis (CFA) [3]. Both methods include automatic dosage of the sample into a flow system (manifold) where the analytes in the sample react with the reagent solutions on their way through the manifold. The sample preparation may be integrated in the manifold. The reaction product is measured in a flow detector.

Phenol index is an analytical convention. It represents a group of aromatic compounds which under the specific reaction conditions form coloured condensation products. The analytical result is expressed in terms of phenol concentration.

This International Standard describes two methods: the determination of phenol index (without distillation) after extraction, and the determination of phenol index (without extraction) after distillation.

It should be investigated whether and to what extent particular problems will require the specification of additional marginal conditions.

Water quality — Determination of phenol index by flow analysis (FIA and CFA)

1 Scope

This International Standard specifies two methods for the determination of the phenol index in waters of different origin (such as ground waters, surface waters, seep waters, and waste waters) in mass concentrations of 0,01 mg/l to 1 mg/l (in the undiluted sample). In particular cases, the range of application may be adapted by varying the operating conditions. Clause 3 describes the determination of phenol index (without distillation) after extraction, and in clause 4 the determination of phenol index (without extraction) after distillation is given.

2 Normative references

The following normative documents contain provisions which, through reference in this text, constitute provisions of this International Standard. For dated references, subsequent amendments to, or revisions of, any of these publications do not apply. However, parties to agreements based on this International Standard are encouraged to investigate the possibility of applying the most recent editions of the normative documents indicated below. For undated references, the latest edition of the normative document referred to applies. Members of ISO and IEC maintain registers of currently valid International Standards.

ISO 3696:1987, *Water for analytical laboratory use — Specification and test methods.*

ISO 5667-3:1994, *Water quality — Sampling — Part 3: Guidance on sample handling and preservation.*

ISO 6439:1990, *Water quality — Determination of phenol index — 4-Aminoantipyrine spectrometric methods after distillation.*

3 Determination of phenol index (without distillation) after extraction

3.1 Principle

The sample is fed into a continuously flowing carrier stream and mixed with also continuously flowing solutions of 4-aminoantipyrine and potassium peroxodisulfate. Phenolic compounds in the sample are oxidized by potassium peroxodisulfate, and the resulting quinones react with 4-aminoantipyrine, forming coloured condensation products. These are extracted in a flow extraction unit from the aqueous phase into chloroform. The chloroform phase is separated by a suitable phase separator (e.g. a hydrophobic semipermeable membrane), and the absorbance of the organic phase is measured spectrometrically in a flow spectrometer at 470 nm to 475 nm. More information on this analytical technique is given in the references [6 to 9].

It is absolutely essential that the test described in this International Standard be carried out by suitably qualified staff.

3.2 Interferences

3.2.1 Chemical interferences

Under the prevailing reaction conditions, aromatic amines will also form condensation products with 4-aminoantipyrine, leading to positive bias.

Interferences can occur when the sample, after the addition of the reagent solutions, does not reach a pH of 10,0 to 10,5. In particular this may occur in the cases of strongly acidic, strongly alkaline and buffered samples. In these cases, the sample is adjusted to a pH between 5 and 7 prior to addition of the reagent solutions.

Further information on interferences is given in [5].

3.2.2 Physical interferences arising from applying CFA and FIA

If the samples contain particulate matter, refer to 3.5 (last paragraph). Turbid samples do not cause interferences with the determination. In the event of coloured samples, check whether the colour can be extracted with chloroform, and determine the sample blank without the addition of reagents R1 and R2. The difference in response between the two measurements shall be taken into account with the evaluation (according to 3.7).

The interlaboratory trial (see clause 6 and annex A) has shown that detergents in waste water can strongly influence the determination, because the foam produced in the flow system can disturb on the one hand the steam distillation of volatile phenols (phenol index after distillation, see clause 4, and on the other hand the phase segmentation and phase separation procedures (phenol index after extraction, see clause 3). In general such interferences can easily be discovered.

In the case of significant detergent content, this International Standard is only applicable for phenol mass concentrations above 0,1 mg/l.

3.3 Reagents

Use only reagents of recognized analytical grade quality. The reagent blank value shall regularly be checked (see 3.6.3). The solutions used for the flow system shall be degassed. If not stated otherwise, it is recommended to degas the solutions under reduced pressure, because by this procedure the solutions are simultaneously purified.

WARNING — Phenol is toxic and can easily be absorbed through the skin. Chloroform is toxic and cancerogenic. Waste containing these substances should be disposed of appropriately.

3.3.1 Water, of grade 1 in accordance with ISO 3696

3.3.2 Potassium hydroxide, KOH

3.3.3 Sodium hydrogencarbonate, $NaHCO_3$

3.3.4 4-aminoantipyrine (4-amino-2,3-dimethyl-1-phenyl-3-pyrazolin-5-one), $C_{11}H_{13}N_3O$

3.3.5 Potassium peroxodisulfate, $K_2S_2O_8$

3.3.6 Phenol, C_6H_5OH

3.3.7 Boric acid, H_3BO_3

3.3.8 Ethanol, C_2H_5OH, 96 % mass fraction

3.3.9 2-Propanol, C_3H_7OH, 100 % mass fraction

3.3.10 Sulfuric acid, $\rho(H_2SO_4)$ = 1,84 g/ml

3.3.11 Hydrochloric acid, HCl, 50 % mass fraction

3.3.12 Potassium hydroxide solution, c(KOH) = 1,0 mol/l

3.3.13 Buffer solution

Dissolve in a 1 000 ml graduated flask in approximately 500 ml of water (3.3.1): 23 g of sodium hydrogencarbonate (3.3.3), 27 g of boric acid (3.3.7), and 35 g of potassium hydroxide (3.3.2) and make up to volume with water.

The pH of the buffer solution is approximately 10,3. The solution is stable for 1 month.

3.3.14 Carrier solution (symbol C in Figure 1)

Use water (3.3.1) degassed under reduced pressure.

3.3.15 4-Aminoantipyrine solution I (symbol R1 in Figures 1 and 2)

Dissolve in a 100 ml graduated flask 0,5 g of 4-aminoantipyrine (3.3.4) in approximately 50 ml of buffer solution (3.3.13), and make up to volume with buffer solution (3.3.13).

Degas the solution, e.g. by membrane filtration.

Prepare fresh solution every day.

3.3.16 Potassium peroxodisulfate solution (symbol R2 in Figures 1 and 2)

Dissolve in a 100 ml graduated flask 5 g of potassium peroxodisulfate (3.3.5) in approximately 90 ml of water (3.3.1), adjust to pH 11 with potassium hydroxide solution (3.3.12) and make up to volume with water.

Degas the solution, e.g. by membrane filtration.

Prepare fresh solution daily.

3.3.17 Chloroform, $CHCl_3$ (symbol Org in Figures 1 and 2)

Degas the chloroform solution either by membrane filtration or for 3 min in an ultrasonic bath.

3.3.18 Phenol stock solution, ρ = 1 000 mg/l

Dissolve in a 1 000 ml graduated flask 1,000 g of phenol (3.3.6) in water (3.3.1) and make up to volume with water. Use only colourless phenol crystals.

The cooled solution (2 °C to 5 °C) is stable for one month.

3.3.19 Phenol standard solution I, ρ = 10 mg/l

Pipette 1 ml of the stock solution (3.3.18) into a 100 ml graduated flask, and make up to volume with water (3.3.1).

The cooled solution (2 °C to 5 °C) is stable for one week.

3.3.20 Phenol standard solution II, ρ = 1 mg/l

Pipette 10 ml of the standard solution I (3.3.19) into a 100 ml graduated flask, and make up to volume with water (3.3.1).

The cooled solution (2 °C to 5 °C) is stable for one week.

3.3.21 Calibration solutions

Prepare the calibration solutions according to the origin of the sample and the expected concentrations by diluting the phenol standard solution I or II respectively (3.3.19 or 3.3.20).

Prepare a minimum of at least five calibration solutions per working range.

Proceed as follows for working ranges I and II, if using e.g. six calibration solutions:

a) Working range I, (0,1 mg/l to 1 mg/l):

Into each of a series of 100 ml graduated flasks pipette 1 ml, 3 ml, 5 ml, 6 ml, 8 ml and 10 ml respectively of the standard solution I (3.3.19), and make up to volume with water (3.3.1).

The concentration of phenol in these calibration solutions is 0,1 mg/l, 0,3 mg/l, 0,5 mg/l, 0,6 mg/l, 0,8 mg/l and 1,0 mg/l, respectively.

b) Working range II (0,01 mg/l to 0,1 mg/l):

Into each of a series of 100 ml graduated flasks pipette 1 ml, 3 ml, 5 ml, 6 ml, 8 ml, and 10 ml respectively of the standard solution II (3.3.20), and make up to volume with water (3.3.1).

The concentration of phenol in these calibration solutions is 0,01 mg/l, 0,03 mg/l, 0,05 mg/l, 0,06 mg/l, 0,08 mg/l and 0,1 mg/l, respectively.

Prepare fresh calibration solutions each day.

3.4 Apparatus

3.4.1 Flow injection analysis system (FIA)

The flow injection system (FIA) shall comprise the following components (see Figure 1):

a) reagent reservoirs;

b) low pulsation pump with specific pump tubing, for flowrates as shown in Figure 1, as an example;

c) displacement bottle for the feeding of the chloroform;

d) sample injector with suitable injection volumes;

e) extraction cell with phase segmentor and phase separator (e.g. hydrophobic semipermeable membrane of PTFE).

EXAMPLES Membrane thickness: 150 µm to 200 µm; pore size: 0,5 µm to 2 µm; porosity: 75 %.

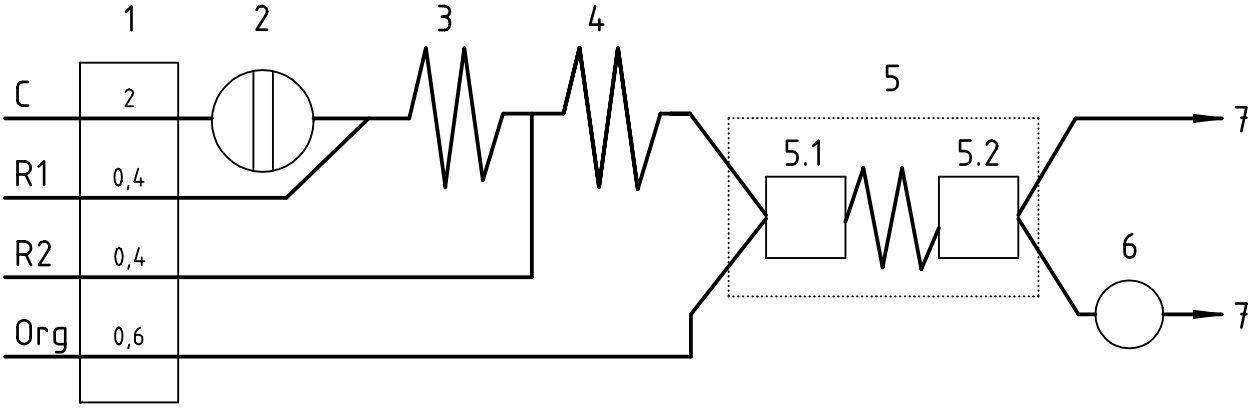
f) transport tubes and reaction coils, internal diameter 0,5 mm to 0,8 mm, tube connections and T-connections of chemically inert plastic, with a minimal dead volume;

g) spectrometric detector with flow cell, of optical path length 10 mm, wavelength 470 nm to 475 nm.

h) recording unit (e.g. strip chart recorder, integrator or printer/plotter).

NOTE In general, peak heights are measured.

i) autosampler, if required.



Key

C: Carrier solution (3.3.14)
R1: 4-aminoantipyrine solution I (3.3.15)
R2: Potassium peroxodisulfate solution (3.3.16)
Org: Chloroform (3.3.17)
1 Pump (flowrates in ml/min)
2 Injector
600 µl [working range 0,01 to 0,1 mg/l phenol]
200 µl [working range 0,1 to 1,0 mg/l phenol]
3 Reaction coil: 60 cm/∅ int. 0,5 mm
4 Reaction coil: 80 cm/∅ int. 0,5 mm
5 Extraction unit: 160 cm/∅ int. 0,7 mm
5.1 Phase segmentor,
5.2 Phase separator
6 Detector: optical pathlength: 1 cm, wavelength: 470 nm to 475 nm
7 Waste

Figure 1 — Example of a flow injection system for the determination of 0,01 mg/l to 1,0 mg/l phenol index without distillation and with extraction (according to 3.4.1)

3.4.2 Continuous flow analysis (CFA)

The continuous flow analysis system shall comprise the following components (see Figure 2):

a) autosampler allowing a reproducible introduction of the sample or of the carrier liquid;

b) reagent reservoirs;

c) low pulsation pump with specific, chemically inert pump tubes, with flowrates as shown in Figure 2, as an example;

d) displacement bottle for the feeding of the chloroform, if required;

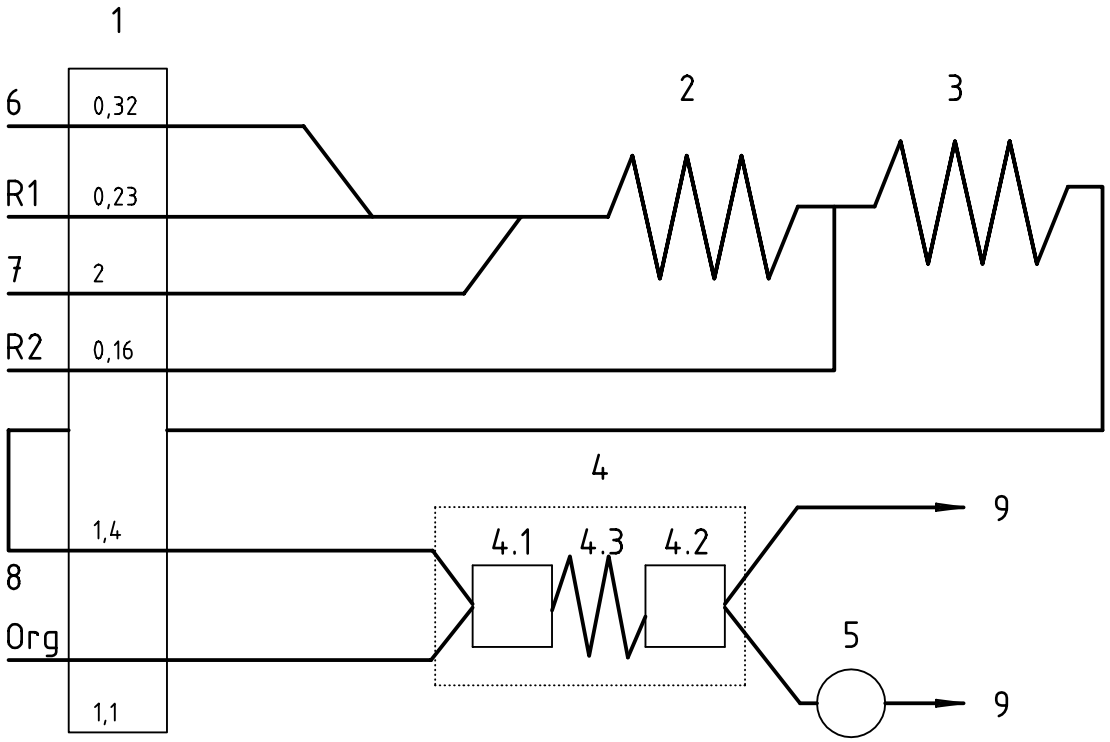
e) manifold with highly reproducible gas bubble, sample, and reagent introduction, with appropriate transport systems and extraction systems, and connection assemblies, e.g. of glass, chemically inert plastics or metal, and with appropriate separator for the separation of the organic phase from the aqueous phase;

f) spectrometric detector with flow cell, optical pathlength 0,5 cm to 5 cm, wavelength 470 nm to 475 nm

g) recording unit (e. g. strip chart recorder, integrator or printer/plotter).

NOTE 1 In general, peak heights are measured.

NOTE 2 A CFA system with an internal diameter of 2 mm is described in Figure 2. Other internal diameters (e.g. approximately 1 mm) may also be used.



Key

R1	4-aminoantipyrine solution I (3.3.15)	4.2	Phase separator
R2	Potassium peroxodisulfate solution (3.3.16)	4.3	Reaction coil: 150cm/∅ int. 2 mm
Org	Chloroform (3.3.17)	5	Detector: optical pathlength: 0,5 cm to 5 cm, wavelength: 470 nm to 475 nm
1	Pump (flowrates in ml/min)	6	Segmentation gas (air)
2	Reaction coil: 40 cm/∅ int. 2 mm	7	Sample
3	Reaction coil: 40 cm/∅ int. 2 mm	8	Resample
4	Extraction unit	9	Waste
4.1	Phase segmentor		

Figure 2 — Example of a continuous flow system for the determination of 0,01 mg/l to 1,0 mg/l of phenol index without distillation and with extraction (according to 3.4.2)

3.4.3 Additional apparatus

a) **Graduated flasks**, 100 ml and 1 000 ml;

b) **Graduated pipettes**, 1 ml to 10 ml;

c) **Membrane filter assembly** with membrane filters, pore size 0,45 µm;

d) **pH measuring device** (e. g. pH electrode).

3.5 Sampling

Use glass or polytetrafluoroethylene (PTFE) containers for sampling.

Prior to use, rinse all containers and devices with which the sample may come into contact, with sulfuric acid of pH approximately 2.

Analyse the samples immediately after their collection. Alternatively, adjust to a pH of approximately 2 with sulfuric acid (3.3.10 or diluted solution) or hydrochloric acid (3.3.11 or diluted solution), store in the dark at a temperature of 2 °C to 5 °C, and analyse within 24 h.

In exceptional cases, after acidification and membrane (pressure) filtration of the sample, a storage of up to two weeks is possible. The applicability of this preservation method shall be checked for the individual case of examination. For more information on sample preservation, see ISO 5667-3 and [10].

Filtration of the sample prior to measurement is necessary, if there is a risk of clogging the transport tubes.

3.6 Procedure

3.6.1 Preparation for measurement

Prior to measurement, continuously run the reagent solutions C (3.3.14), R1 (3.3.15), R2 (3.3.16) and Org (3.3.17) through the flow analysis system, wait for the baseline to stabilize, and zero the baseline.

Consider the system is ready to operate, when the baseline remains stable (no drift). A satisfying signal-to-noise ratio should be obtained.

Verify that a signal-to-noise ratio is obtained that has no significant effect on the results.

The most frequent reasons for a poor signal-to-noise ratio are defective separator membranes or traces of water at the walls of the cell. Traces of water adhering to the walls of the cell can be removed by rinsing the cell with ethanol (3.3.8) or 2-propanol (3.3.9).

Monitor the blank of the reagent and control the membrane function as described in 3.6.3. Carry out the calibration according to 3.6.4.

3.6.2 Checking of the flow system

With the measuring system adjusted to working range II, using a calibration solution (3.3.21) with a concentration of 0,05 mg/l, an absorbance per 1 cm cell length of at least 0,01 cm^{-1} shall be obtained. Otherwise the flow system is not suitable, and it shall be replaced by a system which fulfils this requirement.

NOTE If the photometric detector (3.4.1, 3.4.2) is not designed for measurement of absorbance values, the absorbance can be determined by an external photometer designed to measure absorbance values.

3.6.3 Checking of the reagent blank

Wait for the baseline to stabilize.

In place of the reagent solutions R1 (3.3.15) and R2 (3.3.16), run water through the system until a stable signal is obtained. Record the change in the absorbance.

If the absorbance (per centimetre cell length) decreases by more than 0,05 cm^{-1}, it can be assumed that self-condensation products have been formed. In this case the preparation of the solutions, the checking of the flow system (see 3.6.2.) and the monitoring of the reagent blank (see 3.6.3) shall be repeated.

Subsequently, transport reagent solutions R1 (3.3.15) and R2 (3.3.16) again.

3.6.4 Calibration

Select working range I or II as appropriate, and prepare the calibration solutions for the working range selected. Carry out a separate calibration for each working range.

Use for working range I and FIA (3.4.1), for example, an injection volume of 200 µl, and for working range II, for example, an injection volume of 600 µl.

For the working ranges I and II and CFA (3.4.2), choose the cell length and the flowrate to obtain the highest possible response for the calibration solution of the highest concentration.