

Irish Standard I.S. EN 15851:2010

Foodstuffs - Determination of aflatoxin B1 in cereal based foods for infants and young children - HPLC method with immunoaffinity column cleanup and fluorescence detection

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This document replaces:	This document is based on: EN 15851:2010	<i>Publish</i> 7 April	
This document was published under the authority of the NSAI and comes into effect on: 6 May, 2010			ICS number: 67.060

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**EUROPEAN STANDARD** 

EN 15851

NORME EUROPÉENNE

**EUROPÄISCHE NORM** 

April 2010

ICS 67.060

# **English Version**

# Foodstuffs - Determination of aflatoxin B₁ in cereal based foods for infants and young children - HPLC method with immunoaffinity column cleanup and fluorescence detection

Produits alimentaires - Dosage de l'aflatoxine B<sub>1</sub> dans les produits pour nourrissons et jeunes enfants à base de céréales - Méthode de chromatographie liquide haute performance avec purification sur colonne d'immunoaffinité et détection par fluorescence

Lebensmittel - Bestimmung von Aflatoxin B<sub>1</sub> in Säuglingsund Kleinkindernahrung auf Getreidebasis - HPLC-Verfahren mit Reinigung an einer Immunoaffinitätssäule und Fluoreszenzdetektion

This European Standard was approved by CEN on 27 February 2010.

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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 CEN Management Centre has the same status as the official versions.

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# EN 15851:2010 (E)

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EN 15851:2010 (E)

# **Foreword**

This document (EN 15851:2010) 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of an identical text or by endorsement, at the latest by October 2010, and conflicting national standards shall be withdrawn at the latest by October 2010.

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 has been prepared under a mandate given to CEN by the European Commission and the European Free Trade Association.

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.

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EN 15851:2010 (E)

# 1 Scope

This European Standard specifies a method for the determination of aflatoxin  $B_1$  in baby food by high performance liquid chromatography (HPLC) with immunoaffinity cleanup and fluorescence detection. This method has been validated in an interlaboratory study via the analysis of both naturally contaminated and spiked samples ranging from 0,07  $\mu$ g/kg to 0,18  $\mu$ g/kg.

For further information on the validation, see Clause 9 and Annex B.

# 2 Normative references

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

EN ISO 3696:1995, Water for analytical laboratory use - Specification and test methods (ISO 3696:1987)

# 3 Principle

A test portion is extracted with a mixture of methanol and water. The extract is filtered, diluted with phosphate buffered saline (PBS) to a specified solvent concentration, and applied to an immunoaffinity column containing antibodies specific to aflatoxin  $B_1$ . Aflatoxin  $B_1$  is purified and concentrated on the column and removed from the antibodies using methanol as eluent. Aflatoxin  $B_1$  is quantified by reverse-phase high performance liquid chromatography (RP-HPLC) with post column derivatization (PCD) involving bromination followed by fluorescence detection.

The post column derivatization is achieved with either electrochemically generated bromine or with pyridinium hydrobromide perbromide (PBPB).

# 4 Reagents

### 4.1 General

Use only reagents of recognized analytical grade and water complying with grade 1 of EN ISO 3696:1995, unless otherwise specified. Solvents shall be of quality for HPLC analysis, unless otherwise specified. Commercially available solutions with equivalent properties to those listed may be used.

WARNING — Dispose of waste solvents according to applicable environmental rules and regulations. Decontamination procedures for laboratory wastes have been reported by the International Agency for Research on Cancer (IARC), see [4].

- 4.2 Helium purified compressed gas.
- 4.3 Nitrogen.
- **4.4 Disodium hydrogen phosphate**, Na<sub>2</sub>HPO<sub>4</sub> anhydrous or Na<sub>2</sub>HPO<sub>4</sub>·12 H<sub>2</sub>O.
- 4.5 Potassium bromide.
- 4.6 Potassium chloride.
- 4.7 Potassium dihydrogen phosphate, KH<sub>2</sub>PO<sub>4</sub>.
- 4.8 Sodium chloride.



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