SLOVENSKI STANDARD
SIST EN 12396-2:1999

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Živila brez maščob - Določevanje ostankov ditiokarbamatov in tiuram-disulfidov - 2. del: Plinskokromatografska metoda

Non-fatty foods - Determination of dithiocarbamate and thiuram disulfide residues - Part 2: Gas chromatographic method

Fettarme Lebensmittel - Bestimmung von Dithiocarbamat- und Thiuramdisulfid-Rückständen - Teil 2: Gaschromatographisches Verfahren

Aliments non gras - Détermination des résidus de dithiocarbamates et de bisulfures de thiurame - Partie 2: Méthode par chromatographie en phase gazeuse

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67.050	Splošne preskusne in analizne metode za živilske proizvode	General methods of tests and analysis for food products

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Non-fatty foods - Determination of dithiocarbamate and thiuram disulfide residues - Part 2: Gas chromatographic method

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Fettarme Lebensmittel - Bestimmung von Dithiocarbamat- und Thiuramdisulfid-Rückständen - Teil 2: Gaschromatographisches Verfahren

This European Standard was approved by CEN on 2 October 1998.

CEN members are bound to comply with the CEN/CENELEC Internal Regulations which stipulate the conditions for giving this European Standard the status of a national standard without any alteration. Up-to-date lists and bibliographical references concerning such national standards may be obtained on application to the Central Secretariat or to any CEN member.

This European Standard exists in three official versions (English, French, German). A version in any other language made by translation under the responsibility of a CEN member into its own language and notified to the Central Secretariat has the same status as the official versions.

CEN members are the national standards bodies of Austria, Belgium, Czech Republic, Denmark, Finland, France, Germany, Greece, Iceland, Ireland, Italy, Luxembourg, Netherlands, Norway, Portugal, Spain, Sweden, Switzerland and United Kingdom.

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

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Foreword

This European Standard has been prepared by Technical Committee CEN/TC 275 "Food analysis - Horizontal methods", 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 April 1999, and conflicting national standards shall be withdrawn at the latest by April 1999.

This European Standard EN 12396 „Non-fatty foods - Determination of dithiocarbamate and thiuram disulfide residues“ consists of three parts:

Part 1: Spectrometric method

Part 2: Gas chromatographic method

Part 3: Xanthogenate method

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 European Standard specifies a gas chromatographic method for the determination of residues of dithiocarbamates and thiuram disulfides, which release carbon disulfide under the described conditions (e. g. mancozeb, maneb, propineb, thiram, zineb). It is applicable to such compounds in and on fruits and some vegetables but also in and on cereals and other foodstuffs of plant origin.

Only the quantification of the whole group is possible using this method not the identification of individual compounds. Generally the maximum residue limits (MRLs) are expressed in terms of carbon disulfide.

2 Normative references

This European Standard incorporates, by dated and undated reference, provisions from other publications. These normative references are cited at the appropriate places in the text and the publications are listed here after. 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.

EN 12393-1:1998	Non-fatty foods - Multiresidue methods for the gas chromatographic determination of pesticide residues - Part 1: General considerations
EN 12393-3	Non-fatty foods - Mutiresidue methods for the gas chromatographic determination of pesticide residues - Part 3: Determination and confirmatory tests
EN 12396-1	Non-fatty foods - Determination of dithiocarbamate and thiuram disulfide residues - Part 1: Spectrometric method
ISO 1750	Pesticides and other agrochemicals - Common names

3 Principle

The sample is heated with hydrochloric acid and tin(II)chloride in a gas-tight flask to release carbon disulfide from any dithiocarbamates and/or thiuram disulfide present. The quantity of carbon disulfide collecting in the headspace of the flask is determined by gas chromatography (GC) with an electron capture detector (ECD) or with a flame-photometric detector (FPD) in the sulfur mode. For further information on the principle of this method, see [1] to [4].

4 Reagents

4.1 General

Unless otherwise stated, use reagents of recognized analytical grade, preferably for pesticide residue analysis, and only distilled or demineralized water.

Label all standard containers with name and purity of all pesticides. For the full chemical names and structures, see ISO 1750.

Take every precaution to avoid possible contamination of water, solvents, inorganic salts etc. by plastics and rubber materials or even by air. Use only glass containers for storage and handling of all water and reagents.

4.2 Acetone.

4.3 Light petroleum, boiling range 40 °C to 60 °C.

4.4 Carbon disulfide, colourless, at least 99 % of mass concentration. If stored at - 20 °C it is stable for 2 years to 3 years.

4.5 Hydrochloric acid, concentrated, $\rho_{20}(HCl) = 1{,}16$ g/ml.

4.6 Tin(II)chloride - hydrochloric acid solution, $\rho(SnCl_2 \cdot 2H_2O) = 15$ g/l[1]).

Dissolve 15 g of tin(II)chloride dihydrate in 400 ml of concentrated hydrochloric acid (4.5) and dilute to 1 000 ml with water.

4.7 Carbon disulfide stock solution, $\rho(CS_2) \approx 25$ mg/ml.

Weigh to the nearest 10 mg a stoppered 50 ml volumetric flask with ground glass neck containing 40 ml of acetone (4.2) or light petroleum (4.3). Pipette into this about 1 ml of carbon disulfide (4.4) (according to about 1,25 g), close the flask immediately and weigh again to the nearest 10 mg in order to determine the exact mass of carbon disulfide taken. Dilute to the mark with light petroleum or acetone and mix well. Prepare freshly for each calibration curve.

4.8 Carbon disulfide standard solutions

4.8.1 Carbon disulfide standard solution I, $\rho(CS_2) \approx 10$ mg/ml.

Pipette 20 ml of carbon disulfide stock solution (4.7) into a 50 ml volumetric flask and dilute to the mark with light petroleum (4.3) or acetone (4.2) as appropriate. Prepare freshly for each calibration curve.

4.8.2 Carbon disulfide standard solution II, $\rho(CS_2) \approx 1$ mg/ml.

Pipette 5 ml of carbon disulfide standard solution I (4.8.1) into a 50 ml volumetric flask and dilute to the mark with light petroleum (4.3) or acetone (4.2), as appropriate. Prepare freshly for each calibration curve.

5 Apparatus

5.1 General

Thoroughly clean glassware shall be used.

See 5.1 of EN 12393-1:1998 for the cleaning of glassware.

Usual laboratory equipment and, in particular, the following:

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[1] ρ is the mass concentration.

5.2 Digestion flask: 250 ml glass bottle with a screw cap which has been drilled with a 3 mm hole and fitted with a polytetrafluoroethylene (PTFE) septum.

5.3 Water bath, capable of being controlled at approximately 70 °C.

5.4 Gas-tight injection syringes, 100 µl and 1 000 µl. Check the gas tightness of the syringes regularly.

5.5 Gas chromatograph, equipped with an electron capture detector (ECD) or a flame-photometric detector (FPD) in the sulfur mode (filter 394 nm).

A suitable GC system, preferably equipped with separate heaters for injector, detector and column oven, should be used. See annex B.

Variation in the detector sensitivity should be checked periodically by verifying the calibration curve using carbon disulfide standard solutions.

For the determination of carbon disulfide, packed glass columns are mostly used. For stationary phases and support materials see EN 12393-3.

6 Sampling

Prepare the laboratory sample according to a generally recommended method of sampling to achieve a representative part of the product to be analysed.

NOTE: Sampling procedures for the official control of pesticide residues in and on fruits and vegetables are given in EEC directive 79/700/EEC [5].

7 Preparation of the samples

7.1 Test sample

If the sample reaches the laboratory frozen, store it at - 20 °C before analysis.

Where possible, carry out the analysis of fresh samples immediately after their arrival in the laboratory. Do not analyse a laboratory sample which is wholly or extensively spoiled.

For analysis take only the portion of the laboratory sample to which the maximum residue limit applies. No further plant-parts may be removed. A record of the plant-parts which have been removed shall be kept. The sample thus prepared is the analytical sample.

If the sample cannot be analyzed immediately, store it at 0 °C to 5 °C for no longer than 2 days before analysis.

The reduction of the analytical sample shall be carried out in such a way that representative portions are obtained (e.g. by division into four and selection of opposite quadrants). When the samples are in small units (e.g. small fruits, vegetables, cereals), the analytical sample shall be thoroughly mixed before weighing out the test portion. When the samples are in larger units, take wedge-shaped sections (e.g. large fruits and vegetables) or cross sections (e.g. cucumbers) which include the outer surface from each unit.

NOTE: The residues of dithiocarbamate and thiuram disulfide, which are on the surface of the plant-parts and are not systemic, decompose rapidly especially in chopped samples. Therefore precautions should be taken to avoid decomposition.

If samples have to be stored for more than 2 days, they shall be deep-frozen at - 20 °C. To ensure that even after thawing representative samples can be taken, prepare portions of the product which are each sufficient for one analysis.

7.2 Test portion

Weigh out test portions of masses up to 200 g to an accuracy of ± 1 %. After weighing out the test portion, remove any parts which would interfere with the analytical procedure. In the case of stone fruits, the stones are removed after weighing out. The basis for the calculation of the residue mass fraction is the mass of the original test portion (with stones).

The test portion shall not be cut or reduced to smaller pieces than can just pass the neck of the reaction flask, as the residues of dithiocarbamate and thiuram disulfide fungicides fall the more the test portion is cut.

Analyse the test portion immediately after cutting.

8 Procedure

8.1 Safety aspects

WARNING: Many dithiocarbamates, thiuram disulfides and carbon disulfide are toxic by various routes of exposure, especially in concentrated form. When working with dithiocarbamates, thiuram disulfides and carbon disulfide, consult safety data sheets of the manufacturer for information.

Vapours from some volatile solvents are toxic. Several of these solvents can easily be absorbed through skin. Use effective fume hoods to remove vapours of these solvents as they are set free.

8.2 Preparation of blanks

Prepare reagent and commodity blanks. Spiked recovery tests at levels appropriate to the maximum residue limits should be carried out as specified in annex A and shall lead to satisfying results.

Test the purity of the reagents and solvents by performing reagent blanks. Test solvents by gas chromatography whether they are free from any interfering impurity whose concentration is at the limit of determination. Ensure that the laboratory is free from atmospheric contamination that would give a background response for carbon disulfide, by injecting a few hundred microlitres of air into the GC column. The chromatograph shall not show any interfering impurity.

NOTE 1: Analysts should thoroughly familiarize themselves with the method before starting the analyses.

NOTE 2: Some vegetables (e. g. those of the family *Cruciferae*) contain naturally occurring compounds which release carbon disulfide under the conditions described in this European Standard. Therefore the analysis of such vegetables can lead to false positive results.

8.3 Gas chromatographic operating conditions

Pure (oxygen-free) and dry (water-free) nitrogen (this is especially important when using an electron capture detector (ECD)), or an argon/methane mixture (in case of a pulsed ECD) shall be used as the carrier gas for packed columns. The flow rate depends on the size and the type of the column used. Molecular sieve filters shall be installed for all gas supplies and regenerated regularly.

Make sure that the GC conditions (column length, stationary phase type, injector, detector and column temperatures, gas flow rates, etc.) are such that the separation of the carbon disulfide likely to be present from interfering peaks originating from the samples is as complete as possible.

> NOTE: Capillary GC has become an important technique with a separation power superior to that of packed columns. This capillary technique is recommended especially in the case of complex substrates.

Typical GC operating conditions are presented in annex B.

8.4 Preparation of the calibration curve

Place 50 ml of water into each of four digestion flasks and add 100 ml of tin(II)chloride - hydrochloric acid solution (4.6). Fit the screw caps containing the PTFE septa. Inject through the different septa, into the water, aliquot portions of carbon disulfide standard solutions (4.8) to provide a suitable range of concentrations [e.g. 1 µl, 5 µl, 10 µl, 20 µl, 40 µl, 60 µl, 80 µl and 100 µl of 1 mg/ml solution (4.8.2) corresponding to 0,02 mg/kg; 0,1 mg/kg; 0,2 mg/kg; 0,4 mg/kg; 0,8 mg/kg; 1,2 mg/kg; 1,6 mg/kg and 2,0 mg/kg of residue as carbon disulfide in a 50 g sample]. Place the spiked digestion flasks in the water bath (5.3) for 2 h and follow the procedure described in 8.5.2.

Plot a calibration curve from the peak areas or peak heights against amounts of carbon disulfide added.

> NOTE: As the response from the flame photometric detector follows a square function, the chart recoder readings should be plotted as logarithmic to give linearity.

8.5 Sample measurement

8.5.1 Preparation of the digestion flask

Introduce a 50 g test portion into the digestion flask (5.2), add 100 ml of tin(II)chloride-hydrochloric acid solution (4.6) and immediately close the flask tightly with the PTFE septum and screw cap. Place the flask into the thermostatically controlled water bath (5.3) for 2 h. Shake vigorously the flask for 2 min at least 3 times during the digestion stage, in order to fully release carbon disulfide from any dithiocarbamate and thiuram disulfide present. Remove the flask and allow to cool to room temperature.