

Foodstuffs - Determination of vitamin B1 by high performance liquid chromatography

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English Version

Foodstuffs - Determination of vitamin B1 by high performance liquid chromatography

Produits alimentaires - Détermination de la teneur en vitamine B1 par chromatographie liquide haute performance

Lebensmittel - Bestimmung von Vitamin B1 mit Hochleistungs-Flüssigchromatographie

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

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Foreword

This document (EN 14122:2014) 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of an identical text or by endorsement, at the latest by December 2014 and conflicting national standards shall be withdrawn at the latest by December 2014.

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1 Scope

This European Standard specifies a method for the determination of vitamin B₁ in food by high performance liquid chromatography (HPLC) with enzymatic treatment and pre- or post-column derivatization. This method has been validated in two interlaboratory studies. The first study was for the analysis of samples of whole meal flour, milk powder/spray dried milk, freeze-dried mixed vegetables and freeze-dried pig's liver ranging from 0,295 mg/100 g to 0,807 mg/100 g. The second study was for the analysis of samples of tube feeding solution, baby food with vegetables, powdered milk, meal with fruits, yeast, cereal, chocolate powder and food supplement ranging from 0,11 mg/100 g to 486 mg/100 g. Vitamin B₁ is the mass fraction of total thiamin including its phosphorylated derivatives.

For further information on the validation, see Clause 8 and Annex B.

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

Thiamin is extracted from food after acid hydrolysis followed by dephosphorylation using an enzymatic treatment and quantified by HPLC with pre- or post-column derivatization to thiochrome. An external standard is used for quantification. For further information see [1] to [7].

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, or double distilled water.

- 4.1 **Methanol**, mass fraction $w(\text{CH}_3\text{OH}) \geq 99,8 \%$, HPLC grade.
- 4.2 **Acetic acid solution**, substance concentration $c(\text{CH}_3\text{COOH}) = 0,02 \text{ mol/l}$.
- 4.3 **Isobutanol**, $w(\text{C}_4\text{H}_{10}\text{O}) \geq 98 \%$.
- 4.4 **Sodium dihydrogen phosphate**, $w(\text{NaH}_2\text{PO}_4) \geq 99,8 \%$.
- 4.5 **Hydrochloric acid solution**, $w(\text{HCl}) = 36 \%$.
- 4.6 **Hydrochloric acid solution**, $c(\text{HCl}) = 0,1 \text{ mol/l}$.
- 4.7 **Sulfuric acid solution**, $c(\text{H}_2\text{SO}_4) = 0,05 \text{ mol/l}$.
- 4.8 **Sodium hydroxide**, $w(\text{NaOH}) \geq 99 \%$.
- 4.9 **Sodium hydroxide solution**, mass concentration $\rho(\text{NaOH}) = 150 \text{ g/l}$.
- 4.10 **Sodium hydroxide solution**, $\rho(\text{NaOH}) = 200 \text{ g/l}$.