SLOVENSKI STANDARD
SIST EN 13368-2:2002

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Gnojila - Določ evanje sredstev za kelatiziranje v gnojilih z ionsko kromatografijo - 2. del: EDDHA in EDDHMA

Fertilizers - Determination of chelating agents in fertilizers by ion chromatography - Part 2: EDDHA and EDDHMA

Düngemittel - Bestimmung von Chelatbildnern in Düngemitteln durch Ionenchromatographie - Teil 2: EDDHA und EDDHMA

Engrais - Détermination des agents chélatants dans les engrais par chromatographie ionique - Partie 2: EDDHA et EDDHMA

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65.080	Gnojila	Fertilizers

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EUROPEAN STANDARD

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English version

Fertilizers - Determination of chelating agents in fertilizers by ion chromatography - Part 2: EDDHA and EDDHMA

Engrais - Détermination des agents chélatants dans les engrais par chromatographie ionique - Partie 2: EDDHA et EDDHMA

Düngemittel - Bestimmung von Chelatbildnern in Düngemitteln durch Ionenchromatographie - Teil 2: EDDHA und EDDHMA

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EUROPEAN COMMITTEE FOR STANDARDIZATION
COMITÉ EUROPÉEN DE NORMALISATION
EUROPÄISCHES KOMITEE FÜR NORMUNG

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Content

Foreword

This European Standard has been prepared by Technical Committee CEN/TC 260 "Fertilizers and liming materials", the secretariat of which is held by DIN.

This European Standard shall be given the status of a national standard, either by publication of an identical text or by endorsement, at the latest by July 2001, and conflicting national standards shall be withdrawn at the latest by July 2001.

EN 13368 consists of two parts dealing with the quantitative determination of chelating agents in fertilizers by ion chromatography :

- Part 1 : EDTA, HEDTA and DTPA

- Part 2 : EDDHA and EDDHMA

The annexes A and B are informative.

According to the CEN/CENELEC Internal Regulations, the national standards organizations of the following countries are bound to implement this European Standard: Austria, Belgium, Czech Republic, Denmark, Finland, France, Germany, Greece, Iceland, Ireland, Italy, Luxembourg, Netherlands, Norway, Portugal, Spain, Sweden, Switzerland and the United Kingdom.

1 Scope

This method describes the procedure for the ion chromatographic determination of the total amount of each of the individual ortho-ortho isomer of the chelating agents EDDHA and EDDHMA in fertilizers containing one or both of these substances. The method allows the identification and the determination of the total water soluble fraction of each of these chelating agents. It does not allow to distinguish between the free form and the metal bound form of the chelating agents.

NOTE 1 EDDHA and EDDHMA are abbreviations used in this standard for the sake of simplicity. For complete names see annex A.

NOTE 2 The substances EDDHA and EDDHMA exist under several different isomeric forms. Positional isomers (ortho, meta, and para forms) as well as stereo isomers (meso and dl forms) are known.

This method applies to fertilizers containing chelates of one or more of the following micro-nutrients: cobalt, copper, iron, manganese, zinc and with a mass fraction of at least 0,1 %.

NOTE 3 At present, analytically pure standards only exist for ortho-ortho EDDHA. All other substances being unavailable as a standard, the influence of their eventual presence in the samples (with respect to the sensitivity and the selectivity of this method) has not been studied. For EDDHMA, the method has been developed with samples of iron EDDHMA of uncertain purity and composition, any standard product being unavailable.

NOTE 4 In the chromatographic conditions of this method, the stereo isomers of EDDHA and EDDHMA are eluting together as a single peak. Using an eluent with a lower exchange strength, the meso and the dl forms of EDDHA and EDDHMA can be separated.

2 Normative references

This European Standard incorporates by dated or undated reference, provisions from other publications. These normative references are cited at the appropriate places in the text and the publications are listed hereafter. For dated references, subsequent amendments to or revisions of any of these publications apply to this European Standard only when incorporated in it by amendment or revision. For undated references the latest edition of the publication referred to applies (including amendments).

EN 1482, *Sampling of solid fertilizers and liming materials*

EN ISO 3696, *Water for analytical laboratory use – Specification and test methods (ISO 3696 : 1987)*

3 Principle

The micro-nutrients associated with the chelating agents present in an aqueous extract of the sample are replaced by iron (III). The iron chelates are separated and determined by ion chromatography. The separation is based on anion exchange, by elution with an acidified sulfate solution. The detection is based on photometry at 520 nm.

4 Interferences

Some substances can interfere, to a degree largely dependent on the type of column used. With the column described in 6.2, the following phenomena have been observed.

a) Injection of solutions having high concentrations of salts can cause shifts in the retention times, mostly decreasing the retention when compared to the standard solutions. In these cases, the identity of the peaks can be confirmed by standard addition.

b) High concentrations of chloride, nitrate, carbonate, and sulfate do not interfere. On the other hand, the resolution and the sensitivity of the chelating agents are severely depressed in the presence of high amounts of phosphate.

c) Compounds, related to the group of polyamino polycarboxylic acids, do not interfere. No signals are detected for EDTA, HEDTA, NTA, CDTA, and EDDHSA. DTPA gives a weak non-interfering signal close to the void volume.

NOTE EDTA, HEDTA, NTA, CDTA, EDDHSA and DTPA are abbreviations used in this standard for sake of simplicity. For complete names see annex A.

d) No signals have been detected for the following complexing agents : citrate, oxalate, tartrate, phthalate, and 20 naturally occuring amino acids.

5 Reagents

5.1 General

a) all reagents should be of recognized analytical grade ;

b) all water should conform to EN ISO 3696 and be degassed by boiling before use.

5.2 Nitric acid, $c(HNO_3)$ = 7,2 mol/l

Nitric acid, diluted 1 + 1 with water.

5.3 Sodium hydroxide solution, $c(NaOH)$ = 0,5 mol/l

Dissolve 20 g of NaOH in pellet form in a 1 litre volumetric flask with water. Dilute to the mark and homogenize.

5.4 Sulfuric acid solution, $c(H_2S0_4)$ = 0,1 mol/l

Dilute 2,75 ml of sulfuric acid (96 % H_2S0_4 , ρ = 1,84 g/ml) to 500 ml with water.

5.5 EDDHA solution, c(EDDHA) = 2 mmol/l

Dissolve 72 mg of ethylenediamine-di(o-hydroxyphenyl)acetic acid (EDDHA) in 2 ml of NaOH (5.3) in a 30 ml beaker. After dissolution, transfer quantitatively into a 100 ml volumetric flask, dilute to the mark with water and homogenize. Prepare the solution immediately before use.

5.6 EDDHMA solution, c(EDDHMA) = 2 mmol/l

Dissolve 78 mg of ethylenediamine-di(o-hydroxy-p-methylphenyl)acetic acid (EDDHMA) in 2 ml of NaOH (5.3) in a 30 ml beaker. After dissolution, transfer quantitatively into a 100 ml volumetric flask, dilute to the mark with water and homogenize. Prepare the solution immediately before use.

NOTE In the absence of a pure standard product, a solution of maximally 2 mmol/l can be prepared by dissolving 88 mg of an iron EDDHMA sample in 100 ml of water. For a sample of 100 % pure Fe-EDDHMA, a concentration of exactly 2 mmol/m is obtained.

5.7 EDDHA/EDDHMA standard mixtures

Into a set of 3 volumetric flasks of 100 ml, pipette respectively 2,5 ml, 5 ml and 10 ml of each solution of 2 mmol/l (5.5 and 5.6). Dilute to the mark with water and homogenize. These solutions contain a mixture of EDDHA and EDDHMA, in concentrations of respectively 0,05 mmol/l, 0,1 mmol/l and 0,2 mmol/l. Prepare the solution immediately before use.

5.8 Eluent sulfate (50 mmol/l), acidified

Dissolve 7,10 g of anhydrous sodium sulfate (Na_2S0_4) in a mixture of 800 ml of water and 9 ml of sulfuric acid solution (5.4) in a 1 litre volumetric flask. Dilute to the mark with water and homogenize. Adjust the pH of the eluent to 3,15 ± 0,20. Before use, filter the solution through a 0,45 µm membrane filter (6.3).

5.9 Iron (III) nitrate solution

Dissolve 5 g of ferric nitrate nonahydrate ($Fe(NO_3)_3 \cdot 9H_2O$) in a mixture of 800 ml of water and 21 ml of nitric acid (5.2) in a 1 litre volumetric flask. Dilute to the mark with water and homogenize. Store in a plastics bottle.

NOTE Nitric acid is added in order to stabilize the solution and to allow the complete replacement of other chelated micro-nutrients by iron (III).

6 Apparatus

Usual laboratory equipment, glassware, and :

6.1 Rotary shaker

Rotary shaker capable of operating at a rotational speed of about 35 min^{-1} to 45 min^{-1}.

6.2 Ion chromatograph

Ion chromatograph equipped with :

a) an isocratic pump delivering the eluent (5.8) at a flow rate of 1 ml/min ;

b) an injection valve with an injection loop of about 50 µl.

NOTE 1 An automatic injection valve can malfunction, and its use is not recommended because the column, described in item c), is a short guard column creating a low backpressure.

c) an anion exchange separator column with reduced length 10 µm particles – 2 % substrate *x* – linking – 100 meq capacity (per column) – alkyl quaternary ammonium – medium high (hydrophobicity)[1] ;

NOTE 2 The column mentioned in c) is not exclusive. Any anion exchange column with comparable capacity, hydrophobicity, and selectivity can probably be used ;

NOTE 3 columns can, in their native state, exhibit some sensitivity towards various anions (e.g. chloride, nitrate, sulfate, phosphate) which can disappear after treatment with excess iron. It can be necessary to precondition the column prior to its use by repeated injections, at least 10, of the iron (III) solution (5.9), followed by equilibration under eluent flow for at least 6 h ;

NOTE 4 after many injections, especially of products having complex matrices, the column may lose some of its capacity and its separating efficiency. Substances like humic acids can be strongly adsorbed on the column particles. Suspended and colloidal matter can block the column entrance and disturb the eluent flow. The column manufacturer cleaning instructions are consulted for the suitable cleaning procedure. Generally the column efficiency can be restored by rinsing the column in the reverse direction with 0,5 mol/l of sodium hydroxide, then with water, then with 0,5 mol/l nitric acid, and finally with the eluent for equilibration ;

NOTE 5 the injection of undissolved matter severely decreases the lifetime of the column. Solutions should always be allowed to equilibrate, and then filter through a 0,2 µm membrane filter before injection ;

d) a UV/VIS detector with a 520 nm filter, output range set at an absorbance of 0,1 ;

e) an integrator.

6.3 Membrane filters

Micromembrane filters resistent to aqueous solutions, with porosity of respectively 0,45 µm and 0,2 µm.

7 Preparation of the sample

Prepare the sample according to EN 1482.

NOTE 1 Sample may also be prepared according to method 1 (see [1] of bibliography).

NOTE 2 For the size reduction of samples with a high amount of chelating agents, it is not recommended to use a high speed laboratory mill. It is more convenient to grind the sample in a mortar to a particle size less than 1 mm.

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[1] IonPac AG 7 guard column, serving as a separator, from Dionex Co, Sunnyvale, CA USA, or equivalent are examples of suitable products available commercially. This information is given for the convenience of users of this European Standard and does not constitute an endorsement by CEN of these products.

8 Procedure

8.1 Preparation of the sample solution

Weigh 5 g of the sample (m), to within 1 mg, into a 250 ml volumetric flask (V_0). Add 200 ml of water, and put on the rotary shaker (6.1) for 1 h. Dilute to the mark with water, homogenize, and filter through a paper filter. If necessary, dilute the filtrate with water, in order to obtain a concentration of chelating agent between 0,02 mmol/l and 0,2 mmol/l. Let D be the dilution factor. Pipette 20 ml of the solution into a 100 ml beaker. Add 5 ml of the iron (III) solution (5.9), homogenize, and allow to stand for 15 min.

NOTE The addition of iron may cause precipitation, especially if phosphate is present in the sample solution. For this reason, the precipitate should always be allowed to settle and the solution to equilibrate for 15 min.

8.2 Preparation of the standard solutions

Pipette 20 ml of each of the EDDHA/EDDHMA standard mixtures of respectively 0,05 mmol/l, 0,1 mmol/l and 0,2 mmol/l (5.7) into a set of 100 ml beakers. Add 5 ml of the iron (III) solution (5.9), homogenize, and allow to stand for 15 min.

8.3 Ion chromatographic analysis

Immediately before injection, all solutions shall be filtered through a 0,2 µm membrane filter (6.3). Inject the standard solutions (8.2) into the chromatographic system (6.2). The retention times of the iron chelates are in the order EDDHA < EDDHMA. Adjust the attenuation on the integrator, in order to obtain about 75 % of the output range for the EDDHA peak from the most concentrated standard solution. See typical chromatogram in annex B. Measure the retention times and the peak areas for the two chelating agents. Inject the sample solution (8.1). After elution, identify the chelating agent by the retention time, and measure the corresponding peak area. For each appropriate chelating agent, draw a calibration graph with the values of the peak area of the standard solutions versus the corresponding chelating agent concentration (C_s) (mmol/l). Determine the concentration of the chelating agent in the sample solution (mmol/l), using the corresponding calibration graph.

9 Expression of results

The mass fraction in percent of the chelating agent (EDDHA or EDDHMA), expressed as free acid, in the fertilizer is equal to :

$$\text{Chelating agent} = \frac{C_s \cdot D \cdot M_w \cdot V_0}{10^4 \cdot m} \quad \text{in \%} \qquad (1)$$

where :

C_s is the concentration of the chelating agent in the sample solution (mmol/l), determined with the calibration graph (8.3) ;

D is the dilution factor (8.1) ;

M_w is the molar mass in grams per mol of the chelating agent in the acid form, being for :

EDDHA : 360

EDDHMA : 388

V_0 is the total volume of the extract (8.1), in millilitres ;

m is the mass of the sample taken for extraction, in grams.