

Foodstuffs - Determination of vitamin E by high performance liquid chromatography - Measurement of α -, β -, γ - and δ -tocopherol

EESTI STANDARDI EESSÕNA

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English Version

Foodstuffs - Determination of vitamin E by high performance
liquid chromatography - Measurement of α -, β -, γ - and δ -
tocopherol

Produits alimentaires - Détermination de la teneur en
vitamine E par chromatographie liquide haute performance
- Dosage des α -, β -, γ - et δ -tocophérols

Lebensmittel - Bestimmung von Vitamin E mit
Hochleistungs-Flüssigchromatographie - Bestimmung von
 α -, β -, γ - und δ -Tocopherol

This European Standard was approved by CEN on 17 April 2014.

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Foreword

This document (EN 12822:2014) 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of an identical text or by endorsement, at the latest by December 2014 and conflicting national standards shall be withdrawn at the latest by December 2014.

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

This document supersedes EN 12822:2000.

Annexes A, B and C are informative.

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WARNING — The use of this standard can involve hazardous materials, operations and equipment. This standard does not purport to address all the safety problems associated with its use. It is the responsibility of the user of this standard to establish appropriate safety and health practices and determine the applicability of regulatory limitations prior to use.

Introduction

This European Standard provides the base for the analytical methods. It is intended to serve as a frame in which the analyst can define his own analytical work in accordance to the standard procedure.

As the method in this European Standard deals with the measurement of the mass fraction of α -, β -, γ - and δ -tocopherol in food, reference is made to the literature for the calculation and expression of the vitamin E content in terms of biological activities. For further information see [1], [2], [3] and [4]. The differentiation of *RRR*-tocopherol and all racemic tocopherols is not possible with this method.

1 Scope

This European Standard specifies a method for the determination of vitamin E in foods by high performance liquid chromatography (HPLC). The determination of vitamin E content is carried out by measurement of α -, β -, γ - and δ -tocopherol. This method has been validated in two interlaboratory studies. The first study was for the analysis of α -tocopherol in margarine and milk powder ranging from 9,89 mg/100 g to 24,09 mg/100 g. The second study was for the analysis of α -, β -, γ - and δ -tocopherol in milk powder and of α -, and β -tocopherol in oat powder ranging from 0,057 mg/100 g (β -tocopherol) to 10,2 mg/100 g (α -tocopherol).

NOTE The vitamin E activity can be calculated from the tocopherol content assuming appropriate factors as given in [1], [2], [3] and [4].

2 Normative references

The following documents, in whole or in part, are normatively referenced in this document and are indispensable for its application. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendments) applies.

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

3 Principle

α -, β -, γ - and δ -tocopherol are determined in a sample solution by HPLC separation and subsequent photometric (UV-range) or preferably fluorometric detection. In most cases a saponification of the test material followed by an extraction is necessary. Identification is carried out on the basis of retention times and quantitative determination by the external standard method using peak areas or peak heights. Internal standard methods can also be used if the corresponding recovery tests have proven the same behaviour of the internal standard during the analysis as the analyte itself, for more information see [4] to [14].

NOTE Using normal phase columns, the separation of tocopherols and tocotrienols is also feasible.

4 Reagents

During the analysis, unless otherwise stated, use only reagents of recognized analytical grade and water of at least grade 1 according to EN ISO 3696.

4.1 Methanol.

4.2 Ethanol absolute, volume fraction $\varphi(\text{C}_2\text{H}_5\text{OH}) = 100\%$.

4.3 Ethanol, $\varphi(\text{C}_2\text{H}_5\text{OH}) = 96\%$.

4.4 Sodium sulfate, anhydrous.

4.5 KOH solution, for saponification, in suitable mass concentrations, for example $\rho(\text{KOH}) = 50\text{ g/100 ml}$ or $\rho(\text{KOH}) = 60\text{ g/100 ml}$ or alcoholic solutions, for example 28 g of KOH in 100 ml of a mixture of 9 parts per volume of ethanol and 1 part per volume of water.

4.6 Antioxidants, such as ascorbic acid (AA), sodium ascorbate, pyrogallol, sodium sulfide (Na_2S), hydroquinone or butylated hydroxytoluene (BHT).